PROXIMATE COMPOSITIONS AND TOTAL PHENOLIC CONTENTS OF SELECTED EDIBLE SEAWEED FROM SEMPORNA, SABAH, MALAYSIA

[1,2]Fisal Ahmad, [1]Mohd Rosni Sulaiman,
[1]Welzan Saimon, [1]Chye Fook Yee, & [1]Patricia Matanjun

[1]School of Food Science and Nutrition,
Universiti Malaysia Sabah, Jalan UMS, 88400 Kota Kinabalu, Sabah, Malaysia.
[2]Food Science Department, Faculty of Agrotechnology and Food Science,
Universiti Malaysia Terengganu, 21030 Kuala Terengganu, Terengganu, Malaysia.

ABSTRACT. *In this study proximate compositions and total phenolic contents in extracts of 15 seaweeds from Semporna, Sabah, were determined. In general, results of proximate analysis for all seaweeds showed that moisture content (75.95-96.03%) was the most abundant in seaweed, followed by carbohydrate (26.86 - 74.10% dry weight basis), crude fiber (4.03 - 34.71% dry weight), and ash (6.05 - 45.04% dry weight), crude protein (5.22 - 17.28% dry weight), and crude fat content (0.15 - 0.84% dry weight). The total phenolic contents were determined using Folin-Ciocalteu reagent method based on the standard calibration curve of phloroglucinol measured at 740 nm using UV-Visible Spectrometer (Perkin Elmer). Overall, the total phenolic contents for all seaweeds of methanolic extract were between 9.40 - 51.87 mg/g phloroglucinol equivalents (PGE) of dried sample. The results of the present study showed significant differences ($p<0.05$) in proximate compositions and total phenolic contents among several species of red, green and brown seaweeds. The findings on total phenolic contents and proximate compositions of the seaweeds in this study can be further used as a basis for more advance research on seaweed antioxidant capability and nutritional information guideline, respectively.*

KEYWORDS. Edible seaweed, proximate compositions, total phenolic contents

INTRODUCTION

Macro-algae or "seaweeds" are multicellular plants growing in salt or fresh water. They belong to the lower plants, meaning that they do not have roots, stems and leaves. Instead they are composed of a thallus (leaf-like) and sometimes a stem and a foot (holdfast). Some species have gas-filled structures to provide buoyancy. They are often fast growing and can reach sizes of up to 60 m in length (McHugh, 2003). They are classified into three broad groups based on their pigmentation: i) brown seaweed (Phaeophyceae); ii) red seaweed (Rhodophyceae) and iii) green seaweed (Chlorophyceae) (Mohamed *et al.*, 2012). Red algae is the most abundant group (6000 species), followed by brown (2000 species) and green (1200 species) (Venugopal, 2011).

Seaweeds live in a harsh environment where they are exposed to a wide range of environmental stress such as light, rapid fluctuations in temperature, osmotic stress and desiccation. These factors can lead to the formation of free radicals and other strong oxidizing agents but seaweeds seldom suffer any serious photodynamic damage. This fact implies that seaweed cells have some protective mechanisms and compounds (Matsukawa *et al.*, 1997). A greater diversity in biochemical composition of seaweeds paves the way to explore a variety of compounds in their body composition with a wide range of physiological and biochemical characteristics, many of which are rare or absent in other taxonomic groups (Holdt & Kraan, 2011). These beneficial effects can be attributed to the complex mixture of

phytochemicals which possess antioxidant, antimicrobial, anticancer and antiviral activity. The compounds responsible for these activities include phenolic compounds, sulphated polysaccharides and organic acids (Liu, 2003; Podsedek, 2007).

