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A comparison of physiological responses between attached and pelagic populations of *Sargassum horneri* under nutrient and light limitation

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ABSTRACT

Large-scale *Sargassum* blooms have been increasingly observed in coastal zones in recent years. *Sargassum horneri* (Turner) C. Agardh blooms (pelagic) have been observed in Jeju Island (Korea) and the southwest of the Korean Peninsula, causing serious problems for seaweed and abalone farms as well as for fisheries, tourism and recreational industries. The present study explored the physiological responses of attached and pelagic *S. horneri* populations cultivated under different nutrient concentrations (HN: 50 μM of nitrogen and 5 μM of phosphorus; LN: 5 μM of nitrogen and 0.5 μM of phosphorus) and photosynthetically active radiation (PAR) (H-PAR: 250; M-PAR: 150; L-PAR: 50 μmol photons m^{-2} s^{-1}) for 25 days. Relative growth rates (RGR) were significantly lower in the pelagic population than that in the attached population. All thalli from the pelagic population died within 20 days. Chlorophyll *a and c*, and carotenoids were significantly higher at HN than at LN, and decreased as PAR increased for both populations. For the attached population, photosynthetic rate, tissue nitrogen, and carbon and nitrogen removal were also significantly higher at HN than at LN. These results suggest that high nutrient and lower PAR increased the biomass accumulation of attached populations in coastal areas. Nutrient limitation and high PAR may accelerate senescence of the pelagic populations while traveling on the sea surface from their point of origin.

1. Introduction

Pelagic *Sargassum* was first documented in the Sargasso Sea in the 15th century (Fine, 1970; Wang et al., 2019). Recently, pelagic *Sargassum* rafts have become major issues in the Gulf of Mexico and Caribbean Sea (Cruz-Rivera et al., 2015). Pelagic *S. horneri* (Turner) C. Agardh rafts have also been reported in the Yellow Sea and East China Sea since 2000. These blooms did not receive much attention because they drifted into the open sea without causing serious damage to coastal areas (Zhuang et al., 2020). However, some unusual events happened since 2010s. For example, large amounts of *S. horneri* rafts were observed in the southern part of East China Sea (northern coast of Taiwan and on Tarama Island in the Ryukyu Archipelago) in 2012 (Komatsu et al., 2014a). These blooms even transported to the offshore of Jiangsu Province, China in 2017, affecting nearly 200 km^2 of *Neopyropia* farms (Zhuang et al., 2020). These blooms caused a significant reduction of *Neopyropia* production. Xing et al. (2017) estimated the economic loss due to these blooms was about US$73 million in 2017. *Sargassum horneri* blooms have also occurred in Korea since 2013 (Kim et al., 2019). A significant amount of *S. horneri* biomass was removed from Shinan-gun, Jeonnam Province (>5000 tons) and Jeju Island (>20,000 tons) between January and May 2015 (Hwang et al., 2016). These *Sargassum* blooms caused serious problems to seaweed and abalone farms (Hwang et al., 2016; Liu et al., 2021). Shore-based activities such as fisheries, tourism, recreation activities, etc., were also impacted by these pelagic *S. horneri* rafts (Choi et al., 2020). Furthermore, toxic

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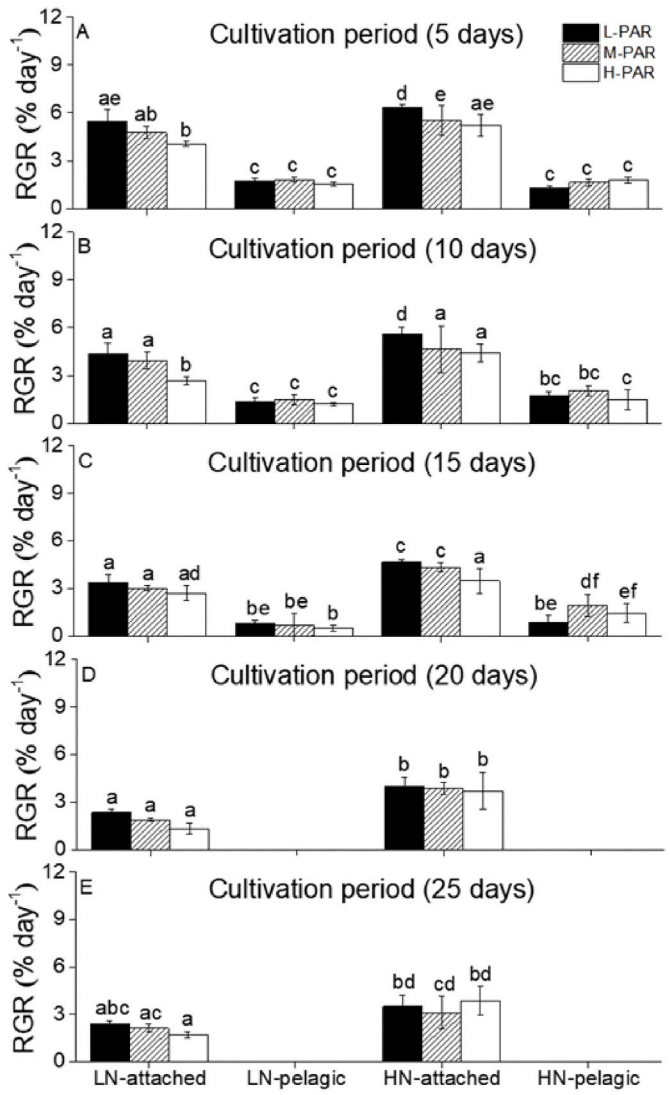

Fig. 1. Relative growth rate (% day^{-1}) (RGR) of attached and pelagic populations of *Sargassum horneri* cultivated under various conditions for 25 days. Attached: attached population of *S. horneri*; pelagic: pelagic population of *S. horneri*; L-PAR: 50 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1}; H-PAR: 250 μmol photons m^{-2} s^{-1}; LN: 5 μmol L^{-1} nitrogen and 0.5 μmol L^{-1} phosphorus; HN: 50 μmol L^{-1} nitrogen and 5 μmol L^{-1} phosphorus. Data show mean values ± SD (n = 3). Three-way analysis of variance (ANOVA) was used to analyze the statistical differences in different PAR and nutrient conditions. Different letters represent significant differences ($P < 0.05$) among different treatments.

compounds such as hydrogen sulfide and ammonia are produced when seaweeds decompose (Sfriso and Facca, 2013; Resiere et al., 2018). However, pelagic *S. horneri* rafts can provide some economic benefits by providing spawning and nursing grounds for fish (e.g. flying fish, Pacific saury) and invertebrate fauna (Abé et al., 2013; Komatsu et al., 2014b). The pelagic *Sargassum* is a good habitat for yellowtail juveniles (Xu et al., 2016). The seaweed biomass from seaweed blooms can also be used for biofuels, fertilizer and nutraceutical products (Milledge et al., 2016; Silva et al., 2019; Thompson et al., 2020). *Sargassum* spp. have been used as functional material in traditional medicine for thousands of years, and as part of nutritional supplement, for their vitamins, amino acids and polysaccharides (Sanjeewa et al., 2017).

