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STUDY OF NUTRIENT CONTENT (NITROGEN ELEMENT) IN *Kappaphycus alvarezii* THALLUS IN RELATION TO FILAMENTOUS ALGAE ATTACHMENT IN THE BONE-BONE COASTAL WATERS, BAUBAU CITY

Studi Kandungan Nutrien (Unsur N) pada Talus *Kappaphycus alvarezii* Kaitannya dengan Penempelan Alga Filamen di Perairan Pantai Bone-Bone Kota Baubau

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ABSTRACT

This study aimed to determine the nitrogen content in the thallus of K. alvarezii in relation to filamentous algae attachment. The research location was determined using the purposive sampling method. This study used a horizontal net (horinet) as a cultivation tool. Sampling of K. alvarezii thallus was conducted over 35 days with a 7-day interval. A total of 20 thallus were collected during the study, consisting of 10 thallus with attached filamentous algae and 10 thallus without attached filamentous algae. The nitrogen content in the thallus was calculated using the Kjeldahl method, which involves three main stages: digestion, distillation, and titration. The results showed that the types of attached filamentous algae included one species from the class Chlorophyta, namely Chaetomorpha crassa, and one species from the class Rhodophyta, namely Neosiphonia apiculata. The highest average nitrogen content in the thallus when filamentous algae were attached was 1.121%, and the lowest was 0.863%. In contrast, in thallus without attached filamentous algae, the highest average nitrogen content was 1.274%, and the lowest was 0.942%. The results of the one-way ANOVA test analysis indicated no significant difference in nitrogen content between thallus with and without attached filamentous algae.

Keywords: Filamentous algae, Horizontal net, Kappaphycus alvarezii, Nitrogen.

ABSTRAK

Penelitian ini bertujuan untuk mengetahui kandungan nitrogen pada talus *K. alvarezii* hubungannya dengan penempelan alga filamen. Penentuan lokasi penelitian berdasarkan metode *purposive sampling* atau dilakukan secara sengaja. Penelitian ini menggunakan horizontal net (horinet) sebagai alat budidaya, pengambilan sampel talus *K. alvarezii* dilakukan

selama 35 hari dengan interval waktu 7 hari, sampel *K. alvarezii* yang diambil selama penelitian berjumlah 20 talus, dimana 10 talus dengan kondisi alga yang menempel dan 10 talus lainnya alga filamen tidak menempel. Perhitungan kandungan nitrogen pada talus menggunakan metode kjeldahl dengan 3 tahapan utama yaitu destruksi, destilasi dan titrasi. Hasil penelitian menunjukkan bahwa jenis alga filamen yang menempel diantaranya 1 jenis kelas Chlorophyta yaitu *Chaetomorpha crassa* dan 1 jenis kelas Rhodophyta yaitu *Neosiphonia apiculata*. Rata-rata kandungan nitrogen tertinggi pada talus saat alga filamen menempel sebesar 1,121 % dan terendah sebesar 0,863, sedangkan pada talus saat alga filamen tidak menempel rata-rata kandungan nitrogen tertinggi sebesar 1,274% dan terendah sebesar 0,942%. Hasil analisis uji *one way anova* menunjukkan tidak terdapat perbedaan kandungan nitrogen antara talus dengan alga filamen yang menempel maupun tidak.

Kata Kunci: Alga filamen, Horizontal net, *Kappaphycus alvarezii*, Nitrogen.

INTRODUCTION

Nutrients are essential nutrients in waters to support the growth and development of potential marine ecosystem resources (Handayani et al., 2016). The primary source of nutrients is naturally obtained from the waters themselves through the processes of decomposition, weathering, and decomposition of plants, dead organisms, and waste such as domestic, industrial, agricultural, and livestock waste, as well as feces and leftover feed from fisheries (Silvi *et al*., 2022). Nutrients in water exist in both macro and micro forms. Macro nutrients consist of C, H, O, N, S, P, K, Mg, Ca, Na, and Cl, which are needed in large quantities, while those included in the micro form consist of Fe, Co, Zn, Si, Mn, and Cu, which are needed in small amounts (Widiardja et al., 2021). One of the most essential nutrients for organisms is nitrogen, which is essential for phytoplankton growth, and is commonly used as an indicator of water quality and fertility (Iklima *et al*., 2019).

