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SEAWEED AS BIOADSORBENT FOR NITROGEN AND PHOSPHORUS REMOVAL

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Abstract

Eutrophication has become a serious environmental problem because of the excessive amounts of nitrogen and phosphorus in the water. Aquaculture waste is one of the drivers of eutrophication. Seaweed is known for its ability to remove nutrients from the water. In Indonesia, research about the efficiency of seaweed in decreasing nutrient concentration in wastewater is still rare. This article reviewed the use of seaweed as an adsorbent for nitrogen and phosphorus removal. This review aims to summarize the efficiency of nutrient removal in various genera of macroalgae. The comparing bioremediation potentials of macroalgae, including growth, nutrient bioaccumulation capacity, and potential nutrient uptake, are discussed. The factors influencing nutrient uptake will also be addressed in this study. The literature was collected from ScienceDirect and Google Scholar databases. This paper found that red algae from the genus *Gracilaria* were the most widely used as bioremediation agents compared to other genera. This article is expected to be useful as a basis for selecting seaweed to be used as a bioremediation agent. We hope that there will be more research on seaweed as a bioadsorbent in Indonesia.

Keywords: Bioadsorbent; Macroalgae; Nitrogen; Phosphorus; Seaweed.

1. Introduction

The increase in world population and human activities (industrial, agricultural, and domestic waste) causes environmental problems, especially eutrophication, which will cause adverse effects (Niu et al., 2022). The main drivers of eutrophication in coastal waters are anthropogenic sources (de Raús Maúre et al., 2021), while the open sea is affected by wind and salinity (Vigouroux et al., 2021). Eutrophication is an excess of nutrient input in the waters which causes an increase in primary productivity resulting in changes in the structure and function of the ecosystem (Smith & Schindler, 2009; Wu et al., 2015c). Eutrophication stimulates harmful algal blooms (Liu et al., 2013). It may cause oxygen depletion (Zhu et al., 2011), water transparency decreases, and fish mortalities (Anderson et al., 2021; Smith & Schindler, 2009).

Eutrophication is also caused by waste from intensive cultivation (Camargo & Alonso, 2006; Kang et al., 2011; Marinho-Soriano et al., 2011; Yu et al., 2019). For instance, in

Dapeng Cove, South China Sea, the seawater quality has decreased because of mariculture activities (Yu et al., 2019). The primary contaminants are nitrogen and phosphorus from fertilizer, feed, and metabolic waste of cultivated animals (Marinho-Soriano et al., 2009). Troell et al. (1997) said that only 30% of nutrients are absorbed by the fish, and the rest is released to the environment as uneaten feed, fish excretion, and respiration. This absorption caused a change in the N/P ratio in the waters and increased ammonium toxicity, harming aquaculture (Kang et al., 2011). In this case, the main challenge is to develop a non-pollution strategy that minimizes the adverse effects of these activities. The excess nutrient from the effluent should be treated before it reaches the sea (Marinho-Soriano et al., 2009).

One of the best methods for nutrient removal is the adsorption method using macroalgae or seaweed (Kotta et al., 2022). For instance, *Sargassum fusiforme* effectively reduces eutrophication levels by up to 36%, and *U. lactuca* has a high capacity to remediate pollutants (Areco et al., 2021; Tian et al., 2023). This method is promising and attractive because of its eco-friendly and inexpensiveness (Arumugam et al., 2018; Fei, 2004). Seaweed is an adsorbent because it requires nutrients, especially nitrogen, and phosphorus, for its growth. It needs during the photosynthesis process (Bews et al., 2021). Seaweed also can store high concentrations of nitrogen in its tissues (He et al., 2008). Usually, nitrogen is the limiting factor for seaweed growth (Roleda & Hurd, 2019; Smith & Schindler, 2009). Another advantage of using macroalgae is inhibiting harmful algal growth, such as *Ulva lactuca* (Tang & Gobler, 2011). Therefore, this method is useful for bioremediation and has economic value.

In Indonesia, research about macroalgae as nutrient adsorption is still rare. Most of the studies focused on cultivation and adsorbent for heavy metal adsorption. For instance, macroalgae could remove heavy metals such as cadmium (Cd) (Kuncoro et al., 2017; Putri & Syafiq, 2019), lead (Pb) (Putri & Syafiq, 2019; Supriyantini et al., 2018), and mercury (Putri & Syafiq, 2019). Besides, macroalgae can remove methylene blue (Pratiwi et al., 2019). In this article, the macroalgae as an adsorbent for removing nitrogen and phosphorus has been reviewed. This review aims to summarize the efficiency of nutrient removal in various genus of macroalgae. The assessment of bioremediation potentials of macroalgae, including growth, nutrient bioaccumulation capacity, and potential uptake of nutrients, are discussed. The factors influencing nutrient uptake will also be addressed in this study. This article is expected to be useful as a basis for selecting seaweed to be used as a bioremediation agent.

2. Methods

This review paper used the literature from Science Direct and Google Scholar databases. The Science Direct database covered the white literature, while the Google Scholar database covered the literature published in national journals. This approach was conducted to get better coverage, especially within the national context of Indonesia. The retrieved articles were published during the period from 1996 to 2021. The literature was retrieved by using the keywords “seaweed,” “macroalgae,” “bioadsorbent,” “nutrient removal,” “nutrient uptake,” and “bioremediation”. Article selection was conducted using the PRISMA protocol (<https://prisma-statement.org/>). The first selection was conducted by reading the titles and abstracts. Only literature that fit the criteria was included in the further process.

The criteria for the first selection were the context of the literature that fits the purpose of this review. For instance, the literature about ‘freshwater seaweed’ or ‘microbe bioremediation’ were excluded. The publication year was not used for selection criteria since the present study attempted to cover all available literature about seaweed bioadsorbent.

Therefore, some of the old literatures were included in the present review paper. The second selection was conducted by deep reading the content and arranging it into the table. The second selection criteria were based on seaweed genera with the most research as a nutrient biofilter. Three hundred papers were filtered, and 110 papers were reviewed and used in the present study (Figure 1).

