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Antioxidant Activity Against Methanol Extraction of *Eucheuma cotonii* and *E. spinosum* Collected From North Sulawesi Waters, Indonesia

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Abstract

Eucheuma cotonii and *E. spinosum* cultivated in Arakan waters, North Sulawesi, Indonesia, were tested the antioxidant activity of fresh and dry samples through maceration in 60%, 70%, and 80% methanol solvent. The tested antioxidants were total phenol, DPPH, FRAP, and total carotene, respectively.

Fresh *E. spinosum* dissolved in 60% methanol had the highest total phenol, 5.87±0.15 mg GAE/g, the highest DPPH, 75.27 ± 0.29 %, the highest FRAP, 44.52 ± 1.27 mg/l, respectively. The highest total carotene, 9.40±0.35 µg/g, was recorded in fresh *E. cotonii,* followed by fresh *E. spinosum,* 8.73±0.23 µg/g, which were also dissolved in 60% methanol. Therefore, the best antioxidant activity was found in fresh *E. spinosum* macerated in 60% methanol.

Keywords: Alga, antioxidant activity, phenol, DPPH, FRAP, carotene

1. Introduction

Algae are fisheries products utilized as food materials, medicines, and cosmetics due to their bioactive compound richness, for instance, vitamins, minerals, food fibres, and antioxidants, such as polyphenol, carotene, and flavonoid (Ganesan, *et al*., 2008; Chew, *et al.,* 2008; Shahidi, 2009). Important minerals contained in algae are useful for body metabolisms, such as iodium, calcium, and selenium. The important vitamins for human diets are vitamin A, B, folic acid (B9), C, and E (Andarwulan, *et al*, 2010; Matanjun, *et al.,* 2009; Anonymous, 2010). Algae are very slowly damaged since their cells possess antioxidative mechanisms and antioxidant compounds (Shanab, 2007).

Eucheuma cotonii and *E. spinosum* belong to red algae that have been cultured and utilized as food materials, such as source of carrageen and antioxidant (Hotchkiss, 2007; Gerung, 2002). Ismael and Tan (2002), showed that the processed commercial algae exhibited different antioxidant acitivity level. It likely resulted from chlorophyll, carotene, and phenol content (Shanab, 2007).

Marine algae of *Rhodophyceae* (red alga) contain phycoerythrine pigment, carotenoid, chlorophyll, organic and inorganic compounds. Previous study showed that carotene in algae was an antioxidant to protect various diseases and stresses (Jimenez-Escrig, *et al*., 2001).

Antioxidants are compounds capable of passing one electron to the free redicals to be neutral (Winarsi, 2007). These are also crucial to take care of the body health since functioning as an inhibitor against free radicals formed in human body. Food materials which become natural antioxidant sources are spices, leaf, cacao, bean, fruit, vegetables, and algae (Hernani, 2006). Several antioxidant compounds were found in macroalgae belonging to phenolic group, terpenoid, taondiol, polyphenolic group, catekin, flavonoid, sulphated polysaccharides group, fucoidan, alginic acid, sulphated galactan, porphyran, carotenoid group, fucoxanthin, B-carotene, antheraxanthin, lutein xanthiphylls zeaxanthin in red algae (Cornish and Garbary, 2010).

Studies on antioxidant activity and total phenol have been enormously done. It was reported that green and brown algae families collected from Pulau Seribu and red algae from Central Java contained antioxidant activities (Suryaningrum, *et al*, 2006; Santoso, 2010). *Padina antilatirum* (brown alga) has antioxidant and total phenol content higher than those of *Eucheuma cotonii* and *Caulerpa rasemosa* taken from Malaysia waters (Chew, *et al.*, 2008).

The antioxidant activity test could be carried out using a variety of extraction methods, and a general technique used is maceration (Suryaningrum, *et al*, 2006; Devi, *et al*., 2008; Kumar, *et al*., 2008; Anonymous, 2011).

