Study of *Actinotrichia fragilis* Indonesian Red Seaweed as Raw Material for Healthy Salt

Nurjanah Nurjanah[1*], Agoes Mardiono Jacoeb[1], Asadatun Abdullah[1], Joko Ahadi Priyanto[1], Naufal Muharam Nurdin[2], and Anggrei Viona Seulalae[1]

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[1] Aquatic Products Technology Department, Faculty of Fisheries and Marine Science, Bogor Agricultural University (IPB University), Bogor 16680, Indonesia

[2] Nutrition Science Department, Faculty of Human Ecology, Bogor Agricultural University (IPB University), Bogor 16680, Indonesia

* Corresponding Author:
nurjanah@apps.ipb.ac.id

Received: 6 March 2023

Accepted: 23 May 2023

Published: 31 May 2023

Academic Editor: Dr. Endar Marraskuranto

 Accreditation Number:148/M/KPT/2020. ISSN: 2089-5690, e-ISSN: 2406-9272. https://doi.org/ 10.15578/squalen.753

Abstract

Healthy seaweed salt is low sodium salt from seaweed that offers health benefits for hypertension patients. Indonesian seaweed has the potential to produce healthy seaweed salt. Research to date still focuses on green and brown seaweed but there is still no report for red seaweed. *Actinotrichia fragilis* is one of red seaweed species that has been discovered in Indonesia's seawater and has not yet been utilized. Thus, this study aimed to determine the chemical composition and antioxidant activity of *A. fragilis* flour and the optimum ratio for producing seaweed salt with a high yield, optimum %NaCl, Na/K ratio, and antioxidant activity. Seaweed salt production treatment was the ratio of seaweed flour and distilled water 1:3, 1:5, and 1:10 (w/v), extracted at 40°C for 10 minutes. The mixture was filtered, then dried at 60°C for 30 hours. Data analysis was performed by analysis of variance. The raw material for dried *A. fragilis* seaweed has a high ash and low-lipid content. Then the ethanol extract had a total phenolic content value of 84.34 mg GAE/g and an antioxidant activity value of 98.22 mg/L. Furthermore, the antioxidant capacity of the ethanol extract was 60.15 μmol ascorbic acid/g and 552.21 μmol Fe^{2+}/g. The best treatment for producing *A. fragilis* salt is 1:10 with yield of 12.76±0.13%, %NaCl 47.22±1.38%, Na/K ratio 3.32±0.18, IC_{50} with DPPH and ABTS method 113 mg/L and 87.27 mg/L, total antioxidant capacity 38.21 μg/mL ascorbic acid/g, and 304.32 μmol Fe^{2+}/g. Furthermore, *A. fragilis* can be used for the production of healthy seaweed salt.

Keywords: Antioxidant, hypertensive, low sodium, red seaweed, sea salt

Introduction

Seaweed has the potential as raw material for the production of low-sodium salt (Magnusson et al., 2016). Seaweed contains bioactive compounds, for example, phenolic compounds, natural pigments, polysaccharides, fiber, and other bioactive compounds that have been studied for antioxidants (Diachanty et al., 2017; Nufus et al., 2017; Nurjanah et al., 2019; Nurjanah et al., 2020c). On the other hand, seaweed also contains macro minerals (Na, K, Ca, Mg) and micro minerals (Zn, Fe) to coordinate physiological functions for health maintenance (Manteu et al., 2018). Healthy seaweed salt study in Indonesia has been done using green seaweed (Nurjanah et al., 2018b; Nurjanah et al., 2020a; Kurniawan et al., 2019) and brown seaweed (Nurjanah et al., 2020b; Nurjanah et al., 2022a; Manteu et al., 2021; Nurjanah et al., 2021a; Nurjanah et al., 2021b; Nurjanah et al., 2022b). Previous research showed that green and brown seaweed salt has a low yield, 10-27% for green seaweed and 20-26% for brown seaweed, antioxidant activity with IC_{50} value was classified as moderate 110.49±19.42 mg/L (green seaweed) and 144.41 mg/L (brown seaweed) then antioxidant capacity 92.50±0.35 μmol Fe^{2+}/g (green seaweed) and 27.65±4.30 μg/mL ascorbic acid/g (brown seaweed) (Nurjanah et al., 2018; Nurjanah et al., 2021b). There is still a limited sources to know the best type of seaweed for seaweed salt production with good quantity and quality. Therefore, identifying characteristics of seaweed salt from other types (red seaweed) is necessary.