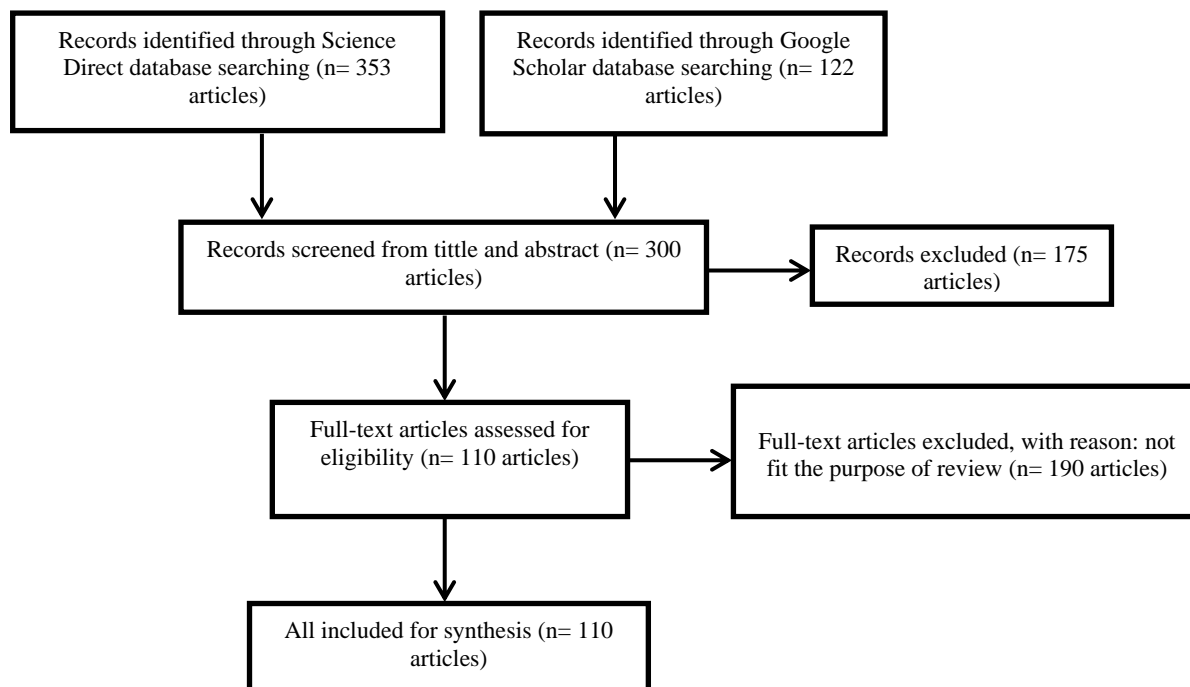


Figure. 1 A flow chart of the review process

3. Results and Discussions

3.1. Macroalgae/seaweeds

Macroalgae or seaweeds are low-level plants that can grow up to 60 m quickly in fresh or salt water. Seaweed has a variety of shapes, sizes, colors, and compositions (Makkar et al., 2016; McHugh, 2003). Based on the pigmentation, seaweed is classified into three types, namely brown (Phaeophyceae), red (Rhodophyceae), and green (Chlorophyceae) (Bharathiraja et al., 2015; Makkar et al., 2016).

Macroalgae play an essential ecological role in the ecosystem (Yang et al., 2006). They act as bioremediate agents to decrease nutrient concentration. Thus, it can protect the ecosystem (Schnerer et al., 2012; Wei et al., 2017). In China, seaweed aquaculture can remove 60.31 t N km⁻² year⁻¹ of nitrogen and 7.60 t P km⁻² year⁻¹ of phosphorus (Xiao et al., 2017). The uptake efficiency of seaweed is 60-87% and 30-43% for ammonia and phosphorus, respectively (Kang et al., 2021). Table 1 shows the research conducted using seaweed as a bioremediation agent to reduce nitrogen and phosphorus content.

Table 2. Parameter estimates (β) and odds ratio of ordinal logistics regression of fertility intention: indonesia susenas 2017

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
<i>Gracilaria lemaneiformis</i>	Red seaweed	Aquaculture water (Bay water)	t = 1–35 d	Co-culture with the fish <i>Pseudosciaena crocea</i>	Nitrogen and Phosphate	N = 21.0%	Wei et al. (2017)
				Cage aquaculture Seawater with salinity of 26–29 (24–27 during low tide) Surface water T = 18.4–26.0 °C Surface water pH = 7.43–7.83 t = 20 d		P = 28.6% N content=3.98% and P content= 0.49%	
<i>Gracilaria lemaneiformis</i>				t= 23 days		NH ₄ -N=85.53%, PO ₄ -P= 65.97%	Yang et al. (2006)
				t=40 days		NH ₄ -N=69.45%, PO ₄ -P= 26.74%	
<i>Gracilaria lemaneiformis</i>				Co-culture with the scallop <i>Chlamys farreri</i> t=3 weeks		NH ₄ -N=83.7% PO ₄ -P= 70.4%	Mao et al. (2009)
<i>Gracilaria lemaneiformis</i>				1 ha cultivation, co-culture with fish <i>Sebastes fuscescens</i>		0.22 t N and 0.03t P, growth rate11.03% d-1	Zhou et al. (2006)

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
<i>Gracilaria tikvahiae</i>				t = 90 d		28 and 94 kg N ha ⁻¹ at the LIS and BRE sites, a growth rate of 16.5% day ⁻¹ at BRE and 4.8% day ⁻¹ at the LIS site, C:N ratio= 5.3-8.6 (BRE) C:N ratio= >16 (LIS)	Kim et al. (2014)
<i>Gracilaria tikvahiae</i>	Red seaweed	Shrimp wastewater	t = 7–18 d	Co-cultured with Pacific white shrimp <i>Litopenaues vannamei</i> Salinity 30.4–34.8g/kg T = 18–33 °C pH = 7.4–7.9 t = 18 d	Nitrogen	N = 35% (Recovery in seaweed) growth rate= 4.8% d-1 N assimilation= 0.83 g N m ⁻² d ⁻¹	Samocha et al. (2015)
<i>Gracilaria chouae</i>	Red seaweed	Aquaculture water (Bay water)	t = 1–47 d	Co-cultured with the black sea bream <i>Sparus macrocephalus</i> The salinity of 28.33–31.07 T = 16.61–22.68 °C pH = 8.16–8.2	Nitrogen and Phosphate	N = 41.2% (NO ₃ -N = 37.76%, NO ₂ -N = 36.99%, NH ₄ -N = 29.27%) P = 46.2%	Wu et al. (2015a)

