



Research Article

PHYTOCHEMICAL AND BIOCHEMICAL ANALYSES OF *SYRINGODIUM ISOETIFOLIUM* (ASCH.) DANDY

¹S. Velmani, ¹C. Santhosh, ¹P. Nithya, ²V. Balamurugan, ¹L. Archana, ¹G. Lavanya,
¹M. Sowmiya, ³B. Anjalidevi, ⁴M. Viji and ^{1*}A. Maruthupandian

¹Ethnopharmacological and Algal Biotechnology Laboratory, Department of Botany, School of Life Sciences,
Periyar University, Periyar Palkalai Nagar, Salem-636 011, Tamil Nadu, India.

²Molecular Biology Laboratory, Department of Botany, School of Life Sciences, Periyar University, Periyar Palkalai
Nagar, Salem-636 011, Tamil Nadu, India

³Department of Botany, Sri Vijay Vidyalaya College of Arts & Science, Kandhikuppam,
Bargur, Krishnagiri, Tamil Nadu, India
Department of Botany, Thiagarajar College, Madurai-625009, Tamil Nadu, India

Article History: Received 19th August 2022; Accepted 20th September 2022; Published 5th November 2022

ABSTRACT

Seagrasses are the flowering plant found in marine ecosystem. It has many bioactive compounds with various applications including pharmaceutical and nutraceutical. The present study has been aimed to evaluate the nutritional composition and antioxidant potentials of the Seagrass *Syringodium isoetifolium*. The Seagrass exhibits significant amounts of biochemical and phytochemical compositions. In *in-vitro* antioxidant, DPPH, ABTS and Hydroxyl radical scavenging assay showed that *S. isoetifolium* was an excellent scavenger for free radicals. The results suggest that the Seagrass *S. isoetifolium* may be used as a very good renewable marine resource for potential biomedical applications in future.

Keywords: Seagrasses, Phytochemical, Antioxidant, Nutrition and Minerals.

INTRODUCTION

Diverse marine species have produced a number of chemically distinct chemicals, some of which are being studied for their potential use in the creation of novel medications (Ghareeb *et al.*, 2020). Seagrasses, a convention of marine blossoming plants, possess the flowing and sub-flowing regions of shallow and protected areas of oceans, inlets, bayous, backwaters, tidal ponds, and estuaries around on mild and tropical shorelines of the world (Meena *et al.*, 2022). Globally, 72 species under 4 families (Zosteraceae, Hydrocharitaceae, Posidoniaceae and Cymodoceae) of Seagrasses has been found of which the seagrasses is one of its true marine flowering plants (Short *et al.*, 2016 and Jeyapragash *et al.*, 2021). Seagrasses are eukaryotic organisms that can be found, with the exception of the Polar Regions, in shallow water sections of moderate, subtropical, and tropical oceans (Mehari Ghebretinsae *et al.*, 2019). The most species are

found in the Gulf of Mannar and Palk Bay, followed by the Andaman & Nicobar and Lakshadweep islands. Seagrasses offer a variety of ecosystem services to meet human requirements, either directly or indirectly (Jaya Durga Jaisankar and Arumugam Ramasubramanian, 2022). The marine environment is an extraordinary source of novel bioactive chemicals, which frequently have different structural and chemical properties from naturally occurring substances on land (Sawssen BelMabrouk *et al.*, 2020). Seagrasses are the only group of flowering plants that successfully recolonize the sea by evolving specialized above and below-water organs. They are unique submerged marine angiosperms (Puntip Wisesongp *et al.*, 2022). Recently, it was discovered that *P. oceanica* secondary metabolites increased in response to seagrass stress brought on by human activity and climate change (particularly phenolic compounds) (Mannino and Micheli, 2020).

