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Physicochemical Characteristics and Quality of Carrageenan from *Eucheuma cottonii* Seaweed Origin Waters of West Seram District

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ABSTRACT

Eucheuma cottonii is a species of red seaweed known for its high carrageenan content. Carrageenan plays a vital role as a stabilizer, thickener, gelling agent, emulsifier, protective colloid, coagulant, and crystallization inhibitor. These functional properties make it widely used in various industries, including food, pharmaceuticals, cosmetics, textiles, paints, and toothpaste. This study aimed to evaluate the physicochemical characteristics and quality of carrageenan extracted from *Eucheuma cottonii* collected from two coastal locations in West Seram District: Nuruwe and Wael. Carrageenan extraction was conducted using a 2% KOH solution followed by precipitation with isopropyl alcohol (IPA). The results showed that carrageenan extracted from *E. cottonii* from Nuruwe demonstrated better physicochemical properties, with a yield of 22.53%, moisture content of 11.80%, ash content of 12.64%, viscosity of 44.37 cps, gel strength of 322.03 g/cm², and sulfate content of 41.51%. FTIR spectral analysis revealed characteristic peaks corresponding to sulfate groups at 1259.52 cm⁻¹, glycosidic bonds at 1045.42 cm⁻¹, 3,6-anhydro-D-galactose at 931.62 cm⁻¹, D-galactose-4-sulfate at 846.68 cm⁻¹, and hydroxyl (OH) groups at 3255.84 cm⁻¹. In comparison, carrageenan from Wael yielded 15.24%, with a moisture content of 14.72%, ash content of 14.18%, viscosity of 34.49 cps, gel strength of 254.29 g/cm², and sulfate content of 30.41%. FTIR analysis indicated similar functional groups, with sulfate groups at 1269.45 cm⁻¹, glycosidic bonds at 1045.42 cm⁻¹, 3,6-anhydro-D-galactose at 931.62 cm⁻¹, D-galactose-4-sulfate at 848.68 cm⁻¹, and OH groups at 3161.33 cm⁻¹. These findings indicate that the quality of carrageenan from Nuruwe is superior in terms of yield, gel strength, and sulfate content, suggesting its greater potential for industrial applications.

INTRODUCTION

Indonesia is a maritime country with vast coastal areas rich in biological resources and diverse ecological potential, including those found in the Maluku region. Maluku

Province is renowned as an archipelagic region, covering an area of 712,480 km^2, consisting of 92.4% marine waters (658,295 km^2) and only 7.6% land area (54,185 km^2), resulting in an extensive coastline and broad coastal zones **(Center for Data and Information, Ministry of Marine Affairs and Fisheries / *Pusdatin KKP*, 2022)**.

One of Maluku's abundant marine resources is seaweed, with several economically valuable species such as *Eucheuma cottonii*, *Eucheuma edule*, *G. coronopifolia*, *Gracilaria* spp., and *Gelidium* spp. *Eucheuma cottonii*—also known as *Kappaphycus alvarezii*—belongs to the red algae group (Rhodophyceae) and is widely cultivated in various coastal regions of Maluku, including West Seram, East Seram, Aru Islands, and Southeast Maluku. The identified potential area for seaweed cultivation in Maluku reaches 19,509.29 hectares. However, the actual area currently utilized remains limited, including 929.9 hectares in West Seram, 1,241.20 hectares in East Seram, 453.24 hectares in Buru, 117.48 hectares in Ambon, 9,228 hectares in Central Maluku, 216.60 hectares in Southeast Maluku, 5,202.64 hectares in Southwest Maluku, and 1,587 hectares in the Aru Islands. This shows that there is still significant potential to expand seaweed cultivation, particularly for *Eucheuma cottonii*, which could contribute substantially to increased production in the province **(Center for Data and Information, Ministry of Marine Affairs and Fisheries, 2022)**.

According to data from the Maluku Provincial Department of Marine Affairs and Fisheries, seaweed production in the region reached 71,928 tons in 2022 **(Maluku Provincial Department of Marine Affairs and Fisheries, 2022)**. Currently, the harvested seaweed is mostly sold in dried form at prices ranging from IDR 10,000 to IDR 18,000 per kilogram. However, processing *Eucheuma cottonii* into semi-finished or finished products—such as carrageenan—would significantly increase its economic value, thereby enhancing the income of local seaweed farmers.

Seaweed farming is widely developed due to its relatively simple and low-cost cultivation techniques, high productivity, low risk of crop failure, and short harvesting cycle—typically every 45–60 days or up to four times a year. The relatively high selling price of seaweed also serves as a motivating factor for its cultivation. Additionally, seaweed farming absorbs a large workforce and generates considerable economic multiplier effects **(Salim & Ernawati, 2015)**.