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
				t = 28 d		(PO ₄ -P = 40.64%) growth rate 7.43 ± 0.37% d ⁻¹	
<i>Ulva lactuca</i>	Green seaweed	Reject water from anaerobically digested sewage sludge	t = 1–18 d	The salinity of 20% from artificial seawater	Nitrogen and Phosphorus	N = 22.7 mg N g DW ⁻¹ d ⁻¹ P = 2.7 mg P g DW ⁻¹ d ⁻¹	Sode et al. (2013)
				T = 15 oC pH = 7.9–8.9 t = 18 d			
<i>Ulva lactuca</i>				t = 14 d		NH ₄ -N=45% growth rate=3.6 % day ⁻¹	Lavania-Baloo et al. (2014)
<i>Ulva lactuca</i>						growth rate=7.4 % day ⁻¹ , TAN= 59-81%, PO ₄ -P, C:P=41-55= 50-55%	Khoi & Fotedar (2011)
<i>Ulva pertusa</i>				Co-cultured with the black rockfish (<i>Sebastes schlegelii</i>)		NH ₄ -N= >80%	Kang et al. (2011)

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
<i>Chondrus crispus</i>	Red seaweed	Finfish culture effluent	T = 6 and 13 °C	Land-based Atlantic halibut (<i>Hippoglossus hippoglossus</i>) farm	Nitrogen	Net N = 2.0 kg m ⁻² (at T = 6 and 13 °C), C:N ratio=6.3-7.2, C content= 30.7-32.4, N content= 4.3-5	Kim et al. (2013)
<i>Palmaria palmata</i>			T = 6 and 16 °C	t = 28 d each trial		Net N = 2.0 kg m ⁻² (at T = 6 °C), C:N ratio=7.5-9.3, C content= 35.1-38, N content= 4.0-4.7 Net N = 4.0 kg m ⁻² (at T = 16 °C)	
<i>Gracilaria vermiculophylla</i>	Red seaweed	Aquaculture effluents	t = 1 month each trial	Land-based pilot scale system Salinity of 30 ppm Mean T oscillates between 10.96 ± 0.19 °C and 20.17 ± 0.03 °C pH = 7.2–8.9 t = 1 month	Nitrogen	N = 40.54 ± 2.02 gm ⁻² month ⁻¹ growth rate= 8% day ⁻¹	Abreu et al. (2011)
<i>Gracilaria caudata</i>	Red seaweed	Aquaculture effluents	t = 72 h	Co-cultured with microcrustacean <i>Artemia franciscana</i> T = 28 oC	Nitrogen and Phosphorus	NO ₂ = 100% NO ₃ = 72.4%	Marinho-Soriano et al. (2011)

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
				Salinity = 35 PSU t = 72 h		DIN = 44.5% PO ₄ increase >100%	
<i>Gracilaria birdiae</i>	Red seaweed	Shrimp wastewater	t = 4 weeks	Salinity of 30.1–30.7 PSU T = 27.2–29.4 oC pH = 7.9–8.1 t = 4 weeks	Phosphate (PO ₄ ³⁻) and Nitrate (NO ₃ ⁻)	PO ₄ ³⁻ = 93.5 % NO ₃ ⁻ = 100 % NH ₄ ⁺ = 34% of growth rate= 2.6% day-1	Marinho-Soriano et al. (2009)
<i>Gracilaria caudata J. Agardh</i>	Red seaweed	Shrimp wastewater	t = 75 d	Co-cultured with in-situ shrimp pond Salinity of 33 PSU Mean T = 29 °C pH = 8.07–8.26 t = 4 h	Nitrogen and Phosphorus	NO ₃ -N = 49.6% PO ₄ -P = 12.3%	Marinho-Soriano et al. (2009)
<i>Gracilaria verrucosa</i>				Co-cultured with the fish <i>Pseudosciaena crocea</i>		PO ₄ -P= 58% NO ₂ -N=48% NH ₄ -N = 61% NO ₃ -N= 47%	Huo et al. (2012)
<i>G. Chilensis</i>				Co-cultured with salmon		NH ₄ -N=95%	Buschmann et al. (2004)

Seaweed	Type	Type of Wastewater	Studied Parameters	Treatment Conditions	Pollutants	Treatment Performance /Maximum reduction efficiency	References
						PO ₄ -P= 32%	
<i>G. Chilensis</i>						growth rate=4% day-1	Buschmann et al. (2008)
<i>Gracilaria edulis</i>				t= 14 d		NH ₄ -N=70% growth rate=4% day-1	Lavania-Baloo et al. 2014
<i>P. yezoensis</i>	red alga					N= 92-93%	Carmona et al. (2006)
						P= 72-85%	
<i>P. yezoensis</i>						DIN= 55.2%	He et al. (2008)
						DIP=54.6%	
<i>P. yezoensis</i>						NH ₄ -N= 50-94%	He et al. (2008)
						NO ₂ -N=42-91%	
						NO ₃ -N= 21-38%	
						PO ₄ -P= 42-67%	
<i>Laminaria Japonica</i>						N=44%, P=40%	Xu et al. (2011)
						N/P ratio = 7.4	

3.1.1. Red seaweed

3.1.1.1. Genus *Gracilaria*

Gracilaria is reported have 197 species (Guiry & Guiry, 2012). *Gracilaria* is commonly used for phycocolloids products (Huo et al., 2012). It grows mostly in tropic and subtropic waters, and they prefer to grow in temperature ranges of 15 - 30 °C (Yang et al., 2006). Most of them, tolerance to salinities, such as *G. vermiculophylla*, can grow at 5-30 S (Kim et al., 2016). They contain polysaccharides with sulfate ester and hydroxyl groups, the primary binding sites (Rathod et al., 2014).