In this study, the methanol solvent was selected as a common solvent utilized in polar, semi-polar, and non-polar compound extraction. *Eucheuma cotonii* and *E. spinosum* cultured in North Sulawesi waters have not been studied and published their antioxidant content yet. Hence, the antioxidant activity characteristics of *E. cotonii*

dan *E. spinosum* cultured in Arakan waters, North Sulawesi, is necessary to study to obtain the best methanol solvent concentration.

2. Material and Method

2.1 Materials and Equipment

Major raw materials of the study were algae *E. cotonii* and *E. spinosum.* These materials were collected from the algae farmer in Arakan, South Minahasa Regency, the Province of North Sulawesi, Indonesia. The algae were tested in both fresh and dry forms.

The equipment used in this study were bottle for maceration, digital balance (denver M-310), freeze dryer, cabinet dryer, UV spectrophotometer, autoclave, centrifuge, glass materials, GC-MS (Gas Chromatography-Mass Spectrometry) or LC-MS (Liquid Chromatography-Mass Spectrometry).

2.2 Test Material Preparation

In this study, the extraction of algal antioxidant compounds, *E. cotonii* and *E. Spinosum,* was carried out in fresh and dry forms through maceration in 3 different methanol concentrations, 60%, 70%, and 80%, respectively.

The fresh samples were initiated with cleaning and rinsing with seawater to remove various attached materials. Furthermore, these were washed with running freshwater, water removed and put into the plastic bag of each 50 g. The fresh samples were kept in frozen form at -20^{o}C for further analysis. In extraction phase, 500 g of fresh sample was weighed and then macerated at room temperature for 3x24 hours using 60%, 70%, and 80% methanol solvents as much as 500 ml per day. The macerat produced was then filtered in filter paper, evaporated in a vacuum rotary evaporator at 40^{o}C until obtaining the liquid extracts. These were then dried in the oven at 40^{o}C. Coarse extracts obtained were then used in the analyses of the antioxidant activity, total phenol, DPPH, FRAP, and total carotene. Before that, the water content of the fresh sample was measured.

For dry samples, *E. cotonii* and *E. spinosum* were cleaned and washed with seawater to remove all attached materials, then washed with freshwater and air-dried on the table for 4 days, then further dried in the cabinet dryer for 24 hours at 40-50 ^{o}C until 10 times the fresh sample weight loss achieved. These dry samples were milled in a waring blender for 3 minutes. The extraction was then carried out through maceration of 50 g dry sample in 3x50ml of 60%, 70% and 80% methanol, respectively, at room temperature for 3x24 hours. The macerat produced was filtered in filter paper, evaporated in a vacuum rotary evaporator at 40^{o}C until a liquid extract obtained and then dried in the oven at 40^{o}C. Dry macerat was stored for total phenol, DPPH, FRAP, and total carotene analysis.

2.3 *Total phenol Analysis*

Total phenol content was measured with a spectrophotometer using the *Folin-Ciocalteau* reagent (Ganesan, *et al,* 2008) with some modification. As much as 0.1 g of extract was weighed and dissolved in 10 ml of methanol *p.a* in the flask. The extract solution was pippetted 0.1 ml and added 1 ml of 1: 2 ratio-*Folin Ciocalteau-aquadest* then left for 5 minutes. It was then added 1 ml of 7% sodium carbonate, homogenized and incubated at room temperature for 30 minutes in the dark. The total phenol content was measured with a UV – Vis spectrophotometer at λ 750 nm. Total phenol content was interpreted as mg equivalent to galic acid/g extract (= mgGAE/g extract).