In Malaysia, Sabah is the main seaweed producer and most of the total production comes from Semporna, which is located on the East coast of Sabah (Sade *et al.*, 2006). According to Matanjun *et al*. (2009) the chemical composition of edible seaweeds from some regions of the world has been well documented, but no reports are available on the nutritive value of the tropical seaweeds from North Borneo and the data from her publication only focus on *Eucheumacottonii, Caulerpalentillifera* and *Sargassumpolycystum* (Matanjun *et al*., 2009) and again in 2011 she only published data on *Kappaphycusalvarezii, Eucheumadenticulatum, Gracilariachangii, Gracilariaedulis, Caulerpalentillifera* and *Sargassumpolycystum* (*Matanjun*, 2011). From literature, it can be concluded that reports on chemical composition of Semporna, Sabah edible seaweeds are still lacking and the proximate compositions depend on species specific differences, growth environments, geographical locations and harvesting season (McDermid & Stuercke, 2003; Ortiz *et al.*, 2006; Renaud & Luong-Van, 2006; Marsham *et al.*, 2007; Chakraborty & Santra, 2008; Matanjun *et al*., 2009; Venugopal, 2011). Furthermore, to evaluate the functional properties of seaweeds require a clear idea about their biochemical composition, and which can provide a platform for identification of the molecules responsible for the different biological activities (Mendis & Kim, 2011). Besides that, there is a large amount of algal biodiversity that has been poorly studied (Gressler *et al.*, 2011). Therefore, this study was carried out to determine the proximate composition and total phenolic content of green, red and brown seaweeds that are found in Semporna, Sabah, Malaysia.

MATERIALS AND METHODS

Sample Collection and Preparation

A total of 15 types of seaweed consisting of two green seaweeds (*Caulerpalentillifera* and *Caulerparacemosa*), four brown seaweeds (*Sargassumpolycystum, Hormophysacuneiformis, Padinagymnospora* and *Turbinariaconoides*) and nine red seaweeds (*Kappaphycusalvarezii* var. aring-aring, *Kappaphycusalvarezii* var. green tambalang, *Kappaphycus striatum* var. Sacol [Katunai green], *Kappaphycus striatum* var. Sacol [katunai brown], *Kappaphycus striatum* var. Sacol [katunai yellow], *Eucheumadenticulatum* [var.yellow], *Gracilariaverrucosa, Laurencia sp*. [yellow] and *Laurencia sp*. [brown]) were collected from Semporna, Sabah in December 2011 for analysis in the present study. The seaweed identification was based on the morphological characteristics. Immediately after collection, the seaweed samples were cleaned and washed with seawater to remove sand, debris, epiphytes and other extraneous matter and transported to the laboratory in an ice cooler box to maintain the low temperature and moisture during the journey. In the laboratory, the samples were sorted and then thoroughly cleaned by rinsing with distilled water and dried with tissue paper to remove excess water. The moisture of the fresh samples was immediately analyzed. The remaining cleaned seaweed samples were then oven dried at 40^{o}C. After reaching constant weight, the dried samples were ground (for 5 min) into a fine powder using a Warring blender before being packed and stored in a freezer at -20°C until further chemical analysis. All chemical analyses of seaweed samples were carried out in triplicate.

Proximate Analysis

Moisture Content Analysis

Moisture content of seaweeds was determined according to the method described by AOAC (2000) with slight modifications. Samples (2 g) were put in a crucible and dried in a universal oven (Binder GmbH, Germany) at 105°C until constant weights were obtained.

Ash Content Analysis

Ash content of seaweeds was determined according to the method described by AOAC (2000) with slight modifications. Dried samples obtained from the moisture content analysis were burnt and ashed in a muffle furnace (Carbolite, United Kingdom) at 525°C overnight.

Crude Protein Analysis

Crude protein content of seaweeds was determined according to the method described by AOAC (2000) with slight modifications as recommended by Kjeltec 2300 (Foss Analytical, Denmark). Briefly, a 2 gram sample was weighed into digestion tubes. Two Kjeltabs Cu 3.5 (catalyst salts) was added into each tube. About 12 ml of concentrated sulphuric acid (H_2SO_4) was carefully added into the tube and then shaken gently. Digestion procedure was performed using pre-heated (420°C) digestion block of InKjel 625M (Behr, Germany) for 60 minutes until clear blue/green solution was obtained. Digested samples were cooled for 10-20 minutes. Distillation was then performed using distillation unit of Kjeltec 2300 (Foss, Denmark) and the percentage of protein was calculated by multiplying the percent of nitrogen found with a factor of 6.25.

Crude Lipid Content Analysis

Crude lipids were extracted from the seaweed powder following the method described by AOAC (2000) using the Soxtec 2050 System (Foss, Denmark) with petroleum ether as the solvent. The contents of crude lipids were determined gravimetrically after oven-drying (80^oC) the extract overnight.

Crude Fibre Content Analysis

Crude fibre was determined by sequential extraction of seaweed samples with 1.25% H_2SO_4 and 1.25% NaOH using the fibre-bag as a container. For drying and ashing, the crucible with sample was dried in an oven for 5 hours at 105°C and ashed in the muffle furnace (Carbolite, United Kingdom) at 525°C overnight. The weight of crucible with sample after drying and ashing was recorded and the crude fibre content was calculated (AOAC, 2000).