In 2018, *Gracilaria* was the third highest world production reaching 3.4 million tonnes among other aquatic algae (Food and Agriculture Organization (FAO), 2020). *Gracilaria* is a good choice for bioremediation due to its easy propagation, high growth rates (Gorman et al., 2017; Kim et al., 2016), reach high biomass, wide tolerance, and potential commercial value (Kim et al., 2014; Marinho-Soriano et al., 2009; Ohtake et al., 2020; Yang et al., 2006; Zhou et al., 2006). Red algae have a high capability to store nitrogen in red pigment. Hence they can support when nitrogen is lacking (Zhou et al., 2006). So, *Gracilaria* has higher efficiency in removing nutrients and producing new biomass (Huo et al., 2012).

A study conducted by Troell et al. (1997) showed that *Gracilaria chilensis* co-cultures with salmon reduce 5% of nitrogen and 27% of phosphorus. Another study reported that *G. chilensis* can remove 95% ammonium and 32% orthophosphate (Buschmann et al., 2004). Abreu et al. (2011) reported that *G. vermiculophylla* can reduce N components in the water. *G. vermiculophylla* can reduce ammonium to 63%. *G. vermiculophylla* has a high uptake rate of ammonium. When the ammonium concentration in water is high, the nitrogen tissue accumulation is high (Abreu et al., 2011). *G. vermiculophylla* (Abreu et al., 2011) and *G. changii* efficiently removed nitrate from waste (Badraeni et al., 2020). Another study reported that ammonium and nitrate uptake by *Gracilaria changii* was 71% and 56.8%, respectively, with growth rates of 4.1% day⁻¹ (Mawi et al., 2020).

The removal efficiencies nutrient from cultivation of *G. chouae* and *S. macrocephalus* (DIN=34.67%, DIP= 40.64%) (Wu et al., 2015a) are lower than cultivation *Gracilaria caudata* co-cultured with *Artemia franciscana* (DIN= 44.5%) (Marinho-Soriano et al., 2011) and *Gracilaria verrucosa* co-cultured with fish *Pseudosciaena crocea* (DIN=52%, DIP= 58%) (Huo et al., 2012).

Another study reported that *Gracilaria tikvahiae* McLachlan, species native to New England, could remove 28 and 94 kg N ha⁻¹ at Long Island Sound and the Bronx River Estuary, USA. The growth rate and tissue N contents are higher in the Bronx River Estuary than in Long Island Sound, with a growth rate reach to 16.5% day⁻¹ (Kim et al., 2014). The growth rate was higher than *Gracilaria tikvahiae* co-cultured with shrimp *L. vannamei* (mean 4.8 % day⁻¹) (Samocho et al., 2015). The high growth rate in BRE was because N content was rich by 33 μmol L⁻¹ and the ratio of C: N was low < 9 (Kim et al., 2014). In addition, *Gracilaria tikvahiae* growth rate was the highest compared to other species. The growth rate of *G. chouae* was 7.43 % d⁻¹ (Wu et al., 2015a), and *G. lemaneiformis* was 13.9% d⁻¹ (Yang et al., 2006). The average contents of N and P of *G. chouae* were 2.39% and 0.32% dry weight, respectively (Wu et al., 2015a). Therefore, the N content of *G. lemaneiformis* increased by about 1.85% (Zhou et al., 2006). The highest N content has been reported for *Gracilaria tikvahiae*, about 5.7% (Samocho et al., 2015).

Many studies about nutrient bioremediation by *G. lemaneiformis* have been reported. *G. lemaneiformis* is an effective adsorbent that could remove NH₄⁺-N and PO₄-P by 85.53% and 65.97%, respectively (Yang et al., 2006), and they can also remove DIN by more than 90% (Zhou et al., 2006). Besides that, many studies have been conducted using *Gracilaria*

lemaniformis with various animals, such as scallop *Chlamys farreri* (Mao et al., 2009), *P. crocea*, *C. gigas* and *A. japonicus* (Wu et al., 2015b), and with fish *Pseudosciaena crocea* (Wei et al., 2017). The highest reduction efficiency of nutrients was found when *Gracilaria lemaneiformis* was cultivated with scallop *Chlamys farreri* by decreasing 83.7% NH_4^+ and 70.4% P with uptake rate of NH_4^+ and P was 6.3 and 3.3 $\mu\text{mol g}^{-1} \text{DW h}^{-1}$ (Mao et al., 2009). However, *G. lemaneiformis* could remove only 21.0% of DIN and 28.6% DIP when cultivated with fish *Pseudosciaena crocea* with a growth rate is $9.84 \pm 0.39\%/d$. The tissues' average N and P contents were 3.98% and 0.49% dry weight, respectively, with their biomass increasing 5.3 times bigger than the initial weight (Wei et al., 2017). In addition, the reduction efficiency of NH_4^+ , NO_2^- , NO_3^- , and PO_4^{3-} when *G. lemaneiformis* co-cultures with *C. gigas* were 55.56%, 12.59%, 24.22%, and 22.88%, respectively (Wei et al., 2019).

G. vermiculophylla can assimilate nitrogen at high concentrations. They prefer to absorb NH_4^+ to NO_3^- . *G. vermiculophylla* efficiently removes ammonium and phosphate from mussel wastewater with the highest removal efficiencies of 90.5% of ammonium and 81.6% of phosphates (Skriptsova & Miroshnikova, 2011).

Gracilaria birdiae was reported as an efficient biofilter that could remove 93.5% of PO_4^{3-} , 34% of NH_4^+ , and 100% NO_3^- (Marinho-Soriano et al., 2009). Ammonium removal efficiency by *Gracilaria birdiae* was lower than by *Gracilaria edulis*, which can remove ammonium by 70% with growth rates of 4.0% day^{-1} (Lavania-Baloo et al., 2014). It also has been reported that ammonium and nitrate uptake by *Gracilaria edulis* was 72.5% and 58.8%, respectively, with growth rates of 4.3% day^{-1} (Mawi et al., 2020). Du et al. (2013) reported the removal efficiency of nitrogen and phosphate by *G. asiatica*. They showed that *G. asiatica* with high biomass has a lower uptake rate but higher removal efficiency.

