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RESEARCH

Characterization of Nutritional, Antimicrobial and Antioxidant Properties of Five Seaweed Species in Sri Lanka

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ABSTRACT

The present study assessed the nutritional, antimicrobial and antioxidant properties of five seaweed species in northern coast of Sri Lanka: *Kappaphycus alvarezii, Sargassum wightii*, *Turbinaria ornata, Caulerpa peltata* and *Caulerpa racemosa*. The proximate composition was measured in dried seaweed powder. The antioxidant properties were determined in seaweed water extracts as total phenolic content (TPC), total flavonoid content (TFC), 2, 2- diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity and ferric reducing antioxidant power (FRAP). Crude methanol and crude water extracts were examined for antimicrobial properties against four human pathogens; *Escherichia coli, Pseudomonas aeruginosa, Staphylococcus aureus* and *Candida albicans*. The protein, fat, ash and gross energy contents varied from 4.7-26.5%, 0.02-0.05%, 7.6-25.6% and 10-18.7 kJg^{-1}, respectively. The two green seaweeds had the highest ($p<0.05$) protein and gross energy contents, while the other three species had higher ($p<0.05$) ash content. The TPC, FRAP and DPPH half maximal inhibitory concentration (IC_{50}) values ranged from 0.4-1.4 mg Gallic acid equivalent (GAE) g^{-1} dry weight, 2923.3-9193.5 mM $FeSO_4$ g^{-1} dry weight and 1.5-8.5 mg mL^{-1}, respectively. The *S. wightii* showed the highest ($p<0.05$) TPC and FRAP, whereas *K. alvarezii* had the lowest ($p<0.05$) DPPH IC_{50} value. No TFC response was observed. All seaweed methanol extracts showed antimicrobial activity against all four pathogens. The crude water extracts of *K. alvarezii* and *T. ornata* showed antimicrobial activity only against *P. aeruginosa*. Results showed that the selected seaweed species possess high nutritional, antioxidant, and antimicrobial properties. Therefore, they have the potential to be used in the food, pharmaceutical, and packaging industries.

INTRODUCTION

Seaweed or marine macroalgae are a diverse group of organisms thrive in brackish water and marine environments (Gupta and Abu-Ghannam, 2011a). They represent non-flowering and primitive sources of photosynthetic energy (Erulan *et al*., 2009). There are three groups of seaweed based on the nutrient and chemical composition, i.e., red (Rhodophyta), brown (Phaeophyta) and green (Chlorophyta) (Dawczynski *et al*., 2007; Gupta and Abu-ghannam, 2011a). Seaweed are utilized across various industries including food (Ismail and Hong, 2002), feed, medicine, fertilizer (Hafting *et al*., 2012) and cosmetics (Pal *et al*., 2014; Kılınç *et al*., 2014). An increasing number of studies have shown that seaweed are rich sources of protein, carbohydrates, vitamins, minerals and bioactive compounds, with variations observed among different species (Tortorella, 2018; Warnasooriya *et al*., 2024).

Seaweeds possess significant antioxidant metabolites, such as fucoxanthin, polyphloroglucinol, phenolic compounds, and bromophenols. The phenolic rings in seaweed polyphenolic compounds serve as electron traps, enhancing their wide-ranging antioxidant properties. This includes the ability to scavenge hydroxyl radicals, peroxy-radicals, and superoxides (Xu *et al*., 2004; Gupta and Abu-ghannam, 2011b). Algal polyphenols are more powerful antioxidants than plant polyphenols due to the presence of up to eight interlinked rings (Wang *et al.,* 2009). On the other hand, various gram-positive and negative bacteria have demonstrated non-resistance to bioactive compounds present in seaweed (Han *et al*., 2005; Kim *et al*., 2008; Gupta *et al*., 2010). Terpenes, phenolic compounds, and lipophilic chemicals are the components responsible for antimicrobial properties of seaweed (Gupta and Abu-Ghannam, 2011b). Therefore, seaweeds are abundant in nutrients and bioactive compounds responsible for antioxidant and antimicrobial properties, and represent potential sources that can contribute to reducing malnutrition and diet-related non-communicable diseases.

Sri Lanka, being an island with a vast coastline of more than 1700 km long, is abundant in various types of seaweed (Coppejans *et al*., 2011; Kariyawasam, 2016). Over 320 species from various families have been discovered in Sri Lanka's northern, southern, and western coastal regions (Durairathnam, 1961; Premarathna *et al*., 2020). Amongst, brown seaweed belonging to the genus *Sargassum* are the most prevalent type found in Sri Lanka (Jayasuriya, 1990; Premarathna *et al*., 2020). Extensive beds of seaweed are present in Jaffna, Gulf of Mannar, Pearl bank off Silarvathurai, Palk Strait, and along the southwest coast from Ambalangoda to Galle. Mannar, Kalpitiya, Puttalam, and Trincomalee are the primary locations where seaweeds are harvested for commercial purposes. However, seaweed culture is still in its infancy in Sri Lanka, and has no prominent seaweed utilization within the country. Most of the seaweed in Sri Lanka have not yet been characterized for the nutritional, antimicrobial and antioxidant properties. Therefore, the present study was conducted to assess the proximate composition and antimicrobial and antioxidant properties of five seaweed species collected from Northern coast in Sri Lanka [i.e., *Kappaphucus alvarezii* (Dotty Dotty)*, Sargassum wightii* (Gulfweed)*, Turbinaria ornata* (Crowded Sea Bell)*, Caulerpa peltata* (Saucer Algae) and *Caulerpa racemosa* (Sea grapes)].