*Corresponding Author: Dr. A. Maruthupandian, Ethnopharmacological and Algal Biotechnology Laboratory, Department of Botany, School of Life Sciences, Periyar University, Periyar Palkalai Nagar, Salem-636 011, Tamil Nadu, Email pandianmd82@gmail.com. 7

The sea environment has extraordinary biological characteristics that have been important in the identification of numerous therapeutic leads (Selvam *et al.*, 2022). While phytochemical screening presents a foundational understanding of the therapeutic value of plant extracts and serves as a starting point for drug discovery (Efren Tangon *et al.*, 2019). For determining the nutritional value of marine predators determining prospective sources of protein and lipids for industrial usage or potential human intake, it is essential to understand the chemical composition of seagrass. Phytochemical analysis, which gives essential knowledge regarding the use of plant extracts as pharmaceuticals, aids in the discovery of new treatments (EfrenTangon *et al.*, 2021).

Benefits for Seagrasses in medicine

Seagrasses are having therapeutic effects against fevers, mental illnesses, wounds, skin conditions, muscle soreness, and stomach disorders (Gono *et al.*, 2022). Seagrasses have been used as fertilizer, food for cattle and humans, and ethnomedicine (Kavitha *et al.*, 2020). Further the Seagrasses have large range of secondary metabolites with distinctive characteristics that could be used to make powerful and efficient medications for a number of ailments (Gono *et al.*, 2022). The bioactive substances from *S. isoetifolium* were also found to have antioxidant, neuroprotective, antibacterial, anticancer, anti-inflammatory and antidiuretic properties (Kumar and

Pandey, 2013). According to Hutomo *et al.*, (2014), seagrasses are blooming plants (Angiospermae) that are submerged in shallow sea water and have been used to cure wounds, muscle pain, skin conditions and indigestion. By indigenous societies living along the coastal area, consume seagrass biomass has frequently been used as food and medicine (Vieira *et al.*, 2018). Based on the literature, seagrass are the marine biomass have numerous biomedical applications. Therefore, the present study has been selected the seagrass *S. isoetifolium* for analysis of biomedical phytochemical and antioxidant activity.

MATERIALS AND METHODS

Collection and identification of plant material

The *Syringodium isoetifolium* seagrass were collected from Kilakarai (9.2343° N, 78.7836° E), Tamil Nadu, India (Plate.1). The plants were identified at Botany Department, Periyar University, Salem, Tamil Nadu, India. The preserved Seagrass voucher specimen and Ref. no (PU/BOT/HVO.086) was submitted to the herbarium. The collected seaweed material was thoroughly washed with seawater to remove epiphytes, calcareous particles and kept in the laboratory and washed with tap water. A portion of the seagrass was taken for the preparing herbarium as well as preserved in 6% seawater formalin for identification. The remaining sample used for extraction.



***Syringodium isoetifolium* (Asch.) Dandy**

Plate 1. Collected seagrass.

Phytochemical analysis

The following procedures of Harborne (1994), phytochemical analysis were performed to determine the presence of active phytochemical elements in the extracts.

Proximate analysis

The proximate composition of *S. isoetifolium* was determined by (AOAC, 2000, Matanjun *et al.*, 2009). The moisture and ash content was followed by using

gravimetric methods (World Health Organization, 1998). Carbohydrates, lipids, and dietary fibre (AOAC, 1995, James, 1995; Siddique and Aktar, 2011). The nitrogen of protein was followed by Kjeldahl method, (AOAC, 2000).

Determination of Minerals

The mineral composition of *S. isoetifolium* was examined by using ICAP200. In terms of the sample dry weight, the mineral value was reported as mg/100g.

Antioxidant activity: DPPH assay

The DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging activity of the *S. isoetifolium* ethanol extract was investigated. 20 to 100 mg/mL was the range of concentrations. Ascorbic acid was also added. The mixture of *S. isoetifolium* ethanol extract and 0.1 mM DPPH solution in ethanol was allowed to react for 30 minutes at room temperature and in the dark. A spectrophotometer set at 517 nm was used to measure the absorbance of the reaction mixtures (Vino *et al.*, 2020).

ABTS assay

The ethanol extract of *S. isoetifolium* was examined for its capacity to scavenge ABTS radicals. Then, 7 mM ABTS and 2.45 mM potassium persulfate were combined to create the ABTS stock solution, which was then incubated for 16 hours in the dark. After incubation, the stock solution was diluted with ethanol to create the working solution at various concentrations. The absorbance was measured at 734 nm after 30 minutes of incubation at room temperature. Ascorbic acid was employed as a baseline (Hemlata *et al.*, 2020).