3.1.1.2. Genus *Pyropia*/ *Porphyra*

Porphyra is a red seaweed that grows in the temperate zone. Recently, many species of *Porphyra* have been reclassified as *Pyropia* (Kerrison, 2017). For instance, *Pyropia yezoensis* is the potential for bioremediation because they grow fast and can remove nutrients (Kim et al., 2007; Wu et al., 2015c). *Porphyra* also has high tolerance at nitrogen concentrations $>100 \text{ mg m}^{-3}$ (Fei, 2004; Wu et al., 2015c). Fei (2004) reported that good harvest and good quality of the cultivation of *Porphyra yezoensis* Ueda in China reached when nitrogen concentration over 7.2 mM. The quality and yield decrease when the N-nutrient level is below 3.6 mM. *Porphyra* can remove ammonium over 17% with an uptake rate of 3.44 $\mu\text{moles.g}^{-1} \text{. dw. min}^{-1}$ (Chung et al., 2002).

Wu et al. (2015c) compared nutrient concentrations between *Porphyra* cultivation and non-cultivation. The concentration of ($\text{NH}_4\text{-N}$, $\text{NO}_2\text{-N}$, $\text{NO}_3\text{-N}$, and $\text{PO}_4\text{-P}$ = 3.59, 0.71, 18.77, and 1,00 $\mu\text{mol L}^{-1}$, respectively) in cultivation area were lower than in non-cultivation ($\text{NH}_4\text{-N}$, $\text{NO}_2\text{-N}$, $\text{NO}_3\text{-N}$ and $\text{PO}_4\text{-P}$ were 6.46, 2.06, 36.57, and 1.83 $\mu\text{mol L}^{-1}$, respectively) (Wu et al., 2015c). Wu et al. (2017) reported that *P. yezoensis* could remove 3688 and 106 tons of nitrogen and phosphorus, respectively. The tissue nitrogen and phosphorus content were increased by 6.84% DW and 0.18% DW, respectively (Wu et al., 2017).

He et al. reported that the range reduction of $\text{NH}_4\text{-N}$, $\text{NO}_2\text{-N}$, $\text{NO}_3\text{-N}$, and $\text{PO}_4\text{-P}$ by *Porphyra* was 50–94%, 42–91%, 21–38%, and 42–67%, respectively. They measured the removal of nutrients in the open water system. *P. yezoensis* could remove 55.2% of DIN and 54.6% of DIP (He et al., 2008). However, these reduction values are lower than in indoor culture experiments (92–93% of N and 72–85% of P) (Carmona et al., 2006). *P. amplissima* has the highest specific growth rate among other species by 17.9 % day^{-1} , but the highest uptake rate was found in *P. purpurea* and *P. haitanensis*. The highest removal efficiency was

found at an ammonium concentration of 150 μM . *P. umbilicalis* was the highest P removal efficiency by 88% (Carmona et al., 2006).

3.1.1.3. Genus Palmiria

Palmaria palmata has a wide and large morphology. The stocking density could affect nutrient uptake and growth (Abreu et al., 2011; Kim et al., 2013). *P. palmata* could decrease nitrogen concentration to 12% (Sanderson et al., 2012). Kim et al. 2013 compared growth rate N removal at different densities. The growth rate was highest at the lower density (0.2 kg m^{-2}), but the ability to remove N was higher at the high density 4 kg m^{-2} . The highest ammonia and phosphate removal were 211 $\mu\text{mol N L}^{-1} \text{d}^{-1}$ and 20 $\mu\text{mol P L}^{-1} \text{d}^{-1}$, respectively. The tissue nitrogen content of *P. palmata* is affected by time and culturing system. The stocking density increased by 6.9 kg m^{-3} in a year. The mean tissue nitrogen of *P. palmata* is 0.5-0.7% DW higher compared to control. The growth rate reached 1% d^{-1} and the highest nitrogen removal achieved was 1.1% (Corey et al., 2014).

3.1.1.4. Genus Kappaphycus

Kappaphycus is red algae with only six species (Guiry & Guiry, 2011). The study of the efficiency of three species of *Kappaphycus* for reducing ammonium in fish wastewater was reported. The result showed the percentage of efficiency ammonium in the 41 to 66 % range, with the growth rate in the range 2.75-4.41%. *K. striatum* can easily absorb ammonium than *K. alvarezii*. It can be said that *K. alvarezii* need low N (Rodríguez & Montaña, 2007). *K. alvarezii* can potentially remove phosphate from water (Rathod et al., 2014). Recently, nutrient removal of *K. alvarezii* in the IMTA system has been reported with reducing ammonium up to 34% and phosphate up to 30%. The removal efficiency is lower compared to the previous study. The growth rate of *K. alvarezii* was 3.33% d^{-1} (Kambey et al., 2020).

3.1.2. Green seaweed

Ulva is green seaweed that can be used as a biofilter because they grow faster, are easy to culture, and have a higher uptake rate of nutrients (Henriques et al., 2017). For instance, *Ulva rotundata* could remove DIN until 54% (Hernández et al., 2005). *Ulva lactuca* co-cultured with *Penaeus latisulcatus* could remove ammonia and phosphate until 81% and 55%, respectively (Khoi & Fotedar, 2011). The removal efficiency was higher than the study by Lavania-Baloo et al. (2014) which the ammonium removal rate for *Ulva lactuca* was 45% with growth rates of 3.6% day^{-1} . *U. lactuca* also has high nitrate and phosphate uptake of $12.7 \pm 5.1\%$ and $14.4 \pm 9.3\%$, respectively (Tremblay-Gratton et al., 2018).

Kang et al. (2011) investigated the growth and bioremediation potential of *Ulva pertusa* from black rockfish effluents. The biofiltering efficiency of *U. pertusa* for NH_4^+ is more than 80% from effluents. *U. pertusa* has a higher biomass yield than *E. stolonifera*, *S. japonica*, and *G. chorda* but lower than *C. fragile*. Tissue N and P content in *U. pertusa* were 2.1 % and 0.1 %, respectively. *U. pertusa* has the highest uptake rate and can remove ammonia above 80% (Kang et al., 2021).

