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Biochemical Profile of Macroalgae: Analysis of Primary Metabolites and Hydrocolloid Content in Oenaek Waters During the Dry Season

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ABSTRACT

Macroalgae are important components in coastal ecosystems, acting as primary producers, oxygen producers through photosynthesis, supporting biodiversity, and providing habitat for marine organisms. In addition to their ecological functions, macroalgae have great economic potential due to their bioactive compound content, including hydrocolloids and nutrient metabolites, which are utilized in the food, pharmaceutical, and cosmetic industries. Biochemical content such as protein, carbohydrates, and hydrocolloids in macroalgae varies greatly depending on species and environmental conditions, so site-specific research is needed. This study analyzed primary metabolites and hydrocolloid content of ten macroalgae species collected from the waters of Oenaek Village during the dry season. The parameters tested included water, ash, fat, protein, carbohydrate, and agar (in red algae) and alginate (in brown algae) content. The results showed that the water content ranged from 1.51% to 29.16%, ash ranged from 0.05%-0.58%, fat ranged from 5.4%–28%, protein ranged from 1.82%–8.54%, and carbohydrates ranged from 50.3%-87.92%. The highest agar content was found in Gracilaria gracilis (19.6%) and the highest alginate in Sargassum polycystum (33.96%). Green algae do not contain hydrocolloids but have the highest carbohydrate content. The biochemical profile indicates that macroalgae from Oenaek have great potential for industrial applications, especially as a source of hydrocolloids and nutritional compounds.

INTRODUCTION

Macroalgae are autotrophic organisms that play an important role in maintaining the balance of aquatic ecosystems. As primary producers, macroalgae function as the basis of the food chain in aquatic environments and make a major contribution to primary productivity in coastal areas (Reeder, 2017). In addition to acting as producers, macroalgae also provide habitat and protection for various types of marine organisms, thus supporting biodiversity in

waters (Andirasdini *et al.*, 2023). The existence of macroalgae provides benefits not only from an ecological perspective, but also from an economic perspective. Photosynthesis activity in macroalgae produces oxygen which is very much needed to maintain water quality, and contains various nutritional compounds that make it potential as a food ingredient or a source of active ingredients in the health sector (Amaranggana & Wathoni, 2017). In addition, several types of macroalgae have also been used as raw materials for industry, food, natural preservative products, and even raw materials for medicines (Marianingsih *et al.*, 2013).

One of the regions in Indonesia that has great potential in the production and utilization of macroalgae is the Province of East Nusa Tenggara (NTT). This region, especially Kupang Regency, has developed intensive macroalgae cultivation since the early 2000s, and until now it has become the main source of livelihood for most coastal communities (Turupadang *et al.*, 2021). Although NTT has steep topography and a semi-arid climate that is less supportive of land farming activities, this geographical condition is actually an advantage for the development of mariculture businesses. The coastal waters in this region are generally clear throughout the year, so they are very supportive of macroalgae growth (Oedjoe *et al.*, 2019). However, the survival and productivity of macroalgae are greatly influenced by various environmental factors, such as substrate type, current, temperature, salinity, light intensity, pH, and nutrient availability.

Macroalgae are generally classified into three large groups, namely green algae (Chlorophyta), red algae (Rhodophyta), and brown algae (Phaeophyta), based on the dominant pigments they contain (Dawes, 1981). In addition to its ecological value, macroalgae also have high economic value due to its bioactive compound content, one of which is hydrocolloid. Hydrocolloid is a group of polysaccharides that have the ability to form gels or thick solutions in water, so they are widely used in various industries, including food, pharmaceuticals, and cosmetics. Agar derived from red algae such as *Gracilaria* and *Gelidium*, has the ability to form gels that can be dissolved again when heated (thermally reversible), while alginate from brown algae such as *Sargassum* forms a stable gel and does not melt (Bixler & Porse, 2010). Green algae such as *Caulerpa* are also increasingly consumed as functional foods due to their high nutritional content (Holdt & Kraan, 2011).

In addition to hydrocolloids, macroalgae are also rich in primary metabolites, namely basic compounds such as water, ash, protein, fat, and carbohydrates, which play an important role in the metabolism and growth of macroalgae themselves. The content of these proximates varies between species and can be influenced by the environmental conditions where macroalgae grow (Kurniawati *et al.*, 2024). Environmental factors such as temperature, light, and nutrients are known to greatly affect the levels of primary metabolites in macroalgae tissue (Holdt & Kraan, 2011). Therefore, it is important to study the biochemical content of macroalgae in depth, especially in areas with high potential such as the waters of Oenaek, Kupang Regency, especially during the dry season when environmental conditions are relatively stable.