METHODOLOGY

Collection of seaweed

Five species of seaweed were collected from different locations in the northern coast of Sri Lanka in September 2022. An identification guide was used to identify the seaweed specimens (Coppejans *et al*., 2011). The specimens were deposited at the Department of Animal Science, Faculty of Agriculture, University of Peradeniya, Sri Lanka for future references. Table 1 contains detailed information and voucher numbers of the seaweed, and their photographs are presented in Figure 1.

Table 1: Seaweed species, voucher number, type, and location and date of collection.

Seaweed Species	Voucher number	Type	Collected location	Location coordinates	Collected date
Kappaphycus alvarezii (Dotty Dotty)	SW10	Red	Commercial farms, Thraiyur, Jaffna	9°37'43.22"N 79°53'1.09"E	07 Sep 2022
Sargassum wightii (Gulfweed)	SW11	Brown	Coastal line, Madaitivu, Jaffna	9°35'51.58"N 79°58'53.51"E	07 Sep 2022
Turbinaria ornata (Crowded Sea Bell)	SW12	Brown	Coastal line, Allaipiddy, Jaffna	9°36'49.04"N 79°57'45.49"E	07 Sep 2022
Caulerpa peltata (Saucer Algae)	SW13	Green	Coastal line, Madaitivu, Jaffna	9°35'51.58"N 79°58'53.51"E	07 Sep 2022
Caulerpa racemosa (Sea Grapes)	SW14	Green	Sea cucumber farm, Thraiyur, Jaffna	9°37'43.22"N 79°53'1.09"E	07 Sep 2022

Preparation of seaweed powder

Seaweeds were washed with cleaned water six times thoroughly to remove dirt, adhered particles and other debris. The cleaned seaweeds were first air-dried and then oven dried at 55 ^{0}C until reaching a constant weight. The dried seaweeds were ground using a grinding miller to get a fine and homogeneous powder. The powdered seaweed samples were packed in labeled airtight bags until further analysis.

Preparation of seaweed water extracts

The powdered seaweed and distilled water were mixed at a ratio of 1:20 in a conical flask. The slurry was shaken on an electronic shaker at 75 rpm for 8 h. The supernatant was passed through Whatman No. 1 filter paper and the residual pellet was re-extracted two times. Water extracts of the first two supernatants of each sample were pooled in labeled Eppendorf tubes and used for antioxidant assays. The third supernatants of the replicates were stored at -21 °C until use for the analysis of antimicrobial properties. The seaweed extraction procedures were carried out in triplicates.

Preparation of seaweed methanol extracts

The powdered seaweed and methanol were mixed at a ratio of 1:10 in a conical flask. The slurry was shaken on an electronic shaker at 75 rpm for 8 h. The supernatant was passed through Whatman No. 1 filter paper and the residual pellet was re-extracted two times. The solvent was evaporated with a rotary evaporator. The crude extracts were collected into labeled Eppendorf tubes and used for antimicrobial assays. All the extractions were done in triplicates.

Proximate composition analysis

The proximate composition of dried seaweed powder was measured according to AOAC, (2005). Dry matter content was determined by oven drying at 104 °C until the weight remained constant. Ash content was measured by combustion at 550 °C. Protein content was measured using Kjeldhal method and conversion factor of five was used to convert nitrogen in seaweed into crude protein (Angell *et al*., 2016). Fat content was measured by Soxhlet extraction method. Energy content was measured using a bomb calorimeter (IKA C7000, Karnataka, India).

Figure 1: Five seaweed species. *Kappaphycus alvarezii* **(A),** *Sargassum wightii* **(B),** *Turbinaria ornata* **(C),** *Caulerpa peltata* **(D)** *and Caulerpa racemosa* **(E).**

Antioxidant assays

The antioxidant properties were determined in water extracts of seaweed as total phenolic content (TPC), total flavonoid content (TFC), 2, 2- diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity and ferric reducing antioxidant power (FRAP). The TPC of seaweed extracts was measured and expressed as Gallic acid equivalents (GAE) by Folin-Ciocalteu method (Singleton and Rossi, 1965). A Gallic acid standard series was prepared using 0.05 mg mL-1 Gallic acid solution in a microplate, by mixing different volumes of Gallic acid (0 µL, 20 µL, 25 µL, 30 µL, 35 µL, 40 µL, 45 µL, 50 µL), distilled water, and 10% Folin-Ciocalteu reagent in microplate wells. Simultaneously, the remaining microplate wells were filled with 50 µL of sample, distilled water and 10% Folin-Ciocalteu reagent. The plate was then incubated for 3 minutes at room temperature. After the initial incubation, 7.5% Na_2CO_3 was added to each well and incubated for another 30 minutes. The absorbances of standard series and samples were measured at 760 nm wavelength with a Microplate Spectrophotometer (Thermo Scientific™ Multiskan™ GO, 51119200). The TFC of seaweed extracts was measured and expressed as catechin equivalents (Agbo *et al*., 2015). A standard series of catechin was prepared using 200 µg mL^{-1} catechin solution, 5 % $NaNO_2$, 10% $AlCl_3$, 4% NaOH and distilled water. Microplate wells were filled with a series of catechin concentrations (0 mg mL^{-1}, 3.45 mg mL^{-1}, 6.90 mg mL^{-1}, 13.80 mg mL^{-1}, 20.70 mg mL^{-1}, 27.60 mg mL^{-1}, 34.50 mg mL^{-1}), distilled water and 20 µL of $NaNO_2$. Simultaneously, 50 µL samples, distilled water and $NaNO_2$ were added to remaining microplate