3.1.3. Brown seaweed

3.1.3.1. Genus Sargassum

Sargassum is a brown seaweed that commonly grows in tropical and temperate regions. *Sargassum* was also suitable to culture in the pond. The optimal salinity and temperature for the growth of most *Sargassum* is 24-42 psu and 24–30°C (Yu et al., 2013). *Sargassum sp.* is renewable, cheap, and effective as a biofilter to remove nutrients in water (Saldarriaga-

Hernandez et al., 2020). *Sargassum hemiphyllum* and *S. henslowianum* are potential for bioremediation. Both *Sargassum* spp have high growth rates and bioaccumulation capacities on inorganic nutrients (Yu et al., 2014). *S. hemiphyllum* grows in shrimp ponds and has a growth rate of 1.65% d⁻¹ (Yu et al., 2013). Another study reported that *S. hemiphyllum* was more suitable to grow on oyster farms than on fish farms because of higher growth rates and nutrient bioaccumulation. The specific growth rate of *S. hemiphyllum* in oyster farm reach up to 7.6% d⁻¹ with total N content in the range of 1.79-2.52% (Yu et al., 2016). This value is higher than *Sargassum macrocarpum* in bio-filtered wastewater with a specific growth rate (0.025 day⁻¹), and total N and P contents were 1.03% DW and 0.55% DW, respectively (Ohtake et al., 2020). Yu et al. (2019) compared *Sargassum henslowianum*'s bioaccumulation efficiency in fish and oyster farms. The results showed that *S. henslowianum* could remove nitrogen higher in oyster farms than in fish farms. The highest specific growth rate of *S. henslowianum* in oyster farms is 6.9% d⁻¹.

3.1.3.2. Genus Laminaria

Laminaria is a brown seaweed that can remove nutrients from the water. *Laminaria japonica* could remove 44% of N and 40% of P. The uptake rates varied from 0.54 to 270 μmol N g⁻¹ DW h⁻¹. The optimum N/P ratio for nutrient uptake was 7.4 (Xu et al., 2011).

3.2. Integrated multi-trophic aquaculture (IMTA)

Starting with research on wastewater from fish cultivations which can be used as nutrients for seaweed growth, an integrated multi-trophic aquaculture (IMTA) system was developed (Chopin et al., 2001; Troell et al., 1997). This system has been suggested as a suitable method for reducing nutrient levels in aquaculture effluent (Reid et al., 2013; Scherner et al., 2012). IMTA combination between the cultivation of two species, namely fed species (fish) and extractive species (seaweed) (Buschmann et al., 2009; Holdt & Edwards, 2014; Kang et al., 2008; Kang et al., 2013). Seaweed will use nutrients from the excretion of animals for growth so that this process reduces the effluent's nutrients and increases Both seaweeds and cultivated animals can be produced (Abreu et al., 2011; Kim et al., 2017; Nobre et al., 2010; Wei et al., 2017; Wu et al., 2015b). Ease of cultivation is essential in choosing the type of seaweed suitable for IMTA. Seaweed species should also be selected based on high nutrient absorption efficiency (Skriptsova & Miroshnikova, 2011). In addition, the suitability of ecophysiological characteristics such as biofiltration capacity, biochemical composition, and growth rate with environmental conditions must be considered (Kang et al., 2008). The method base on index calculation has been developed to select suitable seaweed species in IMTA. The species with the highest index value will be selected (Kang et al., 2013). The study of bioremediation efficiency was conducted by Kang et al. (2021). They compared five species of seaweed in black rockfish effluents. According to the results, *Ulva pertusa* was the highest uptake rate among others. In 2015, the model was also developed to increase IMTA system efficiency. The model simulated interaction parameters in the IMTA system and varying environmental factors (Lamprianidou et al., 2015). The aeration can influence nutrient uptake in the IMTA system. For instance, the total ammonia uptake by *Ulva lactuca* is higher using continuous aeration than intermittent aeration (Ben-Ari et al., 2014).

In 1996, salmon and *Gracilaria chilensis* cultivation succeeded in reducing nutrient levels from effluent (Buschmann et al., 1996). The nutrient sequestration from the IMTA system can also be estimated, as reported by Reid et al. (2013). In another study, Yu et al. (2016) reported that *Sargassum hemiphyllum* cultivated in fish or shrimp ponds had greater biomass than those grown in natural habitats. In 2020, the IMTA method was used in tropical

environments. That method integrated *Solieria filiformis* with fish and sea cucumber (Felaco et al., 2020). Compared to monoculture, the IMTA method can reduce higher levels of nutrients. For instance, co-culture *Litopenaeus vannamei* with *Ulva linza* reduces 99% nitrogen and 98.8% phosphorus (Gao et al., 2022). Besides, the red seaweed *Agardhiella subulata* co-cultured with *Lutjanus campechanus* can absorb nitrate and phosphate more than 90% (Lohroff et al., 2021). Recently, the integration of green seaweed, *E. intestinalis*, with *C. chanos* and *P. vannamei* caused a decrease in nitrogen and phosphorus concentration (Naskar et al., 2023). So, it can be said that the IMTA method is an effective method for bioremediation. A stable isotope could be used in IMTA to determine the extractive species that are more efficient in absorbing nutrients (Park et al., 2021).

3.3. Factors affecting macroalgae growth rate and nutrient uptake

Many factors influence seaweed growth. The seaweed growth is related to the nutrient tissue levels. The nutrients will be stored in the tissue with high nutrient levels. The factors are described below (Ohtake et al., 2020).

3.3.1. Temperature

Temperature is the physical factor affecting seaweed's metabolism and physiology (Ohtake et al., 2020; Roleda & Hurd, 2019)—for example, enzyme regulation, constant chemical reaction, and nutrient diffusion rate. In addition, temperature affects the rate of nutrient absorption (Roleda & Hurd, 2019). Seaweed nutrient uptake rate increases with increasing temperature. At low temperatures, the absorption of nutrients will be slow due to the inhibition of protein transport through the cell membrane. On the other hand, high temperatures promote rapid growth and require more energy, thereby increasing nutrient uptake. For example, *G. asiatica* can remove nutrients effectively at high temperatures (Du et al., 2013), the growth rate of *Gracilaria tikvahiae* increase when temperature increase from 15 to 25 °C (Kim et al., 2016; Samocha et al., 2015), *G. lemaneiformis* grow well in the range temperature 12-23 °C (Yang et al., 2006), *U. prolifera* and *U. lactuca* prefer grow at 8-10 °C (Luo et al., 2012; Tremblay-Gratton et al., 2018), and *G. cervicornis* has the highest growth rate at temperature 28 °C (Kim et al., 2016). The biosorption of *K. alvarezii* increased with increasing temperature (Rathod et al., 2014). In contrast, the growth rate of *Palmaria palmata* decreased at temperatures over 14 °C (Corey et al., 2014), the growth rate and tissue N content of *G. vermiculophylla* decreased with increasing temperature, and *Gracilaria tikvahiae* cannot survive above 29 °C (Gorman et al., 2017) and under ten °C (Kim et al., 2016). However, *Sargassum* prefers to grow at 15-18 °C (Sfriso & Facca, 2013). Besides, the growth rate decreases when the temperature decreases in *Gracilaria tikvahiae* (Kim et al., 2014). Another study found that temperature influenced the P uptake in *L. japonica* (Xu et al., 2011). In addition, Samocha et al. (2015) also found that the temperature is positively correlated with C:N ratio.

