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Development of a Seaweed Species-Selection Index for Successful Culture in a Seaweed-Based Integrated Aquaculture System

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Abstract Integrated multi-trophic aquaculture (IMTA) has been proposed as a concept that combines the cultivation of fed aquaculture species (*e.g.*, finfish/shrimp) with extractive aquaculture species (*e.g.*, shellfish/seaweed). In seaweed-based integrated aquaculture, seaweeds have the capacity to reduce the environmental impact of nitrogen-rich effluents on coastal ecosystems. Thus, selection of optimal species for such aquaculture is of great importance. The present study aimed to develop a seaweed species-selection index for selecting suitable species in seaweed-based integrated aquaculture system. The index was synthesized using available literature-based information, reference data, and physiological seaweed experiments to identify and prioritize the desired species. *Undaria pinnatifida*, *Porphyra yezoensis* and *Ulva compressa* scored the highest according to a seaweed-based integrated aquaculture suitability index (SASI). Seaweed species with the highest scores were adjudged to fit the integrated aquaculture systems. Despite the application of this model limited by local aquaculture environment, it is considered to be a useful tool for selecting seaweed species in IMTA.

Key words seaweed; IMTA; species-selection model; water temperature; physiological characteristics; uptake rate

1 Introduction

Seaweed-based integrated aquaculture techniques serve environmental sustainability by integrating fed aquaculture (*e.g.*, finfish and shrimp) with inorganic and organic extractive aquacultures (*e.g.*, seaweed and shellfish) (Buschmann *et al*., 2001; Chopin *et al*., 2001). These techniques have been proposed as a means for developing environmentally sound aquaculture practices and resource management through a balanced coastal ecosystem approach, which removes excessive nutrients and contributes to economic diversification by producing additional value-added products (Chopin *et al*., 2008). Traditionally, seaweed-based integrated aquaculture techniques are used to provide a feed component for co-cultured fish in Korea (Fig.1). Additionally, the governments of several other countries have begun to emphasize the role of seaweed as a biofilter in estuarine and coastal areas, where it prevents nutrient enrichment from effluents (Buschmann *et al*., 2009).

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The use of macroalgae as a biofilter material in seaweed-based integrated aquaculture has been proved to be an excellent example of ecotechnology (Neori *et al*., 2004; Hernández *et al*., 2006). Examples of such systems as reviewed by Troell *et al*. (2003), are integrated salmon-*Gracilaria*, integrated seabream-*Ulva*, integrated salmon-*Laminaria* (currently *saccharina*), and integrated fish-*Pophyra*. Thus, systematic and comprehensive data based on the ecophysiological characteristics of seaweeds are required for establishing guidelines or standards for the selection of optimal seaweed species (NFRDI, 1994; Neori *et al*., 2004; Kang *et al*., 2008).

The choice of seaweed species in seaweed-based integrated aquaculture should be made according to their ecophysiological characteristics and market value (Neori *et al*., 2004). The ecophysiological characteristics such as biofiltration capacity, biochemical composition, and growth rate can be identified as the major limitations to successful macroalgae aquaculture (Santos, 2006). Primarily, optimal seaweed species for IMTA should exhibit high uptake rates and uptake efficiency to remove nutrients from effluents (Chopin *et al.*, 2008). In addition, they should show fast growth rates and be able to accumulate large amounts of nutrients, especially nitrogen (N). Seaweed should be less affected by the type of environmental stresses encountered during culture, and should maintain health for considerable periods of time. In addition, the ease of cultivation is a prerequisite when selecting seaweed species suitable for IMTA.