METHODS

Location and Time of Research

This research was conducted in June and July 2024 in Oenaek Village, West Kupang District, Kupang Regency, East Nusa Tenggara Province. Sampling was carried out in 5 waters, namely coordinate point one at latitude -10.281849° and longitude 123.497128°, coordinate point two at latitude -10.281017° and longitude 123.496173°, coordinate point three at

Demoor *et al.* (2025)

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latitude -10.279810° and longitude 123.495145°, coordinate point four at latitude -10.282240° and longitude 123.495101°, and coordinate point five at latitude -10.279567° and longitude 123.495187°. The samples were then tested in the Fisheries Laboratory of the Faculty of Marine Animal Husbandry and Fisheries, and the Chemistry Education Laboratory of the Faculty of Teacher Training and Education, Nusa Cendana University.

Tools and Materials

The tools used in this study include buckets, sample plastic, refractometers, pH meters, thermometers, Dissolved Oxygen (DO) meters, analytical scales, centrifuges, cameras, stationery, scissors, hot plates, stirrers/spatulas, Erlenmeyer flasks, beakers, filter cloths, petri dishes, blenders, furnaces, magnetic stirrer bars, filters, aluminum foil, refrigerators/freezers, and GPS cameras. And the materials used in this study include macroalgae obtained from the research location, HCL, 96% ethanol, NaOH solution, 4% KOH solution, distilled water, 3% Na₂CO₃ solution, NaOCL solution, and methanol solution.

Research Design

Data were obtained through macroalgae sampling in the waters of Oenaek Village. Hydrocolloid analysis was conducted at the Fisheries Laboratory of FPKP, Nusa Cendana University, and primary metabolite analysis at the Chemistry Education Laboratory of FKIP, Nusa Cendana University. Supporting data were obtained from the results of water quality measurements at the research location. The data collection technique was carried out by purposive sampling, namely the selection of locations and species was carried out deliberately based on research objectives. Furthermore, survey and snowball sampling methods were used to expand sampling points, based on field information, fishermen, journals relevant to the research topic and the existence of macroalgae populations in coastal areas. Snowball sampling was applied to reach macroalgae population locations that were not easily found directly, by relying on information from the initial sampling point to subsequent points that had ecological similarities. This is in line with the opinion of (Naderifar et al., 2017), which stated that snowball sampling is effective when the target population is difficult to reach using conventional methods. Samples were taken directly from predetermined locations in the waters of Oenaek, with the help of a GPS camera to map the coordinate points. A total of 10 species of macroalgae were successfully collected for further analysis, and data analysis was carried out qualitatively with descriptive descriptions. Laboratory test data were arranged in the form of tables and graphs, then described narratively.

Research Procedures

The stages that will be carried out during the research include the first is determining the location of the research. After the location is determined, preparation and collection of tools and materials needed to support the smooth running of the research process are carried out. Next, samples of macroalgae found at the location are taken. After being taken, the macroalgae are cleaned, identified, and documented before being put into sample plastic. At this stage, measurements of water quality parameters are also carried out, such as temperature, pH, dissolved oxygen, and salinity. In this study, there were 10 species of macroalgae consisting of red, brown, and green macroalgae. The next stage is sample handling, namely by drying the macroalgae in the sun for 2 to 3 days. After drying, the samples are stored and taken to the laboratory to be analyzed for primary metabolite content and hydrocolloid content.

Data Analysis

The data will be analyzed qualitatively and described in descriptive form in the form of tables, graphs, images and narratives to describe the levels of primary metabolites and hydrocolloids in the dry season.

RESULTS

Primary Metabolite Content of Macroalgae in the Waters of Oenaek Village

The waters of Oenaek Village have quite high macroalgae diversity. Based on the survey results, 10 species of macroalgae were found from three large groups, namely red algae (Rhodophyta), brown algae (Phaeophyta), and green algae (Chlorophyta). The macroalgae species found during the study were spread across 5 locations in the Oenaek waters. The different distribution patterns at each sample point indicate that each location has unique habitat conditions. This difference is related to the environmental factors of the waters that affect the number and types of macroalgae that grow. Factors such as temperature, salinity, type of water base, and other physical-chemical conditions also determine the variation of species found at each point (Hurrey et al., 2013; Han & Liu, 2014). The ten species were then analyzed for their primary metabolite content in the laboratory. Primary metabolites are the chemical composition or nutritional content in a material that includes carbohydrates, proteins, fats, water content, and ash content. The results of the analysis of primary metabolite content in macroalgae found in the waters of Oenaek Village are presented in Table 1.