Sargassum horneri grows abundantly on solid substrata and forms underwater forests in sublittoral regions (Yatsuya, 2007). Mature *S. horneri* contains a large amount of spherical gas-filled bladders, allowing the thalli to stand upright from the bottom to the surface of seawater (Yoshida, 1963; Wang et al., 2014; Sanjeewa et al., 2018). When mature, some *S. horneri* thalli can be detached from the substratum due to the force of wave and currents, and gas-filled bladders allow the thalli to float away from their point of origin. These pelagic populations can grow vegetatively for a period of time (Xu et al., 2016, 2018). Recent studies suggested that spring *Sargassum* blooms in the East China Sea originated from the Zhejiang coast, transported to the northeast via Kuroshio current and Taiwan warm current, and reached Jeju Island, Jeonnam Province, Korea, and Japanese coastal waters by the early April (Fig. S1A) (Qi et al., 2017; Byeon et al., 2019; Zhang et al., 2019; Zhuang et al., 2020). Zhuang et al. (2020) also reported that the local attached *S. horneri* in Jeju can also be detached and help form the floating golden tides. A more recent study suggests that the spring bloom in 2020 probably originated from the eastern end of Shandong Peninsula or even Dalian and Changdao in the northern Yellow Sea (Fig. S1B) (Yuan et al., 2022).

The pelagic population of seaweeds including *Sargassum, Macrocystis, Durvillaea,* etc. experience a lot of environmental stresses (high or low PAR, nutrient limitation, etc.) during their journeys from their origins to distant shores (Norton and Mathieson, 1983; Rothäusler et al., 2011a, 2012; Tala et al., 2017). Most pelagic seaweeds start to grow in benthic habitats during their early life stages. After being detached, pelagic populations continue to grow if environmental conditions are suitable, and travel considerable distances with winds and currents (Fraser et al., 2009). At the sea surface, however, increased photosynthetically active radiation (PAR) and UV radiation, and nutrient limitation will affect growth and other physiological functions of pelagic seaweeds (Lapointe, 1995; Rothäusler et al., 2012). Therefore, the pelagic populations may have adapted to environmental stresses and might have enhanced growth capacity when compared to the local attached populations. To date, the *S. horneri* bloom studies have mostly focused on geographical distributions (Yoshida et al., 2004) and their ecological and economic impacts (Qi et al., 2017). We are not aware of any studies that have compared physiological responses between the attached and pelagic populations of *S. horneri*. In this study, attached and pelagic *S. horneri* populations were cultivated under different regimes of nutrients and photosynthetically active radiation (PAR) to explore physiological differences between these populations.

2. Material and methods

2.1. Collection of Sargassum horneri and sample preparations

The attached *S. horneri* samples (30–50 cm length) were collected from Munseom, Jeju Island, Korea (33°13′ 38″N; 126°34′ 04″E) on April 8th, 2020. The pelagic samples were collected from Donggui, Jeju Island (33°29′11″N; 126°24′28″E) on April 9th, 2020. The pelagic samples were not originally from the sampling site since no local populations of *S. horneri* were found near that location. Our local population survey also indicated no evidence of detachment from Jeju before we made our collections. East China Sea or northern Yellow Sea may be the origin of this population according to the satellite images and literature (Yuan et al., 2022). However, no firm evidence was found to confirm the origin. The collected samples were brought to the laboratory in a cooler with ice. The apical tips (3–4 cm long) were cut and washed carefully with artificial seawater to remove sediments and epiphytes. Air bladders were observed in both populations (Fig. S2), but receptacles were not. The *S. horneri* samples were then acclimated in von Stosch enriched (VSE) medium (Ott, 1965) for 5 days, 30 psμ of salinity, 80 ± 10 μmol photons m^{-2} s^{-1} of photosynthetically active radiation (PAR) provided by light-emitting diode (LED) lamps, and a photoperiod of 12 h light-12 h dark. Temperature was maintained at 10 °C.

2.2. Experimental setup

After acclimation, healthy algal tips were selected randomly and cultivated in 250 mL conical flasks. The stocking density was 1.0 g L^{-1}. *Sargassum horneri* was exposed to three levels of PAR (H-PAR: 250 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1} and L-PAR: 50 μmol photons m^{-2} s^{-1}) and two different nutrient concentrations (HN: 50 μM of nitrogen and 5 μM of phosphorus and LN: 5 μM of nitrogen and 0.5 μM of phosphorus). The nutrient levels were determined based on the concentrations in different seasons/regions when/where *S. horneri* blooms occur (Wu et al., 2015; Kang and Chung, 2017). The PAR values were determined based on optimal (M-PAR) or sub-optimal (L-PAR and H-PAR) conditions in literature (Zhang et al., 2014; Wang et al., 2021). *Sargassum horneri* were cultivated in von Stosch-enriched (VSE) medium (Ott, 1965), with adjustment of nitrogen and phosphorus, for 25 days. Each treatment had three replicates. The culture media were renewed every 5 days. *Sargassum horneri* thalli were cultivated in an Intelligent Illumination Incubator (BF400-PEC, Biofree, Korea). Different PAR levels were provided by light-emitting diode (LED) lamps with the accuracy of ±10 μmol photons m^{-2} s^{-1}. All other conditions were same as mentioned above for acclimation.

2.3. Growth rate

Fresh weight (FW) of *S. horneri* was measured after blotting the thalli dry with paper towels every 5 days, when the medium was renewed. The seaweed biomass in each flask was reduced to initial stocking density (1.0 g L^{-1}) at each weighing by removing entire fronds without cutting fragments whenever possible. Relative growth rate (RGR) was calculated as follows:

$$\mathrm{RGR}\ (\%\ \mathrm{day}^{-1}) = \mathrm{In}\ (W_t/W_0)/t \times 100$$

Where W_t and W_0 are the FW at days t and 0, respectively.

2.4. Photosynthesis and respiration

The net photosynthetic and dark respiration rates of *S. horneri* were measured with an optical DO sensor at the end of experiment (Pro ODO-BOD, YSI, USA). Approximately 0.25 g FW of *S. horneri* from each treatment were placed in a 100 mL BOD bottle containing seawater. The temperature was maintained at 10 °C and PAR was set as same as each treatment. The dissolved oxygen content (mg L^{-1}) was recorded every 30 s in the BOD bottle during light or dark condition. The net photosynthetic and dark respiration rates were described as mg O_2 L^{-1} g FW^{-1} h^{-1}.