wells. The plate was then incubated for 6 minutes. After the initial incubation, all the wells containing standards and the samples were added with 20 µL of $AlCl_3$ and 200 µL of NaOH, and incubated for 15 minutes. The absorbances of standard series and samples were measured at 510 nm wavelength with a Microplate Spectrophotometer (Thermo Scientific™ Multiskan™ GO, 51119200). The ability of the extracts to neutralize the DPPH radical was measured according to Brand-Williams *et al* in 1995. In brief, 12 mg of DPPH was dissolved in 100 mL of absolute methanol in a volumetric flask covered with aluminium foil and incubated for 3 h in a dark place to develop the scavenging activity. A sample series was prepared in microplates (final volume of a well as 250 µL) by adding different volumes of samples (0 µL, 30 µL, 60 µL, 90 µL, 120 µL), absolute methanol (150 µL, 120 µL, 90 µL, 60 µL, 30 µL), and DPPH solution (100 µL). The microplate was covered with aluminium foil and incubated for 30 minutes. The absorbances of samples were measured at 517 nm wavelength with a Microplate Spectrophotometer (Thermo Scientific™ Multiskan™ GO, 51119200). The FRAP assay was conducted as described by Al-Farsi *et al* in 2005. The FRAP reagent was prepared by mixing 300 mM sodium acetate buffer (pH 3.6), 10.0 mM tripyridyl triazine (TPTZ) solution and 20.0 mM $FeCl_3.6H_2O$ solution in a volumetric ratio of 10:1:1. The wells in a microplate were filled with 50 µL of each sample at the concentration of 5 mg mL^{-1} and 2 mL of FRAP regent. The reaction mixture was incubated for 4 minutes at room temperature. Additionally, a series of $FeSO_4$ concentrations (1.5 mM) was used to develop the standard curve. The antioxidant capacity, assessed by the sample's ability to reduce ferric ions, was determined using the standard curve and expressed as mM FeSO4 equivalents per gram of dry sample. The increase in absorbance with $FeSO_4$ concentration (mM) was measured at 593 nm with a Microplate Spectrophotometer (Thermo Scientific™ Multiskan™ GO, 51119200).

Antimicrobial assays

The crude methanol extracts of all five seaweed species and crude water extracts of *K. alvarezii* and *T. ornata* were used for the antimicrobial assay. The antimicrobial activity of the seaweed extracts was determined using the well diffusion method (Bauer *et al*., 1996). In brief, stock solutions were prepared by dissolving seaweed extracts in either methanol or sterilized distilled water. The methanolic and aqueous extract concentrations of 50, 100 and 200 mg mL^{-1} were tested against the standard isolates of selected human pathogens, i.e., *Escherichia coli (E. coli)* ATCC 25922*, Pseudomonas aeruginosa (P. aeruginosa)* ATCC 27853*, Staphylococcus aureus (S. aureus)* AT11 43300 and *Candida albicans (C. albicans)* ATCC 10231*,* received from the Division of Microbiology, Faculty of Dental Sciences, University of Peradeniya, Sri Lanka. The turbidity of the bacterial suspensions was adjusted to McFarland 0.05 to standardize the density. The autoclaved (SANYO, Japan) Mueller Hinton Agar (MHA) culture plates were inoculated with 3 mL of bacterial suspensions and allowed the inoculums to spread evenly on the medium. After pipetting out the excess liquid, wells were cut using a No. 8 cork borer. The wells were filled with seaweed extracts, and the relevant solvent as the negative control (either methanol and sterilized distilled water). The culture plates were incubated at 37 °C for 24 hours. The inhibition zone was measured by determining the diameter of the clear area around the well (in mm). The inhibition zones produced by the controls were subtracted from the corresponding extract values to eliminate background effects attributable to the solvents.

Statistical analysis

One way Analysis of Variance was used to analyze proximate composition, and antioxidant and antimicrobial properties, followed by Tukey's mean comparison test. Antimicrobial activities of crude methanol and crude water extracts were compared using t-test. The p value of 0.05 was considered significant and all statistical analysis were done using IBM SPSS software, version 26.

RESULTS

Proximate composition of seaweed

The proximate compositions of the five seaweed species are shown in Table 2. The protein, fat, ash and gross energy contents of seaweed varied from 4.7-26.5%, 0.02-0.05%, 8.4-25.6% and 10-18.7 kJg^{-1}, respectively. The *C. peltata* and *C. racemosa* had higher ($p<0.05$) crude protein (20.1-26.5%), crude fat (0.05%) and energy (17.2-18.7 kJg^{-1}) contents than *K. alvarezii*, *S. wightii* and *T. ornata*. *K. alvarezii* had the lowest ($p<0.05$) crude protein (4.7%) and energy (10 kJg^{-1}) contents. The major component of *K. alvarezii, S. wightii* and *T. ornata* was ash (14.7-25.6%).

Antioxidant properties of seaweed extracts

The antioxidant properties exhibited by the five seaweed species are presented in Table 3. Significant variations in antioxidant activities were observed among the species. The TPC, FRAP and DPPH IC_{50} values of seaweed ranged from 0.4-1.4 mg GAE g^{-1} dry weight, 2923-9193 mM $FeSO_4$ g^{-1} dry weight and 1.5-8.5 mg mL^{-1}, respectively. The water extracts of *S. wightii* showed the highest ($p<0.05$) TPC (1.4 mg $GAEg^{-1}$) and FRAP (9193.5 mM $FeSO_4$ g^{-1}), while *K. alvarezii* had the lowest ($p<0.05$) DPPH IC_{50} value (1.5 mg mL^{-1}). None of the extracts of seaweed species responded to the TFC assay.