2.4 *Antioxidant Activity Test with DPPH Method*

The antioxidant activity was tested with the DPPH method following Chew *et al* (2008) with some modification. Based on the antioxidant ability in the sample to reduce the free radicals, the DPPH was used. As much as 2 ml of DPPH 93 µM solution (3.6 mg in 100 ml metanol) was added into 0.5 ml extract (50,000 mg/l diluted with methanol). The mixture was shaken and incubated at 37^{O}C for 30 minutes, and the absorbance was measured in a UV-VIS spectrophotometer at λ 517 nm. Similar technique was used for comparison with BHT. The capture activity of the free radicals was set as percent inhibition countable following the equation below:

% inhibition = (Control Abs - Sample Abs)/(Control Abs) x 100

2.5 *Antioxidant test using Ferric Reducing Antioxidant Power (FRAP)*

The antioxidant activity test with Ferric Reducing Antioxidant Power (FRAP) method was conducted following the procedure of Chew *et al.*, (2008) with some modification. The potential reduction of methanol extract was done as the following procedures : 1 ml of 50,000 mg/l extract was mixed with 1 ml of phosphate buffer (0.2 M, pH 6.6) and 1 ml of 1 % potasium ferricyanide [$K_3Fe(CN)_6$] and divortex. The mixture was homogenized and incubated at 50^{0}C for 20 minutes (mixture A). It was then added 1 ml of trichloroasetic acid (10%) and centrifuged for 10 minutes at 3,000 rpm (mixture B). The upper layer of mixture B was taken 1 ml and mixed with 1 ml of destilled water, added 0.5 ml of 0.1% $FeCl_3$. The absorbance was measured at λ 700 nm. The FRAP value was interpreted as mg equivalent to galic acid/g extract.

2.6 *Total Carotene Measurement*

Total carotene was measured according to Cagampang and Rodriguez, (1980) and Dere, *et al,* (1989); with

some modification. As much as 0.1 g of algal extract was weighed and dissolved in 5 ml of 1:1 ratio-80% acetone- Petrolium Eter, homogenized in a homogenizer at 1000 rpm for 5 minutes. The supernatant was taken and measured the absorbance at λ 470 nm.

2.7 Statistical Analysis

Experiments were run with 3 treatments of different methanol concentrations, 60 %, 70%, and 80% using a Complete Randomized Design with 3 replications of each algal species and sample condition. If there was difference in treatment effect, the least significant difference test was done to know the inter-treatment difference. Data analysis used the version 20-SPSS software.

3. Results and Discussion

3.1 Total phenol

The total phenol content of the four sample conditions was given in Table 01.

Table 01. Total phenol content of *Eucheuma sp*

Methanol Concentration (%)	**Total phenol (mg GAE/g extract)**			
	Fresh E. cotonii	*Dry E. cotonii*	*Fresh E. spinosum*	*Dry E. spinosum*
60	3.96 ± 0.06 (b)	1.84 ± 0.07 (a)	5.87 ± 0.15 (b)	4.90 ± 0.07 (c)
70	2.96 ± 0.23 (a)	1.76 ± 0.15 (a)	4.98 ± 0.02 (a)	4.75 ± 0.04 (b)
80	3.70 ± 0.12 (b)	1.83 ± 0.13 (a)	5.76 ± 0.11 (b)	4.49 ± 0.02 (a)

Notes: The same alphabethical code in the same column indicates no significant difference of the treatment ($p > 0.05$), and the different alphabethical code in the same column indicates significant difference of the treatment ($p < 0.05$)

Table 01 shows no difference in total phenol content ($p > 0.05$) for dry *E. cotonii* in the three different methanol concentrations. However, other sample condition exhibits different total phenol content ($p < 0.05$), in which the fresh *E. spinosum* possesses the highest total phenol as shown in Fig. 01.

