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Research Article

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Antioxidant activities from different parts of *Sargassum polycystum* thalli through ultrasound-assisted extraction (UAE) method

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ABSTRACT

Sargassum polycystum is well known as macroalgae that contain active compounds with great function as antioxidants. The antioxidants content of the seaweed has links closely to phenolic compounds. The study is aimed to determine active compound quality from different thalli parts of *S. polycystum* extracted by Ultra-sound-assisted extraction (UAE). Fresh samples were prepared into three parts, which are apical, middle, and base thallus. Extraction was carried out by ultrasonication method and using 90% acetone as solvent. Levels of total phenols were analyzed using the Reagent Folin-Ciocalteu. Antioxidant activities were analyzed using DPPH, FRAP, and CUPRAC methods. The highest total phenolic content was found at the apical of the thallus, about 875.64 mg GAE/g. The most excellent DPPH antioxidant activities in *S. polycystum* were found from the apical part of the thallus with an IC 50 value of 38.49 ppm. The FRAP and CUPRAC antioxidant's capacity showed the highest in the apical part of the thallus, which was 989.93 mol Fe (II)/g and 555.52 µmol Trolox/g, respectively. The extraction results of different parts of the thallus show highly potent active compounds of alkaloids, steroids, phenols, flavonoids, and potent antioxidants activity.

Keywords: antioxidants, phytochemical, ultrasonication, S. *polycystum*

ABSTRAK

Sargassum polycystum mengandung senyawa aktif yang berfungsi sebagai antioksidan. Kandungan antioksidan dari rumput laut memiliki keterkaitan erat dengan kandungan senyawa fenolik. Penelitian ini bertujuan menentukan pengaruh perbedaan bagian talus terhadap kandungan senyawa aktif dalam *S. polycystum*. Sampel segar dipreparasi menjadi tiga bagian, yaitu ujung, tengah, dan pangkal. Ekstraksi dilakukan dengan metode ultrasonik dan menggunakan pelarut aseton 90%. Kadar total fenol dianalisis menggunakan pereaksi Folin-Ciocalteu. Aktivitas antioksidan dianalisis menggunakan metode DPPH, FRAP, dan CUPRAC. Total fenolik tertinggi terdapat pada bagian ujung talus sebesar 875.64 mg GAE/g. Aktivitas antioksidan DPPH tertinggi pada *S. polycystum* terdapat pada bagian ujung talus dengan nilai IC_{50} sebesar 38.49 ppm. Kapasitas antioksidan FRAP dan CUPRAC tertinggi terdapat pada bagian ujung talus berturut-turut sebesar 989.93 µmol Fe(II)/g dan 555.52 µmol troloks/g. Ekstrak *S. polycystum* pada ketiga bagian talus terdeteksi mengandung senyawa aktif berupa alkaloid, steroid, fenol, dan flavonoid serta memiliki aktivitas antioksidan yang kuat.

Kata kunci: antioksidan, fenol, *S. polycystum*, senyawa aktif

1. Introduction

Sargassum polycystum is one of the species of brown seaweed (Phaeophyta) whose available widely in tropical waters, including in Indonesian waters. *S. polycystum* generally grows in the subtidal and intertidal zone at a depth of 0. 5-5 m below the surface of the sea and is also located in tidal zones. Its morphology was divided into a holdfast that functions as a basal structure, a stipe or pseudo-stem, and a blade-shaped like a leaf and can grow to a length of 12 m (Widyartini et al., 2017). It is shaped flattened, many branching, leaves wide form,

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serrated and tapered on the part of leaves, there is a point a little black on the leaves, stems main round rather rough, and has a water bladder which serves as a float (Dawes, 2016). According to Zailanie and Sukoso (2014), the *S. polycystum* is characterized by having dominant pigment that carotenoids are a class of β-carotene, fucoxanthin, and xanthophyll that cover the color green of the pigment chlorophyll a and c, the result of photosynthesis. Fucoxanthin of brown seaweed has the potential to be developed as an ingredient nutraceutical, primarily as an antioxidant and an agent chemo-preventive caused by its ability to inhibit radical free (Miyashita et al., 2020).