3.3.2. Nutrient availability

Nutrient availability affects the physiological response of seaweed (Abreu et al., 2011). Nitrogen is necessary for seaweed growth and is usually a limiting factor (Alstyne, 2016; Kang et al., 2011). However, some seaweed, like *Gracilaria coronopifolia*, is phosphorus-limited (Tsai et al., 2005). Nitrogen in the form of nitrate and ammonium is needed for seaweed growth. Nitrate has a higher energy requirement than ammonium for absorption and assimilation. It is indicated that nitrate is more efficient as a nitrogen source when the light is limited, and the growth is slow (Pritchard et al., 2015). The preference for N absorbed can be

influenced by environmental parameters (oxygen, pH, CO₂, and light), nutrient concentrations and chemical species and depends on the seaweed species itself (Abreu et al., 2011; Johnson et al., 2014; Roleda & Hurd, 2019). For instance, *G. vermiculophylla* (Skriptsova & Miroshnikova, 2011), *G. asiatica* (Du et al., 2013), *U. pertusa* (Kang et al., 2011), *P. yezoensis* (Carmona et al., 2006), and *Anotrichium crinitum* (Pritchard et al., 2015) prefer to absorb ammonium than nitrate. *Gracilaria* could absorb ammonium until 90% of the total dissolved nitrogen (Buschmann et al., 2008; Buschmann et al., 2009). Besides, *Ulva pertusa* can utilize ammonium until 100% (Kang et al., 2021). The *Ulva fasciata*'s nutrient uptake rate is faster when ammonium is only a nitrogen source (Shahar et al., 2020). Seaweeds usually take up NH₄⁺, which can be directly incorporated into amino acids, although they utilize various forms of nitrogen (NH₄⁺, NO₃⁻, NO₂⁻, and urea) in seawater. Ammonium is directly used in the assimilation process while nitrate should be deoxidated to amine by nitrate reductase before metabolism (Du et al., 2013).

However, some species prefer NO₃⁻ over NH₄⁺ or utilize both forms equally (Kang et al., 2021). For instance, *Saccharina japonica*, *Gracilariopsis chorda* (Kang et al., 2011), and *L. japonica* prefer to absorb nitrate than ammonium (Xu et al., 2011). In addition, *Sargassum* is preferred to grow in an environment with high nitrate concentration (Sfriso & Facca, 2013). Another study reported that the specific growth rate of *Ulva lactuca* cultivated in NO₃⁻ was higher than in NH₄⁺ (Sode et al., 2013).

Nutrient uptake rate increases when the nutrient concentration is high (Du et al., 2013). For instance, nitrate concentration in water affects the nitrate uptake rate of *Porphyra*. When nitrate concentration is 30 μM, the uptake rate is higher than three μM nitrate concentration (Pedersen et al., 2004). The NH₄⁺ uptake rates of *Gracilaria vermiculophylla* increased with the increasing NH₄⁺ (Abreu et al., 2011).

When the nitrogen content is high, the tissue nitrogen concentration in *Porphyra* species is also high (Kang et al., 2009). The amount of nitrogen and phosphorus in tissue describes seaweed's ability to remove nutrients (Neori et al., 2004).

3.3.3. Water motion/ movement

Water motion is important because it regulates the supply of nutrients (Roleda & Hurd, 2019). Nagler et al. (2003) had been reported that water motion in the fish tank was lower than in the lagoon. The water motion can affect the growth rate. For example, the growth rate of *Gracilaria parvispora* in the tank was lower than in the lagoon (Nagler et al., 2003). The growth rate of *Gracilaria parvispora* increased with water motion (Ryder et al., 2004)—nutrient uptake rate increases as water flow increases. For instance, nutrient uptake rates of *Gracilaria vermiculophylla* and *Undaria pinnatifida* were higher in the continuous flow system than in the static system (Skriptsova & Miroshnikova, 2011). On the other hand, water motion can be a limiting factor for *K. alvarezii* growth (Kambey et al., 2020).

3.3.4. Light

Light is essential in photosynthesis and seaweed growth, so a lack of light will inhibit the growth rate (Roleda & Hurd, 2019). For example, the nutrient uptake rate of *Saccharina japonica* increased during the daytime when there were many irradiances (Kang et al., 2011). In contrast, *Anotrichium crinitum* has a higher growth rate under low light (Pritchard et al., 2015). Besides that, the nutrient uptake of *L. japonica* is affected by irradiance. The N uptake was higher at 18 μmol photons m⁻² s⁻¹, and P uptake rate was the highest at 144 μmol photons m⁻² s⁻¹ (Xu et al., 2011). Light also affected ammonium uptake by *Ulva prolifera* (Sun et al., 2015). *U. prolifera* prefer grow at 50-500 μmol photons m⁻² s⁻¹ (Luo et al., 2012).

3.3.5. Salinity

Salinity affects the growth of *G. vermiculophylla* and *Gracilaria tikvahiae*. *Gracilaria tikvahiae* did not grow at a salinity under 20, but *G. vermiculophylla* can grow well in the range of 15-30 (Kim et al., 2016). The optimal salinity for *S. hemiphyllum* is 30-34 (Yu et al., 2016). In addition, salinity and temperature affected the nitrate uptake rate in *Kappaphycus alvarezii*. The nitrate uptake rate was higher at higher salinities and temperatures (Mandal et al., 2015). In contrast, salinity did not influence *U. lactuca* growth (Bews et al., 2021). Brown seaweeds are the most tolerant to low salinities among all types of seaweed (Tanaka et al., 2020).