H₂O₂ Radical Scavenging Assay

The method of *S. isoetifolium* ethanol extract capacity to remove H₂O₂ was assessed (Ruch Cheng and Klanuing, 1989). The absorbance of hydrogen peroxide at 230 nm was measured after 10 min. Against a blank solution made up of phosphate buffer without hydrogen peroxide. The extracts (1.0 ml) were added to a hydrogen peroxide solution made in 0.1 M phosphate buffer saline (pH 7.4 40 mM and 0.6 ml).

The percentage of scavenging was calculated by using following formula;

$$\text{Percentage scavenged (H}_2\text{O)} = (A - A_1) / A \times 100$$

RESULT AND DISCUSSION

The qualitative phytochemical analysis of ethanol extracts of *S. isoetifolium* were recorded in Table-1. The results showed alkaloids, phenols, sterols, cardiac glycosides, tripenoids, protein and amino acid in the ethanol extract of *S. isoetifolium*. The similarly were observed Selvam *et al.*, 2022. Alkaloids and secondary metabolites, including phenol, fatty acids, and sterols, are abundant in seagrass (Tangon *et al.*, 2021). Research on secondary metabolites is frequently moving in the direction of bioactivity-focused medication discovery. Numerous intricate and unique chemical entities are produced as a result of research into the marine ecology (Kim *et al.*, 2021). Considering that seagrass has a high concentration of antioxidants such polyphenols, terpenoids, flavonoids, tannins and saponins. Recent study has increased the hunt for new compounds derived from seagrass (Ansari and Ghanem, 2019). Triterpenoids glycosides known as saponins are present in many different types of plants. Numerous studies have been done on saponins immunostimulant, permeability, hypocholesterolemic, and anticancerogenic characteristics (Selvam *et al.*, 2022). There have been reports of some seagrass groups of phenolic compounds found in both marine and terrestrial species, while others, such bromophenols and phlorotannins, are solely found in marine sources (Raquel Mateos *et al.*, 2020).

Table 1. Preliminary Phytochemical screening of *S. isoetifolium*.

Phytochemicals	<i>S. isoetifolium</i>
Alkaloids	+
Carbohydrates	-
Flavonoids	-
Phenols	+
Saponins	-
Tannins	-
Glucosides	-
Sterols	+
Cardiac glycosides	+
Resins	-
Terpenoids	+
Protein and amino acids	+
Triterpenoids	-
Reducing Sugar	-

Present (+); Absent (-)

Table 2. Nutrition factors of *S. isoetifolium*.

Biochemicals	<i>S. isoetifolium</i> (% of DW)
Carbohydrate	08.52±0.097
Protein	06.23±0.072
Lipid	01.59±0.063
Ash	18.30±0.065
Crude fiber	17.07±0.085

The results represented Mean ± SD from triplicates experiments

The results of protein, carbohydrate, lipid, ash, and fibre compositions of Seagrasses *S. isoetifolium* is presented in Table 2. The *S. isoetifolium* nutrient factors for highly present for ash, crude fiber and carbohydrate. Since 80% of the world population focuses on these conventional medicines to manage various diseases, scientists are concentrating on understanding the phytochemicals included in these medicinal plants (Kim *et al.*, 2021). The reported that macro and micro elements, protein, carbohydrate, lipids, ash and fiber from seagrass for better nutritional values analysis (Jeyasanta *et al.*, 2018).

Determination of Minerals

The determination of minerals was analysed *S. isoetifolium* from dry materials. The results were presented in Table- 3 *S. isoetifolium* have been good macro and micro elements such as Ca, Mg, K and N compared to other macro and micro elements. Marine organisms have been considered as good resource for nutrients and bioactive compounds (Zhong *et al.*, 2019). The macronutrients-phosphorus, potassium, calcium, magnesium, sodium and micronutrients-nickel, zinc, iron, chromium, copper, cadmium, while *S. isoetifolium* had a lower composition level, higher lipid and ash concentrations

Table 3. Mineral composition of *S. isoetifolium*.