The active components of brown algae have been widely studied and are related to natural sources of antioxidants. *S. polycystum* already assessed are widely demonstrated antioxidant potency that is high in vitro. Phenolic and phlorotannin compounds are the most effective antioxidant that contained in this organism. Antioxidant activity is positively correlated with total phenolic compound (Pramesti et al., 2019). Phlorotannin compounds is a group of derived phenolic compounds were detected on brown algae *S. polycystum*. Phlorotannin has a very strong antioxidant activity which protect cell membrane proteins from reactive oxygen activity (ROS). It also play a role in UV radiation protection, reproduction, and protective mechanism against other biotic factors (Wijesekara et al., 2011).

Antioxidants are bioactive compounds that can inhibit radical free activity by donating one or more electrons. Free radical is an atom, molecule, or compound with an electron without paired to be reactive and quickly react with other substances (Kelman et al., 2012; Miyashita et al., 2020). The antioxidants capacity of fucoxanthin produced by *Sargassum* seaweed is determined by the quantity of phenolic, flavonoid, and alkaloids as active compounds (Johnson et al., 2019). The fucoxanthin content is also has strong correlation with physiological condition of the seaweed, which vary in quantity by species and affected by season as well as location (Terasaki et al., 2017). Various studies also find that the method of extraction of both conventional and modern with the approach to the extraction of the friendly environment (green extraction methods) affect the quantity of the compound that is obtained (Kadam, Álvarez, et al., 2015; Pangestuti & Siahaan, 2018; Tiwari, 2015).

Various descriptions of the above, the potential of antioxidants from brown seaweed *S. polycystum* is remarkable, but the study of the quantity based on a different part of the thallus is not much to do. The use of green extraction methods, such as Ultrasound-assisted extraction (UEA) also needs to be introduce because of its valuable in time efficiency, environmental-friendly solvent used, and high extraction yield. Therefore, this study aimed to determine the effectiveness of antioxidants produced from different parts of the *S. polycystum* thallus, namely the base, middle, and apical. Thus, more effective, and efficient utilization can be carried out in the future.

2. Materials and Methods

Sample collection and preparation

Samples were taken from the coast of Sebesi Island, South Lampung, in October 2019. A 3 kg sample of seaweed was cleaned with clean sea water to remove unwanted materials such as sand, mud, and silt, then preserved using 95% methanol solution until the sample was submerged in Ziplock plastic. Samples were sent to the laboratory in a cool box with ice, then stored in a freezer at a temperature of -23 °C after all methanol were drained and then wait for further experiment. Identification of seaweed species based on previous research on identification of *S. polycystum* species by Widyartini et al., (2017) and Wong et al., (2004). A photograph of sample and differentiation of thallus parts is shown in Figure 1.

Extraction of bioactive compounds

The extraction process uses the ultrasound-assisted extraction (UAE) method, which refers to the research Kadam et al., (2015) with modified. An 80 g of fresh seaweed (no methanol remain) were put into a baker glass and 400 ml of 90% acetone was added as a solvent. The samples then placed in the Ultrasonic LC60H water bath by setting the temperature of 15 °C and 35 kHz for 60 minutes. Temperature control by adding ice into water bath and kept the temperature about 15 °C. The extraction results then centrifuged at the speed of 4000 rpm for 20 minutes at a temperature of 4 °C. Evaporation of the solution (supernatants) was done under a vacuum evaporator at a temperature of 40 °C for 30 minutes. The crude extract yield was calculated, phytochemical testing, total phenol, and antioxidants assessment. 90% of acetone was found as the most efficient ratio to obtain antioxidant compound (Sudhakar et al., 2013).

Proximate composition

Fresh of *S. polycystum* sample was then analyzed for proximate composition, including analysis moisture content refers to the SNI (1992) which was calculated by gravimetry