Nitrogen is an essential macronutrient crucial for plant growth because it is a component of plant cells (Rahmadani *et al*., 2020). A deficiency in N will inhibit seaweed growth because it is an element used in photosynthesis (Kushartono *et al*., 2009). Nitrogen in water occurs in the form of organic and inorganic nitrogen. Inorganic nitrogen consists of ammonia, ammonium, nitrate, nitrite, and molecular nitrogen, while organic nitrogen consists of proteins, amino acids, and urea (Iklima *et al*., 2019).

Proses Seaweed absorbs nutrients through osmosis diffusion throughout its body (Harahap *et al*., 2022). The cell membrane, the outermost layer after the cell wall, acts as a protective barrier for the cell's contents, regulating the movement of nutrients into and out of the cell. The semi-permeable nature of the cell membrane selects which substances can enter the cell. The amount of nutrients osmotically absorbed into the cell depends on the nutrient levels inside and outside the cell, according to its needs (Pramesti, 2013).

One type of epiphyte found on seaweed thallus during cultivation is filamentous algae. Filamentous algae, a type of epiphyte, are plants that attach themselves to seaweed and can act as competitive pests and cause diseases in seaweed (Bunga *et al*., 2018). Filamentous algae are a source of degradation that can lead to decreased quality and quantity in seaweed cultivation. This is because epiphytic algae and cultivated algae share similarities in terms of meeting nutritional needs for survival (Wulandari *et al*., 2024).

Based on the description above, nutrients play a crucial role, directly influencing the growth and survival of seaweed. This study aimed to determine the nitrogen content in the thallus of *K. alvarezii* when the filamentous algae were attached and unattached. The results are expected to contribute to the optimal management and utilization of fishery resources, particularly seaweed.

RESEARCH METHODS

Time and Place

This research was conducted from October to November 2025 in the coastal waters of Bone-Bone, Batupoaro District, Baubau City, Southeast Sulawesi Province. The research location is at -5°27'56.05" South Latitude and 122°35'5.13" East Longitude, approximately 250 meters from land.

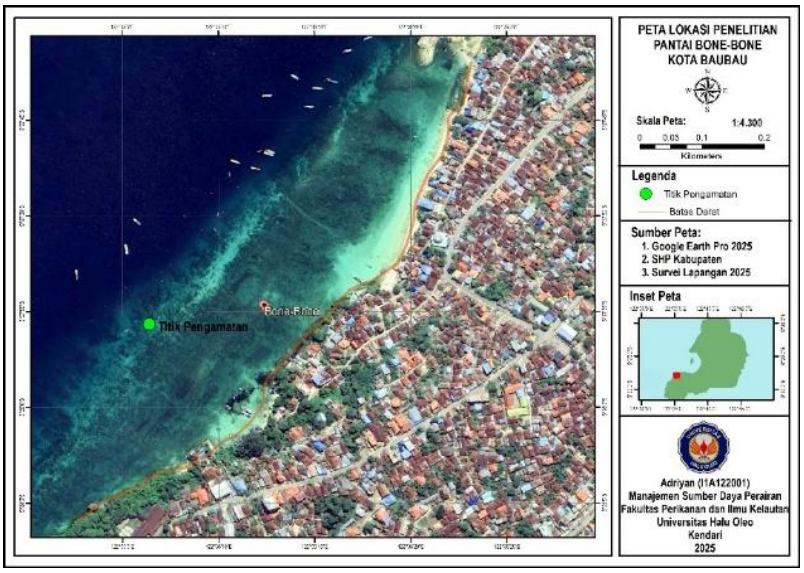


Figure 1. Research Location Map

Placement of Cultivation Tools

This study used horizontal nets (horinet) as a cultivation tool (Figure 2). The research location consisted of one observation point. Two horizontal nets were used. Each horizontal net was labeled A and B for ease of observation. Each horizontal net contained 15 K. alvarezii thallus, each weighing 20 grams.