Antimicrobial properties of seaweed extracts

Antimicrobial properties of crude water and crude methanol extracts of seaweeds are shown in Tables 4 and 5, respectively. Methanol extracts of all the five seaweed species demonstrated antimicrobial activity against all tested pathogens, but the crude water extracts of two tested seaweed species, *K. alvarezii* and *T. ornata* showed antimicrobial activity only against *P. aeruginosa.* In comparison of crude water and methanol extracts of *K. alvarezii* and *T. ornata,* crude water extracts had higher ($p<0.05$) antimicrobial activity compared to crude methanol extracts. At 200 mg mL^{-1}, water extract of *T. ornata* had higher ($p<0.05$) antimicrobial activity against *P. aeruginosa* compared to *K. alvarezii*. The crude methanol extracts of both *K. alvarezii* and *T. ornata* showed higher ($p<0.05$) activity against *P. aeruginosa* compared to other three seaweed species. At 200 mg mL^{-1}, *S. wightii* had the highest ($p<0.05$) antimicrobial activity against *C. albicans*. Amongst the seaweed methanol extracts, at 200 mg mL^{-1}, *T. ornata* had higher ($p<0.05$) antimicrobial activity against *E-coli* compared to *K. alvarezii, C. peltata* and *C. racemosa*. Nevertheless, *T. ornata* showed lower ($p<0.05$) antimicrobial activity against *S. aureus* compared to *K. alvarezii, S. wightii* and *C. racemosa*.

Table 2: Proximate composition of five seaweed species.

Seaweed species	Dry matter (%)	Ash (% of DW)	Crude protein (% of DW)	Crude fat (% of DW)	Energy (kJ g^{-1})
Kappaphycus alvarezii	92.0^{b}±0.07	25.6^{c}±0.01	4.66^{a}±0.24	0.02^{a}±0.00	10.0^{a}±0.00
Sargassum wightii	91.0^{a}±0.15	14.7^{b}±0.48	8.88^{c}±1.00	0.02^{a}±0.00	12.9^{c}±0.07
Turbinaria ornata	93.1^{c}±0.26	25.0^{c}±0.15	6.91^{b}±0.00	0.02^{a}±0.00	11.6^{b}±0.05
Caulerpa peltata	95.1^{d}±0.11	8.43^{a}±0.07	26.5^{e}±0.01	0.05^{b}±0.00	17.2^{d}±0.21
Caulerpa racemosa	96.2^{e}±0.06	7.59^{a}±0.27	20.1^{d}±0.00	0.05^{b}±0.00	18.7^{e}±0.13
p value	***	***	***	***	***

DW: Dry weight. Values are expressed as means ± standard deviation (n=3). Values in the same column with different superscripts are significantly different at P<0.05 according to Tukey's multiple comparison test. *** *P* <0.001.

Table 3: Antioxidant activities of water extracts of five seaweed species.

Seaweed species	TPC (mg GAE g^{-1} DW)	FRAP (mM $FeSO_4$ g^{-1} DW)	DPPH IC_{50} value (mg mL^{-1} DW)
Kappaphycus alvarezii	0.74^{b}±0.08	2923.3^{a}±201.24	1.51^{a}±0.50
Sargassum wightii	1.36^{c}±0.16	9193.5^{d}±70.80	2.06^{ab}±0.21
Turbinaria ornata	0.76^{b}±0.11	5841.7^{c}±383.32	8.52^{b}±2.94
Caulerpa peltata	0.40^{a}±0.06	4064.5^{b}±68.47	3.40^{ab}±2.18
Caulerpa racemosa	0.62^{ab}±0.05	3374.6^{a}±307.06	1.89^{ab}±0.17
p value	***	***	*

TPC = Total Phenolic Content; FRAP = Ferric Reducing Antioxidant Power; DPPH = 2, 2- diphenyl-1-picrylhydrazyl radical scavenging activity. GAE = Gallic acid equivalents. DW: Dry weight. Values are expressed as means ± standard deviation (n=3). Values in the same column with different superscripts are significantly different at P<0.05 according to Tukey's multiple comparison test. * P<0.05, *** P<0.001.

Table 4: Antimicrobial properties (Diameter of the zones of inhibition in mm) of crude water extracts of two seaweed species against *Pseudomonas aeruginosa.*

Seaweed species	Concentration of crude seaweed extracts		
	50 mg mL^{-1}	100 mg mL^{-1}	200 mg mL^{-1}
Kappaphycus alvarezii	7.5±0.71	10.0±0.00	21.0^{a}±0.00
Turbinaria ornata	5.5±0.71	8.0±0.00	27.5^{b}±0.71
p value	NS	NS	**

Values are expressed as means ± standard deviation (n=3). Values in the same column with different superscripts are significantly different at P<0.05. NS = not significant, ** P<0.01.

Table 5: Antimicrobial properties (Diameter of the zones of inhibition in mm) of crude methanol extracts of five seaweed species against the tested human pathogens.