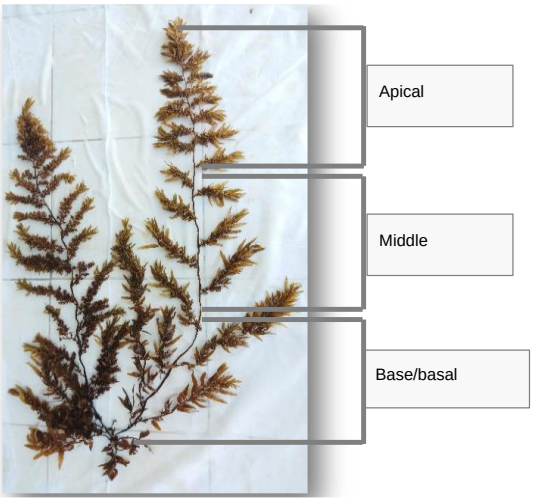

Figure 1. Different part of *S. polycystum* thallus used in this study

method. Analysis of ash content refers to SNI (1992), with calculation based on total ashing result percentage. Fat content Analysis was referring to SNI (2006) and calculated as extracted weight percentage. Protein content analysis refers to the AOAC (2005) which was nitrogen composition multiply by 6.25. Analysis of crude fiber content refers to the AOAC (2005) with H_2SO_4 as solvent continued by destruction using NaOH. The result of crude fiber was calculated based on residual and initial weight percentage. Carbohydrate's content was determined through by-difference method which is Carbohydrates (%) = 100% - (water content + ash + protein + fat + fiber).

The yield of the extraction process

Yield calculated by comparing the weight of dried results extract (pasta) of *S. polycystum* after the process of vacuum evaporation by weight of wet samples, namely through the following formula:

$$Yield = \frac{weight\ of\ dried\ extract}{weight\ of\ wet\ sample}\ X\ 100\%$$

Phytochemical analysis

Qualitative phytochemical tests were carried out to determine the active compounds contained in the sample extract from the apical, middle, and base of the thallus. The phytochemical analysis including alkaloid test, which is based on Dragendorff, Meyer, and Wagner reagent. The second one is steroid test by employed chloroform, acetic anhydrous solution, and H_2SO_4 to produce color indicator. Next is saponin test by using HCl solution as reagent, phenol test is performed under ethanol (PA) and added by $FeCl_3$ 5%, tannin test is using $FeCl_3$, and flavonoid test under magnesium powder, amyl alcohol, and alcohol solutions.

Total Phenolic content

Analysis of total phenols was measured with a spectrophotometer using a reagent Folin-Ciocalteau which refers to the study (Agbor et al., 2014). A total of 1 mL *S. polycystum* sample extract was taken (1:1 w/v) and added 1 mL of 96% ethanol, and 5 mL of distilled water. The extract was added with 0.5 mL of Folin-Ciocalteu 50%, homogenized, and allowed to stand for 5 minutes. The extract was added with 1 mL of 5% Na_2CO_3 and allowed to stand in the dark for 60 minutes. Then the absorption wavelength was read at 725 nm.

DPPH analysis

Methods of analysis of DPPH refer to the study (Molyneux P, 2004), namely activity test of antioxidant carried by the ability of the sample to reduce the radical free 2,2-diphenyl-1-picrylhydrazyl (DPPH). 0.0001 M (Sigma-Aldrich) Reagent DPPH plus ethanol 96%, followed by solving blanks plus DPPH solution of 0.5 mL in 3.5 mL of acetone 90%. Sample solutions with 100, 125, 150, 175, and 200 ppm concentrations were added with 0.5 mL and 90% acetone, then allowed to stand for 30 minutes. The absorbance of the sample reaction was measured at a wavelength of 517 nm using a spectrophotometer. The equation calculates antioxidant activity as follow:

$$\text{DPPH radical-scavenging activity}(\%) = \frac{\text{A blank - A sampel}}{\text{A blank}}\ \text{x}\ 100\%$$

A blank = Absorbance resulting from the reaction of 96% ethanol solutions and DPPH

A sample = Absorbance of sample reaction and DPPH

The concentration and inhibition values of the extract were plotted on the x and y axes,

respectively, in the linear regression equation. Equation of a line obtained in the form y = b (x) + a is used to look for the value of IC (inhibitor concentration), with a stated value of y at 50 and x as the IC 50. Values of IC 50 states concentration solution of the sample required to reduce 50% of radical free DPPH.

FRAP (Ferric Reducing Antioxidant Power) method

This analysis done refers to Benzie & Strain, (1996), which was preceded by the preparation of the FRAP reagent, explicitly the measurement of the amount of Sodium acetate trihydrate, TPTZ, and $FeCl_3.6H_2O$ of 187, 150, and 270 mg, respectively. Absorbance measurements used 0.05 mL of sample, 0.6 mL of distilled water, and 3 mL of FRAP reagent. The sample mixture with FRAP reagent was homogenized with a vortex and then incubated for 30 minutes at 37 C, and the absorbance was measured at a wavelength of 593-595 nm. The Standard that is used is $FeSO_4.7H_2O$ were reconstituted with distilled water. The capacity of antioxidants with the method is expressed in mol Fe (II) g-1 extract.