Carbohydrate Content

Carbohydrate content was calculated based on difference calculation [%Carbohydrate =100% - (%moisture + %ash + %crude fibre + %crude protein + %fat)].

Determination of Total Phenolic Content

Total phenolic was determined by the Folin-Ciocalteu method in accordance with a protocol described by Turkmen *et al.,* (2005) with some modifications. Standard solution stock (1,000 mg L^{-1}) of phloroglucinol (Sigma) was prepared by accurately dissolving 1.0 g of phloroglucinol with methanol and then the volume was made up to 10 mL. The stock solution was transferred to a dark vial and kept cool at 4°C prior to use. Working standard solutions were prepared by appropriate dilution of the stock solution. The contents of total phenolics of the seaweed samples were determined by the Folin-Ciocalteu method using phloroglucinol as a standard compound. The sample extract (1 mL) was mixed with 5 mL of the Folin-

Ciocalteu reagent (Sigma) (10% in distilled water in a test tube). After 5 min, 4 mL of 7.5% (w/v) $Na_2CO_3$was added to each tube, the test tubes were cap-screwed and vortexed (20 sec.). After incubation at room temperature for 2 hours in the dark, the absorbance of the reaction mixture was measured at 740 nm using UV-Visible Spectrometer(Perkin Elmer) against the blank sample which contained the same mixture without the sample extract. Using a six-point calibration curve (20 - 120 mg L^{-1}), the total phenolics were determined by comparison of the values obtained with the calibration curve of phloroglucinol. The results were expressed as phloroglucinol equivalents (PGE) in milligrams per grams of dried sample.

Statistical Analysis

Data collected in this study was analysed using SPSS (Statistical Package for the Social Sciences) version 17.0. One way ANOVA test was used to compare differences in the means of the moisture, ash, crude protein, carbohydrate, crude fibre, fat and total phenolic of different species and varieties of seaweeds. This was followed by Duncan multiple range test analysis to determine the differences between species. A significant difference was considered at the level of $p<0.05$.

RESULTS AND DISCUSSION

Proximate Compositions

The moisture content of the fresh seaweed samples ranged between 75.95% and 96.03% (Table 1). Among the 9 red seaweeds studied, *Laurencia sp.* (var. yellow) was found to have the highest ($p<0.05$)moisture content (96.03%) followed by *Laurencia sp.* (var. brown) (93.59%), *Gracilariaverrucosa* (85.45%), *Eucheumadenticulatum* (var.yellow) (84.54%), *Kappaphycusalvarezii* (aring-aring) (79.78%), *Kappaphycus striatum* var. sacol (Katunai green) (79.70%), *Kappaphycus striatum* var. sacol (katunai yellow) (76.69%) and *Kappaphycus striatum* var. sacol (katunai brown) (75.95%). The highest ($p<0.05$) moisture content between two species of green seaweeds was *Caulerparacemosa* (92.00%) and *Caulerpalentillifera* (90.84%). Meanwhile, the highest ($p<0.05$) moisture content among four species of brown seaweed was *Hormophysacuneiformis* (86.86%) followed by *Padinagymnospora* (84.54%), *Turbinariaconoides* (83.79%) and *Sargassumpolycystum* (83.51%), respectively. The moisture content obtained in *C. racemosa* was close to the values previously reported by Kumar *et al*. (2011), where the moisture content of fresh *C. racemosa* was very high (91.53%) and was the most abundant component in all fresh seaweeds. In general, there was a significant difference ($p<0.05$) of moisture content among seaweed samples except a few seaweed samples in similar classes. There was no significant difference in moisture values ($p>0.05$) between *Eucheumadenticulatum* (var.yellow), *Padinagymnospora* and *Turbinariaconoides*.

Table 1. Moisture (% fresh sample), Carbohydrate (% dry weight) and Ash content (% dry weight) of seaweed samples.

