

CHARACTERIZATION OF A PRODUCT-DEVELOPED FROM THE *Ulva reticulata*(RIBBON SEA LETTUCE)EXTRACT

ABSTRACT

Humans have a strong reliance in using petroleum-based plastics which take several decades to degrade and cause a lot of environmental problems such as pollution. This study intended to develop bioplastics from *Ulva reticulata* (Ribbon sea lettuce) and to determine the bioplastic's physicochemical properties. The sample was collected in Allen, Northern Samar, Philippines, water samples were also collected. After the extraction, development of bioplastic from the sample extract commenced. The developed bioplastic underwent several test to check the stability of the product – its includes physicochemical analysis, tensile strength, thickness, moisture content, and soil degradation test. The Ribbon sea lettuce bioplastic solution was slightly acidic. The seaweed bioplastic have an average thickness of 0.30mm. The Ribbon sea lettuce bioplastic sample was both insoluble in the three solvents. The sample bioplastic can handle an average load of 55.12g. An average moisture content of 51.534% have been observed in the three trial of the seaweed bioplastic sample. The seaweed bioplastic sample naturally degraded during the soil biodegradation test and have lost an average weight percentage of 82.05% after 21 days of being buried in soil. The results showed that the seaweed bioplastic has a potential as an alternative to the non-biodegradable plastic and can be used in agricultural, industrial and economic purposes.

Keywords: Bioplastic, environmental sustainability, physicochemical properties, soil degradation test, *Ulva reticulata*

I. INTRODUCTION

Petroleum-based plastics have become pervasive in modern life despite their well-documented environmental harm. Their slow decomposition and release of toxic chemicals pose significant threats to ecosystems worldwide. In response, research has turned to bioplastics, derived from renewable biomass sources such as plants, bacteria, and seaweeds. Seaweeds, rich in polysaccharides, offer a promising avenue for bioplastic production due to their biodegradability and eco-friendliness.

Among seaweeds, *Ulva reticulata*, commonly known as Ribbon Sea lettuce, stands out for its potential in bioplastic synthesis. This green macroalga proliferates in coastal areas, often causing disruptions in ecosystems if left unchecked (Abu *et al.*, 2022). Rather than allowing it to degrade and disturb environmental balance, this study explores utilizing *Ulva reticulata* as a raw material for bioplastic production.

Ulva species, belonging to the genus Ulvaceae, have diverse applications ranging from food supplements to biomedical uses. *Ulva*'s key component, soluble fiber ulvan, possesses film-forming properties crucial for bioplastic manufacturing. Combined with its cellulose content, *Ulva* sp. presents a compelling option for bioplastic development, offering an environmentally

friendly, cost-effective, and non-toxic alternative to traditional plastics (Wilmar *et al.*, 2020; Kidgell *et al.*, 2019; Lim *et al.*, 2021).

This study focuses on harnessing *Ulva reticulata*'s potential to produce bioplastics, aiming to contribute to sustainable plastic alternatives while mitigating the environmental impact of seaweed proliferation. Through careful analysis and experimentation, we aim to elucidate *Ulva*'s viability as a source of bioplastic material, paving the way for eco-friendly solutions to plastic pollution.

II. METHODOLOGY

Collection of Samples

The *Ulvareticulata* (Ribbon sea lettuce) seaweed sample and the seawater samples were collected from Allen, Northern Samar, Philippines. The researcher developed bioplastics from the Ribbon sea lettuce and were tested to identify the physicochemical properties.