Red seaweed in Indonesia generally has been known as the main economically important seaweed for export commodities such as *Kappaphycus alvarezii*, *Eucheuma spinosum*, *Gracilaria* sp., and *Gelidium* sp.,

whereas in Indonesia today has been identified 911 species of seaweed but only a few species have been explored and known for their potential. *Actinotrichia fragilis* is one of species from red seaweed that has been discovered in Indonesia seawater such as Seribu Island, Badi Island, Lombok, and Malang (Wiriyadamrikul et al., 2013). *Actinotrichia fragilis* is less well-known and has not yet been utilized. *Actinotrichia fragilis* contains high mineral Ca, Cu, Fe, Mg, and Ni can be considered as potential health-promoting resources for human consumption (Rondevaldova et al., 2023). On the other hand, this seaweed also can be a good source of natural antioxidants (Rajakumaran, 2019). The potential of this seaweed can be used as raw materials for producing seaweed salt, and it can expand its added value. Research on seaweed salt from red seaweed has never been reported. Therefore, the objectives of this study were to determine the chemical composition and antioxidant activity of *A. fragilis* flour and the optimum ratio of seaweed flour and distilled water for producing seaweed salt with a high yield, optimum %NaCl, Na/K ratio, and antioxidant activity.

Materials and Methods

Materials

The raw material used was *Actinotrichia fragilis* red seaweed collected from Air Island, Kepulauan Seribu, Jakarta, Indonesia on March 2019. The materials used purified water, Kjeldahl tablet (Merck), ethanol 70% (Bratachem), L-ascorbic acid (Merck), 2,4,6- Tri(2-pyridyl)-s-triazine (TPTZ) (Sigma Aldrich), neocuproine (Sigma Aldrich), Folin-Ciocalteu reagent (Merck), gallic acid (Merck), (±)-6-hydroxy-2,5,7,8-tetramethylchromane-2-carboxylic acid (Trolox) (Merck), copper II chloride dihydrate (Sigma Aldrich), acetic acid (Merck), sodium acetate (Merck), potassium chromate (Merck), and 1,1-diphenyl-2-picrylhydrazyl (DPPH) (Merck). The tools used were a chopper blender (Miyako CH-501, China), water bath shaker (WSB 18L. South Korea), UV-VIS spectrophotometer (UV-2500, Japan), dehydrator (B-One Ov-60), and atomic absorption spectroscopy (AAS) (Shimadzu, Japan).

Preparation of Raw Materials

The preparation of *A. fragilis* seaweed was conducted according to Nurjanah et al. (2018b). The first stage was the preparation of the seaweed at the harvesting place in Air Island, Kepulauan Seribu. The raw material was washed with sea water repeatedly until it was clean from sand and foreign matter. The cleaned seaweed was then placed in a container, drained, and wind-dried for 24 hours at 25-30°C. Furthermore, seaweed was saved in a cool box and taken to the laboratory. In the second stage, in the laboratory, seaweed was then placed again in a container and wind-dried for 3-5 days at 25-30°C. The dried seaweed (moisture content <10%) was cut into small pieces (1-5 cm) and crushed using a blender for around 1 minute until smooth as flour and sieved with a size of 30 mesh. Seaweed flour was analyzed for proximate and heavy metal content (Association of Official Analytical Chemists [AOAC], 2005).

Extraction of *A. fragilis* Flour

The maceration extraction process was carried out according to Diachanty et al. (2017). As many as 50 g of *A. fragilis* flour was extracted using 300 mL ethanol 70% (v/v) for 24 hours. The homogenate was filtered using filter paper 500 microns and then filtered again using filter paper Whatman Number 42. Furthermore, the filtrate was evaporated using a rotary vacuum evaporator at 50°C for 12 hours to obtain the sample in extract form. The extract was analyzed for total phenolic content (Apostolidis & Lee, 2010) and antioxidant activity with the DPPH method (Salazar-Aranda et al., 2011), Cupric Reducing Antioxidant Capacity (CUPRAC) method (Apak et al., 2007), and Ferric Reducing Antioxidant Power (FRAP) (Benzie & Strain, 1996).