Elements	<i>S. isoetifolium</i> (mg /kg ⁻¹ DW)
Ca	18.30±0.368
Mg	24.50±0.400
N	17.26±0.041
P	17.46±0.066
K	26.30±0.378
Cu	0.120±0.075
Fe	0.526±9.636
Mn	0.143±0.032
Zn	0.142±0.070

The results represented Mean ± SD from triplicates experiments

Ca-Calcium, Mg-Magnesium, N-Nitrogen, P-Phosphorus, K-Potassium, Cu-Copper, Fe-Iron, Mn-Maganese, Zn-Zinc

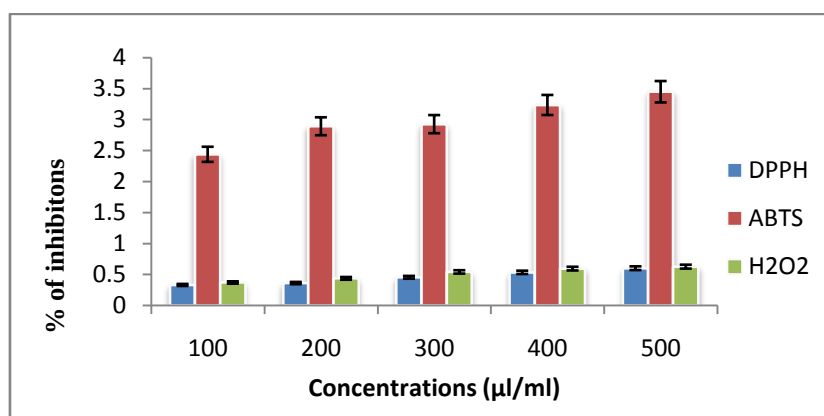


Figure 1. Antioxidant activity of *S. isoetifolium*.

Antioxidant Activity

The ethanol extract of *S. isoetifolium* tested in the different concentrations at 100, 200, 300, 400 and 500 µg/ml. All the three antioxidant activity in DPPH, ABTS and H₂O₂ assays. The highest inhibition was showed in 500 µg/ml in all the three assays (DPPH (0.603), ABTS(3.45), H₂O₂ (0.628) of ethanol extractly of *S. isoetifolium* (Figure1). The similar studies was reported that *Halophila ovalis* seagrass (Athiperumalsami *et al.*, 2010). Reducing power activity was indicated that antioxidant compounds as electron donors and reduce oxidative process (Yen and Chen, 1995).

Recent studies were analysis the antioxidant properties of seagrasses (Ansari and Ghanem, 2019). The flavonoids have knowingly natural antioxidant properties so reduces oxidative damages and protecting cells and cell organs (Selvam *et al.*, 2022).

Some phenolic chemicals found in seagrasses may function as a chemical defence mechanism and contribute to seagrass adaptability (Puntip Wisespongpan *et al.*, 2022). Their capacity to scavenge free radicals aided in the prevention of oxidative stress-related chronic illnesses like cancer, heart disease, and neurological disorders Bhuyan *et*

al., 2017). The harmful consequences of oxidative stress may be mitigated by endogenous and exogenous antioxidants. Exogenous antioxidant supplementation is the most popular and successful method of reducing oxidative stress (Pisoschi *et al.*, 2021). The maximal antioxidant activity for *S. isoetifolium* was 86.4 percent, while the IC50 value was 122.2 g/ml. *S. isoetifolium* exhibits a broad spectrum of pharmacological actions, including antibiotic, antihemolytic, cytotoxic, antibacterial and antifungal activity according to Arora *et al.*, (2016).

CONCLUSION

According to the current research, the seagrass *S. isoetifolium* contains a large number of phytochemicals. The total amounts of alkaloids, phenol, sterols, cardiac glycosides, terpenoids, protein and amino acids were also measured. The high total phenolic content value gives it very strong antioxidant effects. According to numerous researches, phenols are the main reason why seagrass has such high antioxidant capabilities. The analysis of the antioxidant activity reveals that *S. isoetifolium* ABTS radical movement was a great strategy to decide the antioxidant capacity. Further studies are required to investigate the compounds present in this ethanol extract of *S. isoetifolium* the biological effects of these seagrasses because they have a variety of pharmacological activities.