2.5. Pigment contents

The chlorophyll *a* (chl *a*), chlorophyll *c* (chl *c*) and carotenoids were determined every 5 days following the methods by Barradas et al. (2018). Approximately 0.03 g FW of algal thalli was homogenized with 2 mL of absolute methanol at 4 °C for 24 h in dark. The homogenate was then centrifuged at 4000 g for 30 min at 4 °C. The absorbance of supernatant was determined using a spectrophotometer (Orion AquaMate 8000, Thermo Fisher Scientific Solutions LLC, Korea) at 470, 632 and 665 nm, respectively. Chlorophyll *a* (chl *a*) and chlorophyll *c* (chl *c*) were measured according to the equations used in Ritchie (2008). Carotenoid content was measured based on the coefficients of absorption of chl *a* proposed by Lichtenthaler and Buschmann (2001) and chl *c* proposed by Jeffrey (1963) following the model of Lichtenthaler (1987). The carotenoid contents were calculated based on the equation reported by Barradas et al. (2018).

2.6. Soluble protein

At the end of experiment, the total soluble protein was determined following Bradford protein assay (Bradford, 1976). Briefly, approximately 0.1 g of FW algal tissue from each treatment were homogenized with 5 mL extraction buffer (0.1 mol L^{-1}, potassium phosphate buffer, pH 7.0) containing 0.25% Triton X-100 and 1% polyvinylpyrrolidone, and then centrifuged for 10 min at 12000g. The supernatant was mixed with Bradford's reagent and allowed for 5 min prior to measuring absorbance at 595 nm. Protein contents were calculated with bovine serum albumin as a standard and expressed as mg g^{-1} FW.

2.7. Tissue carbon and nitrogen contents

The algal samples were collected from each treatment at the end of experiment to measure tissue contents (carbon and nitrogen). The *S. horneri* samples were oven-dried at 60 °C until constant weight, and then ground to powder using MM400 Ball Mill (Retsch, Germany). The tissue carbon and nitrogen contents were measured using a CHN analyzer (Series II, CHNS/O 2400 Analyzer, PerkinElmer Inc., USA). Carbon and nitrogen removal of *S. horneri* under each nutrient condition was calculated based on the equation of Kim et al. (2007):

$$\mathrm{C\ (or\ N)\ removal}\ \left(\mathrm{mg\ C(or\ N)g\ DW^{-1}day^{-1}}\right) = \frac{(\mathrm{Wt} - \mathrm{W0})^{*}\mathrm{Tissue\ C\ (or\ N)}}{\mathrm{t}} * \frac{\mathrm{DW}}{\mathrm{FW}}$$

Where Wt and W0 represent the fresh weight at day t and 0, respectively.

2.8. Data analysis

Results are expressed as the mean of triplicate analysis ±standard deviation. Data were processed using Origin 9.0 and SPSS 25.0 software. The data conformed to a normal distribution from each treatment and the variances were considered equal (Levene's test, $P > 0.05$). Three-way ANOVA was used to analyze the effects of nutrient, PAR and populations on RGR and pigments. Two-way ANOVA was used to analyze the effects of nutrient and PAR on net photosynthetic rate, dark respiration, soluble protein, tissue C and N, C:N, and C and N removal. Tukey's honest significant difference (Tukey's test) was used for post hoc investigation. A confidence level of 95% was set for all analyses.

3. Results

3.1. Growth

Relative growth rates (RGRs) of pelagic and attached *S. horneri* populations varied with cultivation periods (Fig. 1). RGRs were significantly influenced by the type of population ($P < 0.001$) and nutrients ($P < 0.05$, Table S1). RGRs of the attached population were significantly higher in comparison to the pelagic population (Fig. 1; $P < 0.001$), regardless of nutrient and PAR levels. All thalli from the pelagic population died within 20 days.

For the pelagic population, both nutrient and PAR had no significant effects on growth ($P > 0.05$, Table S1). In the attached population, RGR decreased as time passed, and the growth rates of the attached population were significantly influenced by nutrients during the entire

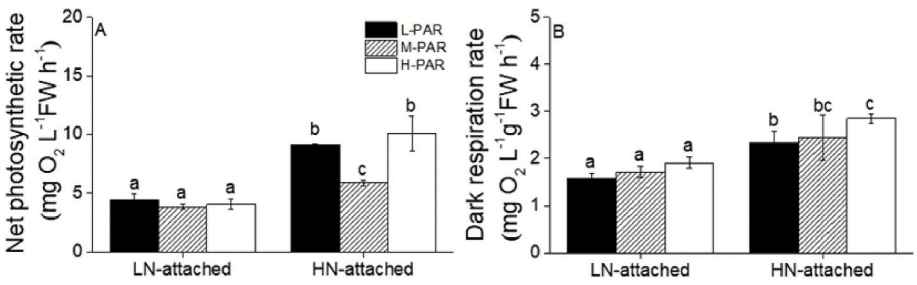

Fig. 2. Net photosynthesis (A) and dark respiration (B) rates of the attached population of *Sargassum horneri* cultivated under various conditions for 25 days. L-PAR: 50 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1}; H-PAR: 250 μmol photons m^{-2} s^{-1}; LN, 5 μmol L^{-1} nitrogen and 0.5 μmol L^{-1} phosphorus; HN, 50 μmol L^{-1} nitrogen and 5 μmol L^{-1} phosphorus. Data show mean values ± SD (n = 3). Two-way analysis of variance (ANOVA) was used to analyze the statistical differences in different PAR and nutrient conditions. Different letters represent significant differences ($P < 0.05$) among different treatments.

cultivation period (Fig. 1, $P < 0.05$). RGRs varied from 4.04 ± 0.15% (LN H-PAR) to 6.31 ± 0.18% (HN L-PAR) on day 5, then decreased to 1.70 ± 0.18% (LN H-PAR) to 3.87 ± 0.92% (HN H-PAR) on day 25 (Fig. 1A, E). At H-PAR the RGR significantly decreased until day 10 (LN) or 15 (HN) in the attached population while RGR was not significantly affected by PAR in the pelagic population ($P > 0.05$).

3.2. Photosynthesis and respiration

Net photosynthetic and dark respiration rates of the attached population of *S. horneri* were significantly influenced by nutrients (Fig. 2, Table S2, $P < 0.001$). HN significantly enhanced both the net photosynthetic and respiration rates, regardless of PAR levels ($P < 0.001$). At HN the photosynthetic rate significantly increased regardless of PAR levels ($P < 0.05$). No differences were observed between L-PAR and H-PAR at both LN and HN conditions (Fig. 2A). Similar to the photosynthetic rate, higher PAR had no significant effects on dark respiration rates at LN ($P > 0.05$). HN increased significantly dark respiration rate in comparison to LN ($P < 0.05$). The rates were also increased as PAR increased at HN ($P = 0.020$, Fig. 2B).

