Research Article

Determination of bioavailability of Cr, Co, I, Ni, Fe, Se, As, and Pb in *Ulva reticulate* Forskall

Maria Rosabel B. Castillo[1], Karina Milagros R. Cui-Lim[1,2*]

[1]*Department of Physical Sciences, College of Science, University of Eastern Philippines, University Town, Northern Samar, Philippines 6400*

[2]*University Research and Development Services, University of Eastern Philippines, University Town, Northern Samar, Philippines 6400*

Received: 8 November 2018 Revised: 21 December 2018 Accepted: 24 December 2018

Abstract

Objective: This study aimed to determine the bioavailability of metals in *Ulva reticulate* Forskall from seaweeds extracts. **Material and methods:** *Ulva reticulata* Forskall *is* a macroalga that was collected from the sea waters of Gamay, Northern Samar. The *U. reticulata* was subjected for physical properties determination; the boiling point, color, odor, density, pH, and solubility. Nutritional values and chemical property of *U. reticulata* undergo several test using gravimetric methods and modern instrumentations. Ash content, carbohydrates content, fat content, moisture content, vitamin C content, and protein content was determined. Also it undergo test for the presence of Cr, Co, I, Fe, Se, Pb, Ni, and As to determine if seaweed is advisable for food consumption or not. Despite of having nutritional value it is important to know the possible contaminants present on it. **Results and conclusion:** All results was done experimentally and obtained the needed data. The results showed that *U. reticulata* is a potential food source needed by the body. Iodine was abundant in the sample followed by Cobalt. Nickel and Lead was detected but below the detection limit.

Keywords: *Ulva reticulata* forskall, bioavailability, macroalga, nutritional values

Introduction

Macro algae are widely distributed in the vast water mass of the earth. These macro algae or we commonly known as seaweeds or sea grasses have various types and species and this multitude of seaweeds contribute also to its countless uses in different industries. Seaweeds are economically important because it is nowadays used in different industries like pharmaceuticals, foods, cosmetics, biofuel, and agriculture and can further be developed and potentially can replace sophisticated and hazardous materials in the near future. Nowadays, people are more careful of what to intake, they are encouraged to choose healthy foods and supplements that their body needs to prevent diseases. Seaweeds are well known for its nutritional value for human dietary requirement and people were consuming it for such purpose. Because of its higher bioaccumulation of essential elements such as Co, Cr, Mo, Ni, Se, V, Mg, Ca, I, and Fe than that of land vegetation, seaweeds are more likely to produce these trace elements in a greater number than other plants in the land. Macro algae acquired metals greatly than other marine life. Other than the essential elements, seaweeds also can accumulate non-essential elements that can be dangerous to one's health if the concentration exceeds its limits and the heavy metals bioavailability ratio from the seaweeds (Abreu, 2015).

Northern Samar have studied floristically different sea creatures including numerous varieties and species of seaweeds but few have studied about its characteristics chemically and its uses beyond what is seen by the naked eye. As the population nowadays grows increasingly in number, the needs of new resources that can accumulate great amount of these important elements also increase.

Address for Corresponding Author:
Karina Milagros R. Cui-Lim
University Research and Development Services,
University of Eastern Philippines, University Town, Northern Samar, Philippines 6400
Email: karina_cui@yahoo.com

DOI: https://doi.org/10.31024/apj.2018.3.6.3

*Ulva reticulata (kulapo) i*s *u*sed in this study for the determination of bioavailability of heavy metals and its nutritional content. *Ulva reticulata* as it is abundant in Gamay, Northern Samar are often neglected by the people and considered merely as not valuable. This study intends to help enhance people's knowledge regarding the edible seaweeds and the essential elements acquired from them that can help increase body functioning and improve humans' health (Cui, et. al, 2017).

Material and methods

Plant collection and Preparation

The *Ulva reticulata* was collected in the Nabunglayan Rock Formation, Gamay, Northern Samar. The sample was washed with seawater and placed in plastic bags and put on an ice box and transported to the laboratory, where the tests were conducted. At the laboratory, samples were washed with sea water to remove sands and other impurities. Then the sample was washed with tap water and rinsed with distilled water and letting the sample dried.

Seaweed extraction

Freshly picked *Ulva reticulata* was washed with tap water and rinsed with distilled water to remove the dirt and other impurities, after, it was dried and extracted using a traditional extraction machine.