Figure 2. Horizontal Net Design
(Source: Kasim *et al*., 2024)

Sampling of *K. alvarezii* and Filamentous Algae

Seaweed sampling was conducted 5 times with a sampling period of every 7 days for 35 days. Seaweed samples were taken randomly or random sampling in each horinet plot of 2

thallus. The seaweed thallus that had been taken were cleaned of filamentous algae using a small brush and cut into 5 cm lengths, then placed in labeled sample plastic and stored in a cool box. A total of 20 thallus samples were taken during the study, where 10 thallus with attached algae and 10 thallus with unattached filamentous algae. Filamentous algae separated from the seaweed thallus were observed and photographed using a camera. The number of filamentous algae stands was counted with the help of a magnifying glass (loupe) and identified using an identification book by Barbara (2009) and Setyobudiandi *et al*., (2009).

Nitrogen Measurement in Talus

Nitrogen analysis is based on the determination of sample distillation using the Kjeldahl method. The Kjeldahl method is basically divided into three stages: destruction, distillation, and titration (Legowo & Nurwantoro, 2004).

1. Destruction Stage

The prepared sample was cut into small pieces, then weighed as much as 0.1 grams, then put into a measuring flask, then the sample was added with a mixture of selenium (*selenium mix*) as much as 1 gram and H2SO4 as much as 10 ml. Next, the sample was destroyed by heating the sample on a hotplate at a temperature of 420°C. The destruction was complete when white steam emerged and a clear extract was obtained (lasts several hours). Then the sample was removed and cooled.

2. Distillation Stage

Next, prepare an Erlenmeyer flask, fill it with 20 ml of BCG indicator, and connect it directly to the distillation apparatus. Fill the distillation flask with the sample extract solution, add 150 ml of distilled water and 50 ml of 40% NaOH solution, and distill until the cross-sectional volume of the 20 ml Erlenmeyer flask becomes 100 ml (blue).

3. Titration Stage

The distillation results are titrated using 0.1 N HCl until the color changes to pink. Then, record the titration volume of the sample (Vs) and blank (Vb) after which calculations are carried out.

Measurement of Water Quality Parameters

Water quality parameters were measured simultaneously with the sampling of *K. alvarezii* thallus, which was carried out five times over 35 days with a 7-day interval. The parameters measured consisted of physical parameters including temperature, current velocity, brightness, and chemical parameters including salinity, nitrate, and phosphate. Total nitrate and phosphate dissolved in the water were analyzed spectrophotometrically according to Kasim *et al*. (2016).

Data analysis

According to Astriana *et al*. (2019), the calculation of the daily growth rate of seaweed refers to Dawes (1981) with the following formula.

$$\text{LPH (\%)} = \frac{\ln Wt - \ln Wo}{t} \times 100\ \%$$

Where:

LPH = Daily growth rate (%)
Wt = Final wet weight (g)
Wo = Initial wet weight (g)
t = Cultivation time (days)

Determination of nitrogen levels refers to Atma (2018) in Sofiana et al. (2024) with the following formula.

$$\% \text{ N} = \frac{(Vs-Vb) \times N\ HCl \times 0{,}014 \times FP}{Sample\ Mass} \times 100\%$$

Where:
Dan = Sample titration volume
Vb = Blank titration volume
AND HCl = HCl normality
FP = Dilution factor

Furthermore, to examine the relationship between nitrogen and the growth rate of K. alvarezii attached and unattached to filamentous algae, as well as environmental factors, the data was statistically processed using a T-test using Statistical Product and Service Solutions (SPSS) software. According to Wulandari *et al*. (2024), a significance value less than 0.05 ($\alpha < 0.05$) indicates a significant difference, and a significance value greater than 0.05 ($\alpha > 0.05$) indicates no significant difference.