Preparation of the Seaweed Bioplastics

The *Ulva reticulata* (Ribbon sea lettuce) was washed to remove unwanted debris. The seaweed was air-dried for a couple of days to remove moisture. After which, the seaweed was heated in an oven for 10-20 minutes at 150°C and grinded to be in powdered form. For the formulation of the bioplastic, the researcher prepared a volume of glycerin, and a volume of polyvinyl alcohol solution to be dissolved in a distilled water. A small amount of gelatin powder was dissolved in a separate distilled water. The solution with glycerin and polyvinyl alcohol solution was heated in a hot plate for 5 minutes and the powdered sample was added and stirred continuously. The solution was heated and stirred for another minute before adding the gelatin solution and allowed the solution to boil. The solution was removed from the hot plate and was poured into the petri dishes with a thickness of 1cm. The solution in the petri dish was allowed to cool for 10-15 minutes until it solidifies and turn into a gel-like sample. The samples were heated in the oven for 30-40 minutes with a 5-minute interval at 150°C to remove excess moisture. The glycerin served as the plasticizer, the gelatin powder and the polyvinyl alcohol solution served as the binding agents in the seaweed bioplastic formulation. This process was based on the study of (Consebit *et al.*, 2022; Dagalea, Lim, 2018) with modifications made.

Water Quality Tests of the Seawater Samples

The temperature, the pH level, the total dissolved solids (TDS), and the salinity of the seawater samples from the location where the seaweed sample was collected were measured using a multiparameter water quality meter.

Physicochemical Characterization of the Seaweed Bioplastics

The pH level of the seaweed bioplastic solution was measured using a digital pH meter. Three trials were performed and the average mean was recorded as the final pH level. Three (3) evaluators optically inspected each seaweed bioplastic sample to identify its color. The observations of the evaluators were recorded.

The thickness of the seaweed bioplastic samples was measured using a micrometer caliper. The sample was 1 square inch in size. Five measurements were performed,

compromising the four edges and a center point. The average mean of the measurements was recorded as the final thickness of the samples. This procedure was based on the study of (Santana *et al.*, 2018) with some modifications made.

Solubility tests was performed to determine the ability of compounds to dissolve in a solvent, which is usually a liquid (Sapkota, 2022). The solubility of the bioplastic was determined against water, ethanol, and hexane. The seaweed bioplastic weighing 0.05g was added to 5mL of each solvent separately. The sample was observed to determine whether any of the seaweed bioplastic was soluble or insoluble in water, ethanol, or hexane.

Tensile tests measure the load required to break a plastic test sample and specimen stretch or elongation to that breaking load. The seaweed bioplastic was hooked with an improvised platform for putting weights. The weights were gradually placed on the platform that was hooked to the sample until it reaches its breaking point. This procedure was based on the study of Mesina (2019) and Caballa (2016).

Moisture content is simply, how much water is in a product. It influences the physical properties of a substance, including weight, density, viscosity, conductivity, and others. It is generally determined by weight loss upon drying (Mermelstein, 2009; Dagalea, Lim, 2021). The seaweed bioplastic sample was conditioned at room temperature for 5 days. Sample was weighed first before drying in an oven for 24 hours at 125°C. The sample was weighed after drying and the loss in weight was reported as moisture. (Official Methods of Analysis of the AOAC, 1984).

The degradation rate of the soil biodegradation test was calculated from weight loss of the sample over time. A direct way to measure the biodegradability of polymers is weight loss. This procedure was based on the study of Amin (2019). Seaweed bioplastic sample of 1 square inch was buried in the soil with a two-inch depth. The soil with seaweed bioplastic samples was placed in the laboratory. The degradation rate of the soil burial test was calculated from weight loss of the sample over time.

III. RESULTS AND DISCUSSION

Water Quality Tests of the Seawater Samples

A Multiparameter water quality meter measured all of the temperature of the seawater samples as 26.8°C. The average temperature of the sea surface is about 20° C (68° F), but it ranges from more than 30° C (86° F) in warm tropical regions to less than 0°C at high latitudes. In most of the ocean, the water becomes colder with increasing depth (Reommich, 2019).

A digital pH meter was used in determining the pH level of the seawater samples and three replications were performed. The average mean was recorded as the final pH.

Table 1. The pH Level of the Seawater Samples

| Seawater Samples | R ₁ | R ₂ | R ₃ | Average pH | Indication |
|--------------------|----------------|----------------|----------------|------------|-----------------------|
| Ribbon Sea Lettuce | 7.793 | 7.803 | 7.793 | 7.796 | <i>Slightly basic</i> |

Findings showed that the seawater samples from where the Ribbon sea lettuce was collected were all slightly basic. This is because the seawater's average pH typically ranges between 7.5 and 8.4, making it slightly basic.