Seaweed species	*Escherichia coli*		*Pseudomonas aeruginosa*		*Staphylococcus aureus*		*Candida albicans*	
	100 mg mL^{-1}	**200 mg mL^{-1}**	**100 mg mL^{-1}**	**200 mg mL^{-1}**	**100 mg mL^{-1}**	**200 mg mL^{-1}**	**100 mg mL^{-1}**	**200 mg mL^{-1}**
Kappaphycus alvarezii	2.5^{a} ± 0.7	4.5^{a} ± 0.7	2.5^{b}± 0.7	5.0^{b}±0.00	3.0^{ab} ± 1.4	7.0^{c}±0.00	7.5 ^{ab} ± 0.7	10.5^{ab} ± 0.7
Sargassum wightii	8.5^{c} ± 0.7	12.0^{cd} ± 1.4	0.0^{a}	0.5^{a} ± 0.7	3.0^{ab}±0.00	9.0^{c} ± 1.4	12.5^{c} ± 2.1	19.5^{d} ± 2.1
Turbinaria ornata	4.5^{ab} ± 0.7	14.5^{d}± 0.7	4.0^{c}±0.00	5.5^{b} ± 0.7	0.5^{a} ± 0.7	2.5^{a} ± 0.7	5.0^{a} ± 1.4	8.5^{a} ± 0.7
Caulerpa peltata	8.0^{bc} ± 1.4	8.5^{bc} ± 0.7	0.0^{a}	0.5^{a} ± 0.7	2.5^{ab} ± 0.7	3.5^{ab} ± 0.7	12.0^{bc}±0.00	15.0^{c}±0.00
Caulerpa racemosa	2.5^{a} ± 0.7	5.5^{ab} ± 0.7	0.0^{a}	0.0^{a}	4.5^{b} ± 0.7	6.0^{bc}±0.00	13.0^{c}±0.00	14.5^{bc} ± 0.7
p value	**	***	***	***	*	**	**	**

Values are expressed as means ± standard deviation (n=3). Values in the same column with different superscripts are significantly different at P<0.05 according to Tukey's multiple comparison test. * P<0.05, ** P<0.01, *** P<0.001.

DISCUSSION

Seaweeds consist of vital nutrients and trace elements necessary for human nutrition (Rupe, 2002). Their compositions are highly diverse depending on the macroalgal class, species, stage of biomass development (i.e., sterile versus fertile tissue), environmental stresses, and season of collection (Garcia-Vaquero *et al*., 2021). The present study further elucidated that the proximate composition of the seaweed can vary depending on the species. As per the previous findings, green seaweeds possess higher nutritional proximate composition, followed by brown and red seaweed (Choudhary *et al*., 2023). Similarly, the present study revealed that the two green seaweed species, *C. peltata* and *C. racemosa*, had higher crude protein, crude fat, and energy contents than red (*K. alvarezii*) and brown (*S. wightii* and *T. ornata*) seaweed, while *K. alvarezii* which is the commercially cultured species in Sri Lanka, had the lowest crude protein and energy contents. The higher ash contents in *K. alvarezii, S. wightii* and *T. ornata* (14.7-25.6%) indicate the presence of high levels of minerals, particularly silica-containing compounds (Liu, 2016). In general, most of the seaweed contain over 20% of ash content, which is higher than that in most vegetables (Sanchez-Machado *et al*., 2004, Warnasooriya *et al*., 2024). All five seaweed species showed lower levels of fat, consistent with previous studies reporting low-fat content in seaweed (Lo *et al*., 2004, Gupta and Abu-ghannam, 2011 and Peinado *et al*., 2014, Warnasooriya *et al*., 2024). However, it is well established that the quality of the fatty acid composition of seaweed is high because of the presence of omega-3 fatty acids (Gamero-vega *et al*., 2020; Stibich, 2022).

According to previous findings, solvent extractions of plant materials are commonly employed for antioxidant isolation due to the diverse antioxidant potentials of substances with varying polarity. Furthermore, the extraction yield, phenolic content, and antioxidant properties of the extracts greatly depend on the solvent employed for the extraction process, with water being identified as the most suitable solvent for extracting plant phenolic compounds (Barchan *et al*., 2014). In the present study, water extracts of seaweed were used for the determination of antioxidant properties. According to the literature, it has been noted that seaweed extracts containing polyphenols demonstrate antioxidant activity (Yan *et al*., 1999). The present results showed that the water extracts of brown seaweed, *S. wightii* had the highest TPC, followed by brown seaweed *T. ornata* and red seaweed *K. alvarezii.* Green seaweed species, *C. peltata* and *C. racemosa* showed the lowest TPC contents. Antioxidant properties were further measured based on ability to reduce ferric ions of seaweeds using the FRAP assay. In accordance with the TPC, FRAP assay also showed that the two brown seaweed species, *S. wightii* and *T. ornata* had higher antioxidant capacity than the red seaweed *K. alvarezii* and the two green seaweed species, *C. peltata* and *C. racemosa*. This may be due to the higher polyphenol content in brown seaweed compared to red and green seaweed as reported by Wang *et al*. in 2009 and Ismail *et al*. in 2020. Phlorotannins, a class of phenolic substances exclusively present in polymers of phloroglucinol, have been identified as a distinctive group of polyphenols found in brown seaweed (Wang *et al*., 2009). Additionally, brown seaweeds also contain fucoxanthin, which is one of the most abundant pigment carotenoid compounds with significant antioxidant activity (Begum *et al*., 2021). The observed higher TPC and FRAP in brown seaweed can, thus, be attributed to the presence of phlorotannins and fucoxanthin.