Water clarity can be analyzed using the following formula from Lubis *et al*. (2020):

$$\text{Water clarity (m)} = \frac{D1+D2}{2}$$

Where:
D1 = Invisible distance (m)
D2 = Visible distance (m)

According to Widyastuti *et al*. (2022), current velocity measurements refer to Alaerts and Santika (1984) with the following formula.

$$v = \frac{s}{t}$$

Where:
v = Current speed (m/s)
s = Distance (m)
t = Time (s)

RESULTS AND DISCUSSION

Types and Number of Filamentous Algae Attached to the *K. alvarezii* Thallus

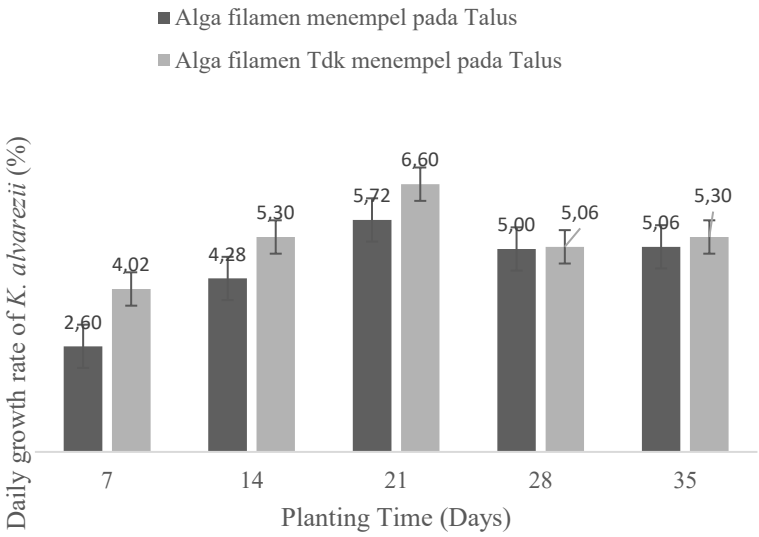
The types of filamentous algae attached to the thallus of K. alvarezii during the study were found to be 2 types, namely 1 type from the Chlorophyta class and 1 other type from the Rhodophyta class, which can be seen in Table 1.

Table 1. Types and Number of Filamentous Algae Attached to the *K. alvarezii* Thallus

No	Class	Spesies	Day No.					Amount
			7	14	21	28	35	
1	*Chlorophyta*	*C. crasaa*	0	11	13	23	17	64
2	*Rhodophyta*	*N. apiculata*	6	5	23	15	30	79
Amount			6	16	36	38	47	143

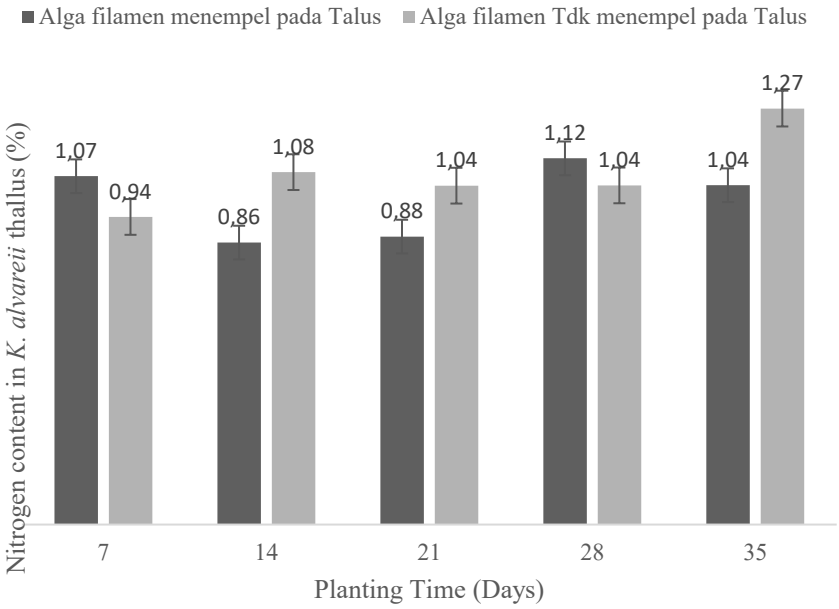
Daily Growth Rate of *K. alvarezii*

The highest average daily growth rate of *K. alvarezii* thallus when filamentous algae were attached occurred on the 21st day at 5.720%, while in conditions where filamentous algae were not attached, the highest average daily growth rate also occurred on the 21st day at 6.601%.