Physical properties determination

For *Boiling point,* about 5ml of sample of *Ulva reticulata* extract was poured into a test tube. The test tube is submerged in an oil bath and the temperature was recorded when the sample extract started to boil. It was done in three replicates. *Color, Ulva reticulata* extract was determined by three respondents using their sense of sight, then the perceived color of *Ulva reticulata* extracts by the respondent's was recorded. *Odor,* the extract was determined by three respondent using their olfactory senses, and the result was recorded. *Density*, 5mL of sample extract was weighed using an analytical balance, the weight of the *Ulva reticulata* extract was divided to the volume used in weighing. The procedure was repeated thrice and the average density was computed. The *pH* was determined in using digital pH meter, it was done in three replicates. *Solubility test* of *Ulva reticulata* extract was tested in three different solvent namely hexane, water and ethanol. Two (2) mL of *Ulva reticulata* extract was poured in three different separated test tubes. Then, add 2ml of hexane, water and ethanol in each separate test tube.

Chemical property determination

Ash content

The three crucible marked then placed 10g of sample and heated in oven for 3 hours in lowered temperature. After heating the three crucible placed in desiccator for 30 minutes and weighed. The crucible containing sample was heated in Bunsen burner until ash resulted. This were removed placed in desiccator for 30 minutes and reweighed. The Ash content was computed using the formula:

$$\text{Ash content } \% = \frac{W3 - W1}{W2 - W1}\ x\ 100$$

Nutritional content determination

Carbohydrates (Wang, et al., 2006)

Calculation of carbohydrates for *Ulva reticulata* (kulapo) extract was determined by adding the moisture content, protein, fat and result was subtracted from 100% carbohydrates content of Kulapo extract was calculated using the equation:

Total carbohydrates = 100 - (% fat + % moisture + % protein)

Fat

To determine the fat content of *Ulva reticulata* the researcher used soxhlet method. Crude fat content was determined by extracting the fat of *Ulva reticulata* using a solvent. The flask washed with hexane and dry in oven for 30 minutes at 102^{0}C. Cooled in the desiccator for 3 minutes, then the flask was placed with 100 mL of hexane. About 10g of powder sample covered with Wattman paper and placed inside of the extraction chamber. The solvent was heated with the flask until it boiled. The kulapo sample was continued to boil for six hours in a hot plate, and let the solvent dripped from condenser into the sample. The flask was removed from the set up and was placed in the oven for continued evaporation of solvent at 102^{0}C for 1 hour. Afterwards, it was placed in the desiccator for 30 min and weigh (De Una, 2016).

Moisture Content

The oven was set to 105^{0}C and was preheated for 30 minutes, 10g of the sample was weighed into pre-dried and pre-weighed crucible. The crucible containing the sample were placed in the oven and dry for 12 hours. After drying in oven, the crucible was placed in the desiccator for 30 minutes and weighed afterwards.

$$\text{Moisture Content } \% = \frac{\text{g sample before drying} - \text{g sample after drying}}{\text{g sample before drying}}\ x\ 100$$

Protein

The crude protein content (N X 4.38) of the samples was determined and analyzed at the University of the Philippines- Los Baños, Laguna.

Fiber (Cockrell et al., 2000)

The determination of Crude fiber has two steps. The first step, the sample was defatted using soxhlet method. Five grams of defatted sample was weighed. In the step two, it was placed in the flask and was added with 67mL boiling sulfuric acid solution. It was boiled for 30 minutes, maintaining the volume of distilled water at constant and swirling the flask periodically to remove the particles adhering to the side of the flask. Buchner funnel was lined with the filter paper, and preheated by boiling water. At the same time, at the end of boiling period, the flask was removed, cooled for one minute and the contents were filtered and carefully suction in the Buchner funnel with filter paper. Filtration was carried out in less than ten minutes. The filter paper was washed with the residue of boiling water. The residue was transferred to the flask using retort containing 67 mL of boiling NaOH solution and boiled for 30 minutes as in step 2.