3.3. Pigment contents

Pigment contents (chl *a*, chl *c* and carotenoid) were significantly influenced by population type ($P < 0.001$), nutrients ($P < 0.001$) and PAR ($P < 0.001$) (Table S2). Chlorophyll *a and c* were significantly influenced by the combination of nutrients and PAR (Chl *a*, $P = 0.030$; Chl *c*, $P = 0.042$) and of nutrient and population (Chl *a*, $P = 0.020$; Chl *c*, $P = 0.017$) (Table S2). In both populations, chl *a* was significantly higher at HN than LN. Chlorophyll *a* decreased as PAR increased (Fig. 3A). Similar to chl *a*, chl *c* was significantly higher at HN than LN. Chl *c* decreased as PAR increased (Fig. 3B). Carotenoids were significantly higher at HN than LN. Carotenoids decreased as PAR increased (Fig. 3C).

3.4. Soluble protein

Soluble protein was significantly influenced by nutrients ($P = 0.017$) and PAR ($P = 0.002$), and the combination of both ($P = 0.010$) (Table S3, Fig. 4). PAR influenced protein concentrations differently at different nutrient conditions: at LN, the soluble protein contents were higher at L-PAR and decreased as PAR increased, significant difference were founded between L-PAR and H-PAR ($P = 0.002$); at HN, the soluble protein contents were not significantly different at different PAR conditions ($P > 0.05$).

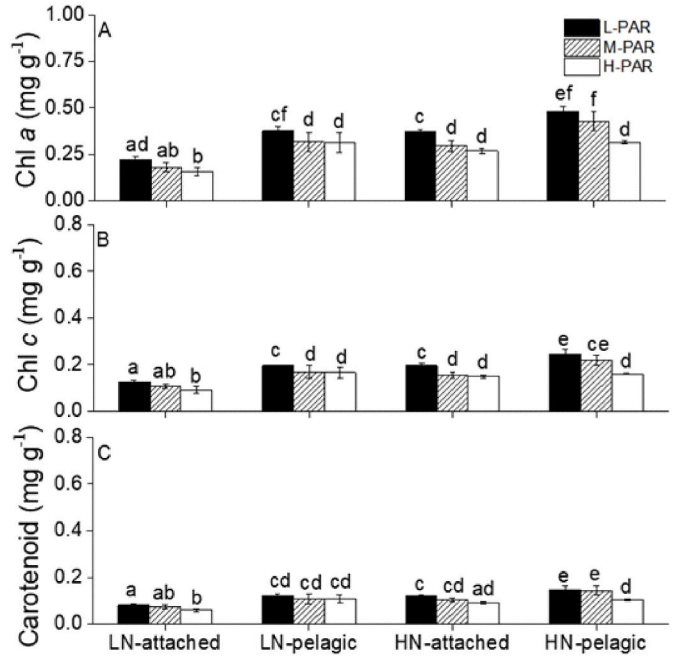

Fig. 3. Pigment contents of attached and pelagic populations (chlorophyll *a* (chl *a*) (A); chlorophyll *c* (chl *c*) (B); carotenoids (carotenoid) (C)) of *Sargassum horneri* cultivated under various conditions. L-PAR: 50 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1}; H-PAR: 250 μmol photons m^{-2} s^{-1}; LN: 5 μmol L^{-1} nitrogen and 0.5 μmol L^{-1} phosphorus; HN: 50 μmol L^{-1} nitrogen and 5 μmol L^{-1} phosphorus. Data show mean values ± SD (n = 3). Three-way analysis of variance (ANOVA) was used to analyze the statistical differences in different PAR and nutrient conditions. Different letters represent significant differences ($P < 0.05$) among different treatments.

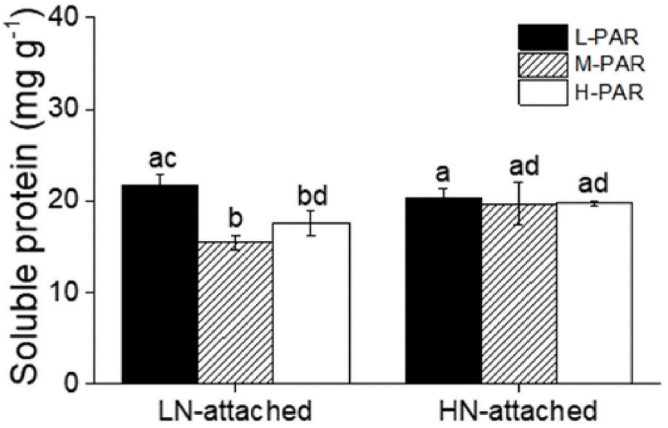

Fig. 4. Soluble protein of attached population of *Sargassum horneri* cultivated under various conditions. L-PAR: 50 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1}; H-PAR: 250 μmol photons m^{-2} s^{-1}; LN: 5 μmol L^{-1} nitrogen and 0.5 μmol L^{-1} phosphorus; HN: 50 μmol L^{-1} nitrogen and 5 μmol L^{-1} phosphorus. Data show mean values ± SD (n = 3). Two-way analysis of variance (ANOVA) was used to analyze the statistical differences in different PAR and nutrient conditions. Different letters represent significant differences ($P < 0.05$) among different treatments.