Table 1. Primary Metabolite Content of Macroalgae in Oenaek Waters

No	Macroalgae Species cies of Red Mac	Image of Macroalgae Species	Water Content (%)	Ash Content (%)	Fat Content (%)	Protein Content (%)	Carbohydrate Content (%)
1	Hypnea	Li Gaigae	6.19	0.42	17.8	7.56	68.03
_	musciformis		0.13	0.42	17.8	7.50	08.03
2	Gracilaria gracilis		9.44	0.21	12.2	5.74	72.41
3	Gracilaria salicornia		16.32	0.3	8	4.76	70.62

Species of Brown Macroalgae

Demoor *et al.* (2025)

https://doi.org/10.29303/jfh.v5i3.7020

No	Macroalgae Species	Image of Macroalgae Species	Water Content (%)	Ash Content (%)	Fat Content (%)	Protein Content (%)	Carbohydrate Content (%)
4	Sargassum polycystum		2.28	0.58	6.6	8.54	80.6
5	Dictyota dichotoma		3.04	0.41	5.4	7.14	84.01
6	Padina australis		16.32	0.35	8	3.08	72.25
Spec	cies of Green M	1acroalgae					
7	Halimeda macroloba		5.13	0.18	28	1.82	64.87
8	Caulerpa lentillifera		1.51%	0.1	8	2.38	87.92
9	Halimeda cylindracea		10.55	0.05	12	2.94	74.46
10	Caulerpa taxifolia		29.16	0.3	18	2.24	50.3

1. Water Content

Moisture content refers to the amount of water contained in a material, which also determines its freshness, storage resistance, and stability. The hygroscopic nature of macroalgae, or easily absorbing water from its surroundings, makes it susceptible to faster deterioration when stored in humid places.

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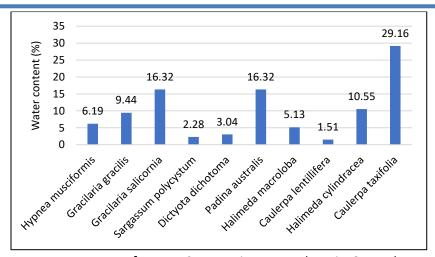


Figure 1. Percentage of Water Content in Macroalgae in Oenaek Waters

2. Ash Content

Ash content is the inorganic residue left after organic material is burned, and is usually used to indicate the amount of minerals contained in a material.

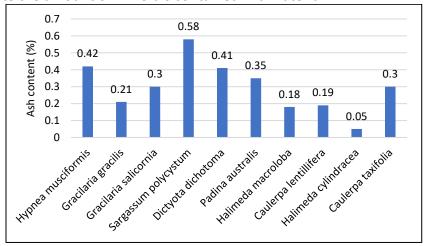


Figure 2. Percentage of Ash Content in Macroalgae in Oenaek Waters

3. Fat Content

Fat content refers to the total amount of lipids in a material, which serve as an energy source and structural component of cells.

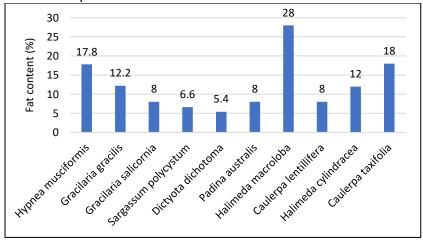


Figure 3. Percentage of Fat Content in Macroalgae in Oenaek Waters

4. Protein Content

Protein levels in macroalgae play a role in growth and metabolism, and are an indicator of the nutritional value of macroalgae.

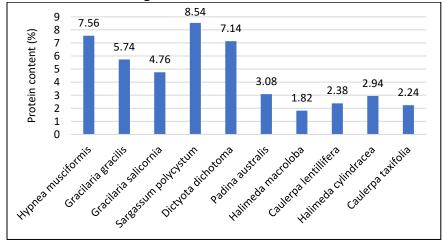


Figure 4. Percentage of Protein Content in Macroalgae in Oenaek Waters

5. Carbohydrate Content

Carbohydrate content is the percentage of carbohydrate content in a food ingredient, which functions as a source of energy and includes compounds such as starch and polysaccharides.

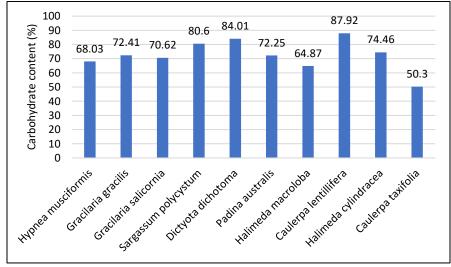


Figure 5. Percentage of Carbohydrate Content in Macroalgae in Oenaek Waters

Macroalgae Hydrocolloid Content in the Waters of Oenaek Village

Hydrocolloids are polysaccharide compounds that are found in abundance in macroalgae and have the property of forming gels or thick solutions when dissolved in water.