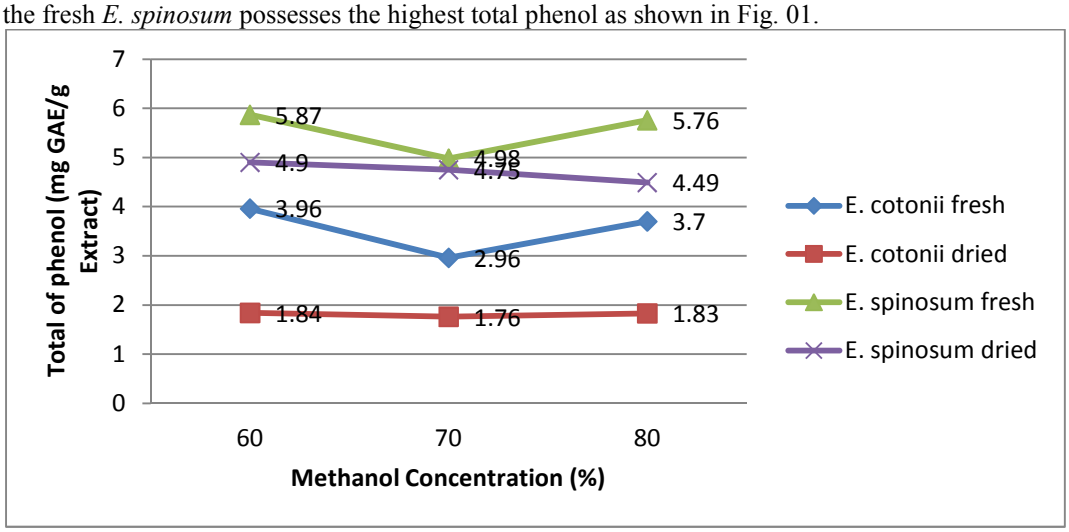

Figure 01. Total phenol content

Figure 01 demonstrates that fresh *E. spinosum* results in the highest total phenol content in all solvent concentrations with the highest recorded in 60 % methanol, 5.87 ± 0.15 mg GAE/g extract.

Harboune (1987) pinpointed that small molecules, including the antioxidant compounds, are relatively easily dissolved in the water than other macromolecules. The phenolic compounds are usually found in plants and averagely reported having biological activities, including the antioxidant characteristics.

Reisce *et al*, (2002) found that compounds belonging to natural antioxidants are tokoferol, ascorbic acid, carotenoid, sterol, phenolic acid, and flavonoid. Phenolic and plavonoid acid are the antioxidant compounds containing phenolic structures and occur in plant in a great number.

3.2 DPPH

The DPPH data of 4 algal sample conditions are given in Table 02.

Table 02. DPPH content in *Eucheuma sp*

Methanol Concentration (%)	**DPPH (%)**			
	Fresh E. cotonii	*Dry E. cotonii*	*Fresh E. spinosum*	*Dry E. spinosum*
60	64.73 ± 1.61 (b)	68.99 ± 1.68 (c)	75.27 ± 0.29 (b)	64.27 ± 1.44 (b)
70	62.72 ± 2.41 (b)	56.59 ± 1.14 (b)	65.19 ± 1.09 (a)	59.32 ± 1.20 (a)
80	58.37 ± 0.76 (a)	52.85 ± 0.53 (a)	66.77 ± 1.08 (a)	58.53 ± 0.61 (a)

Notes: The same alphabethical code in the same column indicates no different effect of the treatment ($p > 0.05$), while the different alphabethical code in the same column indicates different effect of the treatment ($p < 0.05$).

Table 02 shows that there is different DPPH content in all sample conditions ($p < 0.05$) in the three methanol concentrations. The fresh *E. spinosum* has the highest DPPH content as given in Figure 02.

