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Rheological Properties, Moisture Content, and Carrageenan Yield of Macroalga *Kappaphycus alvarezii* Using Freshwater and Marine Water as Pre-Treatment

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ABSTRACT

A variety of seaweed post-harvest techniques have been developed using the same method of washing the seaweed with marine and drying it under the sun; however, a method of washing the seaweed with freshwater has yet to be developed. This study investigates the effect of freshwater and marine water as soaking solutions on the moisture content, carrageenan yield, and rheological properties of macroalga *K. alvarezii*. The seaweed was immersed in freshwater and marine water for 10 minutes with triplicates before being dried in a solar dryer for seven days. Extraction of seaweed was done after drying. Results revealed that the dried *K. alvarezii* soaked in freshwater had significantly lower ($p \leq 0.05$) moisture content than *K. alvarezii* soaked in marine water. Additionally, the carrageenan yield of *K. alvarezii* significantly increased ($p \leq 0.05$) by 14.48% when soaked in freshwater compared to the yield in marine water. Considering the rheological properties of the seaweed, the gelling temperature and melting temperature of *K. alvarezii* soaked in freshwater did not differ significantly ($p \geq 0.05$) from those of *K. alvarezii* soaked in marine water. However, other rheological properties such as the syneresis, viscosity, and gel strength of *K. alvarezii* greatly improved ($p \leq 0.05$) when they were soaked in freshwater with significant increases of 2.21%, 1.84 cPs, and 13.22 g cm^{-2}, respectively. Thus, this study indicates that macroalga *K. alvarezii* immersed in freshwater showed substantial improvements in their carrageenan quality.

KEYWORDS: Carrageenan yield, *Kappaphycus alvarezii*, moisture content, seaweeds, rheological properties

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1. Introduction

Traditionally, seaweeds have been used as a source of food, fodder, and fertilizer, as well as being a medicinal drug source. Moreover, they are the raw materials for making substances such as agar, algin, and carrageenan, which are used in the food industry (Kolanjinathan et al., 2014; El-Beltagi et al., 2022; Yangson et al., 2022). Approximately 28% of the world's seaweed production was attributed to eucheumatoid seaweeds, such as *Eucheuma* and *Kappaphycus* species (FAO, 2022). The seaweeds found in the Philippines can be considered remarkably diverse, with more than 800 species recorded in the country (Silva et al., 1987; Tahiluddin and Terzi, 2021; Valderrama, 2012). Additionally, according to the Fisheries Statistics Organization, the Philippines ranks fourth among the major seaweed producers worldwide in 2020 (FAO, 2022). Among the major income sources for the Philippine economy, seaweeds, particularly *Eucheuma* and *Kappaphycus* species, are the most important aquaculture species (Tahiluddin and Terzi, 2021). Moreover, carrageenan is a hydrocolloid extracted primarily from red seaweeds such as *Eucheuma* and *Kappaphycus* species (Loureiro et al., 2017). A wide variety of raw and semi-processed food products made from seaweed have been developed for use in food manufacturing (Kaliaperumal, 2003). The use of carrageenan is widespread in several industries, including the binding, gelling, and thickening of foods, pharmaceuticals, cosmetics, and commercial products (Vairappan, 2006; Naguit et al., 2009; Necas and Bartosikova, 2013; Rupert et al., 2002).

In general, seaweeds collected from the sea are dried before they are used in nutritional studies or industrial purposes either for research or production (Chan et al., 1997; Pereira, 2011). Moreover, drying seaweeds reduces their water activity, which inhibits microbial growth, preserves the desirable qualities, and reduces the storage volume (Gupta et al., 2011; Vorse et al., 2013). Researchers stated that, in the case of properly dried seaweeds, the gel content of the product can be preserved for an extended period of time without affecting its quality (Chan et al., 1997; Ling et al., 2015). Poncomulyo et al. (2006) have conducted studies on seaweed post-harvest techniques by washing and drying the seaweed with marine water. Additionally, choosing the most suitable post-harvest technique for seaweeds determines the quality of the finished product (Badmus et al., 2019; Vorse et al., 2013). Hence, post-harvest techniques (after cultivation) of seaweed cultivation are essential to ensuring seaweed quality. This study investigates the effect of freshwater and marine water as immersing solutions on the moisture content, carrageenan yield, and rheological properties of seaweed *Kappaphycus alvarezii*.