CUPRAC (cupric ion reducing antioxidant capacity) method

The method used refers to the studies previously by Apak et al., (2004), namely by mixing 50 mL $CuCl_2$ and $2H_2O$ 0.01 M; 50 mL neocuproin (Sigma-Aldrich) 7.5 x 10-3 M; and 50 mL buffer ammonium acetate pH 7. Extract 0.3 mL of reconstituted in ethanol 96%, then added 1 mL of $CuCl_2{\cdot}2H_2O$ 0:01 M, 1 mL neocuproin ethanolic 0.0075 M, 1 mL of buffer ammonium acetate pH 7 1 M, and 0.1 mL of distilled water. The solution was allowed to stand for 30 minutes, and the absorbance was measured at a wavelength of 450 nm. Value absorbance then converted into the activity of antioxidant is expressed in mol Trolox/g. Trolox was used as standard, and number of its concentration show the antioxidant capacity in samples analyzed.

Data analysis

Data retrieval was done through three repetitions on each factor treatment (thallus parts differentiation as follow: the base, middle, and apical). Data were tested for normality (Kolmogorov-Smirnov test and Shapiro-Wilk) and homogeneity. Data that has been tested normality (grades P-value ≥ α (0:05)), then continued using Analysis of Variance (ANOVA) at an interval of confidence of 95%. The design of the experiment is the design of Randomized Complete pattern factorial that are processed using software SPSS24.

3. Results and Discussion

Proximate composition

Proximate analysis of *S. polycystum* in different thallus fresh shows significantly different on every parameter generated (P <0.05). The water content was the most significant component with the highest content in treatment is apical thallus part 85.49%. Furthermore, carbohydrate content was the second-largest component, between 5.58-6.56%, with the apical thallus showing the largest. On the other hand, the ash content was relatively high (4.73-7.52%), with the highest content found at the base of the thallus. Levels of crude fiber and crude protein show the results of the almost identical, which were reaching 1.09- 1.64% and 1.47-1.67%, respectively. However, the different parts of the thallus indicated different highest content: the base part indicated the highest levels of fiber, and the apical showed the highest of protein levels. The minor proximate composition was fat content that reaches only 0.24 to 0.55% with trend composition from the lowest to the highest, namely the base < middle < apical. The analysis of the chemical composition of the thallus of *S. polycystum* is presented in Table 1.

Proximate composition contents generally vary between species and between the same species of seaweed, which is influenced by the condition of the habitat of seaweed habitat, such as sunlight, nutrients, pH, depth, brightness, speed of currents, temperature, and salinity (Bertagnolli et al., 2014; Perumal et al., 2019; Praiboon et al., 2018; Sumandiarsa et al., 2021). In particular, research by Perumal et al., (2019)

Table 1. Chemical composition of *S. polycystum* fresh thallus parts

Parameter (%)	Apical	Middle	Base
Water content (%)	85.49 ± 0.02^a	84.68 ± 0.02^b	83.27 ± 0.02^c
Ash content (%)	4.73 ± 0.03^c	5.43 ± 0.01^b	7.52 ± 0.01^a
Crude protein (%)	1.67 ± 0.03^a	1.53 ± 0.02^b	1.47 ± 0.01^c
Crude fiber (%)	1.09 ± 0.03^c	1.51 ± 0.01^b	1.64 ± 0.02^a
Lipid (%)	0.55 ± 0.01^a	0.34 ± 0.02^b	0.24 ± 0.02^c
Carbohydrates (%)	6.56 ± 0.01^a	6.33 ± 0.01^b	5.58 ± 0.02^c

The different superscripts (a, b, c) in each row show the significant differences (P <0.05).

found that levels of protein and fiber in the thallus seaweed *S. polycystum* differ significantly, with the number of each reach 14.8% and 21.3%, but found from a dry sample.

The content of crude fiber is high on the thallus part of the base can be caused by cellulose content being much dominant, an old tissue, and lots of cells making up the tissue amplifier (Widyartini et al., 2017). The content of cellulose contained in part of the center and at the base of the seaweed is a component of the structural constituent main wall of the cell. According to Srivastava et al., (2017), cellulose can be hydrolyzed using the cellulose enzyme and acts as a cell wall reinforcement in plants. The lipid content in seaweed is generally low or less than 3%, but it is a healthy lipid, such as omega 3 fatty acid. Our study found that *S. polycystum* consist of a comparable lipid with previous research, which about 0.46 % (Muraguri et al., 2016). Wong & Cheung, (2001) stated that seaweed contains very low lipids, so seaweed has food stored and reserves in the form of carbohydrates, especially polysaccharides.