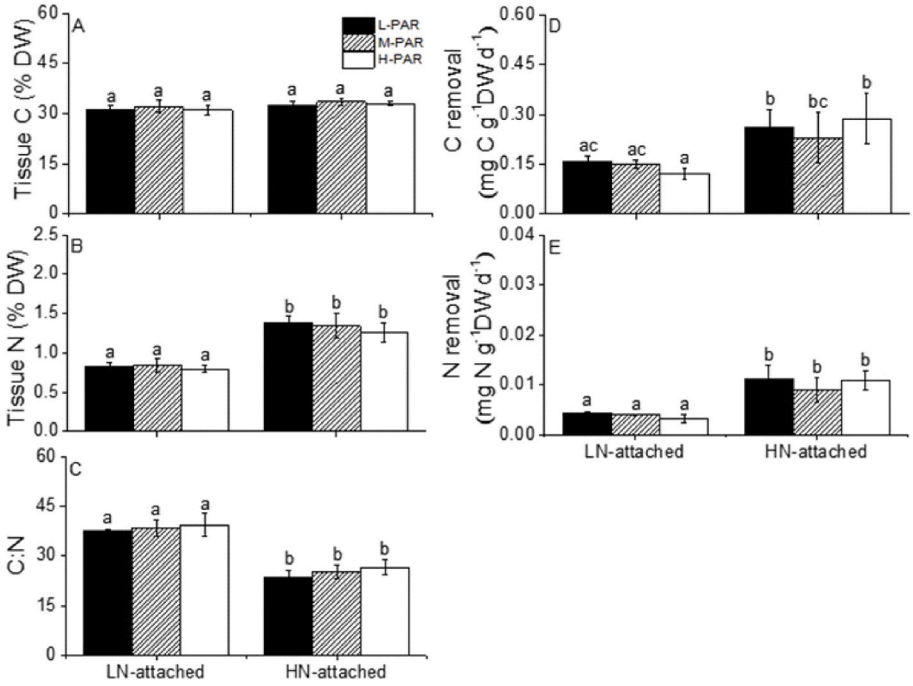

Fig. 5. Tissue carbon (A), nitrogen (B), C:N ratio (C), carbon removal (D) and nitrogen removal (E) of attached population of *Sargassum horneri* cultivated under various conditions. L-PAR: 50 μmol photons m^{-2} s^{-1}; M-PAR: 150 μmol photons m^{-2} s^{-1}; H-PAR: 250 μmol photons m^{-2} s^{-1}; LN: 5 μmol L^{-1} nitrogen and 0.5 μmol L^{-1} phosphorus; HN: 50 μmol L^{-1} nitrogen and 5 μmol L^{-1} phosphorus. Data show mean values ± SD (n = 3). Two-way analysis of variance (ANOVA) was used to analyze the statistical differences in different PAR and nutrient conditions. Different letters represent significant differences ($P < 0.05$) among different treatments.

3.5. Tissue carbon and nitrogen contents

Tissue C ($P = 0.021$) and N contents ($P < 0.001$), C:N ($P < 0.001$), and C and N removal ($P < 0.001$) in the attached population of *S. horneri* were significantly influenced by the level of nutrients, but not by PAR or the combination of both PAR and nutrients ($P > 0.05$) (Table S3). Both HN and higher PAR had no significant influences on tissue C ($P > 0.05$, Fig. 5A). Tissue N contents were significantly higher at HN than LN, regardless of PAR ($P < 0.001$, Table S3). PAR did not significantly affect tissue N at each nutrient level ($P > 0.05$, Fig. 5B). C:N ratio was significantly higher at LN than HN (Fig. 5C, Table S3, $P < 0.001$). PAR did not influence C:N ratio at each nutrient level ($P > 0.05$, Fig. 5C). The C and N removal of the attached population were significantly higher at HN than LN at all PAR conditions (Fig. 5D and E, Table S3, $P < 0.001$). At each nutrient condition, PAR did not influence C and N removal ($P > 0.05$, Fig. 5D).

4. Discussions

4.1. Comparison of responses to environmental factors between attached and pelagic populations

This study examined physiological responses under different environmental stressors of nutrients and PAR that the *S. horneri* blooms may experience during their long journey from the point of origin to the final destination (Qi et al., 2017; Byeon et al., 2019). Two different populations of *S. horneri*, an attached local population in Jeju Island and a pelagic (bloom) population were compared in the present study. It was hypothesized that the pelagic population may have adapted to environmental stresses and therefore, may have enhanced growth capacity when compared to the local attached population. However, the growth rate of pelagic population was significantly lower than the attached population. The pelagic population even died within 20 days of cultivation while the attached population continued to grow. These results suggest that the pelagic population might have been damaged or entered in a period of senescence before it reached Jeju Island, Korea.

The origin of pelagic samples was not confirmed in the present study. The satellite image analysis, local population and literature survey indicated that this specific pelagic population used in this study might not have originated from Jeju but might be from the East China Sea or from the eastern end of Shandong Peninsula or even in Dalian and Changdao in the northern Yellow Sea (Yuan et al., 2022). If the pelagic population came from the East China Sea or the northern Yellow Sea, the journey of this bloom may have taken two or six months, respectively, before they reach Jeju Island. This would provide enough time for *Sargassum* to become senescent.

A previous study showed that *S. horneri* enters into a senescence period after the production of receptacles (Xu et al., 2016). Presence of mature or senescent receptacles may indicate the reproductive status of *S. horneri*. The receptacles were not observed in both populations, but this information cannot confirm if reproduction was completed because only small sections (marginal part) of the plants, instead of whole plants were collected from the field for the experiments. Choi et al. (2008) reported that sexual reproduction of the local (attached) population in Jeju Island is known to occur in April–May, but the attached population used in this study might have not yet entered its reproductive period. This is probably a reason, at least in part, why different growth mechanisms were observed in different populations in this study.

Sargassum horneri undergoes the early shift from its growth stage to sexual reproduction when PAR and temperature increased (Pang et al., 2009). A similar phenomenon was found in other brown algae, such as *S. fulvellum* (Hwang et al., 2006) and *S. fusiforme* (Pang et al., 2005). Immature receptacles (~0.2 mm) can complete their maturation process and discharge gametes within 25 days at suitable conditions (Pang et al., 2009). Some attached *S. horneri* thalli can be detached due to the strong waves and currents, and become pelagic due to their gas-filled bladders (vesicles) (Yoshida, 1963; Sanjeewa et al., 2018). Yatsuya (2007) reported that pelagic *S. horneri* can survive for 4–14 weeks, which is longer than other *Sargassum* species. The pelagic period is mainly dependent on the life stage. If the pelagic thalli were mature, they may survive a shorter period, e.g., 1–8 weeks (Yatsuya, 2007). In addition, some environmental cues such as high temperature, high PAR, etc. may accelerate maturity of immature thalli during this pelagic period (Pang et al., 2009; Yu et al., 2019; Rothäusler et al., 2012).

4.2. Nutrient effects on Sargassum horneri

In the present study, no combined effects of nutrient and PAR were

observed in both populations. Higher nutrient (HN) condition significantly increased the growth rates and photosynthesis of *S. horneri*. The pigment contents were increased at the HN condition. Nitrogen is the key macronutrient component for seaweeds, including the brown seaweeds commonly found floating (Schmid et al., 2020). Pigments are important storage compounds of nitrogen (Etemadian et al., 2017), and therefore, are strongly dependent on the nutrient availability (Friedlander et al., 1991; Barufi et al., 2011). Tissue N contents reflect the nutrient availability of water column and utilization capacity of seaweeds (Yu et al., 2014). Tissue N increased with increased N uptake and assimilation (Huppe and Turpin, 1994). Phosphorus is not a component in chl *a* but is an essential element for protein synthesis and energy (Falkowski, 2007). Even though the effect on soluble protein were not significant in the present study, higher concentrations of phosphorus can provide energy for nitrate uptake and assimilation in *S. muticum* (Xu et al., 2017). Phosphorus can also stimulate chl *a* synthesis-related enzymes in *S. thunbergii* (Nakahara and Gao, 1990). High nutrients availability induced higher chl *a* and chl *c* synthesis to enhance the photosynthetic capacity, increased photosynthetic activity, and consequently, increased the growth rate of *Sargassum* (Hwang et al., 2004; Rothäusler et al., 2012; Yu et al., 2019).