Production of Seaweed *A. fragilis* Salt

Actinotrichia fragilis salt production was carried out according to Magnusson et al. (2016) with modification. The modifications made are the stirring duration during the heating process. *Actinotrichia fragilis* flour 100 g soaked with purified water with different ratios w/v (1:3), (1:5), and (1:10) and heated at 40°C for 10 minutes stirred, using a water bath shaker. The mixture was filtered using calico fabric 85 mesh then filtered again using paper Whatman Number 42, then the filtrate was dried in a dehydrator at 60°C for 30 hours to obtain the seaweed salt. Seaweed salt was analyzed for yield percentage, mineral content (AOAC, 2005), Na/K ratio, %NaCl (Day & Underwood, 1989), total phenolic content (Apostolidis & Lee, 2010), antioxidant activity with the DPPH method (Salazar-Aranda et al., 2011), 2,2-Azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) diammonium salt (ABTS) method (Nantitanon et al., 2007), Cupric Reducing Antioxidant Capacity (CUPRAC) method (Apak et al., 2007), and Ferric Reducing Antioxidant Power (FRAP) (Benzie & Strain, 1996).

Statistical Analysis

The experimental design used a Completely Randomized Design (CRD). Analysis of the data was performed using analysis of variance (ANOVA) with a 95% confidence interval ($\alpha = 0.05$). The significant results were further tested by Duncan's Multiple Range Test (DMRT). Data were processed with Minitab 2016 application.

Results and Discussion

Characteristics of *A. fragilis* Flour

The chemical composition of *A. fragilis* is presented in Table 1. Seven components such as moisture, ash, lipid, protein, crude fiber, and carbohydrate were estimated. *Actinotrichia fragilis* has a high ash content and low lipid content. The moisture content of *A. fragilis* flour is already below the maximum limit of moisture content in dried seaweed (no more than 30%) based on the National Standardization Agency of Indonesia (BSN Indonesia)

Table 1. Chemical composition of *A. fragilis* flour

Parameter	Value (%, dw)
Moisture	9.23±0.01
Ash	61.96±0.27
Lipid	0.46±0.06
Protein	4.23±0.07
Crude fiber	2.20±0.07
Carbohydrate (*by different*)	24.17±0.41

in 2015. Moisture content is related to product damage and quality indicators considered in dried seaweed (Fateha et al., 2019). The ash content of *A. fragilis* from Air Island Indonesia (61.96% dw) was higher than other red seaweeds such *Graciliaria gracilis* from Barru Waters Indonesia (6.78% dw) (Rasyid et al., 2019), *Eucheuma spinosum* from Nusa Penida, Sumenep, and Takalar Indonesia (23.34-24.46% dw) (Diharmi et al., 2019). High ash content is related to macro and micro minerals content in the sample. The seaweed holdfast absorbs excessive micro and macro minerals, which cause high ash content (Diachanty et al., 2017). High ash content was influenced by the habitat of the seaweed like rock and sand. *Actinotrichia fragilis* is one of the species from red seaweed that is laid in the bottom waters with the coral substrate, sand, and weathered rock (Wiriyadamrikul et al., 2013).