Using the multiparameter water quality meter, the total dissolved solid (TDS) of the seawater samples was identified in parts per million or ppm. Triplications were conducted and the average mean (in ppm) of the measurements was recorded as the final TDS amount.

Table 2. The Total dissolved solids (TDS) in the Seawater Samples

| Seawater Samples | R ₁ (ppm) | R ₂ (ppm) | R ₃ (ppm) | Average TDS (ppm) |
|--------------------|----------------------|----------------------|----------------------|-------------------|
| Ribbon Sea Lettuce | 3100 | 2520 | 1029 | 5303 |

Using a multiparameter water quality meter and by performing triplications, the TDS of the seawater sample was determined. Results revealed that the seawater sample from where the Ribbon sea lettuce was gathered had an average TDS of 5303ppm. Freshwater by definition has less than 1500ppm TDS, while seawater was generally more than 5000ppm. If the water is below 1500ppm, it is fine since organic waste also raises TDS and breakdown of organic waste goes through intermediate stages that releases carbohydrates, lipids, proteins, nucleic acids, and others into the water column (Understanding TDS, 2019).

The salinity of seawater was also determined using a multiparameter water quality meter with three replications and the average salinity of the samples in parts per thousand (ppt) were determined.

Table 3 The Salinity of Seawater Samples

| Seawater Samples | R ₁ (ppt) | R ₂ (ppt) | R ₃ (ppt) | Average Salinity (ppt) |
|--------------------|----------------------|----------------------|----------------------|------------------------|
| Ribbon Sea Lettuce | 2.18 | 7.84 | 12.13 | 7.383 |

Salinity is simply the measure of dissolved salts in water. Salinity is usually expressed in parts per thousand (ppt). Coastal waters and surface waters of the ocean far from shore can be less salty than 35 ppt due to freshwater input from land or rain (Low *et al.*, 2020). The findings showed that the average salinity of the seawater samples was below the average salinity of seawater according to Low (2020). This was due to the location where the seaweeds were collected. The coastal area of where the seaweed was collected accumulates a lot of freshwater from land and streams connected to the sea. There are numerous households located near the coastal area which probably dispose most of their wastewaters to the sea. Furthermore, there are streams directly connected to the sea and during rainy season, the coastal waters turn brown in color due to the accumulation of muddy freshwater. These might be the causes why the salinity of the seawater near the coast of the sampling area below the average salinity based on Low (2020).

Table 4. Summary Water Quality Test Results of the Seawater Sample

| Seawater Sample | Temperature (°C) | Average pH Level | Average Total Dissolved Solids (TDS)(ppm) | Average Salinity (ppt) |
|--------------------|------------------|------------------|---|------------------------|
| Ribbon Sea Lettuce | 26.8 | 7.796 | 5303 | 7.383 |

Physicochemical Characterization of the Seaweed Bioplastics

pH level of 7 is neutral, below 7 measurements indicate acidity while above 7 measurements indicate basicity. A digital pH meter was utilized in identifying the pH levels of the Ribbon sea lettuce bioplastic solutions. Three replications were performed, and the average pH levels of the sample was recorded.

Table 5. The pH level of the Seaweed Bioplastic Solution

| Bioplastic Samples | R ₁ | R ₂ | R ₃ | Average pH | Indication |
|---------------------------|----------------|----------------|----------------|------------|------------------------|
| Ribbon Sea Lettuce | 6.80 | 6.77 | 6.89 | 6.82 | <i>Slightly acidic</i> |

Results showed that the Ribbon sea lettuce bioplastic solution were 6.82. According to the EHS Laboratories, a solution is considered hazardous when it has a pH level of less than or equal to 2 or greater than or equal to 12.5 because it is harmful to human and environmental health. The pH levels of the seaweed bioplastic solutions are not hazardous to humans and the environment.

Optical inspection was conducted by three (3) evaluators to identify the color of the seaweed bioplastic samples. The color of the ribbon sea lettuce bioplastic registered a brownish green color. The color of the seaweed bioplastic sample can be attributed to the pigments of the powdered seaweed.