Seaweed is generally traded in several product forms: dried seaweed, directly consumable products, and hydrocolloid-based products (such as carrageenan, agar, and alginate). Globally, approximately 65% of seaweed production is used for direct consumption, 15% for hydrocolloid production, and the remaining 20% for other uses such as fertilizers, paper, and biofuels **(Salim & Ernawati, 2015)**.

Carrageenan, a major hydrocolloid derived from red seaweed, is classified into three types based on the gel it forms: iota-carrageenan (spinosum type), which forms soft, elastic, and durable gels; kappa-carrageenan (cottonii type), which forms strong, firm, and brittle gels; and lambda-carrageenan, which does not form a strong gel but contributes to viscosity. *Eucheuma cottonii* is primarily known for its high kappa-carrageenan content, making it suitable for pharmaceutical applications such as capsule shell production. Carrageenan is considered an ideal material for edible film due to its abundance, non-toxic nature, low cost, and potential for large-scale production **(Milani & Maleki, 2012)**.

This study aimed to determine the characteristics and quality of carrageenan extracted from *Eucheuma cottonii* collected from different waters in West Seram District, Maluku Province.

MATERIALS AND METHODS

Time and location of sample collection

This research was conducted in April 2023. The sampling of *Eucheuma cottonii* seaweed was carried out in two coastal locations: the waters of Wael Hamlet and the waters of Nuruwe Village, both situated in the West Seram Regency, Maluku Province, Indonesia. These sites were selected based on the natural availability of seaweed habitats and the suitability of the surrounding environmental conditions.

The research activities included several main stages: site selection for sampling, measurement of physicochemical parameters of the waters, carrageenan extraction from seaweed samples, quality testing of the extracted carrageenan, and data analysis. Each step was carried out systematically to ensure representative results that support the objectives of the study.

Carrageenan extraction methods

Approximately 200g of seaweed was first washed with fresh water to remove sand, soil, and other impurities. The cleaned seaweed was then cut into small pieces and dried at 60 ºC until completely dry. The dried seaweed was further chopped and ground into powder using a crusher.

About 40g of seaweed powder was placed into a container or pot, then mixed with water at a ratio of 40–50 times the weight of the powder (approximately 1.6L). The pH of the mixture was adjusted to pH 9 by adding approximately 200ml of 0.1N KOH solution. The mixture was then heated or boiled at a temperature of 90– 95ºC for about 1 hour.

After heating, the mixture was filtered using filter cloth. The resulting filtrate was collected in a separate container and then added with 10% NaCl solution, amounting to 5% of the total filtrate volume. The mixture was reheated at 50– 60ºC while stirring continuously for 15 minutes.

Next, isopropyl alcohol (IPA) was added to the filtrate at twice the volume of the filtrate, with constant stirring for 15–30 minutes until coagulation occurs. The coagulated material was then filtered using filter paper. The resulting precipitate was known as carrageenan. To obtain a firmer gel, the coagulated material was re-soaked in 150ml of IPA for 15 minutes and was filtered again using filter cloth.

The fibrous carrageenan clumps were then blended to facilitate powdering, and dried again at 60ºC until getting completely dry. Once dry, the carrageenan was ground using a crusher into powder form and sieved to obtain 80 mesh particle size. The resulting carrageenan powder was stored in sealed plastic containers or bottles.

In this study, two different concentrations of potassium hydroxide (KOH), namely 0.2 and 0.5%, were used during the extraction process to assess the effect of alkali strength on the quality of carrageenan produced. The selection of these concentrations was based on preliminary observations and previous studies indicating that variations in KOH concentration can significantly influence the yield, viscosity, gel strength, and sulfate content of carrageenan **(Desiana & Hendrawati, 2015; Diharmi, 2016; Julaika *et al*., 2017)**. A lower concentration (0.2%) tends to preserve the molecular integrity of polysaccharides and minimize the degradation of carrageenan chains, whereas a higher concentration (0.5%) promotes stronger desulfation and conversion of galactose-6-sulfate into 3,6-anhydrogalactose, which may improve gel formation but reduce total sulfate content. Therefore, comparing these two concentrations provides valuable insights into how alkali treatment intensity affects the physicochemical and structural characteristics of carrageenan derived from *Eucheuma cottonii*.

Data analysis

The data obtained from the carrageenan extraction process were analyzed using descriptive analysis. This approach aimed to describe the characteristics of the extracted carrageenan based on the measured physical and chemical parameters. The parameters analyzed include yield, moisture content, ash content, protein content, fat content, carbohydrate content, sulfate content, viscosity, gel strength, ion sensitivity (K^+ and Na^+), and functional group identification.