The lipid content of *A. fragilis* from Air Island Indonesia was lower than other chemical content. Alvarez-Vinas et al. (2019) stated red seaweed *G. corticate* from Spain has low lipid content (1.41%). However, it is rich in PUFAs (65.6% of total fatty acids). The protein content of *A. fragilis* from Air Island Indonesia was lower (4.23±0.07% dw) than other red seaweed species such as *G. gracilis* from Barru Waters, Indonesia (10.86% dw) (Rasyid et al., 2019) and *E. spinosum* from Nusa Penida, Sumenep, and Takalar, Indonesia (6.04-7.33% dw) (Diharmi et al., 2019). Fleurence et al. (2017) demonstrated the protein content of macroalgae differs according to the species and seasonal cycle. Brown seaweed has high protein content (maximum 47% dw), green seaweed moderate (9-16% dw), and red seaweed low (3-15% dw). The fiber content of *A. fragilis* from Air Island Indonesia was lower (2.20±0.07% dw) than other red seaweed such as *G. gracilis* from Barru Waters, Indonesia (27.48% dw) (Rasyid et al., 2019) and *E. spinosum* from Nusa Penida, Sumenep, and Takalar, Indonesia (15.12-19.89% dw) (Diharmi et al., 2019). *Actinotrichia fragilis* from Air Island Indonesia has lower carbohydrates content than other red seaweed such as *G. gracilis* from Barru Waters, Indonesia (63.13% dw) (Rasyid et al., 2019) and *E. spinosum* from Nusa Penida, Sumenep, and Takalar, Indonesia (69.07-69.66% dw) (Diharmi et al., 2019).

The heavy metal analysis was carried out to determine the contamination of heavy metals in the aquatic ecosystem such as seaweed. The heavy metal content of *A. fragilis* is presented in Table 2. The detected heavy metal Cadmium (Cd), Mercury (Hg), Lead (Pb), and Arsenic (As) are relatively low if compared to the standard based on BSN Indonesia in 2015 and Indonesia Food and Drug Administration (BPOM Indonesia) in 2022. The heavy metal that accumulated in *A. fragilis* was thought to originate from the water column because all parts of *A. fragilis* seaweed are in water and stick to the substrate. Source

Table 2. Heavy metal content of *A. fragilis*

Heavy metal	Value (mg/kg)	Dry seaweed[1]	Food[2]
Cd	<0.004	Max. 0.10	Max. 0.05
Hg	<0.002	Max. 0.50	Max. 0.03
Pb	<0.004	Max. 0.30	Max. 0.20
As	<0.002	Max. 1.00	Max. 1.00

Note: [1](BSN Indonesia, 2015); [2](BPOM Indonesia, 2022).

of heavy metal pollution has emerged due to human activity, especially metal industries, waste dumps, excretion, livestock and chicken manure, use of pesticides, insecticides, and more that flow to an aquatic environment and it is dangerous to human health if accumulated to the body (Briffa et al., 2020).

Total Phenolic Content (TPC) and Antioxidant Activity of *A. fragilis* Ethanol Extract

TPC is one of the important analyses to prove there is an antioxidant compound in the sample. Phenolic compounds play a role for inhibit oxidation reactions (Sofiana et al., 2020). TPC of *A. fragilis* ethanol extract in this study was 84.34±0.57 mg GAE/g extract. This result is relatively high compared to *E. spinosum* ethanol extract (16.47±0.14 mg GAE/g extract) (Sofiana et al., 2020) but relatively low compared to *S. polycystum* ethanol extract with TPC 713 mg GAE/g extract (Manteu et al., 2018). The phenolic compound will increase following the polarity of the solvent (Sofiana et al., 2020; Diachanty et al., 2017).

The antioxidant activity of *A. fragilis* was examined based on three methods such as free radicals inhibition (IC_{50}) (DPPH), antioxidant capacity with CUPRAC, and FRAP. The IC_{50} value of *A. fragilis* ethanol extract with the DPPH method was 98.22±1.04 mg/L. This result indicates the sample has a strong antioxidant activity for inhibiting 50% of free radicals. The IC_{50} value is classified as very strong if the IC_{50} is less than 50 mg/L, strong if the IC_{50} is 50-100 mg/L, moderate if the IC_{50} is 100-150 mg/L, and weak if the IC_{50} is 150-200 mg/L (Molyneux, 2004). The principle of this assay is the ability of antioxidant compounds in a sample to bind free radicals and donor the hydrogen atom (Abdullah et al., 2020). The IC_{50} values of some seaweed extracts have strong antioxidant activity, for instance, brown seaweed *Sargassum polycystum* ethanol extract (77.58±0.27 mg/L) and *Padina minor* ethanol extract (68.38±0.62 mg/L) (Manteu et al., 2018), green seaweed *Caulerpa lentilifera* ethanol extract (47.61 mg/L), *Ulva lactuca* ethanol extract (43.53 mg/L), and *Halimeda opuntia* ethanol extract (95.91 mg/L) (Nufus et al., 2017), and red seaweed *G. gracilis* methanol extract (59.01 mg/L) (Panjaitan & Yuliana, 2022) and *E. cottonii* methanol extract (39.93 mg/L) (Wulandari et al., 2018).