Thickness of seaweed bioplastics was measured using a micrometer caliper at five different spots. The thickness value was measured from the average of five measurements of the seaweed bioplastics.

Table 6. Thickness of the Seaweed Bioplastic

| Bioplastic Samples | Spot 1 (mm) | Spot 2 (mm) | Spot 3 (mm) | Spot 4 (mm) | Center (mm) | Average Thickness (mm) |
|--------------------|-------------|-------------|-------------|-------------|-------------|------------------------|
| Ribbon Sea Lettuce | 0.28 | 0.31 | 0.32 | 0.30 | 0.29 | 0.30 |

According to European Plastic Converters, biodegradable plastics should have a minimum thickness of 0.016mm. The thickness of the seaweed bioplastic sample derived from Ribbon sea lettuce was at 0.30mm. The results showed that the seaweed bioplastic achieved the minimum thickness requirement based on the European Plastic Converters.

Solubility is the ability of a substance to be dissolved in a solvent to form a homogeneous solution. In this study, water, ethanol, and hexane were utilized to determine the solubility of the seaweed bioplastic. The sample was cut into small pieces (1 cm²) and was inserted into different test tubes containing different solvents – water, ethanol, and hexane. A sample with a size of 1 cm² and a weight of 0.05g was added to 5mL of solvent and the samples were allowed to dissolve for 20minutes. This procedure was based on the study of Beevi (2020).

Table 7. Solubility of the Seaweed Bioplastic

| Bioplastic Samples | Solubility | | |
|--------------------|------------|-----------|-----------|
| | Water | Ethanol | Hexane |
| Ribbon Sea Lettuce | Insoluble | Insoluble | Insoluble |

Findings indicated that the Ribbon sea lettuce was insoluble in all of the solvents. This contradicting to the study of Ren *et al.* (2017) that seaweed bioplastic samples contain polysaccharides which are polar molecules and are often soluble in water and insoluble in organic solvents like ethanol and hexane.

To determine the amount of load that the seaweed bioplastics can handle, the researchers conducted a tensile strength test based on the study of Mesina (2019) and Caballa (2016) with some modifications made. Three trials were performed and the average load in grams that the samples can handle was recorded.

Table 8. Tensile Strength of the Seaweed Bioplastic

| Bioplastic Samples | Average Load the sample can handle |
|--------------------|------------------------------------|
| Ribbon Sea Lettuce | 55.12 g |

The tensile strength test revealed that the Ribbon sea lettuce bioplastic held an average load of 55.12g. This means that the developed bioplastic can handle a product to at most 50.00 grams without breaking, this also means that the it could be used as alternative to small scale plastic wrapping.

Moisture content is the amount of water present in a sample. It is determined by weight loss drying. The moisture content of the seaweed bioplastics was identified by the researcher and the results are shown below.

Table 9. Moisture Content (in %) of the Seaweed Bioplastic

| Seaweed Bioplastic Samples | Weight of Samples (g) | | | | | | Average Moisture Content (%) |
|----------------------------|-----------------------|--------|---------|--------|---------|--------|------------------------------|
| | Trial 1 | | Trial 2 | | Trial 3 | | |
| | Before | After | Before | After | Before | After | |
| Ribbon Sea Lettuce | 0.1956 | 0.0989 | 0.1956 | 0.0884 | 0.1956 | 0.0971 | 51.534% |

It was revealed that the Ribbon sea lettuce bioplastic has a moisture content of 51.534%. Absorbed moisture has been shown to act as a plasticizer, reducing the glass transition temperature and strength of plastic – which is a reversible effect. However, absorbed water also can lead to irreversible degradation of the polymer structure. The data gathered indicates high moisture content of seaweed bioplastic. Moisture content can alter the properties of the bioplastics making them unsuitable to various applications.

This procedure was based on the study of Amin (2019) Seaweed bioplastic samples of 1 square inch were buried in the soil with a 2-inch depth. The containers of soil with seaweed bioplastics samples were placed in the laboratory at room temperature for 21 days. The degradation rate of the soil burial test was calculated from weight loss of the sample over time with 7 days interval and three trials were performed.