Seaweeds	Moisture (% f.s.)	CHO (% d.w.)	Ash (% d.w.)
Caulerpalentillifera	90.84±0.46^{d}	53.08±0.10^{e}	14.10±0.76^{g}
Caulerparacemosa	92.00±0.43^{c}	67.40±1.16^{b}	10.64±0.40^{h}
Sargassumpolycystum	83.51±0.27^{h}	34.93±1.34^{g}	21.87±0.40^{ef}
Hormophysacuneiformis	86.86±0.32^{e}	40.57±1.80^{f}	26.81±1.52^{c}
Padinagymnospora	84.54±1.18^{g}	26.86±0.17^{h}	45.04±1.61^{a}
Turbinariaconoides	83.79±0.32^{gh}	41.03±4.00^{f}	21.37±0.60^{f}
Kappaphycusalvarezii(aring-aring)	79.78±0.22^{i}	66.66±0.48^{b}	23.25±0.08^{d}
Kappaphycusalvarezii(green tambalang)	79.65±0.71^{i}	62.50±0.39^{c}	26.25±0.13^{c}
Kappaphycus striatum var. sacol (Katunai green)	79.70±0.70^{i}	66.02±0.70^{b}	22.99±1.04^{de}
Kappaphycus striatum var. sacol (katunai brown)	75.95±0.08^{j}	66.63±0.07^{b}	21.93±0.01^{ef}
Kappaphycus striatum var. sacol (katunai yellow)	76.69±0.33^{j}	67.49±0.62^{b}	22.84±0.25^{de}
Eucheumadenticulatum(var.yellow)	84.54±0.12^{g}	57.79±0.11^{d}	28.79±0.33^{b}
Gracilariaverrucosa	85.45±0.14^{f}	74.11±0.77^{a}	6.05±0.31^{j}
Laurencia sp. (var. yellow)	96.03±0.01^{a}	66.78±0.47^{b}	8.90±0.40^{i}
Laurencia sp. (var. brown)	93.59±0.15^{b}	66.00±0.27^{b}	11.60±0.09^{h}

Values are expressed as mean±sandard deviation, n=3.
Different superscript letters within a column indicate significant differences between samples at the level of $p<0.05$.

Carbohydrate content was a major component in dried seaweed (dry weight basis) (Table 1). All 15 dried seaweeds contained 26.86% to 74.11% d.w. of carbohydrate. Red seaweeds contain the highest amount of carbohydrate (57.79% to74.11% d.w.) followed by green seaweeds (53.08% to 67.40% d.w.) and brown seaweeds (26.86% to 41.03% d.w.). Nguyen *et al*. (2011), reported higher result for carbohydrate content (64.00% d.w.) in *Caulerpalentillifera* as compared with this study of only 53.08% d.w. and this might be due to many factors such as environment, geography, and season (Mendis & Kim, 2011). From statistical analysis, all brown seaweeds showed significant differences ($p<0.05$) in carbohydrate content compared to the other seaweeds (all were lower) and *Padinagymnospora* contained the lowest ($p<0.05$) amount of carbohydrate (26.86% d.w.). Green seaweeds, *Caulerparacemosa* (67.40% d.w.) and *Caulerpalentillifera* (53.08% d.w.) showed a significant difference ($p<0.05$) in their carbohydrate contents. Meanwhile, for red seaweed, only *Gracilariaverrucosa* (74.11% d.w.), *kappaphycusalvarezii* (green tambalang) (62.50% d.w.) and *Eucheumadenticulatum* (var.yellow) (57.79% d.w.) showed significant differences in carbohydrate contents ($p<0.05$).

Results from Table 1 show, ash as the second highest ($p<0.05$) component of dried material for all the twelve seaweed samples except for *Gracilariaverrucosa, Laurencia sp.* (var. yellow) and *Laurencia sp.* (var. brown). These results were comparatively higher than those of terrestrial counterparts with only 5% to 10% d.w. (USDA, 2001). The high ash content is a general feature of seaweeds, and these values are generally much higher than those of terrestrial vegetables other than spinach (Ruperez, 2002). The highest ($p<0.05$) ash value (45.04% d.w.) was recorded in *Padinagymnospora* and the results of this study showed that brown seaweeds contain high amounts of ash (21.37% to 45.04% d.w.) followed by green seaweeds (10.64% to 14.10% d.w.) and red seaweeds (6.05% to 28.79% d.w.). High ash content invariably indicates the presence of appreciable amounts of diverse mineral components (Mantanjun *et al*., 2008). Overall the value of ash varied and did not show any

significant patterns. Moreover, amounts of ash vary with phylum, season, environmental, geographical, and physiological variations (Ito & Hori, 1989; Kaehler & Kennish, 1996; Mendis & Kim, 2011).