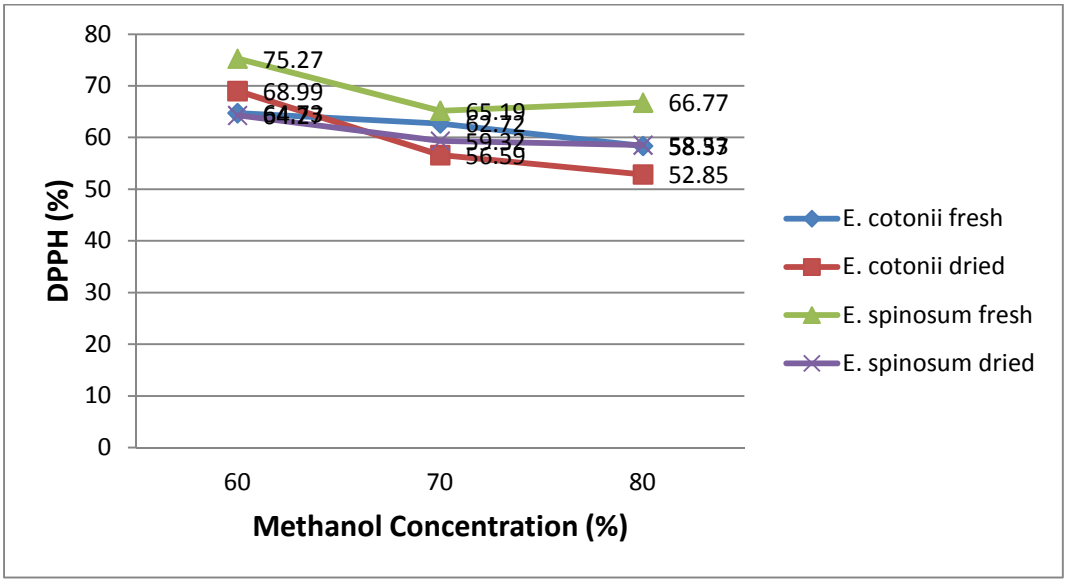

Figure 02. DPPH content

It is apparent in Figure 02 that the fresh *E. spinosum* result in the highest DPPH content in the three methanol concentrations, and the highest is recorded in 60 % methanol, 75.27 ± 0.29.

A 1,1-diphenyl-2-picryhydrazil (DPPH) compound is stable and actified radical by delocating the free electron on a molecule containing free radicals, so that the molecule becomes inreactive. The free radicals are highly reactive and unstable molecules since these have one or more unpaired electrons. Capture mechanism of DPPH radicals by the antioxidant occurs through proton donation to the radical. Therefore, the compound that enables to donate its proton contains strong radical capturing activity (Kumar *et al.,* 2008; Ganesan *et al*., 2008). The compounds belong to phenolic, flavonoid, tanin, and alkaloid groups, and the compounds with many sulfide groups. Proton donation causes the violet colored-DPPH radical turn to colorless non-radical compounds. Thus, the radical capture activity could be counted from DPPH radical scavenging. The remaining DPPH radical content was spectrophotometrically measured at λ 517 nm (Kumar *et al*., 2008; Chew e*t al.*, 2008). The inhibitory ability against the free radicals is affected by the extent of extract concentration. The DPPH activity generally rises with extract increment up to certain concentration. Then it will decrease with more concentration addition. The DPPH test was extensively used in natural product studies for antioxidant isolation and extract and pure compound ability to absorb the radicals.

3.3 FRAP (mg/l)

FRAP content data in 4 different sample conditions are shown in Table 03.

Table 03. FRAP content in *Eucheuma sp*

Methanol Concentration (%)	**FRAP (%)**			
	Fresh E. cotonii	*Dry E. cotonii*	*Fresh E. spinosum*	*Dry E. spinosum*
60	33.43 ± 1.57 (a)	28.17 ± 0.52 (c)	44.52 ± 1.27 (c)	31.05 ± 0.57 (a)
70	35.34 ± 1.18 (a)	25.65 ± 0.54 (b)	31.33 ± 1.21 (a)	30.09 ± 0.79 (a)
80	33.82 ± 1.00 (a)	22.88 ± 1.33 (a)	35.45 ± 0.55 (b)	30.15 ± 0.96 (a)

Notes: The same alphabethical code in the same column indicates no different effect of the treatment ($p > 0.05$), while the different alphabethicalcode in the same column indicates different effect of the treatment ($p < 0.05$).

Table 03 shows no difference in FRAP content ($p > 0.05$) for both fresh *E. cotonii* and dry *E. spinosum*. In other sample conditions, there is difference in FRAP content ($p < 0.05$), in which the fresh *E. spinosum* has the highest FRAP content (Fig. 03).