Each parameter was presented using the average value of the measurements, standard deviation (if available), and compared to standard values or references from relevant literature. This descriptive approach helps assess how well the extracted carrageenan meets technical or industrial quality standards.

Descriptive analysis was also used to identify patterns or tendencies in each parameter that may be influenced by extraction conditions, such as solvent ratio, pH, heating temperature, or the type of solvent used (in this case, isopropyl alcohol).

All results were presented in the form of tables and graphs to facilitate interpretation and to provide a comprehensive overview of the quality of carrageenan produced by the applied extraction method.

RESULTS AND DISCUSSION

Characterization of carrageenan from *Eucheuma cotonii* seaweed

Carrageenan is a sulfated polysaccharide classified as a linear macromolecule composed of repeating units of galactose and 3,6-anhydrogalactose, interconnected through glycosidic bonds. These polymer chains are further substituted with esterified sulfate groups and cations such as potassium and sodium. This molecular structure allows carrageenan to form long chains consisting of more than thousand galactose units, which contribute to the unique physicochemical properties of each carrageenan type **(Kumayanjati & Dwimayasanti, 2018)**.

The physicochemical characterization of carrageenan extracted from *Eucheuma cotonii* includes several important parameters such as yield, moisture content, ash content, viscosity, gel strength, and sulfate content. The experimental results obtained from different extraction conditions and sampling locations are summarized in Table (1).

As presented in Table (1), carrageenan extracted from *Eucheuma cotonii* collected at Nuruwe using a 0.2% solvent concentration yielded the highest extraction percentage at 22.53%, surpassing the FAO minimum standard of 20%. However, the moisture content of samples from Wael exceeded the FAO maximum threshold of 12%, suggesting inadequate drying or storage conditions.

All samples demonstrated viscosity values well above the FAO minimum requirement of 5 cP, with the highest viscosity recorded at 44.37 cP for Nuruwe 0.2%. In terms of gel strength, Nuruwe 0.2% also exhibited the highest value at 322.03 g/cm², indicating excellent gelling capacity, which is desirable for food and pharmaceutical applications.

Sulfate content in the Nuruwe 0.2% sample was found to be 41.51%, slightly exceeding the FAO upper limit of 40%. This may indicate the presence of impurities or the need for further purification steps during extraction. It should be noted that although both ash content and sulfate content in Table (1) appear to have similar FAO standard ranges (15–40%), they represent different quality indicators. The ash content reflects the total mineral composition remaining after combustion, mainly potassium, sodium, and calcium ions, whereas the sulfate content refers specifically to ester sulfate groups attached to the polysaccharide backbone of carrageenan **(Winarno, 1996; FAO, 2007; Distantita *et al.*, 2012)**. Therefore, while their numerical ranges may overlap, these two parameters describe distinct aspects of carrageenan's physicochemical quality.

Variations in physicochemical properties across the samples can be attributed to multiple influencing factors such as harvest age, seaweed species, solvent concentration, extraction temperature and duration, and the geographical origin of the seaweed. Therefore, optimizing the extraction process parameters is critical for producing high-quality carrageenan that meets industry standards.

Table 1. Physicochemical properties of carrageenan extracted from *Eucheuma cotonii* under different extraction conditions

Parameter	Nuruwe 0.2%	Nuruwe 0.5%	Wael 0.2%	Wael 0.5%	FAO Quality Standard
Yield (%)	22.53	20.47	15.24	12.44	≥ 20
Moisture Content (%)	11.80	11.58	14.72	14.13	< 12

Ash Content (%)	12.64	17.19	14.18	17.67	15–40
Viscosity (cps)	44.49	.35.39	34.49	23.34	> 5 cP
Gel Strength (g/cm²)	322.03	249.11	254.29	228.19	-
Sulfate Content (%)	41.51	30.94	30.41	25.54	15–40

Carragenan yield

Research findings indicate that the average carrageenan yield from Nuruwe waters, calculated as the mean of 0.2% and 0.5% KOH treatments, was 21.504%, which was higher than that from Wael waters, recorded at 13.841%. These values represent the mean yields obtained from duplicate extractions at each KOH concentration. Fig. (1) illustrates the average carrageenan yield from both locations, showing that the yield from Nuruwe was significantly higher ($P < 0.05$) than that from Wael, indicating that seaweed from Nuruwe produced superior carrageenan yield overall.