The range of protein content in all three different classes of seaweeds varied and ranged from 5.22% to 17.28%d.w. in red seaweeds followed by green seaweeds (10.52% to 13.24% d.w.) and brown seaweeds (5.93% to 7.78% d.w.) (Table 2). These results were also similar to previous studies by other researchers where generally higher protein contents were found in green and red seaweeds (10% to 47% d.w.) compared to brown seaweeds (5% to 24% d.w.) (Burtin, 2003; Matanjun *et al*., 2009; Polat & Ozogul, 2009; Holdt & Kraan, 2011). They also found that most seaweeds proteins contained all the essential amino acids at levels close to that recommended by FAO/WHO (Matanjun *et al*., 2009; Wong & Cheung, 2000). The red seaweeds, *Laurencia sp.* (var. yellow) contained the highest ($p<0.05$) amount of protein (17.28% d.w.) followed by *Laurencia sp.* (var. brown) (14.80% d.w.) and both were significantly different ($p<0.05$) from the others. Next, *Caulerpalentillifera* (13.24% d.w.), *Gracilariaverrucosa* (11.73% d.w.) and *Caulerparacemosa* (10.52% d.w.), green, red and green seaweeds, respectively. They were also significantly different ($p<0.05$) from each other and the rest. The protein values in the present study were marginally higher than the values reported for other *Caulerpa* species that range from 5.8% to 10.41% d.w. (Matanjun *et al*., 2008; Renaud & Luong-Van, 2006) and previous studies reported that the protein content in seaweeds varied according to the season and the species (Fleurence, 1999; Galland-Irmouli *et al*., 1999; Murata & Nakazoe, 2001).

Eucheumadenticulatum (var.yellow) (7.65% d.w.), *Sargassumpolycystum* (7.78% d.w.) and *Turbinariaconoides* (7.40% d.w.) were not significantly different ($p>0.05$) from each other but significantly different ($p<0.05$) from the rest. Among the brown seaweed samples, *Sargassumpolycystum* and *Turbinariaconoides* contained the highest ($p<0.05$) amount of protein followed by *Hormophysacuneiformis* (6.42% d.w.) and *Padinagymnospora* (5.93% d.w.). Meanwhile, the other samples that showed no significant difference ($p>0.05$) in protein content and also contained the lowest amount of protein were *Kappaphycusalvarezii* (green tambalang) (5.63% d.w.), *Kappaphycus striatum* var. sacol (katunai green) (5.42% d.w.), *Kappaphycus striatum* var. sacol (katunai yellow) (5.40% d.w.), *Kappaphycusalvarezii*(aring-aring) (5.35% d.w.) and *Kappaphycus striatum* var. sacol (katunai brown) (5.22% d.w.).

Table 2. Crude Protein (% dry weight), Crude Fibre (% dry weight) and Crude Lipid (% dry weight) of seaweed samples.

Seaweeds	Protein (% d.w.)	Fibre (% d.w.)	Lipid (% d.w.)
Caulerpalentillifera	13.24 ± 0.11^{c}	19.40 ± 0.78^{d}	0.17 ± 0.05^{f}
Caulerparacemosa	10.52 ± 0.28^{e}	11.29 ± 0.47^{e}	0.15 ± 0.02^{f}
Sargassumpolycystum	7.78 ± 0.05^{f}	34.71 ± 1.99^{a}	0.71 ± 0.04^{ab}
Hormophysacuneiformis	6.42 ± 0.37^{g}	25.36 ± 1.25^{c}	0.84 ± 0.15^{a}
Padinagymnospora	5.93 ± 0.06^{gh}	21.66 ± 1.46^{d}	0.51 ± 0.05^{cd}
Turbinariaconoides	7.40 ± 0.06^{f}	29.61 ± 1.59^{b}	0.59 ± 0.21^{bc}
Kappaphycusalvarezii(aring-aring)	5.35 ± 0.02^{h}	4.50 ± 0.32^{fg}	0.23 ± 0.10^{f}
Kappaphycusalvarezii(green tambalang)	5.63 ± 0.45^{h}	5.45 ± 0.11^{fg}	0.18 ± 0.02^{f}
Kappaphycus striatum var. sacol (Katunai green)	5.42 ± 0.47^{h}	5.34 ± 0.03^{fg}	0.22 ± 0.05^{f}
Kappaphycus striatum var. sacol (katunai brown)	5.22 ± 0.15^{h}	5.96 ± 0.07^{fg}	0.25 ± 0.03^{f}
Kappaphycus striatum var. sacol (katunai yellow)	5.40 ± 0.07^{h}	4.03 ± 0.38^{g}	0.24 ± 0.01^{f}
Eucheumadenticulatum(var.yellow)	7.65 ± 0.14^{f}	5.23 ± 0.40^{fg}	0.54 ± 0.04^{cd}
Gracilariaverrucosa	11.73 ± 1.31^{d}	7.84 ± 0.25^{f}	0.27 ± 0.01^{ef}
Laurencia sp. (var. yellow)	17.28 ± 0.50^{a}	6.64 ± 0.35^{fg}	0.40 ± 0.04^{de}
Laurencia sp. (var. brown)	14.80 ± 0.35^{b}	7.16 ± 0.18^{fg}	0.45 ± 0.07^{cd}