3.3.6. Ratio

C: N or N:P ratios of seaweed tissues determine nutrient limitation. If N:P ratio < 16, nitrogen is a limiting factor; if N:P ratio > 24, phosphorus is a limiting factor for seaweed growth. For instance, *C. fragile*, *U. pertusa*, and *G. chorda* is P limited, while *E. stolonifera* and *S. japonica* are N-limited (Kang et al., 2021). The ratios are varied depends on the species. C: N ratios show high values when nitrogen is limited and decrease when nitrogen concentration is high (Nelson et al., 2001). For instance, the N:P ratio is low in *U. pertusa* and *S. japonica* because of higher tissue N contents. Meanwhile, the N:P ratio of *G. chorda* did not change due to increasing tissue P content. This concludes that the N:P ratio value differs depending on nitrogen and phosphate uptake rates (Kang et al., 2011). N:P ratio affected nutrient uptake rate. The nutrient uptake rate decreased when the N:P ratio increased (Xu et al., 2011). Increasing nutrient concentration can enhance assimilation process that increases seaweed growth. The growth rate of *G. lemaneiformis* increased when the N/P concentration increased and reached the critical point at 400/25 $\mu\text{mol/L}$ (Yu & Yang, 2008).

Seaweed density and cultivation period also affect nutrient reduction efficiency. For instance, *Gracilaria lemaneiformis* has the highest ammonium and phosphorus reduction efficiency when the high seaweed density and the long cultivation (Mao et al., 2009).

3.4. Nutrient uptake, assimilation, and storage

Nutrient uptake mechanisms involve passive and active transport (Pedersen et al., 2004). Inorganic nutrients move over the cell membrane by three mechanisms: (1) passive transport by passive diffusion; (2) facilitated diffusion using a carrier or channel proteins; and (3) active transport that needs energy to fuel membrane transport systems (Roleda & Hurd, 2019).

Nitrate uptake is by macroalgae through active transport, whereas ammonium uptake is facilitated or passive diffusion requires less energy than nitrate. If ammonium concentrations are high, ammonium uptake will become saturated because the supply exceeds the metabolic demand (Pritchard et al., 2015; Roleda & Hurd, 2019).

Seaweed can store and assimilate nutrients in high concentrations when the nutrients needed for growth are in excess (Ohtake et al., 2020). NO_3^- is reduced to NH_4^+ , which involves nitrate and nitrite reductase, and energy from photosynthesis (Phillips & Hurd, 2004; Pritchard et al., 2015). NO_3^- can be stored in inorganic form in cellular vacuoles (Phillips & Hurd, 2004). However, ammonium cannot be stored in high concentrations in cells due to the fast process of transformation into amino acids (Roleda & Hurd, 2019). Nitrogen can be stored in limited light or at low temperatures depending on each species' capabilities (Phillips & Hurd, 2004; Pritchard et al., 2015).

3.5. Capability of seaweed for bioremediation

In this study, we have reviewed the potential capability of seven seaweed genus. The most capable genus for bioremediation was highlighted in Figure 1 (i.e., *Gracilaria*, *Pyropia*, and *Ulva*). Genus *Gracilaria* has been reported as the most widely used genus as a nutrient adsorbent. *Gracilaria lemaneiformis* co-culture with *Chlamys farreri* is the most effective for removing nutrients, but the growth rates are low. *Gracilaria birdiae* has the highest efficiency for nitrate and phosphate removal among all *Gracilaria*. *Pyropia yezoensis* is also effective for bioremediation. *Ulva lactuca* and *Ulva pertusa* can remove nitrogen above 80% (Figure 2).

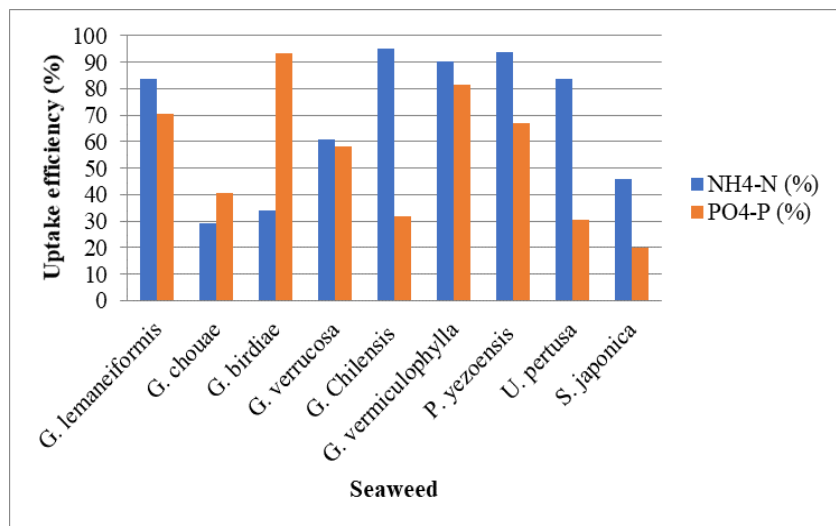


Figure. 2 Uptake Efficiency NH₄-N and PO₄-P from various species of seaweed

4. Conclusion

Seaweeds are potential adsorbents to remove nutrients from the water to act as bioremediation agents. There are more studies on bioremediation using red seaweeds than any other genus. Until now, *Gracilaria* has been the most effective in decreasing nutrients when co-cultured with animals. In general, the cultivation of seaweed can decrease eutrophication in coastal areas. The choice of seaweed as a suitable biofilter must be based on many factors. They have to be easy to cultivate and have higher removal efficiency. We hope this review can give useful information to decide on macroalgae for biofilters to improve the environment and increase economic value. Further studies are needed to evaluate the aquaculture method. It is recommended that principal components analysis (PCA) be used to correlate seaweed growth and environmental factors. This recommendation helps find the best methodology to increase the quality of cultivated seaweed. It is also necessary to explore more genus and species of seaweed that have the ability as a biofilter.

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Author Contribution

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Visualization, H.M.; Supervision, H.M. and A.J.W.; Project Administration, H.M. and A.J.W.; and Funding Acquisition, H.M. and A.J.W.

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