The difference in carrageenan yield between the two locations is presumed to be influenced by the varying environmental conditions, particularly current velocity, water transparency, and nutrient concentrations. The current velocity in Nuruwe waters is 0.31 m/s, slightly higher than in Wael waters at 0.27m/ s. According to **Anggadireja *et al.* (2010)**, the optimal current velocity for cultivating *Eucheuma cottonii* ranges from 0.2 to 0.4m/ s, indicating that both sites fall within the optimal range.

Stronger currents enhance nutrient delivery, thereby facilitating metabolic processes that support growth and increase carrageenan content **(Supriyantini *et al*., 2017)**. In addition, strong currents help remove debris and prevent pest infestations on seaweed thalli.

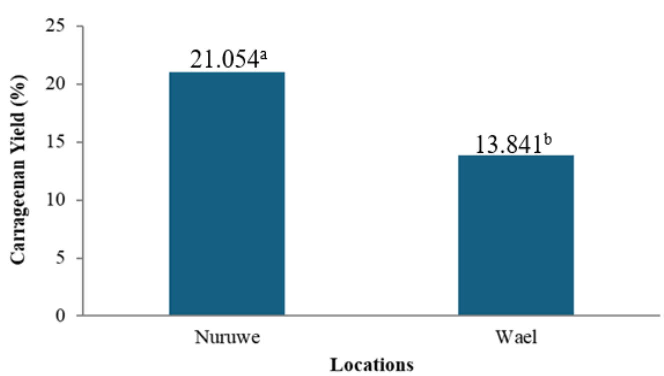

Fig. 1. Average carrageenan yield (%) of *Eucheuma cottonii* from Nuruwe and Wael waters, calculated as the mean of two KOH concentrations (0.2% and 0.5%). Different letters indicate statistically significant differences between locations ($P < 0.05$).

Light penetration or water transparency also plays a crucial role in determining carrageenan yield, as sunlight energy is essential for photosynthesis. This process significantly influences carrageenan synthesis. Transparency levels in Nuruwe waters reach 133.16cm, slightly higher than in Wael waters at 125.60cm. These values fall within the optimal range for seaweed cultivation, which is 113.8– 136.67cm, as specified in the Indonesian Minister of Environment Decree No. 51/2004 on Seawater Quality Standards **(Minister of Environment, 2004)**.

Phosphate and nitrate are important indicators of primary productivity and fertility in aquatic environments **(Effendi, 2003)**. Phosphate concentrations at both study sites were relatively low, measured at 0.036 mg/L in Nuruwe and 0.017 mg/L in Wael, both of which are close to the standard threshold for marine biota (0.015 mg/L), as specified in the Indonesian Minister of Environment Decree No. 51/2004 on Seawater Quality Standards **(Minister of Environment, 2004)**. Nitrate levels are higher in Nuruwe (0.026 mg/L) compared to Wael (0.01 mg/L), potentially due to anthropogenic inputs. Nonetheless, both remain within acceptable limits, ranging from 0– 5mg/ L **(Effendi, 2003)**.

Further research shows that carrageenan yield decreases with increasing KOH concentration (Fig. 2). This finding is consistent with studies conducted by **Herliany *et al*. (2013)**, **Rizal *et al*. (2016)** and **Arto *et al*., (2021)**, which report a negative correlation between KOH concentration and carrageenan yield.

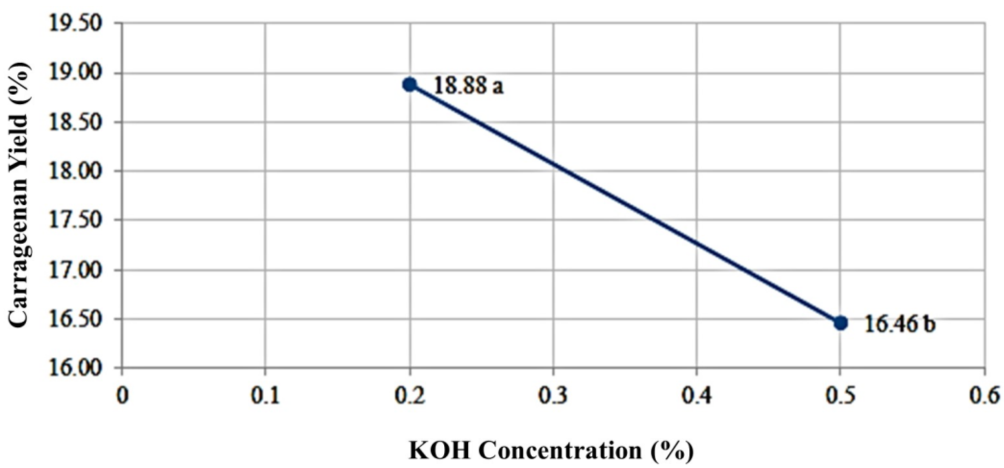

Fig. 2. Effect of KOH concentration on carrageenan yield: The figure shows the decreasing trend in carrageenan yield with increasing KOH concentration. Carrageenan yield was higher at 0.2% KOH (18.88%) compared to 0.5% KOH (16.46%). Different letters (a, b) indicate statistically significant differences between treatments ($P < 0.05$).