Values are expressed as mean±sandard deviation, n=3.
Different superscript letters within a column indicate significant differences between samples at the level of $p<0.05$.

The results showed a segregate pattern of the crude fibre content of seaweeds ranging significantly from 21.66% to 34.71% d.w. for brown seaweed, followed by green seaweed (11.29 to 19.04% d.w.) and red seaweed (5.23 to 7.84% d.w.). The highest value ($p<0.05$) of crude fibre was *Sargassumpolycystum* (34.71% d.w.) followed by *Turbinariaconoides* (29.61% d.w.), *Hormophysacuneiformis* (25.36% d.w.) and *Padinagymnospora* (21.66% d.w.) in the brown seaweeds class and they were also significantly different ($p<0.05$) from the others. Green seaweeds *Caulerpalentillifera* and *Caulerparacemosa* were significantly lower ($p<0.05$) than brown seaweeds with the value of 19.40 % d.w. and 11.29% d.w., respectively. Meanwhile, the red seaweeds that contained significantly lower ($p<0.05$) amounts of crude fibre were *Gracilariaverrucosa* (7.84% d.w.) followed by *Laurencia sp.* (var. brown) (7.16% d.w.), *Laurencia sp.* (var. yellow) (6.64% d.w.), *Kappaphycus striatum* var. sacol (katunai brown) (5.96% d.w.), *Kappaphycusalvarezii* (green tambalang) (5.45% d.w.), *Kappaphycus striatum* var. sacol (Katunai green) (5.34% d.w.), *Eucheumadenticulatum* (var.yellow) (5.23% d.w.), *Kappaphycusalvarezii* (aring-aring) (4.50% d.w.) and *Kappaphycus striatum* var. sacol (katunai yellow) (4.03% d.w.). Overall, these results were lower than the previous studies, which reported that dry weight edible seaweed contained 33% to 62% total fibres. This is higher than the levels found in higher plants, and these fibres are rich in soluble fractions (Lahaye, 1993; Dawczynski *et al*., 2007). Holdt & Kraan (2011) reported that the seaweed dietary fibres contained some valuable nutrients and substances, and there has been a great deal of interest in seaweed meal, functional foods and nutraceuticals for human consumption (McHugh, 2003; Gupta & Abu-Ghannam, 2011) because, among other things, polysaccharides show anti-tumour and anti-herpetitic bioactivity; they are potent as an anticoagulant and decrease low-density lipid (LDL)-cholesterols in rats (hypercholesterolemia); they prevent obesity, large intestine cancer and diabetes; and they have antiviral activities (Lee *et al*., 2004; Murata & Nakazoe, 2001; Amano *et al*., 2005; Athukorala *et al*., 2007; Ghosh *et al*., 2009; Ye *et al*., 2008; Harnedy & FitzGerald, 2011).

The total lipid content was very low in all seaweeds samples (0.15% to 0.84% d.w.) (Table 2) and became a minor proximate component, but this result also fell within the ranges reported previously (Rupérez & Saura-Calixto, 2001; Matanjun *et al*., 2009; Gómez-Ordóñez *et al*., 2010). In contrast, McDermid & Stuercke (2003) reported relatively higher lipid values (7.2% d.w.) for *Caulerpa* species collected from the Hawaiian coast. These differences could be due to the different environmental conditions, season of harvesting and habitat. Generally, the results showed that brown seaweed contained significantly higher ($p<0.05$) lipid as compared to red and green seaweeds ranging from 0.51% to 0.89%d.w., 0.18% to 0.54% d.w. and 0.15% to 0.17% d.w., respectively. Overall, the proximate content varies depending on the season and the area of production (Connan *et al*., 2004; Khan *et al*., 2007; Marinho-Soriano *et al*., 2006; Murata & Nakazoe, 2001; Zubia *et al*., 2008).