Extraction yield

Extraction was carried out to obtain crude extract at the apical, middle, and base thallus of *S. polycystum*, containing bioactive components. The yield of the obtained at 1.26-1.66% with the most significant derived from parts of the base of the thallus. *S. polycystum* thallus differentiation showed a significant effect ($p<0.05$) on the yield of the resulting extract. There are many factors affected the extraction yield, including solvent type and its ratio. (Hernes et al., 2018) stated that the highest yield obtained from acetone solvent with ratio of 1:5, which reached 2.4% of the same seaweed. Besides, the used of extraction equipment such as ultrasonication, showed a significant effect on extraction time than maceration.

The results obtained were extensive due to Ultra-sound-assisted extraction, which has advantages in improving the yield, fewer solvents, is efficient against time, and can be combined with other extraction technique methods (Kadam, Tiwari, et al., 2015). Mittal et al., (2017) in his research stated that ultrasonic extraction was able to increase extraction efficiency about 69.6 to 93%. The use of ultrasound in the extraction, according to Falleh et al., (2012) will break down the membrane in the walls of cells seaweed and accelerate the diffusion through the membrane so that the solvent can extract more quickly and produce content of relatively high antioxidants. The extract yield of the part of the *S. polycystum* thallus is presented in Table 2.

Phytochemical compounds from *S. polycystum* thallus

Testing phytochemical extracts of *S. polycystum* includes alkaloids, steroids, saponins, phenols, tannins, and flavonoids compounds. Identify the components of the extract compounds active aims to provide information of compounds active with testing phytochemicals that are qualitative (Mehdinezhad et al., 2016). The third extract parts of the thallus on the seaweed have demonstrated their alkaloids compounds in Reagent Dragendorff and Wagner but was not

Table 2. Extract yield of the part of the thallus *S. polycystum* by UEA with acetone

Part of the thallus	Yield (%)
Apical	1.26 ± 0.03^{c}
Middle	1.45 ± 0.02^{b}
Base	1.66 ± 0.01^{a}

The different superscripts (a, b, c) in each row show the significant differences (P <0.05).

Table 3. Phytochemical test results of the extract of the thallus *S. polycystum*

Test	Part of the Extract thallus			Color indication
	Apical	Middle	Base	
Alkaloid				
Dragendorff	+	+	+	Orange precipitate
Wagner	+	+	+	Brown precipitate
Meyer	-	-	-	Yellowish green
Steroid	+	+	+	Green color
Saponin	-	-	-	Green color
Phenol	+	+	+	Black color
Tannins	-	-	-	Yellow color
Flavonoid	+	+	+	Yellowish color

(+) : Detected, (-) : Not detected

detected in the Reagent Meyer. Steroids, phenols, and flavonoids compounds are detected in the thallus's three extract parts. However, from the results of the test phytochemical, saponin, and tannins compounds were not detected positive. The test results of phytochemical extracts from *S. polycystum* different thallus parts are presented in Table 3.

The result of this was comparable with the research Gazali et al., (2018), which stated that extract ethanol of *S. polycystum* does not show the results positive in the compound alkaloid reagents Meyer and the compound of saponins, flavonoids, and tannins. It is caused by the differences in a solvent used to influence the level of polar compounds based on the similarity of solvent polarity. Alkaloids contain at least one or more nitrogen atoms in their essential cyclic chains (Waller et al, 1978). Güven et al., (2010) stated the test compound alkaloid conducted by reacting the extract with a reagent Meyer, Wagner, and Dragendorff. The Meyer, Wagner, and Dragendorff alkaloid reagent test results showed that the sample was detected positive according to Bribi, (2018), white, brown, and orange precipitates were formed. These reagents can prove the potential of a compound as an antioxidant and antibacterial.

The seaweed has been known as a source of bioactive with broad utilization and has become an exciting topic to study in various countries. Natural qualities of phytochemicals from compounds extracted demonstrate a specific ability, so it's useful in the field of nutraceuticals such as antibacterial, anti-fungal, healing wounds, and antioxidant, antiviral, anticancer, and anti-tumor. Capabilities are derived from essential compounds such as phenol, saponins, steroids, phlorotannin, and polysaccharide sulfate such as those found in brown seaweed *Sargassum* (Hermund, 2018; Miyashita et al., 2020).