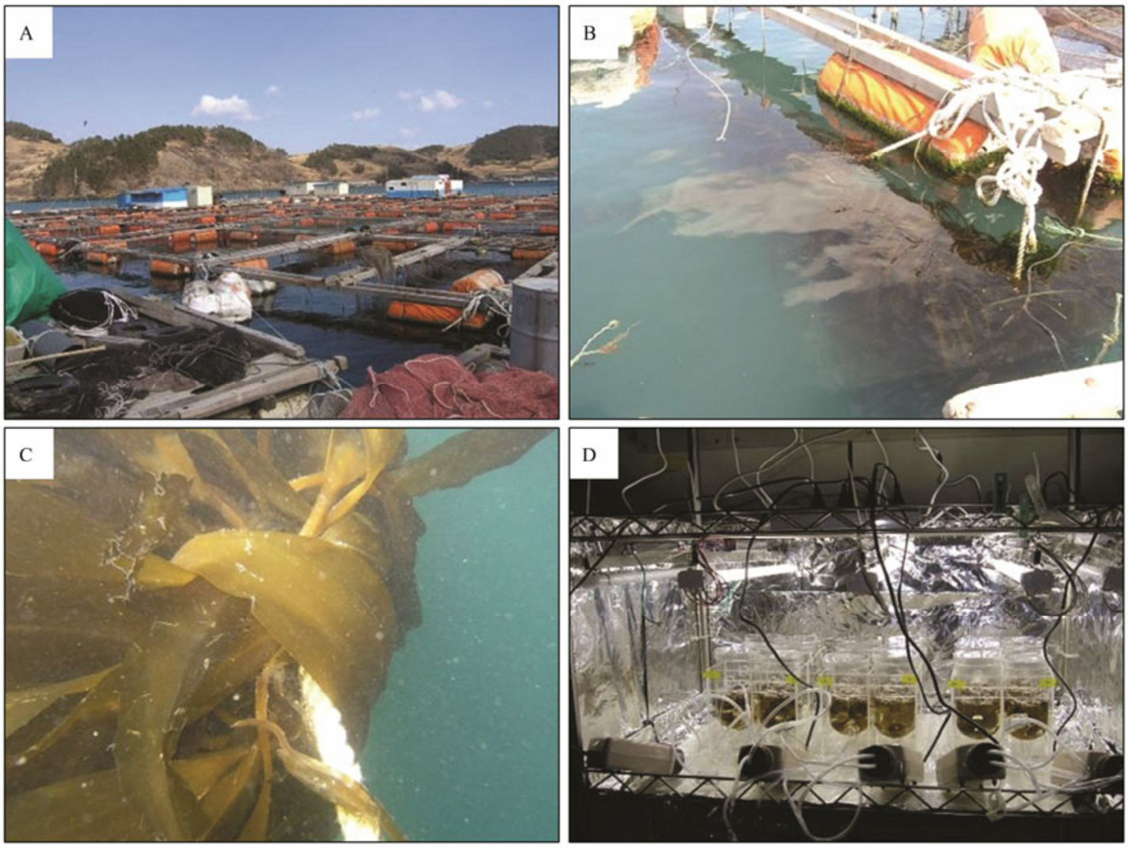

Fig.1 An example of fish-seaweed co-culture around Hawtae Island on the southern coast of Korea and nutrient uptake rate experiments in the laboratory. The brown alga *Saccharina japonica* is cultured using a line method around fish farms: A, a fish farm; B, seaweed around a fish farm; C, brown algae (*S. japonica*); and D, uptake rate experiments in the laboratory.

The economic value of seaweeds may be directly or indirectly related to their physiological health. The health of seaweeds should be monitored to prevent dramatic deterioration in their culture and to improve or maintain their quality. The biochemical composition and photosynthesis of seaweeds have been known to be powerful indices of physiological performance (Vergara *et al*., 1993; Kang *et al*., 2009). Tissue N content may provide an efficient way for removing nutrients from eutrophic seawater (He *et al*., 2008). In addition, responses in the growth and photosynthetic pigment concentrations of seaweeds are correlated with responses in tissue N content (Jones *et al*., 2002). Thus, it is necessary to evaluate the biochemical composition and photosynthesis of seaweeds when selecting optimal seaweed species.

The purpose of this study was to develop a seaweed species-selection index for selecting species suitable for seaweed-based integrated aquaculture. The species-selection index was modified based on the site-selection model developed by Short *et al*. (2002). The index synthesizes available literature-based information, reference data, and physiological seaweed experiments to identify and prioritize species for this kind of aquaculture. The process for obtaining an index was divided into three steps: 1) the available physiological reference data were used to establish a preliminary seaweed-based integrated aquaculture suitability index (PSASI), which was then used to pre-screen and eliminate unsuitable seaweed species; 2) the PSASI-identified priority species were analyzed using laboratory experiments to determine nutrient uptake rate, nutrient reduction efficiency, tissue N content, and photosynthetic pigments; 3) based on the experimental results, a seaweed-based integrated aquaculture suitability index (SASI) score was calculated for each species. The species-selection index allowed us to reduce the necessary cost and effort on selecting seaweed suitable for given culture purposes (*e.g.*, seasonality and profitability).