Nitrogen Content in *K. alvarezii* Thallus

The highest average nitrogen content in the *K. alvarezii* thallus when the filamentous algae were attached occurred on the 28th day at 1.121%, while in conditions where the filamentous algae were not attached, the highest average nitrogen content occurred on the 35th day at 1.274%.



Water Quality Parameters

The physicochemical parameters of the waters measured during the study were temperature, current velocity, clarity, salinity, nitrate, and phosphate. The values ranged from those optimal for *K. alvarezii* growth, as shown in Table 2.

Table 2. Water Quality Parameters at Bone-Bone Beach

No	Parameter	Unit	Range

1	**Physics**		
	Temperature	°C	28,4 – 29
	Current Speed	m/Second	0,117 – 0,252
	Brightness	%	100
2	**Chemistry**		
	Salinity	‰	30 – 32
	Nitrate	mg/L	0,112 – 0,194
	Phosphate	mg/L	0,045 – 0,084

Pearson Correlation Analysis

Based on the results of the correlation analysis, it was found that the relationship between the nitrogen content absorbed by *K. alvarezi*i and the daily growth rate (LPH) both when the filamentous algae were attached and not attached had a low correlation, as can be seen in Table 3.

Table 3. Results of the Correlation Test of Nitrogen Content with Daily Growth Rate of *K. alvarezii*

	N (attached filamentous algae)	N (non-attached filamentous algae)	Information
LPH (attached filamentous algae)	-0,296	-	Low
LPH (non-attached filamentous algae)	-	0,304	Low

*. *Correlation is significant at the 0.05 level* (*2-tailed*)

Based on the results of the correlation test analysis of daily growth rates with water quality parameters that support the growth of *K. alvarezii* thallus, it is presented in Table 4.

Table 4. Correlation Test of Water Quality Parameters with Daily Growth Rate in *K. alvarezi*i Talus

	Temperature	Salinity	Current Speed	Nitrate	Phosphate
LPH (attached filamentous algae)	-0,032	0,089	-0,019	0,410	-0,579
LPH (non-attached filamentous algae)	-0,401	0,275	0,016	0,301	-0,338

*. *Correlation is significant at the 0.05 level* (*2-tailed*).

Based on the results of the correlation test analysis of nitrogen content with water quality parameters that support nitrogen absorption in *K. alvarezii* thallus, it is presented in Table 5.

Table 5. Correlation Test of Water Quality Parameters with Nitrogen Content in *K. alvarezii* Talus

	Temperature	Salinity	Current Speed	Nitrate	Phosphate
Nitrogen content (attached filamentous algae)	-0,270	0,521	0,603	0,139	0,388
Nitrogen content (non-attached filamentous algae)	-0,724	0,826	0,640	0,555	-0,335

*. *Correlation is significant at the 0.05 level* (*2-tailed*).

Types of Filamentous Algae Attached to the Thallus of *K. alvarezii*

Berdasarkan Based on the results of observations conducted on the Talus *K. alvarezii* for 35 days of maintenance, 2 types of filamentous algae from 2 classes were obtained. The most abundant type of filamentous algae was from the Rhodophyta class, type *N. apiculata*, with 79 colonies, and the least abundant was from the Chlorophyta class, type *C. crassa,* with 64 colonies.