One contributing factor to the reduced yield is the extended duration of the extraction process. During alkaline treatment, hydroxide ions from KOH penetrate *Eucheuma cottonii* tissues, removing sulfate groups through a desulfation reaction at the C6 position of galactose units, producing 3,6-anhydro-D-galactose (3,6-AG). Simultaneously, potassium ions (K^+) neutralize the removed sulfate groups, forming potassium sulfate **(Distantita *et al*., 2009; Azefado *et al*., 2015)**.

While prolonged interaction between alkali solutions and *E. cottonii* can enhance extraction efficiency, excessive alkali concentration or overly extended extraction time may lead to carrageenan degradation, ultimately reducing the yield **(Ilias *et al*., 2017)**.

Moisture content

Moisture content in carrageenan is an important component, as water can trigger microbiological activity that affects the shelf life of carrageenan. Moisture content indicates the amount of water and volatile compounds present in the carrageenan. Moisture analysis is conducted to determine the water content in the carrageenan product.

The results showed that carrageenan from Nuruwe waters with 0.2 and 0.5% KOH concentrations had moisture contents of 11.80 and 11.58%, respectively. In contrast, carrageenan from Wael waters at the same KOH concentrations had moisture contents of 14.72 and 14.13% (Table 1). These results indicate that the moisture content of carrageenan from Nuruwe meets the FAO quality standard, which sets a maximum limit of 12%. Meanwhile, the moisture content from Wael waters is still within the range of commercial-grade carrageenan, which is 14.34% ± 0.25% **(Hakim, 2011)**.

Moisture content is also associated with the seaweed's harvest age—longer harvest periods tend to result in higher moisture content **(Harun, 2013)**. In this study, the harvest age of *Eucheuma cottonii* in both locations was the same, 45 days. Analysis of variance showed that there was a highly significant interaction between water location and KOH concentration on the moisture content of carrageenan.

According to **Desiana and Hendrawati (2015)** and **Gerung (2019)**, moisture content is also influenced by extraction time. Extended extraction times tend to reduce moisture content due to the enhanced capability of alkali to extract the seaweed matrix more thoroughly. Furthermore, **Ningsih (2014)** reported that higher alkali concentrations lead to lower moisture levels. This is attributed to the alkaline nature of the solution, which acts as an inhibitor to certain molecular interactions within the carrageenan structure, thus reducing its capacity to retain water.

These findings are consistent with the data in this study, which showed a decrease in moisture content with increasing KOH concentration, suggesting that alkaline concentration plays a significant role in optimizing carrageenan quality.

Ash content

The ash content of carrageenan extracted from *Eucheuma cottonii* at two different KOH concentrations (0.2% and 0.5%) is presented in Table (1). The results show a clear increase in ash content with the rise in KOH concentration for both sampling locations, Wael and Nuruwe.

At a KOH concentration of 0.2%, the ash content of carrageenan from Wael was 14.18%, while that from Nuruwe was slightly lower at 12.64%. When the KOH concentration was increased to 0.5%, the ash content rose to 17.67% in Wael and reached 17.19% in Nuruwe. These findings indicate that higher concentrations of KOH in the extraction process may enhance the release of mineral components into the carrageenan structure, thus increasing the total ash content.

The difference in ash content between the two locations at the same KOH concentration may be attributed to variations in environmental conditions, such as water quality, nutrient availability, and substrate composition, which can affect the biochemical composition of the seaweed. However, the overall trend in both locations suggests a consistent effect of KOH concentration on ash content levels.

These results align with previous studies which report that higher alkali treatment levels can increase ash content in carrageenan due to more extensive breakdown of cell wall structures, allowing more inorganic substances to be retained in the final product.

The results of this study indicate that the ash content percentage of carrageenan increases in line with the rising concentration of the alkaline solvent (KOH) used during extraction. This finding is consistent with the statement of **Ega *et al.* (2016)**, who reported that the use of alkaline solvents such as KOH enhances the ash content due to the increasing presence of potassium ions (K^+) that react with carrageenan during the extraction process. Similarly, **Ningsih (2014)** noted that the use of alkali solvents containing sodium (Na) or potassium (K) can influence the ash content, as these elements bind to the carrageenan during extraction.