The total antioxidant capacity of *A. fragilis* ethanol extract with CUPRAC method (60.15±0.12 μmol ascorbic acid/g extract) and FRAP method (552.21±1.05 μmol Fe^{2+}/g extract. The higher the total antioxidant capacity the higher the antioxidant activity of the extracts. The antioxidant activity of extracts will increase with the increasing concentration of extracts and exhibit maximum activity at 1 mg/mL (Lakshmanan & Padmanabhan, 2022). The CUPRAC method principle is the ability of antioxidant compounds to reduce Cu^{2+}Nc metal to Cu^{+}Nc. This electron donor reaction is characterized by a color change from turquoise color to yellow color in a solid reaction. The FRAP method principle is the ability of antioxidant compounds to reduce Fe^{3+} ions to Fe^{2+}. Iron is one of the pro-oxidant compounds in the body and it has the ability for oxidizing other compounds and create other free radicals (Abdullah et al., 2020).

Characteristics of *A. fragilis* salt

The yield of *A. fragilis* salt treated with different ratios of soaked 1:3, 1:5, and 1:10 was 7.47±0.06%, 9.36±0.12%, and 12.76±0.13%, respectively. The highest yield was 1:10 and the lowest was 1:3. This result indicated the high ratio of sample flour and purified water will increase the yield of seaweed salt. The yield of *A. fragilis* salt is relatively low compared to green seaweed (10-27%) (Nurjanah et al., 2018b; Kurniawan et al., 2019) and brown seaweed (20-26%) (Nurjanah et al., 2020b; Nurjanah et al., 2022a). Yield percentage may be influenced by several factors, such as different species, the chemical composition of raw material, and the mass lost during the filtering process (Manteu et al., 2021).

Macro and micro minerals have an important function in the physiological mechanism of the human body. A total of five mineral elements were examined in *A. fragilis* salt, such as sodium (Na), potassium (K), magnesium (Mg), calcium (Ca), and iron (Fe) (Table 3). The results show that *A. fragilis* salt has high Na content and low Fe content. Notowidjojo et al. (2021) evaluated *E. cottonii* salt that was mixed with ordinary salt has high K content (42,091.56 mg/kg), and Mg content (1,622.87 mg/kg). *Sargassum polycystum* salt and *U. lactuca* salt have high K content (200.33±0.13 mg/g; 56.21±1.44 mg/g) and low Fe content (0.07±0.00 mg/g; 0.04±0.01 mg/g) (Nurjanah et al., 2022a; Nurjanah et al., 2020a). The treatment with ratios of soaked 1:3, 1:5, and 1:10 affects ($p<0.05$) mineral content of seaweed salts. From Table 4, high mineral content was obtained when the ratio of sample flour and purified water increased. The mineral content in seaweed salt may be influenced by different environmental conditions of raw material, the age of raw material when harvested, and the absorption of minerals by seaweed in seawater (Misurcova, 2011). Furthermore, the treatment combination for producing seaweed salt such as mixing with ordinary salt (Notowidjojo et al. 2021), mixing with the residue of seaweed salt (Nurjanah et al., 2021a), mixing with other natural material (Nurjanah et al., 2022a) is also influencing the mineral content of seaweed salt.