Material and Methods

2.1. Study Site

The study was conducted at the Seaweeds Post-Harvest Laboratory, College of Fisheries, Mindanao State University-Tawi-Tawi College of Technology and Oceanography (MSU-TCTO), Philippines (Latitude 5.037107˚, Longitude 119.743357˚). The sample of macroalga *K. alvarezii* was obtained from seaweed farmers of Pasiagan, Bongao, Tawi-Tawi, Philippines. The systematic classification of macroalga *K. alvarezii* is also given below (Figure 1).

2.2. Drying Preparation

A two-step post-harvest technique was followed: one sample of *K. alvarezii* was thoroughly washed with marine water, while the other sample of *K. alvarezii* was thoroughly washed with freshwater. Following this, the samples were immersed in freshwater and marine water separately for 10 minutes and then dried in a solar dryer for seven days. The dried macroalga *K. alvarezii*

Kingdom: Plantae

Phylum: Rhodophyta

Class: Florideophyceae

Order: Gigartinales

Family: Solieriaceae

Genus: *Kappaphycus*

Species: *alvarezii*

Figure 1. Systematic Classification of *Kappaphycus alvarezii* (Photograph was using Huawei nova 7i)

sample immersed in marine water was darker than the dried macroalga *K. alvarezii* sample in freshwater (Figure 2).

2.3 Moisture Content Analysis

Each dried seaweed sample from the four corners of the pile and one at the center was mixed thoroughly. Each treatment with three replicate samples of 3 g weight was analyzed using a moisture analyzer. Moisture content was calculated using the formula below:

$$\text{Moisture Content} = \frac{\text{Initial Weight} - \text{Final Weight}}{\text{Final Weight}} \times 100$$

2.4 Carrageenan Quality Analysis

Extraction for the carrageenan quality was followed by the method of Romero et al. (2000). The rheological properties such as gelling temperature, melting temperature, viscosity, syneresis index, and gel strength were determined after the extraction of carrageenan yield. Carrageenan yield was calculated using the formula below:

$$\text{Carrageenan Yield} = \frac{\text{Weight of Carrageenan}}{\text{Weight of Dried Seaweed}} \times 100$$

In the experiment, the gelling temperature was determined using a laboratory thermometer.

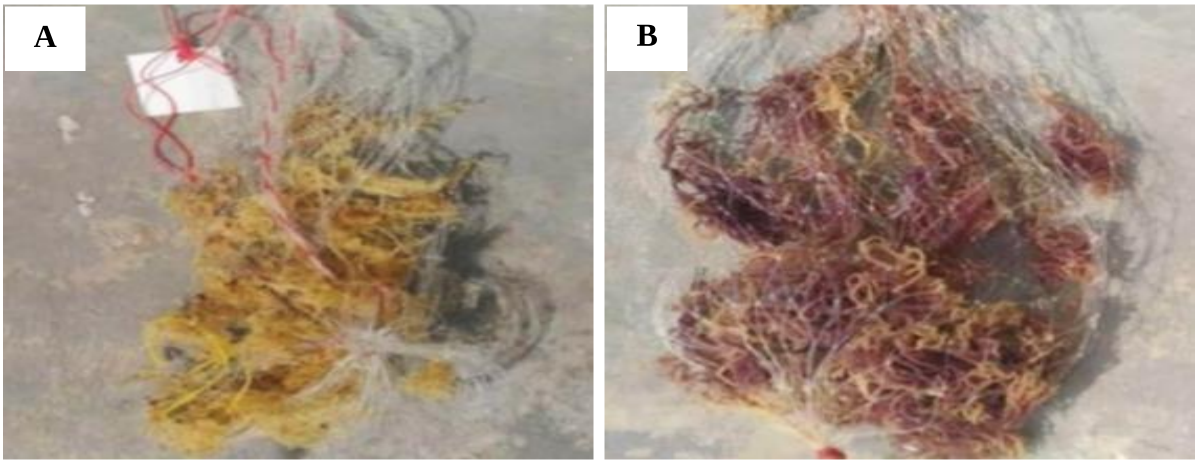

Figure 2. Dried sample of macroalga *K. alvarezii* after 7 days. A, *K. alvarezii* immersed in freshwater; B, *K. alvarezii* immersed in marine water.

It is determined when glass beads with a diameter of 2.85 mm and a weight of 30 mg fail to sink to the bottom at an interval of 0.5℃. In addition, the viscosity was determined using a Brookfield Viscometer (Model LVF) at 75°C running at 30 rpm in an electrolytic beaker measuring 110.3=46 mm in diameter. In the syneresis index, the percent of water released from the cylindrical gels (2.2=3.5cm) in the filter paper after two hours of weight loss was considered. Moreover, a gel strength measurement was performed on the carrageenan solution after it had been allowed to gel at room temperature for 15 hours.