2 Materials and Methods

Compilation of the species-selection index was carried out in three steps. Step I was to identify potential seaweed species according to available information, including previous physiological experimental data on seaweeds, yielding one of the scores that are incorporated into the calculation of the SASI. Step II involved the compilation of physiological characteristic data under various conditions for the highest-scoring species identified in Step I. Step III was the final calculation of the SASI based on the results of Steps I and II.

2.1 Step I: Identification of Potential Seaweed Species and PSASI I Rating

Step I involved reviewing available information on selecting potential seaweed genera for integrated multi-trophic aquaculture (IMTA). The parameters of seaweed selection considered in Step I were economic value, ease of cultivation, and seaweed-based integrated aquaculture application. This information contributed to the PSASI, which was used as the first level of screening in optimal-species selection (Table 1). Information for the PSASI was collected from both published and unpublished literature. Each of the parameters in Step I received a rating (Table 1), and the PSASI I score was calculated as the product of these ratings.

Table 1 Data used in Step I of the preliminary seaweed-based integrated aquaculture suitability index (PSASI) for identifying potential seaweed species

Parameter	PSASI rating	Reference
PSASI I		
Economic value	1 for good	FAO (2004)
	2 for excellent	Oh *et al.* (1990)
Cultivation	1 for field species	FAO (2004)
	2 for candidate species	
Application of seaweed-based aquaculture	1 for not applied IMTA	Troell *et al.* (2003)
	2 for applied IMTA	
PSASI II		
NH_4^+ uptake rate		
V_{max}	1 for <100 μmolg^{-1} DW h^{-1}	Hanisak (1983)
	2 for ≥ 100 μmolg^{-1} DW h^{-1}	Kraemer *et al.* (2004)
K_m	1 for <150 μmolL^{-1}	Lobban and Harrison (1994)
	2 for ≥ 150 μmolL^{-1}	Kraemer *et al.* (2004)

Note: Maximum possible PSASI I and PSASI II scores are 8 and 4, respectively.

The economic value of seaweed is an important consideration when selecting species for IMTA. Nutrients released from fish, shrimps, and bivalves are suitable for seaweed growth, and consequently, seaweed production can be dramatically increased in IMTA (Neori *et al.*, 2004). Thus, for profit maximization, it is important to select seaweeds with high economic value and good market standing. In this study, we gave seaweed a rating of 2 if it is used as a food source or in medical or industrial materials. Otherwise, the seaweed received a rating of 1. The economic value of seaweed was based on data from the FAO (2004) and Oh *et al.* (1990).

Even though seaweed has high market value and a high uptake rate, its cultivation must nonetheless be easy and efficient. Thus, if a seaweed genus could be cultivated easily and efficiently, it was rated 2. Otherwise, it was rated 1. Information on seaweed culture can be derived from the literature (*e.g.*, FAO, 2004). Additionally, if a genus had been previously selected as a candidate for IMTA system application, it was rated 2. Otherwise, it was rated 1 (Troell *et al.*, 2003).

2.2 Step Ⅱ: Physiological Characteristics of Seaweed Species

2.2.1 Seaweed collection

Six species of macroalgae from three divisions, *Ulva compressa* (Chlorophyta), *Porphyra yezoensis* and *Gracilaria incurvata* (Rhodophyta), *Ecklonia cava*, and *Undaria pinnatifida* (Phaeophyta) were collected from the intertidal zone of a wave-exposed rocky shore at Geoje Island and a seaweed farm located in Ilkwang and Gamak bay, Korea. All thalli were rinsed to remove sediment, debris and epiphytes, and then acclimated in filtered seawater in a culture room (30 μmol photons m^{-2} s^{-1}; 12 : 12 light : dark photoperiod; 15–17℃) for 3–4 d (Hanisak and Harlin 1978; Kang *et al.*, 2008). The seawater was collected from the southwest part of the East Sea in October 2006. The average concentrations of nitrate (NO_3^-) and ammonium (NH_4^+) in seawater were less than 0.1 μmol L^{-1}. The seawater was filtered first through Whatman glass-fiber filters (1.2 μm) and then through Advantec MFS membrane filters (0.2 μm).