Keberadaan The presence of filamentous algae (*N. apiculata and C. crasaa*) attached to the thallus of *K. alvarezii* does not have a direct negative impact, but the presence of filamentous algae can be a competitor for its host in obtaining nutrients and sunlight for photosynthesis. The presence of filamentous algae can inhibit productivity, attract predators, inhibit the process of photosynthesis, resulting in slow death of the host plant (Purbiantoro *et al*., 2013). Research on Bone-Bone beach found 4 types of filamentous algae attached to the thallus of *K. alvarezii*, namely *C. crassa, Ulva intestinalis, N. savateri* and *N. apiculata* (Wulandari *et al*., 2024). Epiphytes that are mostly attached to the thallus *of E. cottonii* are mostly *algae* such as *A. spicifera, Hypnea sp, Polysiphonia sp, D. dichotoma, Padina santae* and *C. crassa* (Arisandi *et al*., 2013). The presence of algae attached to seaweed is due to the seaweed being a source of nutrients for the attached algae when nutrient levels in the water are low (Azim *et al*., 2005). Some epiphytic macroalgae are parasitic; in some, the holdfast can penetrate the host thallus and absorb nutrients from it. This condition can result in crop failure in cultivation activities (Nurdiana *et al*., 2016).

Daily Growth Rate of *K. alvarezii* When Filamentous Algae Are Attached and Not Attached

The results of the study showed that the growth of K. alvarezii varied significantly over the 35-day cultivation period. Observations showed that the highest daily growth rate of *K. alvarezii* attached to filamentous algae occurred on day 21 (week 3), with an average value of 5.720 ± 2.027%. The lowest daily growth rate occurred on day 7, with an average value of 2.604 ± 0.000%.

The low growth rate in the first week (day 7) was due to the seaweed still adapting to the aquatic environment and healing wounds from cutting the seedlings. During the first week of seaweed cultivation, seaweed plants experience a lag or adaptation phase (Togatorop *et al*., 2017). During the first week, seaweed growth did not differ significantly from the initial weight because the seedlings were adapting to the natural environmental conditions and healing wounds from cuts during cutting.

The analysis showed that the daily growth rate of *K. alvarezii* was considered quite good. A good daily growth rate for seaweed is 2.03-2.36% (Ariyati *et al*., 2017). Cultivated seaweed growth is considered good when the growth rate exceeds 3% (Patadjai *et al*., 2024). The low growth rate of K. alvarezii in the study by Rachmawati and Abdillah (2019) was suspected to be due to unfavorable weather conditions during the cultivation period, such as extreme temperature fluctuations due to changes from hot to rainy and vice versa within a day. The decrease in growth on days 28 and 35 from the highest growth (day 21) is suspected to be due to the seaweed entering harvest time, resulting in slow growth (Yusran *et al*., 2021).

Based on observations, the highest daily growth rate of K. alvarezii that was not attached to filamentous algae occurred on day 21 (week 3), with an average value of 6.601 ± 2.539%. Meanwhile, the lowest daily growth rate occurred on the 7th day with an average value of 4.017 ± 0.381%. This growth rate can be said to be good because it has a growth value above 3%. This is thought to be due to the absence of filamentous algae (epiphytes) attached to the thallus of *K. alvarezii* so that nutrients are directly used for photosynthesis and growth. This is

because there is no nutrient competition between the host (*K. alvarezii*) and the epiphyte (filamentous algae) (Hosnan *et al*., 2016).

Nitrogen Content in the Thallus of K. alvarezii When Filamentous Algae Are Attached and Not Attached

Based on the analysis results, the highest nitrogen content of *K. alvarezii* attached to filamentous algae occurred on day 28 (week 4) with an average value of 1.121 ± 0.005%. The lowest nitrogen content occurred on day 14 (week 2) with an average value of 0.863 ± 0.096%. Meanwhile, the highest nitrogen content of K. alvarezii without filamentous algae occurred on day 35 with an average value of 1.274%, and the lowest nitrogen content occurred on day 7 (week 1) with an average value of 0.942%.

Research in the coastal waters of Lakeba, Baubau City, found that the N nutrient content in the thallus of *K. alvarezii* attached to epiphytes was 0.964-1.884%, while in the thallus without epiphytes it was 0.875-1.866%. The nitrogen content is influenced by the density of the thallus growth, resulting in competition for nutrients (Alimusra, 2021). Nitrogen uptake by *K. alvarezii* during 45 days of observation using the longline method was 1.11–2.88 µmol/g/day, and this value was influenced by different planting distances (Perdana *et al*., 2022).