Table 2. Hydrocolloid Content of Macroalgae in Oenaek Waters

No	Macroalgae Species	Image of Macroalgae Species	Type of Hydrocolloid Tested	Content (%)		
Speci	Species of Red Macroalgae					

No	Macroalgae Species	Image of Macroalgae Species	Type of Hydrocolloid Tested	Content (%)
1	Gracilaria gracilis		Agar	19.6
2	Gracilaria salicornia		Agar	12.6
3	Hypnea musciformis		Agar	15.2
Spec	cies of Brown Macroalgae			
4	Sargassum polycystum		Alginate	33.96
5	Dictyota dichotoma		Alginate	12.24
6	Padina australis		Alginate	3.00

Based on the data in Table 2, the analysis of hydrocolloid content in the form of agar and alginate was carried out on six species of macroalgae from the red and brown groups. Meanwhile, four species of green macroalgae were not tested because they generally do not contain hydrocolloid compounds such as agar and alginate. This is in line with the opinion of Holdt & Kraan (2011) who stated that red macroalgae, such as *Gracilaria* and *Gelidium*, are the main sources of agar, brown macroalgae such as *Sargassum* are rich in alginate. While green macroalgae, such as *Caulerpa*, are more widely consumed as functional foods because of their high nutritional content.

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1. Agar

Agar is a polysaccharide extracted from red macroalgae (Rhodophyta), especially from the genera *Gracilaria*, *Gelidium*, and *Hypnea*. This compound has the ability to form strong gels, so it is widely used in various industries, such as food, pharmaceuticals, and microbiology.

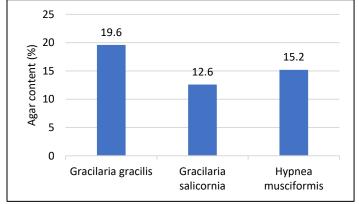


Figure 6. Percentage of Agar Content in Macroalgae in Oenaek Waters

2. Alginate

Alginate is a natural polysaccharide extracted from the cell walls of brown macroalgae (Phaeophyceae), especially from the genera *Sargassum*, *Laminaria*, and *Macrocystis*. Alginate is known as a water-soluble hydrocolloid and has the ability to form a gel, so it is widely used in various industries.

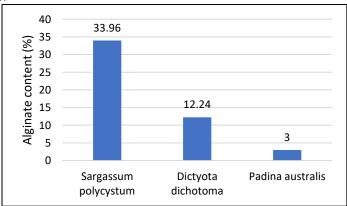


Figure 7. Percentage of Alginate Content in Macroalgae in Oenaek Waters

DISCUSSION

Primary Metabolite Content of Macroalgae in the Waters of Oenaek Village

1. Water Content

Based on the analysis results, the water content of macroalgae species found in Oenaek waters varies between 1.51% and 29.16%. *Caulerpa taxifolia* showed the highest water content of 29.16%, while the lowest content was found in *Caulerpa lentillifera* at 1.51%. In general, green macroalgae have a higher water content compared to brown or red macroalgae. This difference can be caused by the type of species, different cell structures, fiber content, and environmental conditions such as temperature and water salinity. Macroalgae with high water content usually have a softer and wetter texture, while those with low water content tend to have a denser structure or more fiber. In addition, the drying method before analysis also affects the water content of macroalgae. Tapotubun (2018) explained that the drying process greatly affects the water content; drying with a longer time and low temperature

Demoor et al. (2025)

https://doi.org/10.29303/jfh.v5i3.7020

allows water to evaporate slowly, so that the remaining water content is lower than direct drying in the sun.

2. Ash Content

Based on the analysis results, the ash content of macroalgae in Oenaek waters ranges from 0.05% to 0.58%. The species with the highest ash content is *Sargassum polycystum* (0.58%), while the lowest is *Halimeda cylindracea* (0.05%). The low ash content indicates that most of these species have relatively low mineral content. Macroalgae with high ash content usually have higher content of important minerals such as calcium, magnesium, and potassium. Therefore, macroalgae like this are more suitable for use in non-food industries, such as pharmaceuticals, cosmetics, and fertilizers (Astriani et al., 2024). Conversely, macroalgae with low ash content are preferred for food ingredients because of their more neutral taste and better texture (Alwahida *et al.*, 2023).