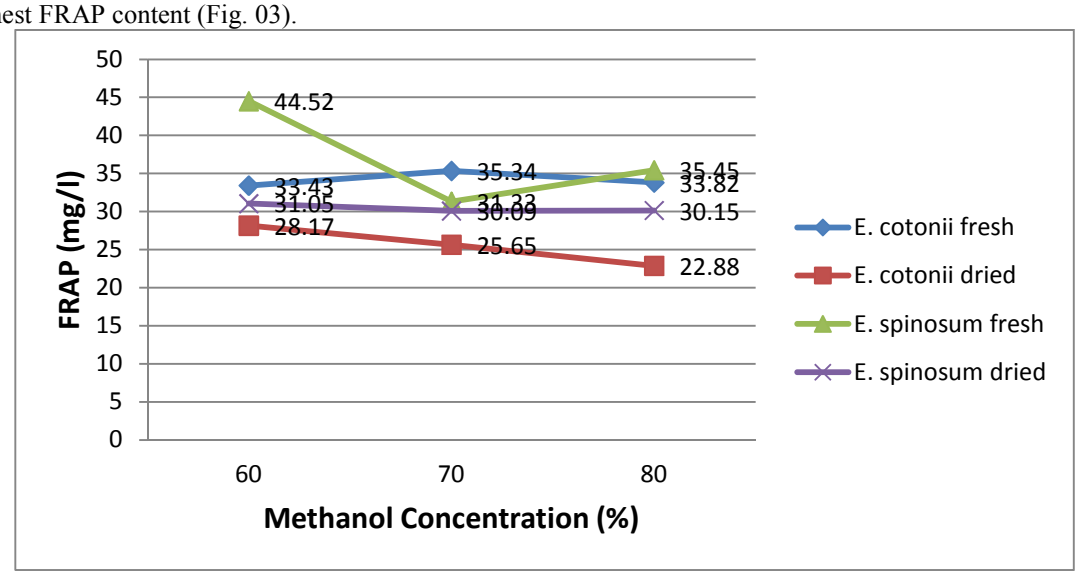

Figure 03. FRAP content (mg/l)

Fig. 03 demonstrates that the fresh *E. spinosum* results in relatively high FRAP content for the three methanol concentrations, and the highest is recorded in 60 % methanol, 44.52 ± 1.27 mg/l.

The determination of total antioxidant content with FRAP method is based on the compound ability to reduce the Fe^{3+} ion to be Fe^{2+} ion using 2,4,6-Tris (1-pyridyl)-5-Triazine (TPTZ). The antioxidant ability of a compound is analogized to the ability to reduce the compound in a reaction of redox-linked colourimetric (Halvorsen *et al*., 2002; Chew *et al*., 2008).

3.4 Total Carotene

Total carotene data in four sample conditions are given in Table 04.

Table 04. Total carotene in *Eucheuma sp*

Methanol Concentration (%)	**Total Carotene** (µg/g)			
	Fresh E. cotonii	*Dry E. cotonii*	*Fresh E. spinosum*	*Dry E. spinosum*
60	9.40 ± 0.35 (b)	7.47 ± 0.12 (a)	8.73 ± 0.23 (b)	6.73 ± 0.12 (a)
70	7.60 ± 0.69 (a)	8.13 ± 0.42 (b)	7.80 ± 0.20 (a)	6.80 ± 0.20 (a)
80	7.60 ± 0.20 (a)	7.93 ± 0.12 (a)	7.73 ± 0.12 (a)	6.87 ± 0.23 (a)

Notes: The same alphabethical code in the same column indicates no different effect of the treatment ($p > 0.05$), while the different alphabethicalcode in the same column indicates different effect of the treatment ($p < 0.05$).

Table 04 indicates no difference in total carotene content ($p > 0.05$) for dry *E. spinosum* in the three methanol concentrations. In other sample conditions, there are differences in total carotene content ($p < 0.05$), in which the fresh *E. cotonii* contains the highest total carotene (Fig. 04).