The crucible was carefully preheated with boiling water. Hydrolyzed mixture was filtered after cooling for one minute. The residue was washed with boiling water, with HCl solution, and then washed again with boiling water. Finishing with 3 washes of hexane, the crucible with sample content was placed in drying oven at 105^{0}C for one hour, cooled in at the desiccator. The crucible was weighed quickly with the residue. It was burned for three hour, then placed in the desiccator for 30 minutes and weighed. Crude fiber content was calculated by the following formula;

$$\% \text{ Fiber} = \frac{\text{weight of ash obtained}}{\text{weight of original sample}} x100$$

Bioavailability of Cr, Co, I, Ni, Fe, Se, As, and Pb in seaweed extract

Total metal determination was performed using Energy Dispersion X-ray (EDX) and Atomic Absorption Spectroscopy (AAS). Six elements have been quantified and standard solutions were used to correct possible matrix effects and signal drift.

Results and discussion

Physical properties determination

The ash content of *Ulva reticulata* determined by burning a given quantity of samples under prescribed condition and measuring residue. The table shows that the average ash content of *Ulva reticulata* is 9.92%. The ash content represents the total mineral content in food and it implies that *Ulva reticulate* has high mineral content.

Chemical property determination

Total ash content was mentioned in the figure 1.

Determination of nutritional content

The determination of Crude fiber has an average of 1.16% in *Ulva reticulata*. Fiber helps lower cholesterol and blood sugar regulation (The Journal of the American Board of family medicine). The fats determined of *Ulva reticulata* after the extraction of the solvent was evaporated, the residue weighed and reported as percent of crude fat. The average fat of *Ulva reticulata* was 0.87%.

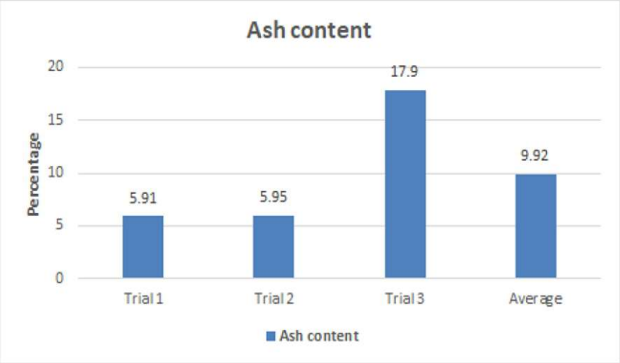

Figure 1. Ash content of *Ulva reticulate* extract

Table 1. Physical properties of the *Ulva reticulate* extract

Properties	Trial 1	Trial 2	Trial 3	Average
Boiling point	86°C	85°C	86°C	86°C
Color	Green	Light green	Light green	Light green
Odor	Unpleasant	Unpleasant	Unpleasant	Unpleasant
Density (g/mL)	4.43	3.99	4.36	4.36
pH	7.61	7.54	7.60	7.58
Solubility:				
Hexane	Immiscible	Immiscible	Immiscible	Immiscible
Water	Miscible	Miscible	Miscible	Miscible
Ethanol	Miscible	Miscible	Miscible	Miscible

Table 2. Nutritional content of the *Ulva reticulate* extract

Properties	Trial 1	Trial 2	Trial 3	Average
Moisture content	31.0%	27.2%	24.4%	27.53%
Crude fiber	0.2%	0.3%	3%	1.16%
Crude fat	0.4%	1.3%	0.9%	0.87%
Crude Protein	0.824	0.756	0.830	0.803

Carbohydrates and Vitamin C

Standard curve for vitamin C (ascorbic acid) at 521 nm wavelength shows a linear curve with increasing absorbance as concentration increases. On the other hand, *U. reticulata* shows a similar linear curve with that of the standard and the same absorbance of 0.11 nm was observed in concentrations 0.06% in the standard ascorbic acid and 0.5% in *U. reticulata*, an indication that *U. reticulata* do contain vitamin C.

Table 3. Percent of Carbohydrates content in *Ulva reticulata*

Total% fat	Total % fiber	Total % Moisture	Total % protein	Total % carbohydrates
0.85%	1.13%	23.9%	0.84%	73.42%