The DPPH assay serves as a widely employed method for assessing free radical scavenging capability of seaweed extracts (Wang *et al*., 2009). The DPPH is a stable free radical with an unpaired electron on one of its nitrogen bridges (Eklund et. al. 2005). When a substrate with a hydrogen atom donor encounters DPPH, the radical DPPH obtains the hydrogen atom and converts into a non-radical form. The radical form of the DPPH gives a dark purple color, while the non-radical form gives a yellowish color (Baliyan *et al*., 2022). The DPPH IC_{50} value represents the amount of sample needed to inhibit 50% of the radicals in the DPPH (Martinez-Morales *et al*., 2020). Hence, the species with a lower IC_{50} value indicates higher antioxidant activity.

The highest antioxidant activity was, thus, demonstrated by red seaweed *K. alvarezii*, whereas the brown seaweed, *T. ornata* showed the lowest activity. However, these findings deviate somewhat from previous research, as brown seaweed species were found to exhibit higher antioxidant activities compared to other seaweed (Wang *et al*., 2009; Saranya *et al*., 2014; Ismail *et al*., 2020), which may be attributed to species differences and habitat variations.

The methanol extracts of seaweed showed antimicrobial activity against the gram-positive bacterium, *S. aureus*; two-gram negative bacteria, *E. coli* and *P. aeruginosa* and the fungus, *C. albicans*. However, the crude water extracts of *K. alvarezii* and *T. ornata* showed antimicrobial activity only against gram-negative *P. aeruginosa.* It is possible that the methanol may have extracted more bioactive compounds especially non-polar substances from seaweed responsible for antimicrobial properties. However, water extracts showed antimicrobial activities even at the concentration of 50 mg mL^{-1}, whereas crude methanol did not show any activity at 50 mg mL^{-1}. Comparing the antimicrobial properties of crude water and methanol extracts of *K. alvarezii* and *T. ornata,* crude water extracts showed higher antimicrobial activities, indicating water may have extracted more bioactive compounds capable of inhibiting *P. aeruginosa* than methanol. It has been recognized that seaweeds harbor antifungal compounds (De Corato *et al*., 2017; Pourakbar *et al*., 2021). In accordance, methanol extracts showed higher antimicrobial activities against *C. albicans,* suggesting that the five seaweeds possess antifungal activities. In particular, *S. wightii* had the highest antimicrobial activity against *C. albicans*, and thereby presenting a potential antifungal agent. Among the five seaweed species, methanol extracts of *T. ornata* exhibited higher antimicrobial activity against *E. coli*, but demonstrated lower activity against *S. aureus*. This suggests that *T. ornata* may be more effective against *E. coli* than against *S. aureus*. However, differences in the antimicrobial effects of methanol and water extracts of *K. alvarezii* and *T. ornata,* warrant further studies of the water extracts of the rest of the seaweed species. Although a positive control (standard antibiotic) was not included in the present study, the observed inhibition zones provide a comparative indication of the relative antimicrobial potential among the tested seaweed extracts.

CONCLUSION

In conclusion, the proximate composition, and antioxidant and antimicrobial properties of the five selected Sri Lankan seaweed species, namely *K. alvarezii*, *S. wightii*, *T. ornata*, *C. peltata*, and *C. racemosa*, exhibited variations across the species. Further, the antimicrobial properties of seaweed extracts were dependent on the type of solvent used for the extraction. The present results indicate that the five selected Sri Lankan seaweeds contain higher nutritional value, as well as antioxidant and antimicrobial properties, suggesting their potential for use as food or food additives, or for packaging and pharmaceutical applications.

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COMPETING INTERESTS

The authors declare that they have no competing interests.

REFERENCES

Agbo, M. O., Uzor, P. F., Nneji, U. N. A., Odurukwe, C. U. E., Ogbatue, U. B., & Mbaoji, E. C. (2015). Antioxidant, total phenolic, and flavonoid content of selected Nigerian medicinal plants. Dhaka University *Journal of Pharmaceutical Sciences*, 14(1), 35–41

Al-Farsi, M., Alasalvar, C., Morris, A., Baron, M., & Shahidi, F. (2005). Comparison of Antioxidant Activity, Anthocyanins, Carotenoids, and Phenolics of Three Native Fresh and Sun-Dried Date *(Phoenix dactylifera L.)* Varieties Grown in Oman. *Journal of Agricultural and Food Chemistry,* 53(19), 7592–7599. DOI:10.1021/jf050579q

Angell, A. R., Mata, L., de Nys, R., & Paul, N. A. (2016). The protein content of seaweeds: a universal nitrogen-to-protein conversion factor of five. *Journal of Applied Phycology*, 28(1), 511–524. DOI: https://doi.org/10.1007/s10811-015-0650-1

AOAC (2005). Official Methods of Analysis of AOAC International, 18th Edition, AOAC International, Gaithersburg, MD, USA.

Baliyan, S., Mukherjee, R., Priyadarshini, A., Vibhuti, A., Gupta, A., Pandey, R.P. & Chang, C.-M (2022). Determination of Antioxidants by DPPH Radical Scavenging Activity and Quantitative Phytochemical Analysis of *Ficus religiosa*. *Molecules*, 27, 1326. DOI: https://doi.org/10.3390/molecules27041326

Barchan, A., Bakkali, M., Arakrak, A., Pagán, R., & Laglaoui, A. (2014). The effects of solvents polarity on the phenolic contents and antioxidant activity of three Mentha species extracts. *International Journal of Current Microbiology and Applied Sciences*, 3(11), 399–412.