The increase in ash content is not solely affected by the concentration of alkali used but may also be influenced by other factors. According to **Wenno *et al*. (2012)** and **Asikin *et al.* (2019)**, the ash content of carrageenan tends to rise with the age of seaweed at harvest. Older seaweed has a greater capacity for mineral absorption, which contributes to higher ash content. In addition, the sampling location plays a role in ash level variation. **Kreckhoff *et al*. (2015)** and **Supriyantini *et al*. (2017)** emphasized that environmental conditions, particularly the mineral composition of the growing waters, can result in different ash content values in carrageenan.

Ash content is influenced by the abundance of mineral elements and the salinity level of the surrounding water **(Syukri *et al*., 2007)**. In this study, the salinity of waters at Nuruwe was higher (33 ppt) compared to Wael (31.6 ppt). It is presumed that macro elements (N, P, K, Mg, Ca, S) and micro elements (Cu, Zn, Fe, Mn, Cl) in the waters of Nuruwe are absorbed more efficiently by the seaweed, contributing to an increased ash content in the extracted carrageenan. **Supriyantini *et al*. (2017)** further stated that seaweeds have a strong capacity to absorb minerals directly from their environment.

Sulfate content

Sulfate content is an important parameter used to determine the type and quality of polysaccharides found in red algae, and it plays a significant role in evaluating the final quality of carrageenan products **(Husna *et al*., 2016)**. Several studies have reported that increasing the concentration of KOH during the extraction process leads to a decrease in sulfate content. **Julaika *et al*. (2017)** found that the use of higher KOH concentrations (6–8%) significantly reduced sulfate levels in carrageenan. Similarly, **Distantina *et al*. (2009)** observed a reduction in sulfate content with increasing KOH concentrations (0.1–0.3 N). **Arto *et al*. (2021)** also reported that the application of alkaline solutions leads to a decrease in sulfate content in carrageenan.

In the present study, the sulfate content of carrageenan extracted from *Eucheuma cottonii* collected in Nuruwe was 41.51% at 0.2% KOH and decreased to 35.94% at 0.5% KOH. In contrast, carrageenan from Wael exhibited sulfate contents of 30.41% and 25.54% at 0.2% and 0.5% KOH, respectively (Fig. 3). These results confirm that higher KOH concentrations are associated with lower sulfate content.

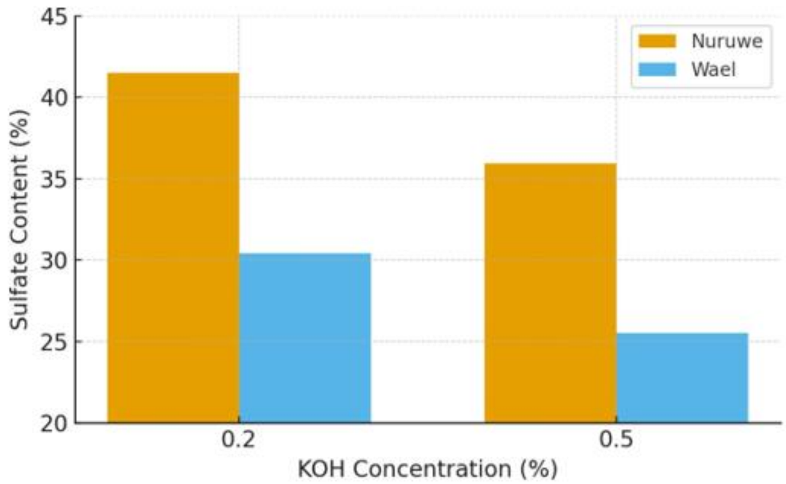

Fig. 3. Sulfate content of carrageenan at different KOH concentrations ($P < 0.05$)

The sulfate content values obtained in this study meet the quality standard set by the FAO, which ranges from 15 to 40%. The observed decrease in sulfate concentration with increasing KOH is likely due to the ability of KOH to bind potassium ions to ester groups while releasing one of the sulfate groups. The release of sulfate groups results in the formation of carrageenan composed of potassium, sodium, magnesium esters, and potassium sulfate, along with galactose and 3,6-anhydro-galactose in a linear chain, producing the compound K_2SO_4. The release of K_2SO_4 during extraction leads to the reduction in total sulfate content in carrageenan **(Winarno, 1996; Julaika *et al*., 2017)**.

Fig. (3) demonstrates that an increase in alkali solution concentration results in a corresponding decrease in sulfate content. This finding aligns with **Oliveira *et al.* (2020)**, who stated that one of the functions of alkaline solutions in carrageenan extraction is to catalyze the removal of 6-sulfate groups from the monomer units by forming 3,6-anhydrogalactose. This implies that KOH solution plays a role in reducing the sulfate content in carrageenan, which subsequently affects its gelling ability.