2.5 Statistical Analysis

IBM SPSS software version 20 was used to analyze the collected data of moisture content, carrageenan yield, and rheological properties of macroalga *K. alvarezii* at $p \leq 0.05$ significance level. Data were presented as mean ± standard error of the mean (SEM). Determination of significant differences was computed through *t*-test.

3. Results

Figure 3 shows the moisture content and carrageenan yield of macroalga *K. alvarezii* immersed in freshwater and marine water. A sample of *K. alvarezii* immersed in marine water showed a moisture content of 10.39 ± 0.82%, significantly lower ($p \leq 0.05$) than a *K. alvarezii* immersed in freshwater, which showed a moisture content of 16.67 ± 0.49%. The yield of carrageenan from *K. alvarezii* immersed in freshwater (55.04 ± 1.33%) was significantly higher ($p \leq 0.05$) than that from *K. alvarezii* immersed in marine water (40.20 ± 0.97%).

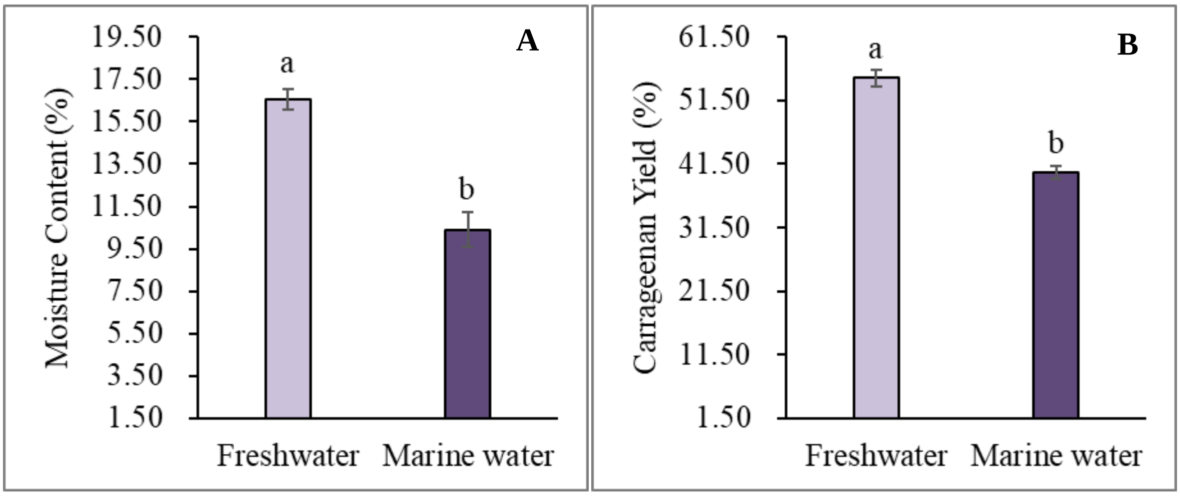

Figure 3. Macroalga *K. alvarezii* immersed in freshwater and marine water. A, Moisture content: B, Carrageenan yield. Differences in the letter are significantly different ($p \leq 0.05$). Error bar in SEM (standard error mean), n=6.

Moreover, Figure 4 shows the rheological properties of *K. alvarezii* immersed in freshwater and marine water. The gelling temperature of *K. alvarezii* immersed in freshwater (37.33 ± 0.33 ˚C) did not significantly differ ($p \geq 0.05$) from *K. alvarezii* immersed in marine water (37.00 ± 0.58 ˚C). *K. alvarezii* immersed in freshwater showed a melting temperature of 42.33 ± 6.67 ˚C, did not significantly different ($p \geq 0.05$) than *K. alvarezii* immersed in marine water, which showed a melting temperature of 48.33 ± 1.35 ˚C. Additionally, the viscosity of *K. alvarezii* immersed in freshwater (5.67 ± 0.44 cPs) was significantly higher ($p \leq 0.05$) than *K. alvarezii* immersed in marine water (3.83 ± 0.17 cPs). The syneresis index from *K. alvarezii* immersed in freshwater (8.31 ±

0.14%) was significantly lower ($p \leq 0.05$) than that from *K. alvarezii* immersed in marine water (10.52 ± 0.52%). Moreover, macroalga *K. alvarezii* immersed in freshwater showed a gel strength of 37.22 ± 0.46 g cm^{-2}, significantly improved ($p \leq 0.05$) than *K. alvarezii* immersed in marine water, which showed a gel strength of 24.00 ± 0.67 g cm^{-2}.