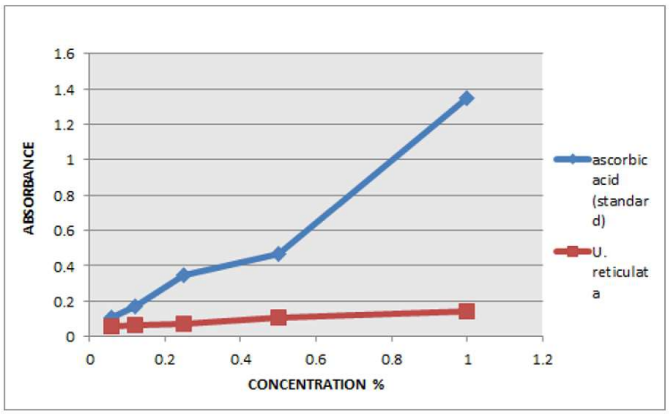

Figure 2. Absorbance of vitamin C and *Ulva reticulata*

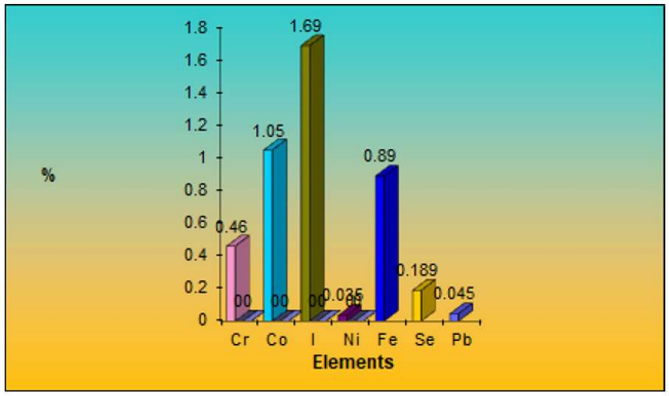

Figure 3. Elemental content of seaweed sample

Bioavailability in seaweed extract

The bioavailability of elements evaluated in the different species of seaweeds reveals that for the seven elements quantified in different species of seaweeds, results have shown that the collected species contain a considerable amount of essential elements, but some species can also accumulate non-essential elements as presented in figure 3.

Conclusion

Ulva reticulate was identified to have a nutritional value through series of test using gravimetric methods. Ash content of *Ulva reticulate* was determined to be 9%, carbohydrates 73.2%, fat content 0.85%, moisture content of 23.9%, crude fiber of 1.13%, crude protein of 0.803% and vitamin C was present in the sample at 521 nm. The bioavailability of elements in *Ulva reticulate* contained a considerable amount of essential element. Thus, *Ulva reticulate* is suitable for human consumption as food source.

Conflicts of interest: Not declared

References

Abreu H. 2015. The Novel Role of Seaweed in Traditional Food Products. Harnessing Marine Bioresources for Innovation in the Food Indusry. RDS Dublin Ireland.

Cockrell I, Francis BHD. 2000. Changes in Nuitritive Value of Concentrate Feeding Stuffs During Storage. Tropical Product Institute. London, UK.

Cui-Lim KMR, Ambida PKP, Alvarez MC, Bangco N, Cortez F, Flores AAU, Perez B, Somoray MJ, Unay OD. 2017. Study on Nutritional and Elemental Analysis of the Seaweeds of Northern Samar, Philippines. Green Chemistry and Technology Letters, 3(1):17-20.

De Una M. 2016. Nutritional Content of *Artocarpos serisicicarpus* Jarett (Gumihan) Seed. College of Science-University of Eastern Philippines.

Erivera J, Cui KMR. 2016. Bioaccumulation of heavy Metals in *Seylla serrata* (Mud crab) collected from Lavezares and Rosario, Northern Samar. International Journal of Chemistry, 5(2):132-135.

Khahed A, Amal H, Halim A, Morry F. 2014. Distribution of Heavy Metals in Seaweeds Collected along Marsa-Matrouh beaches, Egyptian Mediterranean Sea. Egyptian Journal of Aquatic Resources. 40:363-371.

Mamboya A. 2007. Heavy Metal Contamination and toxicity: Studies of Macroalgae from the Tanzanian Coast. Stockholm University. 1-48.

Rybak A, Beata M, Boguslana L. 2013. The Accumulation

of Metal Co, Cr, Cu, Mn, and Zn in fresh water (*Ulva chlorophyta*) and its habitat. Ecotoxicology. 22:558-573.

Sartal C, Alonso M, Pinerio A, Barrera P. 2013. Study of Cooking on the Bioavailability of As, Co, Cr, Cu, Fe, Ni, Se and Zn from Edible Seaweeds. Microchemical Journal. 108:92-99.

Tourmaa T. 2000. Chromium, Selenium and other Trace Minerals in Health Reproduction. Journal of Orthomolecular Medicine. 15:3.