4.3. PAR effects on Sargassum horneri

PAR is a very important environmental factor influencing the growth of *Sargassum* (Hales and Fletcher, 1989, 1990). In the present study, higher PAR reduced the growth of the attached *S. horneri* at least during the 15 days of cultivation. These results suggest that attached *S. horneri* requires lower PAR to support growth in the subtidal zone. Similar results were reported by Choi et al. (2008) who found that growth rate of *S. horneri* was higher at 20 μmol photons m^{-2} s^{-1} than 80 μmol photons $m^{-2}s^{-1}$. Zou et al. (2018) also reported that the maximum growth rate of *S. polycystum* ranged from 20 to 80 μmol photons·m^{-2}·s^{-1}, and then decreased as PAR increased. This phenomenon was probably because pigments (Chl a, Chl c and carotenoids) contents decreased as PAR increased, photo-inhibition might have occurred at higher PAR (>80 μmol photons·m^{-2}·s^{-1}), similar as observed in other floating seaweeds (Rothäusler et al., 2011a; Tala et al., 2017). The decrease in pigments is a well-known phenomenon at high light condition due to photoacclimation, as shown in the floating *Macrocystis pyrifera* (Rothäusler et al., 2011a, b). The capacity to adjust the pigment synthesis may help the photosynthetic apparatus to protect against the high light energy when the seaweed is floating at the sea surface (Demmig-Adams and Adams, 1992). These results suggest that higher PAR may reduce the growth due to the inhibition of pigment synthesis, but the seaweed still maintain the photosynthetic activity (Rothäusler et al., 2011a).

5. Conclusions

The present study suggests that pelagic populations of *S. horneri* may become senescent earlier than the attached population due to the environmental stresses during their long journey from its origin to Jeju Island. Therefore, the growth rate was significantly lower for the pelagic population versus the attached population. Higher PAR and nutrient limitation significantly decreased the growth, photosynthesis and pigment contents of attached *S. horneri*. However, no combined effect of nutrient and PAR was observed in both populations of *S. horneri*. *Sargassum horneri* requires lower PAR to support its growth, suggesting that self-shading may not be a serious problem for pelagic populations of *S. horneri*. In addition to stress due to PAR and nutrient limitations, floating *S. horneri* also experience other environmental stressors (i.e., temperature, UV, etc.) as the pelagic biomass moves towards the Korean coast. These additional environmental stressors should also be evaluated to understand the ecophysiological mechanisms that may contribute to the survival and expansion of *S. horneri* blooms.

Author statement

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

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References

Abé, H., Komatsu, T., Kokubu, Y., Natheer, A., Rothausler, E.A., Shishido, H., Yoshizawa, S., Ajisaka, T., 2013. Invertebrate fauna associated with floating *Sargassum horneri* (Fucales: Sargassaceae) in the east China sea. Species Divers. 18 (1), 75–85.

Barradas, J.I., Chow, F., Dias, G.M., Ghilardi-Lopes, N.P., 2018. The influence of trampling disturbance on the fluorescence and pigment concentration of *Sargassum* beds (Fucales). J. Exp. Mar. Biol. Ecol. 506 (9), 163–170.

Barufi, J.B., Korbee, N., Oliveira, M.C., Figueroa, F.L., 2011. Effects of N supply on the accumulation of photosynthetic pigments and photoprotectors in *Gracilaria tenuistipitata* (Rhodophyta) cultured under UV radiation. J. Appl. Phycol. 23 (3), 457–466.

Bradford, M.M., 1976. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. Anal. Biochem. 72 (1–2), 248–254.

Byeon, S.Y., Oh, H.J., Kim, S., Yun, S.H., Kang, J.H., Park, S.R., Lee, H.J., 2019. The origin and population genetic structure of the 'golden tide' seaweeds, *Sargassum horneri*. In: Korean waters. Sci. Rep-UK, 9, pp. 1–13, 1.

Choi, H.G., Lee, K.H., Yoo, H.I., Kang, P.J., Kim, Y.S., Nam, K.W., 2008. Physiological differences in the growth of *Sargassum horneri* between the germling and adult stages. J. Appl. Phycol. 20 (5), 729–735.

Choi, S.K., Oh, H.J., Yun, S.H., Lee, H.J., Lee, K.H., H, Y.S., Kim, S., Park, S.R., 2020. Population dynamics of the 'golden tides' seaweed, *Sargassum horneri*, on the southwestern coast of Korea: the extent and formation of golden tides. Sustainability-Basel 12 (7), 2903, 2020.

Cruz-Rivera, E., Flores-Díaz, M., Hawkins, A., 2015. A fish kill coincident with dense *Sargassum* accumulation in a tropical bay. Bull. Mar. Sci. 91 (4), 455–456.

Demmig-Adams, B., Adams Iii, W.W., 1992. Photoprotection and other responses of plants to high light stress. Annu. Rev. Plant Biol. 43 (1), 599–626.

Etemadian, Y., Shabanpour, B., Ghaemi, V., Kordjazi, M., 2017. Compare the chlorophyll amount in three brown algae species of the Persian Gulf by using three solvents and applying two formulas. Int. J. Biochem. Biophys. Mol. Biol. 2 (6), 77.

Falkowski, P.G., 2007. Raven J. Aquatic Photosynthesis, second ed. Princeton University Press, New Jersey, USA.

Fine, M.L., 1970. Faunal variation on pelagic *Sargassum*. Mar. Biol. 7 (2), 112–122.