4. Discussion

Carrageenan is primarily derived from red seaweeds and is used for gelling, thickening, and stabilizing purposes (Husin, 2014). Several researchers have stated that carrageenan is an important ingredient in foods, pharmaceuticals, cosmetics, personal care, and other products (Thirumaran et al., 2009; Hayashi et al., 2011).

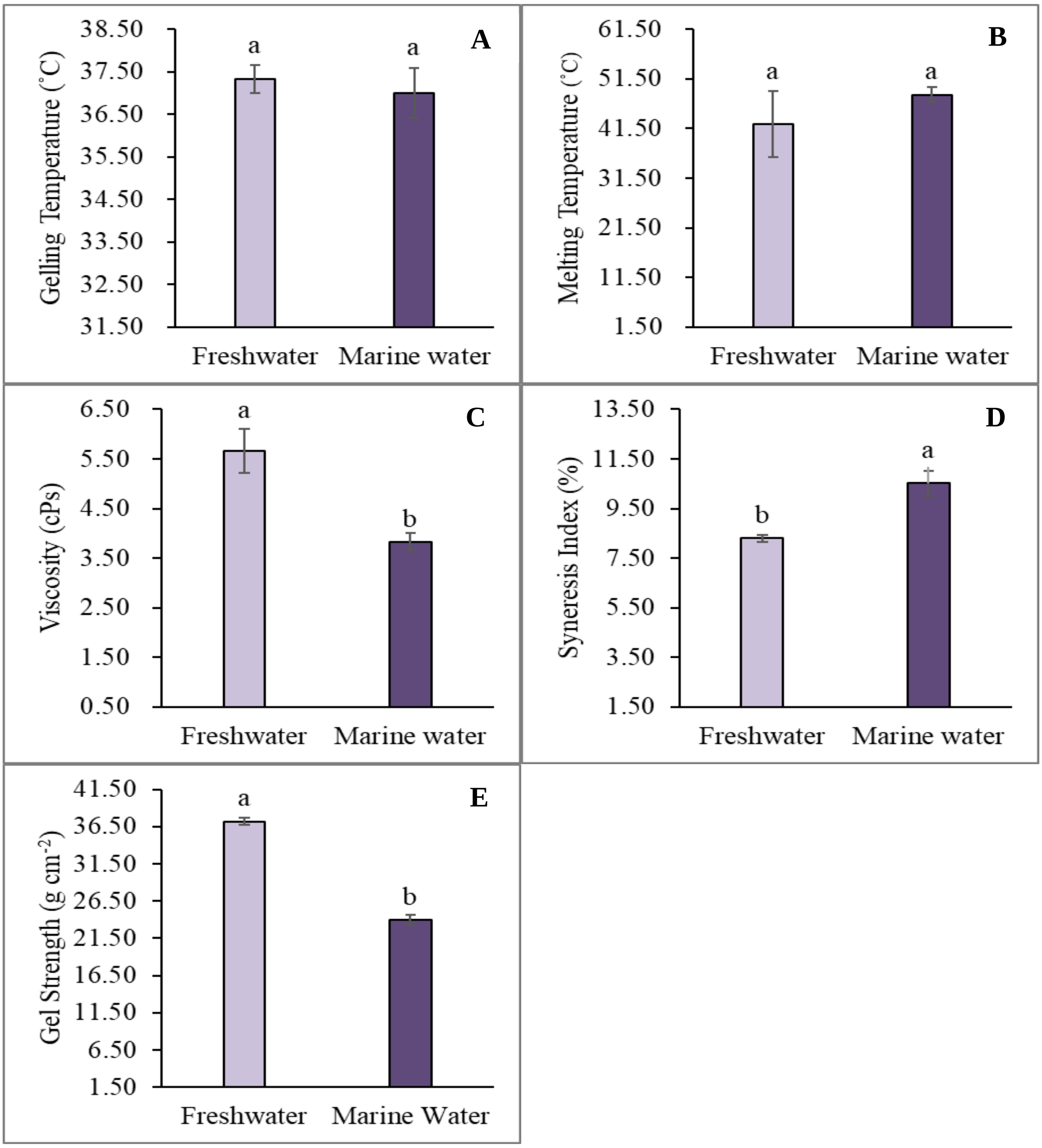

Figure 4. Rheological properties of macroalga *K. alvarezii* immersed in freshwater and marine water. A, Gelling temperature: B, Melting temperature: C, Viscosity: D, Syneresis index: E, Gel strength. Differences in the letter are significantly different ($p \leq 0.05$). Error bar in SEM (standard error mean), n=6.