Total phenolic contents

In general, extracts from the *Sargassum* species have higher total phenolic compound than other brown seaweeds (Nunes et al., 2017). Differences in the extract of *S. polycystum* thalli show a significant different (P <0.05) on the total phenolics content. The highest was found at the apical of 875.64 mg GAE/g, while the lowest was at the base of 817.78 mg GAE/g wet weight samples. The results obtained were very high in comparison with several *Sargassum* species such as *S. miyabei Yendo* (88.97 ± 4.34 mg gallic acid equivalent/g dry weight (dw)) (Baek et al., 2021). Phenolic compounds have linear contributions to the activity of antioxidants, even higher levels of phenolic then antioxidants are produced increasingly powerful (Rice-Evans et al., 1997). Phenolic compounds working through the termination of the chain reaction of radicals and donate atomic hydrogen so radical free which produced a more stable (Nimse & Pal, 2015). According to Baihakki et al., (2015) the extraction of total phenolic compounds from *Sargassum* sp. produce compounds of polyphenols that highest on the type of seaweed *S. polycystum* and potential as a good antioxidant. Besides that, the other active compounds in this type is a phenolic compound in the form of phlorotannin (Agregán et al., 2016). Variation in total phenolic contents were influenced by many factors such as extraction methods that lead different purities of results (Mekinić et al., 2019). The contents of the total phenolic extracted from different parts of the thallus are presented in Figure 2.

It is in line with the results of other studies Abdala-Diaz, (2014), which states that the highest of a total phenolic compound from methanol extract of *Cystoseira tamariscifolia* (Phaeophyceae) contained in apical part, which was 900 mg GAE / g. It was due to young tissue is in that part, as an organ of photosynthesis, and

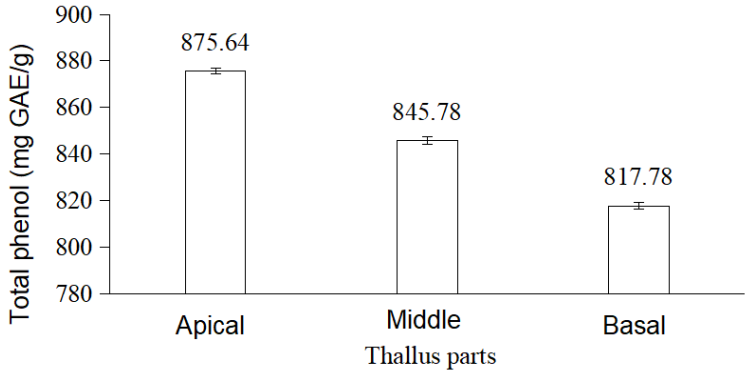

Figure 2. Total phenolic content of the different thallus part of *S. polycystum*. The different superscripts (a, b, c) show the significant differences (P <0.05).

there is an apical meristem tissue as well. The apical meristem tissue functions to produce new cells to accelerate the growth process and the photosynthesis process (Kumar et al., 2004). The apical part of the thallus responds to long-wave radiation Photosynthetically Active Radiation (PAR), a spectrum of radiation used in photosynthesis (Ginneken, 2017). It is concluded that the content of total phenolic extract from different parts of the thallus which are apical, middle, and base *S. polycystum* can be categorized as having the value of the content of total phenols were very strong, which ranged between 817-875 mg GAE / g (Molyneux P, 2004).

Antioxidant activities of *S. polycystum* brown seaweed

The antioxidant activity of the extract from different thallus parts of *S. polycystum* showed significantly different results ($p<0.05$) with the DPPH method. The highest value of IC 50 was at the apical of the thallus, which about 38.49 ppm. In contrast, the lowest of the IC 50 was at the bases part that reached 49.51 ppm. Activities of antioxidants were much weaker than the previous study results, containing of 29.86 ppm from the apical part of *Cystoseira tamariscifolia* macroalgae (Abdala-Diaz, 2014). However, the result was much powerful than studies under Johnson et al., (2019), which was only 183.82 ppm with the acetone solvent in *S. polycystum* and *S. duplicatum*. Based on various part of the thallus, research by Darmawati, (2012) stated that the outer cells are identical to the cells of the thallus tip (apical), wherein it is stated that the apical part consists of assimilator cells or cells that are actively growing. So that the apical is called young tissue which produces new cells for growth and photosynthesis. The antioxidant activities of extract from the different thallus parts of *S. polycystum* with the DPPH method are presented in Figure 3.