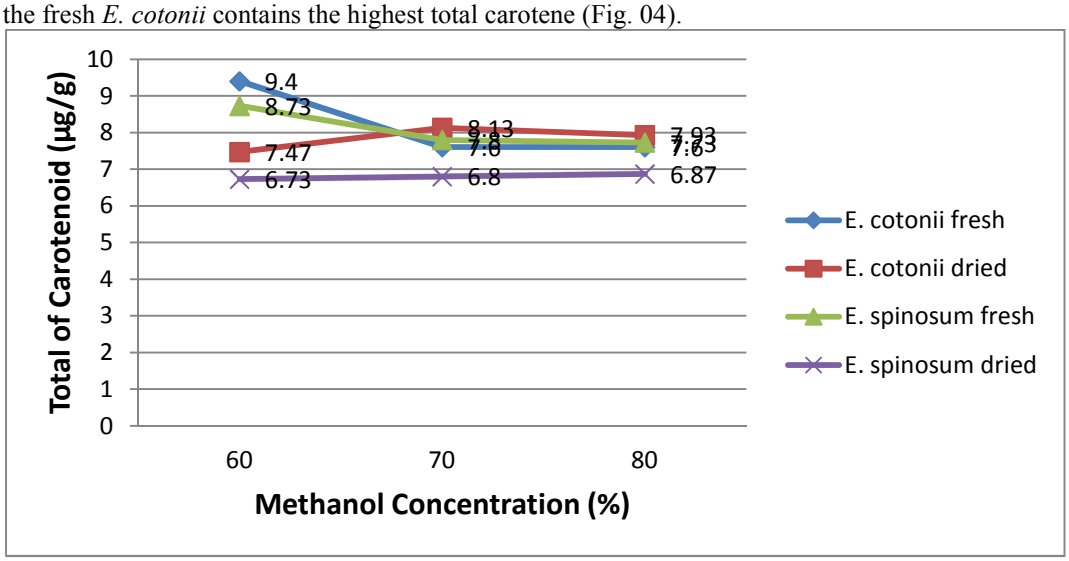

Figure 04. Total carotene content

Fig. 04 exhibits that the fresh *E. cotonii* results in the highest total carotene content (9.40 ± 0.35 µg/g) in 60% methanol, followed by the fresh *E. spinosum* (8.73 ± 0.23 µg/g).

Carotene in algae is an antioxidant that could work to protect various diseases and stresses (Cornish and Garbary, 2010). The measurement of total carotene content reflects that the fresh *E.spinosum* extracted in 60% methanol gives the highest antioxidant activity.

Conclusion

Eucheuma cotonii and *Eucheuma spinosum* cultured in Arakan waters, North Sulawesi, contain antioxidant compounds. The best antioxidant activity was recorded in fresh *E. spinosum* macerated in 60% methanol. This result is as initial information for antioxidant compound characteristic isolation and identification study.

References

Andarwulan, N., R. Batari, D.A. Sandrosari, B. Bolling, and H. Wijaya, 2010. Flavonoid content and antioxidant activity of vegetable from Indonesian. Food chemishy 121, 1231-1235.

Anonymous, 2010. FT-IR Spectroscopy – Introduction & Principles. Thermo Scientific. www.thermo.com/com/cda/category/category-lp/1, 234,00,html.

Anonymous. 2011. Maceration. Wikipedia. www.wikipedia.freeenciklopedia/maceration.html.

Cagampang, G.B., and F.M. Rodriguez. 1980. Methods Of Analysis For Screening Crops of Apropriate Qualities. Analytical Services Laboratory, Institute of Plant Breeding, University of The Philippines at Los Banos.

Chew, Y.L., Y.Y. Lim, M. Omar, and K.S. Khoo. 2008. Antioxidant activity of Three edible seaweeds from two areas in South East Asia. LWT 41 (2008) : 1067 – 1072.

Cornish, L. M., and D. J. Garbary. 2010. Antioxidants From Macroalgae : potensial application in human health and nutrition. James S. Craigie Researh Centre, Acadian Seaplants Limited, Cornwallis, NS BOS 1 HO, Canada. Review. Algae 2010, 25(4): 155 - 171

Dere, S., T. Gross, and R. Sivaci. 1998. Spectrophotometric Determination of Chlorophyll – A, B and Total Carotenoid Contens of Some Algae Species Using Diffrent Solvents. Tr. J. of Botany, 22: 13-17.