ACKNOWLEDGMENT

The authors express sincere thanks to the Ethnopharmacological and Algal Biotechnology Laboratory, Department of Botany, School of Life Sciences, Periyar University, Periyar Palkalai Nagar, Salem-636 011, Tamil Nadu, India for the facilities provided to carry out this research work.

REFERENCES

- Ansari, A. A., Ghanem, S. M., and Naeem, M. (2019). Brown Alga Padina: A review. *International Journal of Botany Studies*, 4(1), 01-03.
- AOAC (1995). *Official methods of analysis* (16th ed.). Washington, DC, USA; Association of Official Analytical Chemists.
- AOAC (2000). Method 992.23, In "Official Methods of Analysis", 17th Ed., AOAC International, Gaithersburg, MD.
- AOAC Assn. of Official Analytical Chemists. (2000). Coffee and tea. In: Official methods of analysis. 17th ed. Gaithersburg, Md: AOAC.
- Arora, P., Narang, R., Nayak, S. K., Singh, S. K., and Judge, V. (2016). 2, 4-Disubstituted thiazoles as multitargeted bioactive molecules. *Medicinal Chemistry Research*, 25(9), 1717-1743.
- Athiperumalsami, T., Rajeswari, V.D., Poorna, S.H., Kumar, V., and Jesudass, L.L., (2010). Antioxidant activity of seagrasses and seaweeds. *Botanica Marina* 53, 251–257.
- Bhuyan DJ, Basu A (2017). Utilisation of bioactive compounds derived from waste in the food industry. In: Vuong QV, editor (Utilization of bioactive compounds from Agricultural and Food Production Waste) CRC Press: Boca Ration FL USA; p. 342-57.
- Kim, D. H., Mahomoodally, M. F., Sadeer, N. B., Seok, P. G., Zengin, G., Palaniveloo, K., and Rengasamy, K. R. (2021). Nutritional and bioactive potential of seagrasses: a review. *South African Journal of Botany*, 137, 216-227.
- EfrenTangon, Elvinia R. Alivio, Jocelyn A. Pajiji and Kingpu O. Ajik (2021). Phytochemical Screening and Proximate Composition of The Seagrass *Halodule Pinifolia* of The Coastal Waters of Carmen, Agusan Del Norte, Philippines, *International Journal of Modern Pharmaceutical Research*, 5(2), 75-80.
- EfrenTangon, Oliva P. Canencia, Romeo M. del Rosario. (2019). Phytochemical screening and proximate composition of the Seagrasses *Enhalusacoroides* and *Thalassia hemprichii* in the Coastal Waters of Carmen, Agusan Del Norte, Philippines. *International Journal of Biosciences, International Network for Natural Sciences*, 15 (1), pp.1-7.
- Ghareeb, M.A., Tammam, M.A., El-Demerdash, A., and Atanasov, A.G. (2020). Insights about Clinically Approved and Pre clinically Investigated Marine Natural Products. *Current Research Biotechnology*. 2, 88-102.
- Gono, C. M. P., Ahmadi, P., Hertiani, T., Septiana, E., Putra, M. Y., and Chianese, G. (2022). A comprehensive update on the bioactive compounds from seagrasses. *Marine Drugs*, 20(7), 406.
- Grayer, R. J., and Harborne, J. B. (1994). A survey of antifungal compounds from higher plants, 1982–1993. *Phytochemistry*, 37(1), 19-42.
- Hemlata, P. R. M., A. P. Singh, and K. K. Tejavath. (2020). Biosynthesis of silver nanoparticles using *Cucumisprophetarum* aqueous leaf extract and their antibacterial and antiproliferative activity against cancer cell lines. *American Chemical Society Omega* 5 (10), 5520-8.
- James, (1995) C.S. James Analytical chemistry of foods. Chapman and Hall, New York.
- Jaya DurgaJaisankar and ArumugamRamasubramanian, (2022) Micromorphological, histological and phytochemical characterization of Seagrasses *Cymodocea serrulata* and *S. isoetifolium* of Palk Bay region, Tamil Nādu, *Journal of Global Resources*, Vol. 08 (02).
- Jeyapragash, D., Saravanakumar, A., and Yosuva, M. (2021). Seagrass Metabolomics: A New Insight towards Marine Based Drug Discovery. In

Metabolomics-Methodology and Applications in Medical Sciences and Life Sciences. Intech Open.