The increase in KOH concentration leads to a reduction in sulfate levels because KOH facilitates the binding of potassium ions to ester groups, thereby releasing one of the sulfate groups. The released sulfate group contributes to the formation of carrageenan compounds consisting of potassium, sodium, and magnesium esters, along with potassium sulfate. These compounds are structured with galactose and 3,6-anhydrogalactose units in a linear chain, ultimately resulting in the formation of K_2SO_4.

The release of K_2SO_4 during extraction is responsible for the observed decrease in sulfate content as KOH concentration increases **(Winarno, 1996; Julaika *et al*., 2017)**.

In addition to KOH concentration, the duration of extraction also significantly influences the sulfate content. **Hidayah *et al.* (2013)** reported that prolonged extraction time with KOH further reduces sulfate levels in carrageenan. This is supported by **Distantina *et al*. (2009)**, who noted that longer extraction periods tend to result in lower sulfate content. This is likely due to the more complete elimination of sulfate groups over time, leading to more thorough sulfate removal during the extraction process.

Viscosity of carrageenan from *E. cottonii*

Viscosity is employed as a key parameter to determine the thickness of a dynamic fluid. It can also be defined as the molecular flow resistance within a solution system. Viscosity testing is conducted to evaluate the thickness level of carrageenan as a solution under specific concentrations and temperatures. In this study, viscosity was measured at 75°C with a carrageenan concentration of 1.5% **(FAO, 2007)**.

The viscosity values obtained in this research showed that carrageenan from Nuruwe waters, with KOH concentrations of 0.2% and 0.5%, had viscosities of 44.37 cP and 40.92 cP, respectively. In contrast, carrageenan from Wael waters, under the same KOH concentrations, exhibited viscosities of 34.49 cP and 23.34 cP (Table 1). The analysis of variance indicated that the interaction between sampling location and KOH concentration significantly affected the viscosity values.

In general, carrageenan viscosity decreased with increasing concentrations of the alkaline KOH solution. This decline in viscosity is attributed to the reduction in sulfate content. According to **Oliveira *et al*. (2020)**, carrageenan viscosity is influenced by several factors, including carrageenan concentration, temperature, dispersion level, sulfate content, and molecular weight.

The viscosity values of carrageenan obtained in this study still comply with the standards established by FAO. **Ega *et al*. (2016)** reported that increased viscosity in carrageenan correlates with higher alkali concentrations, which enhance the solubility of mineral salts in seaweed. This finding is supported by studies from **Husna *et al*. (2016)** and **Nosa *et al*. (2020)**, who observed that increased fluid viscosity is caused by a reduction in carrageenan particle size due to elevated alkali concentration.

Other studies, including those by **Kumayanjati and Dwimayasanti (2018)** and **Arzani *et al*. (2020)** have also shown that extraction temperature and sampling location can influence carrageenan viscosity.

Gel strength

Gel strength, also known as breaking force, is defined as the maximum load required to rupture the polymer matrix under applied pressure. This parameter reflects the quality of carrageenan in forming a gel structure—i.e., its ability to transition from a liquid to a gel state.

Gel strength is closely related to the sulfate content in carrageenan. A faster extraction process tends to increase sulfate content but may lead to a reduction in gel strength. Conversely, prolonged extraction, as reported by **Arzani *et al*. (2020)**, not only enhances the formation of 3,6-anhydrogalactose but also reduces the amount of sulfate groups. This is consistent with **Siregar *et al*. (2016)**, who found that a reduction in sulfate groups could induce cross-linking modifications between polymer chains, thereby increasing gel strength.

Gel strength is not only influenced by harvest age and extraction time. Several other factors, such as alkali concentration and extraction temperature, also significantly affect gel strength **(Supriyantini *et al*., 2017; Kumayanjati & Dwimayasanti, 2018)**.

Previous studies indicate that the gel strength values of carrageenan extracted from Indonesian seaweeds generally do not meet the FAO standard, which is set at >500 g/cm².

In this study, the gel strength of carrageenan extracted from two marine locations—Nuruwe and Wael—was evaluated under two different KOH concentrations: 0.2% and 0.5%. The results showed that the highest gel strength was recorded from the Wael site at 0.2% KOH, reaching 322.03 g/cm². Conversely, the lowest value was observed at the Nuruwe site with 0.5% KOH, at 228.19 g/cm² (Table 1). These findings indicate that not only the geographic origin of the seaweed but also the concentration of alkali used during extraction plays a significant role in determining carrageenan gel characteristics.