Begum, R., Howlader, S., Mamun-Or-Rashid, A. N. M., Rafiquzzaman, S. M., Ashraf, G. M., Albadrani, G. M., Sayed, A. A., Peluso, I., Abdel-Daim, M. M., & Uddin, M. S. (2021). Antioxidant and Signal-Modulating Effects of Brown Seaweed-Derived Compounds against Oxidative Stress-Associated Pathology. *Oxidative medicine and cellular longevity*, 2021, 9974890. DOI: https://doi.org/10.1155/2021/9974890

Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. *LWT - Food Science and Technology*, 28(1), 25–30. DOI: https://doi.org/10.1016/S0023-6438(95)80008-5

Bauer, A. W., Kirby, W. M., Sherris, J. C., & Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. *American journal of clinical pathology*, *45*(4), 493–496.

Choudhary, B., Khandwal, D., Gupta, N. K., Patel, J., & Mishra, A. (2023). Nutrient Composition, Physicobiochemical Analyses, Oxidative Stability and Antinutritional Assessment of Abundant Tropical Seaweeds from the Arabian Sea. *Plants*, 12(12), 1–27. DOI: https://doi.org/10.3390/plants12122302

Coppejans E, Leliaert F, Dargent O, Gunasekara R, D. C. O. (2011). *Sri Lankan seaweeds methodologies and field guide to the dominant species*. *Botanica Marina*, 54(1).

Dawczynski, C., Schubert, R., & Jahreis, G. (2007). Amino acids, fatty acids, and dietary fibre in edible seaweed products. *Food Chemistry*, 103(3), 891–899. DOI: doi:10.1016/j.foodchem.2006.09.041

De Corato, U., Salimbeni, R., De Pretis, A., Avella, N., & Patruno, G. (2017). Antifungal activity of crude extracts from brown and red seaweeds by a supercritical carbon dioxide technique against fruit postharvest fungal diseases. *Postharvest Biology and Technology*, 131, 16–30. DOI: doi:10.1016/j.postharvbio.2017.04.011

Durairathnam, M. (1961). Fisheries research station bulletin no . 10 contribution to the study of the marine algae. Fisheries Research Station Department of Fisheries, Ceylon, 10, 117.

Erulan, V., Soundarapandian, P., Thirumaran, G.& Ananthan, G. (2009). Studies on the Effect of *Sargassum polycystum* (C.Agardh, 1824) Extract on the Growth and Biochemical Composition of Cajanus cajan (L.) Mill sp. *American-Eurasian J. Agric. & Environ. Sci*, 6(4), 392–399.

Gamero-vega, G., Palacios-palacios, M., & Quitral, V. (2020). Nutritional Composition and Bioactive Compounds of Red Seaweed : A Mini-Review. *Journal of Food and Nutrition Research.* 8(8), 431–440. DOI: https://doi.org/10.12691/jfnr-8-8-7

Garcia-Vaquero, M., Rajauria, G., Miranda, M., Sweeney, T., Lopez-Alonso, M., & O'doherty, J. (2021). Seasonal Variation of the Proximate Composition, Mineral Content, Fatty Acid Profiles and Other Phytochemical Constituents of Selected

Brown Macroalgae. *Marine Drugs*, 19(4). DOI: https://doi.org/10.3390/MD19040204

Gupta, S., & Abu-Ghannam, N. (2011a). Bioactive potential and possible health effects of edible brown seaweeds. *Trends in Food Science and Technology*, 22(6), 315–326. DOI: https://doi.org/10.1016/j.tifs.2011.03.011

Gupta, S., & Abu-Ghannam, N. (2011b). Recent developments in the application of seaweeds or seaweed extracts as a means for enhancing the safety and quality attributes of foods. *Innovative Food Science and Emerging Technologies*, 12(4), 600–609. DOI: https://doi.org/10.1016/j.ifset.2011.07.004

Gupta, S., Rajauria, G., & Abu-Ghannam, N. (2010). Study of the microbial diversity and antimicrobial properties of Irish edible brown seaweeds. *International Journal of Food Science & Technology,* 45(3), 482–489. DOI: https://doi.org/10.1111/j.1365-2621.2009.02149.x

Hafting, J. T., Critchley, A. T., Cornish, M. L., Hubley, S. A., & Archibald, A. F. (2012). On-land cultivation of functional seaweed products for human usage. *Journal of Applied Phycology*, 24(3), 385–392. DOI: https://doi.org/10.1007/s10811-011-9720-1

Han, L., Xu, N., Shi, J., Yan, X., & Zheng, C. (2005). Isolation and pharmacological activities of bromophenols from Rhodomela confervoides. *Chinese Journal of Oceanology and Limnology*, 23(2):226-229. DOI:10.1007/BF02894243

Ismail, A., & Hong, T. S. (2002). Antioxidant activity of selected commercial seaweeds. *Malaysian journal of nutrition*. 8. 167-77.

Ismail, G. A., Gheda, S. F., Abo-Shady, A. M., & Abdel-Karim, O. H. (2020). In vitro potential activity of some seaweeds as antioxidants and inhibitors of diabetic enzymes. *Food Science and Technology* (Brazil), 40(3), 681–691. DOI: https://doi.org/10.1590/fst.15619

Jayasuriya P. M. A., the status of culture and utilization of seaweeds in Sri Lanka, FAO Report of the Regional Workshop on the Culture & Utilization of Seaweeds (online), Available at https://www.fao.org/3/ab727e/AB727E06.htm; Accessed 11 Nov 2022.