- Jeyasanta, I., Sathish, M.N., and Patterson, J. (2018). Antimicrobial susceptibility patterns of *Aeromonashydrophila* from seafood sources from fish market in Tuticorin, South East coast of India. *European Journal of Biomedical* 5, 365–373.
- Kavitha, D., P. Kalaivani and V. Vanitha. 2020. Areview on phytochemicals and biologicalactivities of seagrass. *Journal of Critical Reviews* 7(7): 257-264.
- Kumar, S., and Pandey, A. K. (2013). Chemistry and biological activities of flavonoids: an overview. *The scientific world journal*.
- Macreadie, P. I., Atwood, T. B., Seymour, J. R., Fontes, M. S., Sanderman, J., Nielsen, D. A., and Connolly, R. M. (2019). Vulnerability of seagrass blue carbon to microbial attack following exposure to warming and oxygen. *Science of the total environment*, 686, 264-275.
- Mannino, A.M., and C. Micheli. (2020). Ecologicalfunction of phenolic compounds from Mediterranean Fucoid algae and Seagrasses: An overview on the genus *CystoseiraSensu Lato* and *Posidoniaoceanica* (L.) Delile. *Journal of Marine Science and Engineering*, 8(1): 19.
- Marine Pharmacology. Available online: <https://www.marinepharmacology.org/> (accessed on 2 May 2022).
- Matanjun, P., Mohamed, S., Mustapha, N. M., and Muhammad, K. (2009). Nutrient content of tropical edible seaweeds, *Eucheumacottonii*, *Caulerpalentillifera* and *Sargassumpolycystum*. *Journal of Applied Phycology*, 21(1), 75-80.
- Meena. B, K Durgadevi., and V.Ramamurthy (2022). Studies on phytochemical and antibacterial potential of *Cymodoceaserrulata*, *International Journal of Botany Studies*, Volume 7, Issue 2, Page No. 556-560.
- Mehari Ghebretinsae, M., Awet Adhanom, B., Henok Ghebremedhin, G., and Biniam Tsegay, B.A (2019). Review on Bioactive Secondary Metabolites of Seagrass of the Southern Red Sea, Eritrea. *International Journal Advance Research*, 7, 306-314.
- Nithya, P., Archana, L., Kokila, P., Viji, M., Murugan, P and Maruthupandian, A. (2022). The Nutritional Assessment on Brown Macro algae *Lobophora variegata* from GOMBR, Tamil Nadu, India. *Journal of Advanced Applied Scientific Research*, 4(2).
- Pisoschi, A. M., Pop, A., Iordache, F., Stanca, L., Predoi, G., and Serban, A. I. (2021). Oxidative stress mitigation by antioxidants-an overview on their chemistry and influences on health status.*European Journal of Medicinal Chemistry*, 209, 112891.
- Puntip Wisespong and., Attawut Khantavong., Preecha Phothong., and Wutthikorn Wanghom (2022). Antimicrobial, Antioxidant and Antifouling Activity from Extracts of above ground and below ground Parts of Seagrasses *Cymodoce arotundata* and *Cymodocea serrulata*, *Journal of Fisheries And Environment*, Volume 46 (1).
- Raquel Mateos, Jose Ricardo Perez-Correa and Herminia Dominguez (2020). Bioactive Properties of Marine Phenolics, *Marine Drugs*, 18, 501, p-1-65.
- Ruch, R.J., Cheng, S.J., and Klaunig, J.E. (1989). Prevention of cytotoxicity and inhibition of intracellular communication by antioxidant catechins isolated from Chinese green tea. *Carcinogenesis*, 10, pp. 1003-1008.
- Sabilu, Y., and Jafriati, R. M. (2022). Test Of Bioactitvity And Antioxidant Activity Of Sea Urchin (*Diadema Setosum*) Gonads As Medicinal Ingredients Based On Marine Biodiversity. *Journal of Southwest Jiao tong University*, 57(1).
- SawssenBelMabrouk, Mariana Reis, Maria Llgia Sousa, Tiago Ribeiro,J oana R. Almeida, Sandra Pereira, Jorge Antunes, Filipa Rosa,VitorVasconcelos, LotfiAchour, AdnenKacem., and Ralph Urbatzka (2020).The Marine Seagrass *Halophilasti pulacea* as a Source of Bioactive Metabolites against Obesity and Biofouling, *Marine*, 18, 88;p 1-16.
- Selvam, M. S., Tresina, P. S., Benjamin, J., and Mohan, V. R. (2022). Assessment of Phytochemical, FT-IR and GC-MS Fingerprint Profiling of Marine Angiosperms *Enhalusacoroides* (Lf) Royle and *Syringodium isoetifolium* (Asch) Dandy, Gulf of Mannar Biosphere Reserve, Tamil Nadu. *Asian Journal of Biological and Life Sciences*, 11(2), 469.
- Short, F, Short, C., and Novak, A (2016).Seagrasses. In The Wetland Book: II: Distribution, Description and Conservation; *Springer Science: New York, NY, USA*, ISBN 978-94-007-6173-5.
- Siddique, M.A.M and Aktar, M., (2011).Changes of nutritional value of three marine dry fishes (*Johniusdussumieri*, *Harpodonnehereus* and *Lepturacanthussavala*) during storage. *Food and Nutrition Sciences*, 2011. boi:10.4236/fns.2011.210145
- Supriadi, S., Kaswadji, R. F., Bengen, D. G., and Hutomo, M. (2014). Carbon Stock of Seagrass Community in Barranglompo Island, Makassar (Stok Karbonpada Komunitas Lamun di Pulau Barranglompo, Makassar).*Ilmu Kelautan: Indonesian Journal of Marine Sciences*, 19(1), 1-10.
- Tangon, E., Canencia, O. P., and del Rosario, R. M. (2019). Phytochemical screening and proximate composition of the sea grasses *Enhalus acoroides* and *Thalassia hemprichii* in the Coastal Waters of Carmen, Agusan Del Norte, Philippines. *International Journal of Biosciences*, 15(1), 1-7.