Devi, K. P., N. Suganthy, P. Kesika, and S.K. Pandian. 2008. Bioprotective Properties of Seaweed: In Vitro Evaluation of Antioxidant Activity and Antimicrobial Activity Against Food Borne Bacteria in Relation to Polyphenolic Content. BMC complementory and Alternative Medicine., 8(38)

Ganesan, P., Chandini S. Kumar, and N. Bhaskar. 2008. Antioxidant Properties of Methanol extract and its solvent fraction obtained from selected Indian red seaweed. J. Bioresource Technology 99 (2008) : 2717 – 2723

Gerung, G. 2002. Seaweeds Resources of Indonesia. Artikel ilmiah. Sam Ratulangi University, Faculty of Fisherias and Marine Science. Manado.

Halvorsen, B.L., K. Holte, M.C.W. Myhrstad, I. Barikmo, E. Hvattum, S.F. Reberg, A.B. Word, D.R. Jacobs, and R. Blomhoff. 2002.. A Systematic Screening of Total Antioxidant In Diietary Plants. J. Nutrition. 132 :

461-471.
Harborne, J.B. 1987. Metode Fitokimia. Penuntun cara Modern menganalisis Tumbuhan. Penerbit ITB, Bandung.
Hernani, M. Rahardjo. 2006. Tanaman Berkhasiat Antioksidan. Berbagai Jenis Tanaman Penangkal Racun. Seri Agrisehat. Penebar Swadaya. Cetakan ke-2. Jakarta.
Hotchkiss, S. 2007. Seaweed rediscovered : Healthy hydrocolloids and beyond. Nutritrional seaweed. Cybercolloid LTD. Carrigaline
Ismael, A., and S. H. Tan. 2002. Antioxidant and Antiprokiferative activities of extract from a variety of Edible seaweed. Food and Chemical Toxicology 44, 1144-1150.
Jimenez-Escrig, A, I. Jimenez-Jimenez, R. Pulido, and F. Saura-Calixto. 2001. Antioxidant activity of fresh and processed edible seaweeds. J. Sci. Food Agric., 81 :530-534.
Kumar, K. S., K. Ganesan, and P.V. Subba Rao. 2008. Antioxidant Potensial of Solvent Extract of *Kappaphycus alvarezii* (Doty) Doty- An edible seaweed. J. Food Chemistry 107 (2008) : 289 – 295.
Matanjun, P., Suhaila Mohamed, Noordin M. Musthapa, and K. Muhammad. 2009. Nutrient Content of Tropical Edible Seaweeds, *Eucheuma cottonii*, *Caulerpa lentillefera,* and *Sargassum polycystum*. J. Appl. Phycol 21:75-80.
Reische, D.W., D.A. Lillard, and R.R. Etenmiller. 2002. Antioxidant. In Akoh, C.C. dan D.B. Min. Food Lipids. 2008. Marcel Dekker. New York. Hal 490 – 500.
Santoso, J. 2010. Aktivitas Antioksidan Ekstrak Metanol, Etil asetat, dan Heksana Rumput Laut Hijau *Caulerpa lentillifera*. Jurnal Ilmu Kelautan. Februari 2010. Vol. 1. Edisi khusus: 1 – 10.
Shahidi, F. 2009. Nutraceuticals and functional foods : whole versus processed foods. Review. Trends in Food Science & Technology 20 (2009) 376 – 387.
Shanab, S. 2007. Antioxidant and Antibiotic Activities of Some Seaweed (Egyptian Isolates). International Journal of Agriculcure & Biology., 9 (2) : 220-225.
Suryaningrum, D.T, T. Wikanta., dan H. Kristiani. 2006. Uji Aktivitas Senyawa Antioksidan dari Rumput Laut *Halymenia harvenya da Euchema cottonii.* Jurnal Pascapanen dan Bioteknologi Kelautan dan Perikanan. Vol I No I.
Winarsi, 2007. Antioksidan Alami dan Radikal bebas. Potensi dan Aplikasinya dalam Kesehatan. Kanisius. Yogjakarta.

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