3. Fat Content

Based on the analysis results, the fat content in macroalgae varies between 5.4% and 28%, with *Halimeda macroloba* having the highest fat content (28%) and *Dictyota dichotoma* the lowest (5.4%). Research by Wajdia (2024) showed variations in fat content in the *Caulerpa* species in the Spermonde Islands, indicating that species differences also affect the fat content in the macroalgae. Differences in fat content between species are also influenced by genetic and environmental factors, such as temperature, salinity, and light intensity, which play a role in lipid biosynthesis (Novianti & Arisandi, 2021). In addition, post-harvest drying methods, such as wind drying, are better at maintaining lipid components than direct drying in sunlight (Tapotubun, 2018).

4. Protein Content

Based on the analysis results, the protein content in macroalgae ranges from 1.82% to 8.54%, with *Sargassum polycystum* having the highest protein content (8.54%) and *Halimeda macroloba* the lowest (1.82%). Protein content is influenced by environmental conditions such as nutrient availability and water quality, as well as genetic factors between species. In general, brown macroalgae have higher protein content, in accordance with Handayani (2019) who stated that protein content in brown macroalgae is in the range of 5–11%. This variation is also influenced by temperature, salinity, light intensity, and post-harvest processing techniques.

5. Carbohydrate Content

Based on the analysis results, the carbohydrate content in macroalgae in Oenaek waters ranges from 50.3% to 87.92%, making it the largest component in primary metabolites of macroalgae. *Caulerpa lentillifera* has the highest carbohydrate content (87.92%), while *Caulerpa taxifolia* has the lowest (50.3%). This variation is influenced by species characteristics and environmental conditions such as nutrients, temperature, depth, seasonal changes, and geographical location (Yuan, 2009). Research by Novianti & Arisandi (2021) states that carbohydrate levels in brown macroalgae can be influenced by light intensity and nutrient availability, which impacts carbohydrate biosynthesis in algal tissue.

Macroalgae Hydrocolloid Content in the Waters of Oenaek Village

1. Agar

Based on the analysis results, variations in agar content in several macroalgae species in Oenaek waters, with *Gracilaria gracilis* (19.6%), followed by *Hypnea musciformis* (15.2%), and the lowest *Gracilaria salicornia* (12.6%). These data indicate variations in agar levels between species. Variations in agar content can be caused by several factors such as differences in species, and environmental factors, such as season, habitat, and extraction methods used (Chapman & Chapman, 1980).

Demoor et al. (2025)

https://doi.org/10.29303/jfh.v5i3.7020

2. Alginate

Based on the analysis results, brown macroalgae species in Oenaek waters, such as *Sargassum polycystum* (33.96%), have the highest alginate content, followed by *Dictyota dichotoma* (12.24%), and the lowest is *Padina australis* (3.00%). Alginate content is influenced by species variation, environmental factors such as temperature, salinity, light intensity, nutrient availability, and extraction methods (Salosso, 2019). Previous studies have shown that environmental factors and extraction techniques, such as Microwave-Assisted Extraction (MAE), can affect the yield and quality of alginate produced, as found in *Padina sp.* with the highest yield of 26.83% (Pasaribu *et al.*, 2020).

CONCLUSION

The macroalgae species found in the waters of Oenaek exhibit significant biochemical diversity, with primary metabolite content varying considerably among species. Water content ranged from 1.51% in Caulerpa lentillifera to 29.16% in Caulerpa taxifolia, ash content from 0.05% in Halimeda cylindracea to 0.58% in Sargassum polycystum, fat content from 5.4% in Dictyota dichotoma to 28% in Halimeda macroloba, protein content from 1.82% in Halimeda macroloba to 8.54% in Sargassum polycystum, and carbohydrate content from 50.3% in Caulerpa taxifolia to 87.92% in Caulerpa lentillifera. Hydrocolloid analysis showed that red algae such as Gracilaria gracilis had the highest agar yield at 19.6%, followed by Hypnea musciformis (15.2%) and Gracilaria salicornia (12.6%). Meanwhile, brown algae demonstrated considerable alginate content, with Sargassum polycystum showing the highest at 33.96%, followed by Dictyota dichotoma (12.24%) and Padina australis (3.00%). These findings indicate that Sargassum polycystum is the most promising species for alginate extraction, and Gracilaria gracilis for agar production. Although green algae do not contain hydrocolloids, their high primary metabolite content—particularly carbohydrates in Caulerpa lentillifera underscores their potential as functional food ingredients. Overall, macroalgae from Oenaek waters offer strong potential for industrial applications in food, pharmaceuticals, and biobased materials.

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