Eklund, P. C., Långvik, O., Wärnå, J., Salmi, T., Willför, S., & Sjöholm, R. (2005). Chemical studies on antioxidant mechanisms and the free radical scavenging properties of Lignans. *Organic and Biomolecular Chemistry*, 3. 3336-47. DOI:10.1039/b506739a

Kariyawasam, I. (2016). Seaweed Mariculture: A Potential "Multi-Million Dollar Industry". *SATH SAMUDURA; Healthy Oceans; Healthy Planet; ISSN: 2279-3208; MArine Environment Protection Authority, Sri Lanka.* 2016. 44-49.

Kim, Y. B., Moon, Y. G., & Heo, M. S. (2008). Antioxidant and antimicrobial activities of seaweed, *Ecklonia cava*. *Journal of Biotechnology*, 136, S598.

Kılınç, B., Cirik, S., & Turan, G. (2014). Organic Agriculture Towards Sustainability. DOI: https://doi.org/10.5772/57033

Liu, K. (2016). Characterization of Ash in Algae and Other Materials by Determination of Wet Acid Indigestible Ash and Microscopic Examination., 208, 515. DOI: https://doi.org/10.54032/2203-008-029-014

Lo, J., Lo, J., & Sa, D. I. (2004). Fatty acids , total lipid , protein and ash contents of processed edible seaweeds. 85, 439–444. DOI: https://doi.org/10.1016/j.foodchem.2003.08.001

Martinez-Morales, F., Alonso-Castro, A. J., Zapata-Morales, J. R., Carranza-Álvarez, C., & Aragon-Martinez, O. H. (2020). Use of standardized units for a correct interpretation of IC50 values obtained from the inhibition of the DPPH radical by natural antioxidants. *Chemical Papers*, 74(10), 3325–3334. DOI: https://doi.org/10.1007/s11696-020-01161-x

Pal, A., Kamthania, M. C., & Kumar, A. (2014). Bioactive Compounds and Properties of Seaweeds. *A Review - OALib*, 01(04), 1–17. DOI: https://doi.org/10.4236/oalib.1100752

Peinado, I., Girón, J., Koutsidis, G., & Ames, J. M. (2014). Chemical composition, antioxidant activity and sensory evaluation

of five different species of brown edible seaweeds. *Food Research International*, 66, 36–44. DOI: https://doi.org/10.1016/j.foodres.2014.08.035

Pourakbar, L., Moghaddama, S. S., Enshasy, H. A. El, & Sayyed, R. Z. (2021). Antifungal activity of the extract of a macroalgae, *Gracilariopsis persica*, against four plant pathogenic fungi in vitro. *Plants*, 10(9), 1–13. DOI: https://doi.org/10.3390/plants10091781

Premarathna, A. D., Jayasooriya, A. P., Amcp, K., Jayasooriya, A. P., De, J., & Adhikari, R. B. (2020). Distribution and Diversity of Seaweed Species in South Coastal Waters in Sri Lanka. *Journal of Oceanography Marine Research*, 7(196), 1–7. DOI: https://doi.org/10.35248/2572-3103.19.7.196

Rupe, P. (2002). Mineral content of edible marine seaweeds. Food Chemisrty, 79, 23–26.

Sanchez-Machado, D.I., Lopez-Cervantes, J., Lopez-Hernandez, J. and PaseiroLosada, P. (2004). Fatty acids, total lipid, protein and ash contents of processed edible seaweeds. *Food Chemistry*, 85: 439-444.

Saranya, C., Parthiban, C., & Anantharaman, P. (2014). Evaluation of antibacterial and antioxidant activities of seaweeds from Pondicherry coast. *Advances in Applied Science Research*, 5(4), 82–90.

Singleton, V. and Rossi, J. (1965) Colorimetry of Total Phenolic Compounds with Phosphomolybdic-Phosphotungstic Acid Reagents. *American Journal of Enology and Viticulture*, 16, 144-158.

Stibich, M., (2022) Seaweed Nutrition Facts and Health Benefits, www.verywellfit.com. Available at: https://www.verywellfit.com/seaweeds-health-claims-2223487#:~:text=In%20addition%20to%20vitamins%20and,diet%20to%20promote%20better%20health; Accessed 16 December 2022.

Tortorella, B. E. (2018). High Nutrition Plants from the Sea to Make Your Bees Healthier and Stronger, IBP-CNR,Naples (Italy).

Wang, T., Jónsdóttir, R., & Ólafsdóttir, G. (2009). Total phenolic compounds, radical scavenging and metal chelation of extracts from Icelandic seaweeds. *Food Chemistry*, 116(1), 240–248. DOI: https://doi.org/10.1016/j.foodchem.2009.02.041

Warnasooriya, V., Gunawardena, S., Weththasinghe, P., & Jayawardana, B. (2024). Nutritional Properties , Antioxidant Activity , and Heavy Metal Accumulation in Selected Marine Macro-Algae Species of Sri Lanka. 50–64.

Xu, X., Song, F., Wang, S., Li, S., Xiao, F., Zhao, J., Yang, Y., & Shang, S. (2004). Dibenzyl Bromophenols with Diverse Dimerization Patterns from the Brown Alga *Leathesia nana*. 1, 1661–1666.

Yan, X.; Chuda, Y.; Suzuki, M.; Nagata, T. Fucoxanthin as the Major Antioxidant in Hijikia Fusiformis , a Common Edible Seaweed. *Biosci. Biotechnol. Biochem*., 1999, 63 (3), 605–607. DOI: https://doi.org/10.1271/bbb.63.605