2.2.2 PSASI Ⅱ experiment: ammonium uptake rate of seaweeds

The PSASI II experiment was conducted to measure the ammonium uptake rates of five species of seaweed. Specimens of 3.0 g fresh weight were put into a 500-mL corn bottle containing 300 mL of nutrient medium, which was prepared in triplicate. The initial NH_4^+ concentrations of each medium were maintained at 0, 10, 50, 100, 200, 300, 500, and 750 μmol L^{-1} (NH_4Cl) with filtered seawater. The experiments were conducted in a controlled-environment room with constant temperature (16℃ ± 0.5℃) and light intensity (100–150 μmol photons m^{-2} s^{-1}) for approximately 10 h. Control experiments were carried out without addition of algal materials. Water samples were collected from the chambers at consistent time intervals (1–2 h) with NH_4^+ concentrations analyzed immediately using the colorimetric technique of Parsons *et al.* (1984). At the end of the experiments, the dry weight (DW) of samples was measured by drying at 60℃ for 48 h. The uptake rate was plotted as a function of nutrient concentration, and NH_4^+ uptake kinetics was derived using the Michaelis-Menten equation:

$$V = V_{max} \times S / (K_m + S),$$

where V is the NH_4^+ uptake rate, V_{max} is the maximum uptake rate, S is the NH_4^+ concentration, and K_m is the half-saturation value, that is, the nutrient concentration where $V = V_{max} / 2$. The uptake rate was normalized to μmol NH_4^+ g DW^{-1} h^{-1}.

Seaweeds with a high V_{max} were considered to be desirable for IMTA applications (Kraemer *et al.*, 2004). Lobban and Harrison (1994) have reported that with low substrate concentrations, a high-affinity system operates, exhibiting a high degree of ion specificity and a low value of K_m, whereas with high substrate concentrations, a low-affinity system is operative, exhibiting much less ion selectivity and a very high value for K_m. Thus, the PSASI II ratings for V_{max} were assigned: 1 for < 100 μmol g^{-1} DW h^{-1}; 2 for ≥ 100 μmol g^{-1} DW h^{-1}. Considering that the NH_4^+ levels in fish farm effluent were approximately 150 μmol L^{-1} (Neori *et al.*, 1996; Carmona *et al.*, 2006), the K_m of a seaweed was rated 1 if the level was less than 150 μmol L^{-1} and 2 if greater than or equal to 150 μmolL^{-1}.

2.2.3 SASI experiments: NH_4^+ reduction efficiency, tissue N content, and chlorophyll *a* content

In the SASI experiments, NH_4^+ reduction efficiency, tissue N content, and chlorophyll *a* content of five seaweed species (*U. compressa*, *P. yezoensis, G. incurvata*, *E. cava*, and *U. pinnatifida*) were measured daily for 7 d at 17℃± 0.5℃ under 100 μmol photons m^{-2} s^{-1}. Specimens of 5.0 g fresh weight were put into a 1 000-mL flask containing 700 mL of medium (150 NH_4^+ μmol L^{-1}). The NH_4^+ concentration of the media was determined based on the N concentration in fish farm effluent (Neori *et al.*, 1996; Carmona *et al.*, 2006). The culture medium was replaced daily during the experimental period. The NH_4^+ reduction efficiency of seaweed was calculated using the difference between the initial and final NH_4^+ concentrations (μmol L^{-1}) in seawater according to the following equation

$$\textit{Reduction efficiency}\ (\%) = (S_0 - S_t) / S_0 \times 100,$$

where S_0 is the initial NH_4^+ concentration, and S_t is the NH_4^+ concentration after *t* d.

For carbon (C) and N analysis, samples were taken before and after the experiments. The samples were dried for 48 h at 60℃, and then ground using a pestle and mortar. Approximately 2–3 mg of ground tissue was placed into a tin to determine the C and N contents using a CHN elemental analyzer (Vario-EL III), and the C : N molar ratios were calculated.

The chlorophyll *a* content was extracted using N, N, dimethylformamide (DMF) following the methodology described by Wellburn (1994). The samples were examined spectrophotometrically after extraction with 4 mL of DMF for 24 h at 4℃ in the dark.