Table 10. Weight Loss Data of Ribbon Sea Lettuce Bioplastic Samples

| Trials | Weight of Ribbon Sea Lettuce Bioplastic Samples (g) | | Weight Loss (%) |
|--------|---|---------------|-----------------|
| | Initial | After 21 days | |

| | | | |
|-------------------------|--------|--------|--------|
| Trial 1 | 0.1601 | 0.0297 | 81.45% |
| Trial 2 | 0.1601 | 0.0281 | 82.45% |
| Trial 3 | 0.1601 | 0.0284 | 82.26% |
| Average Weight Loss (%) | | | 82.05% |

It can be seen that the Ribbon sea lettuce bioplastic lost 82.05% of its weight after 21 days of being buried in soil. In just 21 days, it can be observed that the ribbon sea lettuce is biodegradable, a good characteristic for an environmentally-friendly plastic material.

IV. CONCLUSION

The results of this study provide valuable insights into the potential of *Ulva reticulata* seaweed as a raw material for bioplastic production. Water quality tests of seawater samples revealed that the environment from which the seaweed was collected exhibited slightly basic pH levels, high total dissolved solids (TDS), and salinity below the average seawater salinity, possibly due to freshwater input from nearby land sources.

The physicochemical characterization of the seaweed bioplastics derived from *Ulva reticulata* demonstrated promising properties. The bioplastic solutions exhibited a slightly acidic pH, which falls within safe limits for human and environmental health. Optical inspection revealed a brownish-green color, likely attributed to the pigments of the seaweed. Additionally, the bioplastics met the minimum thickness requirement for biodegradable plastics set by European standards. Solubility tests showed that the seaweed bioplastics were insoluble in water, ethanol, and hexane, contradicting some previous studies but indicating potential differences in bioplastic composition. Tensile strength testing revealed that the bioplastics could withstand an average load of 55.12 grams, suggesting potential applications in small-scale packaging. However, the high moisture content of the seaweed bioplastics poses challenges as it can alter the material's properties and lead to irreversible degradation. Nonetheless, the soil degradation test demonstrated that the *Ulva reticulata* bioplastic degraded significantly, losing 82.05% of its weight after just 21 days, indicating its biodegradability and potential as an environmentally friendly alternative to traditional plastics. These findings highlight the promising attributes of *Ulva reticulata* seaweed for bioplastic production, offering a sustainable solution to mitigate plastic pollution and promote environmental sustainability. Further research and development are warranted to optimize the production process and explore various applications for seaweed bioplastics in different industries.