A general trend of decreasing gel strength was observed as the KOH concentration increased from 0.2% to 0.5% at both locations. This phenomenon is likely attributed to the reduction of sulfate groups caused by higher alkali exposure, which in turn may impair the ability of carrageenan to form cross-links between polymer chains. This aligns with previous findings which suggest that an optimal level of sulfate groups is essential for maintaining gel network integrity and promoting the formation of a stable three-dimensional structure.

Although the gel strength values obtained in this study fall below the FAO standard of >500 g/cm², these results offer a preliminary insight into the potential of local seaweed resources for carrageenan production. Optimization of processing parameters such as extraction time, temperature, and pH may enhance gel strength outcomes. Moreover, marine locations such as Wael, which yielded comparatively higher gel strength, could serve as promising sources for the development of high-quality carrageenan production in the future.

Carrageenan functional group testing

The FTIR spectroscopic analysis revealed the presence of key functional groups characteristic of carrageenan extracted from *Eucheuma cottonii* collected from Nuruwe and Wael waters under two different KOH concentrations (0.2% and 0.5%). At Nuruwe with 0.2% KOH, the carrageenan exhibited sulfate ester (S=O) absorption at 1259.52 cm^{-1}, glycosidic linkage (C–O–C) at 1045.42 cm^{-1}, 3,6-anhydro-D-galactose (C–O) at 931.62 cm^{-1}, D-galactose-4-sulfate (C–O–SO_3 at C_4) at 846.68 cm^{-1}, and hydroxyl (–OH) groups at 3255.84 cm⁻¹. For the 0.5% KOH treatment in the same location, only the sulfate ester and glycosidic bond peaks were observed, at 1259.52 cm^{-1} and 1035.77 cm^{-1}, respectively, indicating possible degradation or modification of other functional groups at higher alkali concentration (Table 2).

Table 6. FTIR absorption bands of carrageenan extracted from *Eucheuma cottonii* with different KOH concentrations and locations

Sample	Ester Sulfate (S=O)	Glycosidic Bond (C–O–C)	3,6-anhydro-D-galactose (C–O)	D-galactose-4-sulfate (C–O–SO_3 at C_4)	Hydroxyl Group (–OH)
Nuruwe KOH 0.2%	1259.52	1045.42	931.62	846.68	3255.84
Nuruwe KOH 0.5%	1259.52	1035.77	—	—	—
Wael KOH 0.2%	1269.45	1045.42	931.62	848.68	3161.33
Wael KOH 0.5%	1259.2	—	—	—	—
Tual, Southeast Maluku (Lewerissa, 2005)	1238.2	1068.5	929.6	848.6	—
Sumenep, Madura (Andarini, 2011)	1222.87	1029.99	1070	852.54	3201.83
Aceh (Nasution, 2019)	1246.9	1074	934	—	3382

Similarly, carrageenan extracted from *E. cottonii* in Wael waters with 0.2% KOH exhibited similar functional group patterns. The sulfate ester group was identified at 1269.45 cm^{-1}, glycosidic linkage at 1045.42 cm^{-1}, 3,6-anhydro-D-galactose at 931.62cm^{-1}, D-galactose-4-sulfate at 848.68cm^{-1}, and hydroxyl groups at 3161.33cm^{-1}. In contrast, the sample treated with 0.5% KOH showed only the sulfate ester absorption band at 1259.2cm^{-1}, suggesting that excessive alkali concentration might have disrupted the structural integrity of certain key moieties.

Based on the presence of the characteristic absorption bands—particularly the strong signal around 930cm^{-1} indicative of 3,6-anhydrogalactose—it can be concluded that the extracted carrageenan belongs to the κ-carrageenan (kappa-carrageenan) type. The observed spectral patterns are consistent with those previously reported for κ-carrageenan, confirming its structural identity. These results are also summarized in Table (6), which highlights the functional group distributions across different treatments and supports the classification and structural integrity of the extracted polysaccharides.

CONCLUSION

The carrageenan extracted from *Eucheuma cotonii* using 2% KOH and isopropyl alcohol (IPA) precipitation demonstrated optimal physicochemical properties, particularly in samples from the Nuruwe waters. At a 0.2% carrageenan concentration, the Nuruwe sample exhibited higher yield (22.53%), greater gel strength (322.03 g/cm^2), higher viscosity (44.37 cps), and sulfate content (41.51%) compared to the Wael sample.

FTIR analysis confirmed the presence of key functional groups—sulfate esters, glycosidic linkages, 3,6-anhydro-D-galactose, D-galactose-4-sulfate, and hydroxyl groups—in both samples, with slightly different wavenumber profiles. These findings suggest that the Nuruwe coastal area provides more favorable conditions for producing high-quality carrageenan from *E. cotonii*.

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