Fraser, C.I., Nikula, R., Spencer, H.G., Waters, J.M., 2009. Kelp genes reveal effects of subantarctic sea ice during the Last Glacial Maximum. Proc. Natl. Acad. Sci. U.S.A. 106 (9), 3249–3253.
Friedlander, M., Krom, M.D., Ben-Amotz, A., 1991. The effect of light and ammonium on growth, epiphytes and chemical constituents of *Gracilaria conferta* in outdoor cultures. Bot. Mar. 34 (3), 161–166.
Hales, J.M., Fletcher, R.L., 1989. Studies on the recently introduced Brown alga *Sargassum muticum*. Bot. Mar. 32, 167–176.
Hales, J.M., Fletcher, R.L., 1990. Studies on the recently introduced brown alga *Sargassum muticum* (Yendo) Fensholt. V. Receptacle initiation and growth, and gamete release in laboratory culture. Bot. Mar. 33 (3), 241–250.
Huppe, H.C., Turpin, D.H., 1994. Integration of carbon and nitrogen metabolism in plant and algal cells. Annu. Rev. Plant Biol. 45 (1), 577–607.
Hwang, E.K., Lee, S.J., Ha, D.S., Park, C.S., 2016. *Sargassum* golden tides in the Shinan-gun and Jeju Island, Korea. Korean J. Fish. Aquat. Sci. 49 (5), 689–693.
Hwang, E.K., Park, C.S., Baek, J.M., 2006. Artificial seed production and cultivation of the edible brown alga, *Sargassum fulvellum* (Turner) C.Agardh: developing a new species for seaweed cultivation in Korea. J. Appl. Phycol. 18 (2), 251–257.
Hwang, R.L., Tsai, C.C., Lee, T.M., 2004. Assessment of temperature and nutrient limitation on seasonal dynamics among species of *Sargassum* from a coral reef in southern Taiwan. J. Phycol. 40 (3), 463–473.
Jeffrey, S.W., 1963. Purification and properties of chlorophyll c from *Sargassum flavicans*. Biochem. J. 86 (2), 313–318.
Kang, J.W., Chung, I.K., 2017. The effects of eutrophication and acidification on the ecophysiology of *Ulva pertusa* Kjellman. J. Appl. Phycol. 29 (1), 2675–2683.
Kim, J.K., Kraemer, G.P., Neefus, C.D., Chung, I.K., Yarish, C., 2007. Effects of temperature and ammonium on growth, pigment production and nitrogen uptake by four species of *Porphyra* (Bangiales, Rhodophyta) native to the New England coast. J. Appl. Phycol. 19 (5), 431–440.
Kim, K.Y., Shin, J.S., Kim, K.Y., Ryu, J.H., 2019. Long-term trend of green and golden tides in the Eastern Yellow Sea. J. Coast Res. 90 (SI), 317–323.
Komatsu, T., Mizuno, S., Natheer, A., Kantachumpoo, A., Tanaka, K., Morimoto, A., Hsiao, S.T., Rothäusler, E.A., Shishidou, H., Aoki, M., Ajisaka, T., 2014a. Unusual distribution of floating seaweeds in the East China Sea in the early spring of 2012. J. Appl. Phycol. 26 (2), 1169–1179.
Komatsu, T., Fukuda, M., Mikami, A., Mizuno, S., Kantachumpoo, A., Tanoue, H., Kawamiya, M., 2014b. Possible change in distribution of seaweed, *Sargassum horneri*, in northeast Asia under A2 scenario of global warming and consequent effect on some fish. Mar. Pollut. Bull. 85 (2), 317–324.
Lapointe, B.E., 1995. A comparison of nutrient-limited productivity in *Sargassum* natans from neritic vs. oceanic waters of the western North Atlantic Ocean. Limnol. Oceanogr. 40 (3), 625–633.
Lichtenthaler, H.K., 1987. Chlorophylls and carotenoids: pigments of photosynthetic biomembranes. Method. Enzymol. 148, 350–382.
Lichtenthaler, H.K., Buschmann, C., 2001. Chlorophylls and carotenoids: measurement and characterization by UV-VIS spectroscopy. Curr. Protoc. Food Anal. Chem. 1 (1). F4. 3.1-F4. 3.8.
Liu, J., Xia, J., Zhuang, M., Zhang, J., Sun, Y., Tong, Y., Zhao, S., H, P., 2021. Golden seaweed tides accumulated in *Pyropia* aquaculture areas are becoming a normal phenomenon in the Yellow Sea of China. Sci. Total Environ. 774 (26), 145726.
Milledge, J.J., Nielsen, B.V., Bailey, D., 2016. High-value products from macroalgae: the potential uses of the invasive brown seaweed, *Sargassum muticum*. Rev. Environ. Sci. Biotechnol. 15 (1), 67–88.
Nakahara, H., Gao, K.S., 1990. Effects of nutrients on the photosynthesis of *Sargassum thunbergii*. Bot. Mar. 33 (5), 375–384.
Norton, T., Mathieson, A.C., 1983. The biology of unattached seaweeds. In: Round, F., Chapman, D. (Eds.), Progress in Phycological Research, vol. 2. Elsevier Sci. Publ. Amsterdam, pp. 333–386, 2.
Ott, F.D., 1965. Synthetic media and techniques for the xenic cultivation of marine algae and flagellate. Va. J. Sci. 16, 205–218.
Pang, S.J., Chen, L.T., Zhuang, D.G., Fei, X.G., Sum, J.Z., 2005. Cultivation of the brown alga *Hizikia fusiformis* (Harvey) Okamura: enhanced seedling production in tumbled culture. Aquaculture 245 (1–4), 321–329.
Pang, S.J., Liu, F., Shan, T.F., Gao, S.Q., Zhang, Z.H., 2009. Cultivation of the brown alga *Sargassum horneri*: sexual reproduction and seedling production in tank culture under reduced solar irradiance in ambient temperature. J. Appl. Phycol. 21 (4), 413–422.
Qi, L., Hu, C., Wang, M., Shang, S., Wilson, C., 2017. Floating algae blooms in the east China sea, 11 Geophys. Res. Lett. 44 (22), 501–511, 509.
Resiere, D., Valentino, R., Nevière, R., Banydeen, R., Florentin, F., 2018. *Sargassum* seaweed on Caribbean islands: an international public health concern. Lancet 392 (10165), 2691.
Ritchie, R.J., 2008. Universal chlorophyll equations for estimating chlorophylls *a, b, c*, and *d* and total chlorophylls in natural assemblages of photosynthetic organisms using acetone, methanol, or ethanol solvents. Photosynthetica 46 (1), 115–126.
Rothäusler, E., Gómez, I., Karsten, U., Tala, F., Thiel, M., 2011a. Physiological acclimation of floating *Macrocystis pyrifera* to temperature and irradiance ensures long-term persistence at the sea surface at mid-latitudes. J. Exp. Mar. Biol. Ecol. 405 (1–2), 33–41.