The ratio of Na/K and %NaCl results are presented in Table 3. The treatment with the ratio of soaked 1:10 has a Na/K ratio close to 1 and the %NaCl is lower than other treatments (1:3 and 1:5). The treatment with the ratio of soaked 1:3, 1:5, and 1:10 affects ($p<0.05$) Na/K ratio and %NaCl content of seaweed salts. The Na/K ratio and %NaCl decreased with a high ratio of soaked. The results show Na/K ratio of *A. fragilis* salt is still high compared to *S. polycystum* salt (0.50) (Nurjanah et al., 2022a), *T. conoides* salt (0.81) (Nurjanah et al., 2020b) and *U. lactuca* salt (1.49) (Nurjanah et al., 2020a). The seaweed species affects the Na/K ratio. Red seaweed is known as a producer of carrageenan. Carrageenan is a hydrocolloid compound that absorbs water consisting of potassium, sodium, magnesium, and calcium sulfate esters with galactose and 3.6 anhydrogalactose (Yasita & Rachmawati, 2009). Positively charged mineral ions (Na^+, Ca^{2+}, Mg^{2+}, and K^+) will bind with carrageenan to produce high ash and are insoluble, therefore the minerals obtained from the seaweed salt production process are not optimal. World Health Organization in 2012 recommended the Na/K ratio approximately one to one, which is considered beneficial for health. A low Na/K ratio and high K content in salt are considered to provide health benefits for consumers (Nurjanah et al., 2020b). The Na/K ratio of 0.60-0.75 is proven to reduce blood pressure of youth aged 17-21 years (Buendia et al., 2015).

The NaCl percentage of *A. fragilis* salt is within the standard of BSN Indonesia in 2016 which is required to be a diet salt (under 60%). This result is appropriate to the NaCl percentage from other seaweed salts such as green seaweed salt from *U. lactuca* (23.90%) (Nurjanah et al., 2020a), brown seaweed salt from *T. conoides* (46.21%), and *S. polycystum* (35.37%). This result indicates, so far, that all the seaweed salts meet the diet salt standard. The NaCl consumption must be limited for diet because it can exceed the NaCl requirement in the body related to high blood pressure and other cardiovascular diseases (Vaudin et al., 2021).

Table 3. Mineral content, Na/K ratio, and %NaCl value of *A. fragilis* salt

Mineral	Ratio 1:3	Ratio 1:5	Ratio 1:10
Na (mg/g)	15.45±0.80^a	14.41±0.53^b	12.03±0.40^c
Ca (mg/g)	2.39±0.31^a	2.47±0.22^a	2.60±0.29^a
Fe (mg/g)	0.08±0.00^a	0.08±0.00^b	0.09±0.00^b
Mg (mg/g)	2.09±0.03^a	2.32±0.05^a	2.34±0.11^b
K (mg/g)	3.64±0.77^a	3.64±0.20^a	4.48±0.49^{ab}
Ratio Na/K	4.25±0.17^a	3.97±0.17^b	3.32±0.18^c
%NaCl	51.16±0.55^a	50.58±0.89^b	47.22±1.38^c

Noted: Superscript font different groups showed a significant difference ($p<0.05$).

Total Phenolic Content and Antioxidant Activity of *A. fragilis* salt

Phenolic compounds are secondary metabolites from natural plants and it contributes to health promotion, for instance as antioxidants (Sethi et al., 2020). The total phenolic content of *A. fragilis* salts is presented in Table 4. The treatment with the ratio of soaked 1:3, 1:5, and 1:10 affects ($p<0.05$) the total phenolic content of seaweed salts. The ratio of 1:10 has the highest total phenolic content than other treatments. This result indicates the higher the soaked ratio the total phenolic content is increasing. The solvent polarity has an important role in increasing phenolic compounds solubility (Rahardhian et al., 2019). The total phenolic content of this study was higher than green seaweed and lower than brown seaweed. A previous study showed the total phenolic content of seaweed salt from *S. polycystum* is 251.88±2.26 mg GAE/g and seaweed salt from a combination of *U. lactuca* and active carbon active is13.72±0.19 mg GAE/ g (Nurjanah *et al.*, 2021b; Kurniawan et al., 2019).

The antioxidant activity of *A. fragilis* salt is presented in Table 4. The treatment with the ratio of soaked 1:3, 1:5, and 1:10 affects ($p<0.05$) the antioxidant activity of seaweed salts. The ratio 1:10 has the lowest IC_{50} value and the highest total antioxidant capacity than other treatments. The antioxidant activity is highly correlated with total phenolic content (Lakshmanan & Padmanabhan, 2022). The total phenolic content of the ratio 1:10 is higher than other treatments.