Total Phenolic Contents

The content of total polyphenols was determined using the Folin-Ciocalteu's method. The calibration curve of phloroglucinol, obtained by representing the absorbance measurements versus the concentration of phloroglucinol, was adjusted to a linear equation $y = 0.1194x - 0.1026$ with a coefficient of correlation of $R^2 = 0.9995$. The linear range was within 5-100 mg L^{-1}. Each point of the calibration curve is the average of two absorbance measurements.

The variation of phenolic content was quite large and significant differences were found ($p<0.05$) among different seaweed species, ranging from 9.41 to 51.87 mg PGE/g dried sample (Table 3). This study, indicated that both green seaweeds and brown seaweeds (not including *Hormophysacuneiformis*) contained higher amounts ($p<0.05$) of polyphenols than red seaweeds. Green seaweeds *Caulerpalentillifera* (51.87 mg PGE/g dried sample), *Caulerparacemosa* (47.88 mg PGE/g dried sample) and also brown seaweed *Turbinariaconoides* (33.51 mg PGE/ g dried sample), *Padinagymnospora* (25.33 mg PGE/g dried sample) and *Sargassumpolycystum* (23.03mg PGE/g dried sample), showed significantly higher ($p<0.05$) phenolic content than all of the red seaweeds and *Hormophysacuneiformis* (14.60 mg PGE/ g dried sample) of brown seaweed. Previous studies also found that the total phenolic contents varies with species and generally the green seaweeds have higher free-radical scavenging properties, followed by the brown seaweed, then the red seaweeds (Chandini *et al*., 2008; Duan *et al*., 2006; Matanjun *et al*., 2008; Santoso *et al*., 2004).

This study, indicated that the antioxidant compounds were significantly different ($p<0.05$) depending on the species of seaweeds. In addition, the selection of the extracting solvent is an important factor for obtaining active compounds in seaweed. Therefore further study can be done to select the best solvent and followed by determination of their antioxidant activities. This is because the antioxidant activity of the extracts from seaweeds is not directly correlated with their total phenolic contents (Lim *et al*., 2002; Chandini *et al*., 2008). The finding on total phenolic content in this study can be used for further research on antioxidant capability.

Table 3. Total phenolic contents (TPC) of seaweed methanolic extracts expressed as phloroglucinol equivalents (mg PGE/g dried sample).

Seaweeds	Total Phenolic Contents(mg PGE/g dried sample)
Caulerpalentillifera	51.87 ± 0.53^{a}
Caulerparacemosa	47.88 ± 1.18^{b}
Sargassumpolycystum	23.03 ± 1.86^{de}
Hormophysacuneiformis	14.60 ± 0.63^{g}
Padinagymnospora	25.33 ± 1.78^{d}
Turbinariaconoides	33.51 ± 2.80^{c}
Kappaphycusalvarezii (aring-aring)	10.47 ± 1.01^{h}
Kappaphycusalvarezii (green tambalang)	10.99 ± 1.95^{h}
Kappaphycus striatum var. sacol (Katunai green)	11.26 ± 1.81^{h}
Kappaphycus striatum var. sacol (katunai brown)	9.76 ± 0.66^{h}
Kappaphycus striatum var. sacol (katunai yellow)	10.63 ± 2.35^{h}
Eucheumadenticulatum (var.yellow)	9.41 ± 1.81^{h}
Gracilariaverrucosa	12.08 ± 2.35^{gh}
Laurencia sp. (var. yellow)	20.51 ± 1.04^{e}
Laurencia sp. (var. brown)	17.52 ± 0.81^{f}

Values are expressed as mean±sandard deviation, n=3.
Different superscript letters indicate significant differences between samples at the level of $p<0.05$.

CONCLUSIONS

The values of proximate and total phenolic contents are diverse depending on the species of seaweeds. These seaweeds could be potentially rich sources of natural antioxidants and the findings on total phenolic contents and proximate compositions of the seaweeds in this study can be used as a basis for more advanced research on seaweed antioxidant capability which will enrich the national food composition database.

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