The antioxidant activity of DPPH in the percent of its inhibition against DPPH radicals is expressed by the IC 50 value. (Molyneux P, 2004) stated that the IC 50 concentrates on a sample solution required to inhibit 50% of radical free DPPH. A compound is categorized as a powerful antioxidant if the IC50 value is <50 ppm, strong if the IC50 value is 50-100 ppm, moderate if the IC50 value is 101-150 ppm, and weak if the IC50 value is >150 ppm. The DPPH antioxidant test on the thallus of the brown seaweed extract of S. polycystum showed an IC50 value of <50 ppm, ranging from 38-49 ppm. It indicates that the antioxidant produced is powerful, and the use of ultrasonication method can be considered as well as the used of time, temperature and solvent that can optimize the results (Kadam, Tiwari, et al., 2015; Tiwari, 2015).

This study found that there were significant differences between parts of the thallus ($p<0.05$) in form of antioxidant production based on FRAP method. The highest antioxidant capacity of the FRAP method was found at the apical of the thallus *S. polycystum* that reached 989.93 µmol Fe (II)/g. In contrast, the lowest antioxidant capacity value was found at the base of the thallus that only about 929.40 µmol Fe (II)/g (Figure 4). In comparison, research under Diachanty et al., (2017) found that *S. polycystum* grown in Seribu island has low antioxidant under FRAP method which was reached 105.36 µmol Fe (II)/g. It is shown that the finding was not much different from this study. The apical part of the thallus gives the highest value since there is an active role of substances phytohormones, which affect the component active in parts of the apical of the thallus, so that the content of phenolic and flavonoid increased and the activity of antioxidant that produced is getting stronger.

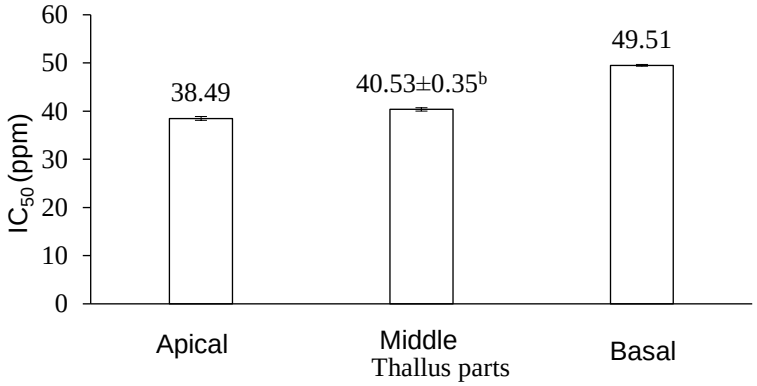

Figure 3. Antioxidant activity of the different thallus part of *S. polycystum d*etermined by the DPPH method. The different superscripts (a, b, c) show the significant differences (P <0.05).

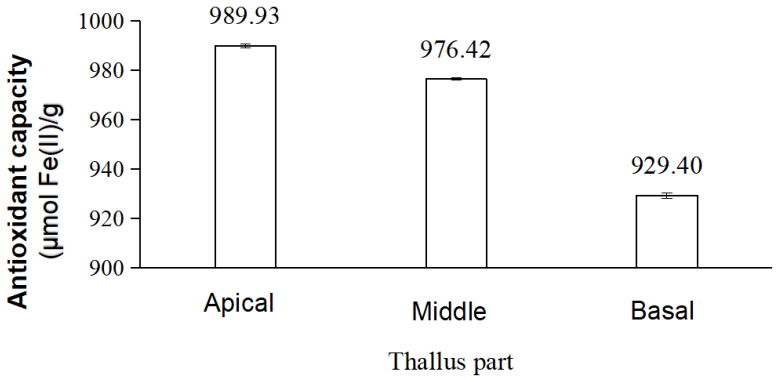

Figure 4. Antioxidant capacity of the different thallus part of *S. polycystum* determined by the FRAP method. The different superscripts (a, b, c) show the significant differences (P <0.05).