References

- Abu NJ, Bujang JS, Zakaria MH, Zulkifly S. Use of *Ulva reticulata* as a growth supplement for tomato (*Solanum lycopersicum*). *PLoS One*. 2022 Jun 27;17(6):e0270604. doi: 10.1371/journal.pone.0270604. PMID: 35759504; PMCID: PMC9236269.
- Behera L, Motanta M, and Thirugnanam A. 2021. Intensification of yam-starch based biodegradable bioplastic film with bentonite for food packaging application. *Environmental Technology & Innovation*, 25 (102180).
- Bhavsat N, Patel J, Soni D, Raol G, Surati V. 2019. Agar-agar bioplastic synthesis and its characterization. *Journal of Emerging Technologies and Innovation Research*, 6(3).
- Caballa FJA, Getalado MC. 2019. Plastic Film from *Dioscorea hispida* Dennst (Korot) Tuber. *International Journal of Trend in Scientific Research and Development*, 3(3):484-486.
- Cindana Mo'o, F.R.; Wilar, G.; Devkota, H.P.; Wathoni, N. Ulvan, a Polysaccharide from Macroalga *Ulva* sp.: A Review of Chemistry, Biological Activities and Potential for Food and Biomedical Applications. *Appl. Sci.* **2020**, *10*, 5488. <https://doi.org/10.3390/app10165488>
- Consebit, K., Dermil, K., Magbanua, E., Racadio, F., Saavedra, S., Abusama, H., & Valdez, A. (2021). Bioplastic from Seaweeds (*Eucheuma Cottonii*) as an Alternative Plastic. *ASEAN Journal of Science and Engineering*, 2(2), 129-132. doi:<https://doi.org/10.17509/ajse.v2i2.37799>
- Cosenza, V.A., Navarro, D.A., Ponce, N.M.A., Stortz, C.A. (2017). Seaweed Polysaccharides: Structure and Applications. In: Goyanes, S., D'Accorso, N. (eds) *Industrial Applications of Renewable Biomass Products*. Springer, Cham. https://doi.org/10.1007/978-3-319-61288-1_3
- Dagalea FMS, Cui-Lim KMR. 2018. Antimicrobial properties of biocomposite films from kappa-Carrageenan (kC) filled with Nanorod-rich Zinc Oxide (ZnO-N). *Advance Pharmaceutical Journal*, 3(2). <https://doi.org/10.31024/apj.2018.3.2.5>
- Dagalea, F. M. S., & Cui – Lim, K. M. R. (2021). Analysis of Biofilms from Biosynthesized Zinc Oxide Nanoparticles. *International Research Journal of Pure and Applied Chemistry*, 22(1), 92–97. <https://doi.org/10.9734/irjpac/2021/v22i130381>
- Lahaye M, and Robic A. 2007. Structure and Functional Properties of Ulvan, a Polysaccharide from Green Seaweeds. *Biomacromolecules*, 8(6):1765-1774.
- Largo, D. (2012) “*Ulva reticulata* (ribbon sea lettuce)” Link: *Ulva reticulata* (ribbon sea lettuce) | CABI Compendium (cabidigitallibrary.org)
- Lim C, Yusoff S, Ng CG, Lim PE, Ching YC. 2021. Bioplastic from seaweed polysaccharides with green production methods. *Journal of Environmental Chemical Engineering*, 9(5).
- Mermelstein NH. 2009. Measuring Moisture Content and Water Activity. *Food Technology Magazine*
- Pointer, K. (2017) “Carrageenan, Safety, Side effects and More” Link: Carrageenan: Safety, Side Effects, and More (healthline.com)
- Rajaram, B. et al, (2022) “Material Analysis for Bioplastics Quality Assurance and Degradation” Link: Material Analysis for Bioplastics Quality Assurance and Degradation - TA Instruments
- Ren, Y. et al, (2017) “The Preparation and Structure Analysis Methods of Natural Polysaccharides of Plants and Fungi: A Review of Recent Development” Link: The Preparation and Structure Analysis Methods of Natural Polysaccharides of Plants and Fungi: A Review of Recent Development - PMC (nih.gov)

- Roemmich, D. (2014) “VOYAGER: HOW LONG UNTIL OCEAN TEMPERATURE GOES UP A FEW MORE DEGREES?” Link: Voyager: How Long until Ocean Temperature Goes up a Few More Degrees? | Scripps Institution of Oceanography (ucsd.edu)
- Santana RF, Bonomo RCF, Gandolfi ORR, Rodrigues LB, Santos LS, Dos Santos Pires AC, de Oliveira CP, da Costa Ilhéu Fontan R, Veloso CM. Characterization of starch-based bioplastics from jackfruit seed plasticized with glycerol. J Food Sci Technol. 2018 Jan;55(1):278-286. doi: 10.1007/s13197-017-2936-6. Epub 2017 Dec 2. PMID: 29358820; PMCID: PMC5756213.
- Shafie, M., Kamal, M., Zulkiflee, F., et al, (2022) “Application of Carrageenan extract from red seaweed (*Rhodophyta*) in cosmetic products: A review” Vol. 99, Issue 9.
- Susmitha B. S., Vanitha, K. P., and Rangaswamy B. E., (2016) “Bioplastic – A Review”, International Journal of Modern Trends in Engineering and Research
- Ulva reticulata* Online Source: *Ulva reticulata* - Encyclopedia of Life (eol.org)
- Wahab, I. F., & Razak, S. I. A. (2016). Polysaccharides as Composite Biomaterials. InTech. doi: 10.5772/65263