Nitrogen absorption in seaweed can be seen from the nitrogen content. Nitrogen absorption by seaweed (*K. alvarezii*) varies over time. The high and low nitrogen content in seaweed is influenced by the cultivation environment (Muktiniati *et al*., 2022).

This study found that nitrogen absorption by the *K. alvarezii* thallus, when attached and unattached, showed almost the same pattern, with no differences. The attachment of filamentous algae (epiphytes) to the K. alvarezii thallus only affected growth. However, the nitrogen absorption values for seaweed showed a tendency for thallus without filamentous algae attachment compared to thallus with filamentous algae attachment.

Water Quality Parameters

Physics

Water quality parameters also play an important role in the growth of *K. alvarezii*. Water temperature was found to range between 28.4-29°C. The optimal temperature for the growth of K. alvarezii seaweed is 28-30°C (Prayudha *et al*., (2024). Current speed ranges from 0.117 - 0.252 m/s. Good current speed for seaweed cultivation ranges from 0.2-0.4 m/s (Tisera & Tanody, 2020). Water clarity is 100% at a depth of ±5 meters. A water clarity value of 100% indicates that the clarity reaches the bottom of the water. A good clarity value for seaweed is more than 1 m (Risnawati *et al*., 2018).

Chemistry

Salinity ranges from 30-32 ppt, nitrate (dissolved) concentrations range from 0.112 to 0.194 mg/L, and phosphate concentrations range from 0.045 to 0.084 mg/L. A suitable salinity level for *K. alvarezii* growth is 30-32 ppt. A nitrate content level of 0.00-1.46 ppt is within the tolerable range and can support seaweed growth (Fikri *et al*., 2015).

According to the Ministry of Environment (KLH, 2004) on Seawater Quality Standards, the tolerable phosphate level for marine biota is 0.015 mg/L. Waters with phosphate levels <0.01 mg/L are considered unsuitable for seaweed cultivation (Edy *et al*., 2017).

Pearson Correlation Test

(Pearson Product Moment Correlation) is a parametric statistical method used to measure the strength and direction of the linear relationship between two continuous variables on an interval or ratio scale.

Table 6. Basic Guiding Values in Decision Making

Pearson Correlation Value	Description
0,00 – 0,20	No correlation
0,21 – 0,40	Weak correlation
0,41 – 0,60	Moderate correlation
0,61 – 0,80	Strong correlation
0,81 – 1,00	Very strong correlation

(Source: Minsas *et al*., 2023)

Based on the results of the correlation analysis of water quality parameters with daily growth rate, it was found that the parameters positively and significantly related to *K. alvarezii* growth were nitrate (r = 0.410) and salinity (r = 0.275). The correlation values obtained were classified as low to moderate. Meanwhile, the water quality parameter with a negative and significant relationship was phosphate (r = -0.622). This correlation value indicates a strong relationship.

Based on the analysis results for attached filamentous algae, a weak negative relationship was found between nitrogen content and daily growth rate, with a correlation coefficient of -0.283 (low). This value statistically indicates that increasing nitrogen content in the environment tends to be followed by a decrease in the daily growth rate of algae attached to the thallus, although the effect is not strong (low). Conversely, in unattached filamentous algae, the relationship is weakly positive, with a correlation coefficient of 0.289 (low). This means that in free conditions (without epiphytes), increasing nitrogen content tends to be followed by an increase in daily growth rate, although the relationship is relatively weak.

CONCLUSION

The nitrogen content of *K. alvarezii* thallus when attached to filamentous algae was relatively low, with an average value ranging from 0.863 to 1.121%. Meanwhile, in *K. alvarezii* thallus when unattached, the nitrogen content was relatively low, with an average value ranging from 0.942 to 1.274%. The nitrogen content of K. alvarezii thallus showed a nearly identical pattern between thallus attached and unattached with filamentous algae, with no significant differences. However, nitrogen content tended to be higher in thallus without filamentous algae attachment. Statistical tests showed a weak relationship between nitrogen content and daily growth rate of *K. alvarezii*.

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