In the present study, *A. fragilis* salt has moderate (DPPH method) and strong antioxidant activity (ABTS method) for inhibiting 50% of free radicals. It must be underlined that the antioxidant activity of samples determined by different antioxidant assays could give different results (Sethi et al., 2020). The smaller IC_{50} value indicates the inhibition of antioxidants is getting stronger, and vice versa (Molyneux, 2004). Based on IC_{50} values, red seaweed salt has a good potential as a source of antioxidants compared to brown and green seaweed salts. A previous study showed that the IC_{50} value of *S. polycystum* salt (159.313 mg/L) (Nurjanah et al., 2021b) and *U. lactuca* salt (93.24 mg/L) (Nurjanah et al., 2018b). The principle of ABTS assay is the ability of antioxidant compounds to stabilize the radical free by the radical proton donor, on the other hand, the principle of DPPH assay is the ability of antioxidant compounds in a sample to bind free radicals and donor the hydrogen atom (Abdullah et al., 2020).

Table 4. Antioxidant activities and total phenolic content (TPC) of *A. fragilis* salt

Seaweed salt treatment	IC50 DPPH method (mg/L)	IC50 ABTS method (mg/L)	CUPRAC (µmol ascorbic acid/g seaweed salt)	FRAP (µmol Fe2+/g seaweed salt)	Total phenolic content (mg GAE/g seaweed salt)
Ratio 1:3	184.52±2.06[a]	120.27±1.07[a]	16.51±0.16[a]	125.37±1.05[a]	11.01±0.87[a]
Ratio 1:5	175.06±2.34[b]	95.67±0.95[b]	18.29±0.36[b]	195.19±0.06[b]	14.25±0.87[b]
Ratio 1:10	113.00±4.17[c]	87.27±0.56[c]	38.21±0.29[c]	304.32±0.91[c]	33.87±0.87[c]

Noted: Superscript font different groups showed a significant difference (p<0.05).

The results of the total antioxidant capacity of *A. fragilis* salt with CUPRAC and FRAP methods show that the ratio 1:10 has the highest value from other treatments (1:3 and 1:10). The higher total antioxidant capacity value indicates a stronger antioxidant activity of the sample (Apak et al., 2007). The FRAP and CUPRAC results are influenced by the presence of flavonoid compounds in the samples that can help the metal ions reduction process such as iron and copper (Benavente et al., 1997). *Actinotrichia fragilis* has total flavonoid content of 3.86±1.02 mg/g of extract (Abd El Hafez et al., 2022). The CUPRAC assay is highly efficient for glutathione and thiol types of antioxidants which cannot be detected in FRAP method (Apak et al., 2005). The FRAP assay has a role as primary and secondary antioxidant from reduce the oxidized intermediate of the lipid peroxidation process (Matanjum et al., 2008). The antioxidant activity and total phenolic content of *A. fragilis* are presented in Table 4. µmol ascorbic acid/g seaweed salt.

Conclusion

The raw material for dried *A. fragilis* seaweed has a high ash content (61.96±0.27%) and low lipid content (0.46±0.06%). Then the ethanol extract of *A. fragilis* had a total phenolic content (TPC) value of 84.34 mg GAE/g extract and an IC_{50} value of 98.22 mg/L. Furthermore, the antioxidant capacity of the ethanol extract *A. fragilis* was 60.15 µmol ascorbic acid/g extract and 552.21 µmol Fe^{2+}/g extract. The best treatment for producing *A. fragilis* salt is 1:10 (w/v) with a yield of 12.76±0.13%, %NaCl 47.22±1.38%, Na/K ratio of 3.32±0.18, and good antioxidant activity with DPPH, ABTS, FRAP, and CUPRAC method. Seaweed *A. fragilis* can be used as raw material for the production of healthy seaweed salt but still need further study to increase the yield percentage with a lower Na/K ratio.

Acknowledgments

The authors express gratitude to the Directorate General of Higher Education, Research, and Technology Indonesia Ministry of Higher Education and Technology for funding this study through the Research Grant PTUPT 1/E1.KP.PTNBH/2021.

Supplementary Materials

Supplementary materials is not available for this article.

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