- Vieira VMNCS, Lopes IE., and Creed JC. (2018). The biomass-density relationship in seagrasses and its use as an ecological indicator. *BMC Ecology*.18(1),44.doi: 10.1186/s12898-018-0200-1, PMID 30340566.
- Vino, P., S. Muruges, M. Viji, C. Santhosh, L. Wenchao, B. Balamuralikrishnan., and A. Maruthupandian.(2020). Phytoconstituents and their potential antimicrobial, antioxidant and mosquito larvicidal activities of *Goniiothalamuswrightii* Hook. F. &Thomson. *Arabian Journal for Science and Engineering* 45,4541–55.
- Wispongpan, P.,Khantavong, A., Phothong, P., and Wanghom, W. (2022). Antimicrobial, Antioxidant and Antifouling Activity from Extracts of Aboveground and Belowground Parts of Seagrasses *Cymodoce arotundata* and *Cymodoce aserrulata*. *Journal of Fisheries and Environment*, 46(1), 37-53.
- World Health Organization, (1998). Quality control methods for medicinal plant materials. World Health Organization.
- Yen, G. C., and Chen, H. Y. (1995). Antioxidant activity of various tea extracts in relation to their antimutagenicity. *Journal of Agricultural and Food Chemistry*, 43(1), 27-32.
- Zhong, Q., Wei, B., Wang, S., Ke, S., Chen, J., Zhang, H., and Wang, H (2019). The Antioxidant Activity of Polysaccharides Derived from Marine Organisms: An Overview. *Marine Drugs*, 17, 674.