This assumption is reinforced by the statement by Abdala-Diaz, (2014) that the apical of the thallus is a young tissue due to getting more sunlight. Photosynthesis and growth are more optimal than other parts, and the active components produced are higher. The positive correlation of phenolic compounds in the tissue and antioxidant activity showed that the thallus extract contained phenolic compounds.

Generally, a material is categorized as a very strong antioxidant if the antioxidant capacity value reaches 500 mol Fe (II)/g. the strong level, if the antioxidant capacity value is about 100-500 mol Fe(II)/g, while the antioxidant capacity value is 10-100 mol Fe (II)/ g, and weak if the antioxidant capacity value is 10 mol Fe(II)/g (Benzie & Strain, 1996). The results of thallus extract at the apical, middle, and base using the UAE method from *S. polycystum* showed antioxidant capacity values between 929-989 mol Fe (II)/g, so it could be categorized as a very strong antioxidant. FRAP analysis results were shown in Figure 4.

Testing under the CUPRAC method on the talus extract of *S. polycystum* had a significant effect ($p<0.05$) on the antioxidant capacity produced. The highest antioxidant value of the test results is located at the end of the thallus 555.52 mol Trolox/g, while the lowest antioxidant value was at the base of 501.27 mol Trolox/g. The results of the CUPRAC antioxidant analysis can be seen in Figure 5. The test results were lower than the extraction of *Ascophyllum nodosum* seaweed which shows the highest antioxidant capacity found at the apical part of 652.10 mol Trolox/g (Apak et al., 2008). CUPRAC analysis results were shown in Figure 5.

The apical part of seaweed is unique. According to Connan et al., (2006), it is dominated by phenolic compounds, ranging from simple but heavy molecular compounds such as phenols, phlorotannin to complex compounds such as tannins and polyphenol derivatives during growth, so that parts that are still in the growth stage contain dominant secondary metabolites and have potential as strong antioxidants. The phenolic compound and

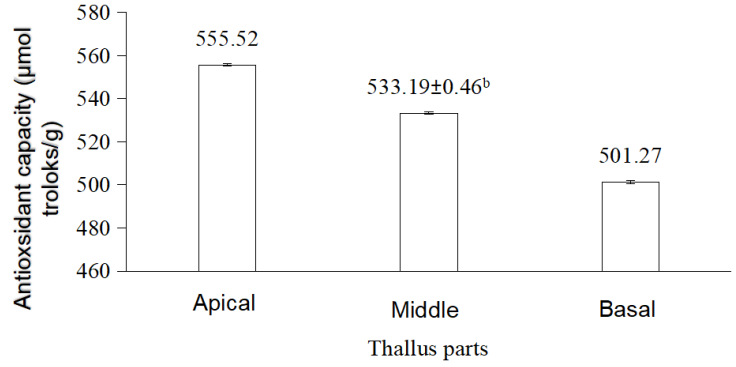

Figure 5. Antioxidant capacity of the different thallus part of *S. polycystum* determined by CUPRAC method. The different superscripts (a, b, c) show the significant differences (P <0.05).

derivatives are believed to be a component of the main compound of antioxidants produced by seaweed species of Phaeophyceae (Agregán et al., 2016). The active compounds that accumulate on every part of the thallus is dominated by parts of pigment of photosynthesis. Molecular components that respond to light are located in chloroplasts and thylakoids, such as photosynthetic pigments including chlorophyll a, d, phycocyanin, phycoerythrin, and carotenoids (Chen et al., 2019). Increasing the rate of photosynthesis with the absorption of nutrients and carbon from the environment until it reaches a constant photosynthesis rate can affect quantitative growth. Its effects include the increase in biomass used by phytoplankton and marine plants in the formation of phycocolloid compounds, pigments, and other active compounds (Masojídek et al., 2013). In general, the study found that *S. polycystum* has prominent potential to be utilized as sources of food ingredient and also bioactive due to its high antioxidant activities.

4. Conclusion

Different thalli parts of *S. polycystum* exert a significant effect on proximate composition, extraction yield under UEA method, total phenolic compounds, and antioxidant activities according to DPPH, FRAP, and CUPRAC methods. This study also detected the contain of alkaloids, steroids, saponins, phenols, tannins, and flavonoids. Future use may consider the UAE as an extraction tool and the seaweed thalli parts also needs to be considered in obtaining extract and the highest bioactive compound.

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