Rothäusler, E., Gómez, I., Karsten, U., Tala, F., Thiel, M., 2011b. UV-radiation versus grazing pressure: long-term floating of kelp rafts (*Macrocystis pyrifera*) is facilitated by efficient photoacclimation but undermined by grazing losses. Mar. Biol. 158 (1), 127–141.
Rothäusler, E., Gutow, L., Thiel, M., 2012. Floating Seaweeds and Their Communities. In: Seaweed Biology. Springer, Berlin, Heidelberg, pp. 359–380.
Sanjeewa, K.K.A., Fernando, I.P.S., Kim, S.Y., Kim, H.S., Ahn, G., Jee, Y.H., Jeon, Y.J., 2018. In vitro and in vivo anti-inflammatory activities of high molecular weight sulfated polysaccharide; containing fucose separated from Sargassum horneri. Int. J. Biol. Macromol. 107 (2), 803–807.
Sanjeewa, K.K.A., Lee, J.-S., Kim, W.-S., Jeon, Y.-J., 2017. The potential of brown-algae polysaccharides for the development of anticancer agents: an update on anticancer effects reported for fucoidan and laminaran. Carbohydr. Polym. 177 (23), 451–459.
Schmid, M., Fernández, P.A., Gaitán-Espitia, J.D., Virtue, P., Leal, P.P., Revill, A.T., Nichols, P.D., Hurd, C.L., 2020. Stress due to low nitrate availability reduces the biochemical acclimation potential of the giant kelp *Macrocystis pyrifera* to high temperature. Algal Res. 47 (3), 101895, 2020.
Sfriso, A., Facca, C., 2013. Annual growth and environmental relationships of the invasive species *Sargassum muticum* and *Undaria pinnatifida* in the lagoon of Venice. Estuar. Coast Shelf Sci. 129 (13), 162–172.
Silva, L.D., Bahcevandziev, K., Pereira, L., 2019. Production of bio-fertilizer from *Ascophyllum nodosum* and *Sargassum muticum* (Phaeophyceae). J. Oceanol. Limnol. 37 (3), 918–927.
Tala, F., Penna-Díaz, M.A., Luna-Jorquera, G., Rothäusler, E., Thiel, M., 2017. Daily and seasonal changes of photobiological responses in floating bull kelp *Durvillaea antarctica* (Chamisso) Hariot (Fucales: Phaeophyceae). Phycologia 56 (3), 271–283.
Thompson, T.M., Young, B.R., Baroutian, S., 2020. Pelagic *Sargassum* for energy and fertiliser production in the Caribbean: a case study on Barbados. Renew. Sustain. Energy Rev. 118 (2), 109564.
Wang, Y., Zhong, Z.H., Qin, S., Li, J.L., Li, J.J., Liu, Z.Y., 2021. Effects of temperature and light on growth rate and photosynthetic characteristics of *Sargassum horneri*. J. Ocean Univ. China 20 (1), 101–110.
Wang, G., Sun, J., Liu, G., Wang, L., Yu, J., Liu, T., Chi, S., Liu, C., Guo, H., Wang, X., Wu, S., 2014. Comparative analysis on transcriptome sequencings of six *Sargassum* species in China. Acta Oceanol. Sin. 33 (2), 37–44.
Wang, M., Hu, C., Barnes, B.B., Mitchum, G., Lapointe, B., Montoya, J.P., 2019. The great Atlantic *Sargassum* belt. Sci 365 (6448), 83–87.
Wu, H.L., Huo, Y.Z., Zhang, J.H., Liu, Y.Y., Zhang, Y.T., He, P.M., 2015. Bioremediation efficiency of the largest scale artificial *Porphyra yezoensis* cultivation in the open sea in China. Mar. Pollut. Bull. 95 (1), 289–296.
Xing, Q., Guo, R., Wu, L., An, D., Cong, M., Qin, S., Li, X., 2017. High-resolution satellite observations of a new hazard of golden tides caused by floating *Sargassum* in winter in the Yellow Sea. IEEE Geosci. Remote S. 14 (10), 1815–1819.
Xu, M., Sakamoto, S., Komatsu, T., 2016. Attachment strength of the subtidal seaweed *Sargassum horneri* (Turner) C. Agardh varies among development stages and depths. J. Appl. Phycol. 28 (6), 3679–3687.
Xu, M., Sasa, S., Otaki, T., Hu, F.X., Tokai, T., Komatsu, T., 2018. Changes in drag and drag coefficient on small *Sargassum horneri* (Turner) C. Agardh individuals. Aquat. Bot. 144 (1), 61–64.
Xu, Z.G., Gao, G., Xu, J.T., Wu, H.Y., 2017. Physiological response of a golden tide alga (*Sargassum muticum*) to the interaction of ocean acidification and phosphorus enrichment. Biogeosciences 14 (3), 671–681.
Yatsuya, K., 2007. Floating period of *Sargassacean* thalli estimated by the change in density. J. Appl. Phycol. 5 (20), 797–800.
Yoshida, T., 1963. Studies on the distribution and drift of the floating seaweeds. Bull. Tohoku Reg. Fish. Res. Lab. 23, 141–186.
Yoshida, Y., Nagakane, K., Fukuda, R., Nakayama, Y., Okazaki, M., Shintani, H., Inoue, S., Tagawa, Y., Suzuki, K., Munck, J.D., Meerbeek, B.V., 2004. Comparative study on adhesive performance of functional monomers. J. Dent. Res. 83 (6), 454–458.
Yu, J., Li, J., Wang, Q., Liu, Y., Gong, Q., 2019. Growth and resource accumulation of drifting *Sargassum horneri* (Fucales, Phaeophyta) in response to temperature and nitrogen supply. J. Ocean Univ. China 18 (5), 1216–1226.
Yu, Z.H., Zhu, X.S., Jiang, Y.L., Luo, P., Hu, C.Q., 2014. Bioremediation and fodder potentials of two *Sargassum* spp. in coastal waters of Shenzhen, South China. Mar. Pollut. Bull. 85 (2), 797–802.
Yuan, C., Xiao, J., Zhang, X., Fu, M.Z., Wang, Z.L., 2022. Two drifting paths of *Sargassum* bloom in the Yellow Sea and East China sea during 2019– 2020. Acta Oceanol. Sin. 41 (1), 1–10.
Zhang, J.H., Ding, X.W., Zhuang, M.M., Wang, S.Y., Chen, L., Shen, H., He, P.M., 2019. An increase in new *Sargassum* (Phaeophyceae) blooms along the coast of the east China Sea and Yellow Sea. Phycologia 58 (4), 374–381.
Zhang, P., Wang, T., Xie, Q., Zhang, H., Yan, X., Chen, M., 2014. Effects of environment factors on growth of *Sargassum horneri* seedlings under indoor conditions. J. Shanghai Ocean Univ. 23 (2), 200–207.
Zhuang, M., Liu, J., Ding, X., He, J., Zhao, S., Wu, L., Gao, S., Zhao, C., Liu, D., Zhang, J., H, P., 2020. *Sargassum* blooms in the east China Sea and Yellow Sea: formation and management. Mar. Pollut. Bull. 162 (1), 111845.
Zou, X.X., Xing, S.S., Su, X., Zhu, J., Huang, H.Q., Bao, S.X., 2018. The effects of temperature, salinity and irradiance upon the growth of *Sargassum polycystum* C. Agardh (Phaeophyceae). J. Appl. Phycol. 30 (2), 1207–1215.