It is common practice to dry seaweeds such as *Kappaphycus* sp. directly after they have been harvested without first being washed with freshwater (Sarri et al., 2022; Tahiluddin et al., 2022). The present study examined the effect of immersing farmed *K. alvarezii* in marine and freshwater on carrageenan quality in seaweed. A moisture content measurement was conducted before extracting macroalgae *K. alvarezii* for the quality of carrageenan. Results revealed that the moisture content from *K. alvarezii* immersed in marine water (10.39 ± 0.82%) was significantly lower than that from *K. alvarezii* immersed in freshwater (16.67 ± 0.49%). As reported by Tiroba (2006), seaweed of acceptable quality should not contain more than 35% moisture. Hence, the moisture content of the present study was within acceptable limits in terms of moisture content.

Moreover, the carrageenan yield from *K. alvarezii* immersed in freshwater achieved 55.04 ± 1.33% was significantly improved than the carrageenan yield from *K. alvarezii* immersed in marine water, which obtained 40.20 ± 0.97%. In other studies, it has been shown that macroalga *Kappaphycus* sp. was directly dried after harvesting without being washed or immersed in freshwater, and then extracted, resulting in a carrageenan yield of 43% (Luhan et al., 2014), 38% (Loureiro et al., 2014), 36% (Sarri et al., 2022), 34% (Robles, 2020), 33% (Loureiro et al., 2014), and 32% (Tahiluddin et al., 2022). In contrast to the present study, in which macroalga *K. alvarezii* immersed in freshwater increased the amount of carrageenan yield, the results suggest that seaweeds should be immersed in freshwater before drying to maximize carrageenan yield production.

In this study, various rheological properties of macroalga *K. alvarezii* immersed in freshwater and marine water have been investigated, such as their gelling temperature, melting temperature, syneresis index, viscosity, and gel strength. It has been revealed that the gelling and melting temperature of macroalga *K. alvarezii* immersed in freshwater was not significantly different than macroalga *K. alvarezii* immersed in marine water. However, the syneresis index, viscosity, and gel strength of macroalga *K alvarezi* immersed in freshwater improved considerably than the macroalga *K. alvarezii* immersed in marine water. In another study, Robles (2020) investigated that the gelling and melting temperatures of farmed macroalga *K. alvarezii* were 35 ˚C and 47 ˚C, respectively. Compared to the present study, which achieved gelling and melting temperatures of 37.33 ˚C and 42.33 ˚C, respectively, for *K. alvarezii* immersed in freshwater, while it reached gelling and melting temperatures of 37.00 ˚C and 48.33 ˚C, respectively, for *K. alvarezii* immersed in marine water. The syneresis index refers to the quantity of water exuded from a given amount of gel (Bryant et al., 1996; Robles, 2020). A lower syneresis index was obtained in macroalga *K. striatus* immersed in freshwater (8.31%), indicating that it improved the quality of carrageenan. Moreover, in the present study, the gel strength and viscosity of carrageenan quality from macroalga *K. alvarezii* immersed in freshwater were significantly improved, reaching 5.67 cPs and 37.22 g cm^{-2}, respectively. This was lower than the other study in which the viscosity and gel strength were achieved at 3.50 cPs and 31.77 g cm^{-2}, respectively (Robles, 2020).

5. Conclusion

In conclusion, macroalga *Kappaphycus alvarezii* immersed in freshwater before drying demonstrated substantial improvements in their carrageenan quality, including their rheological properties. In light of the results of this study, basic information can be given to seaweeds farmers that it is recommended to wash or immerse the seaweeds in freshwater before drying, as this will improve the quality of carrageenan that can be harvested from the seaweeds.

Compliance with Ethical Standards

Acknowledgement

The abstract of this study was presented at the 4th International Congress on Engineering and Life Science (ICELIS) on November 17-19, 2023 at Comrat, Maldova.

Conflict of interest

The authors declared that for this research article, they have no actual, potential, or perceived conflict of interest.

Author contribution

The contribution of the authors to the present study is equal. All the authors read and approved the final manuscript. All the authors verify that the Text, Figures, and Tables are original and that they have not been published before.

Ethical approval

Ethics committee approval is not required.

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Data availability

Data are available upon request.

Consent for publication

Not applicable.

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