2.3 Step III: Calculating the SASI Score

At Step III, the SASI score was calculated, again as a multiplicative index, for each seaweed species (Table 2). The SASI was determined by combining the results of the PSASI I, PSASI II, and SASI, thus reflecting the results of the physiological tests on each seaweed. Table 2 shows the rating for each parameter. A PSASI rating of 1 was assigned if the PSASI score was ≤2 and 2 if the PSASI was over 2. The PSASI II rating was assigned as follows: 1 if the PSASI score was < 2 and 2 if the PSASI was ≥2.

Table 2 Data from Step II in the calculation of the seaweed-based integrated aquaculture suitability index (SASI)

Parameter	SASI rating	Reference
PSASI I	1 for PSASI = 1–2	
	2 for PSASI = 4–8	
PSASI II	1 for PSASI II = 1–2	
	2 for PSASI II = 4	
Removal efficiency	0 for <30%	
	1 for 30%–75%	
	2 for >75%	
N content	1 for < mean – SD or no data	Hernández *et al.* (2002)
	2 for ≥ mean – SD	
Chl *a*	1 for < mean – SD	Costanzo *et al.* (2000)
	2 for ≥ mean – SD	

Notes: The SASI score was calculated by multiplying the SASI ratings (PSASI I × PSASI II × SASI data) for each species for each water temperature; Maximum possible SASI score = 32.

The SASI rating for reduction efficiency was based on NH_4^+ concentrations in a water column observed on day 7. A score of 0 was assigned for < 30% reduction efficiency, 1 for 30%–75% reduction efficiency, and 2 for 75% reduction efficiency. The SASI ratings for the N content of seaweed were assigned as follows: 1 if the N content (mean ± SD) was less than the (mean – SD) of the initial pre-experiment samples or if data were not available for those parameters, and 2 if the N content was greater than or equal to the (mean – SD) of initial pre-experiment samples. The tissue N content is an optional SASI parameter that provides a finer resolution of the index. The chlorophyll *a* content of the seaweed was rated 1 if the content (mean ± SD) was less than the (mean – SD) of the initial samples and 2 if it was greater than or equal to the (mean – SD) of the initial samples.

The SASI score was calculated with the equation:

SASI score = PSASI I × PSASI II × SASI (reduction efficiency × N content × Chlorophyll *a*).

3 Results and Discussion

3.1 PSASI I Data and Ratings: Characteristics of Seaweed Species Suitable for Application to IMTA

Genera were used for calculating the PSASI rating because seaweed species vary with region and local species should be used. The PSASI ratings, shown in Table 3, were derived from both published and unpublished literature. Five genera were collected and evaluated according to the three parameters employed in this study. All genera received a rating of 2 due to the economic values as food source or industrial/medical materials (Oh *et al.*, 1990; FAO, 2004), even though their values are not the same. It is possible that other genera, except *Ecklonia* and *Ulva*, can be relatively easily or industrially cultured. All genera except *Ecklonia* have been found suitable for application to IMTA. Accordingly, *Ecklonia* was rated 1 for both cultivation and the application of seaweed-based aquaculture parameters (Troell *et al*., 2003; FAO, 2004). In contrast, *Ulva* was rated 1 for cultivation. *Porphyra*, *Gracilaria*, and *Undaria* received a rating of 2 for both parameters. Based on the PSASI I scores for these ratings, *Porphyra, Gracilaria*, and *Undaria* had the highest PSASI I values (score = 8) of all tested genera, whereas *Ecklonia* had the lowest PSASI I value (score = 2). *Ulva* had PSASI I values of 4.

Table 3 Ratings of the PSASI I parameters and final PSASI I scores for seaweed species

Species	Economic value	Cultivation	Application of seaweed-based aquaculture	PSASI score
Porphyra	2	2	2	8
Gracilaria	2	2	2	8
Undaria	2	2	2	8
Ecklonia	2	1	1	2
Ulva	2	1	2	4

Note: maximum possible PSASI I = 8.

3.2 PSASI II Data and Ratings: Ammonium Uptake Rates of Seaweeds

The NH_4^+ uptake rates were determined for five seaweed species (*U. pinnatifida*, *E. cava*, *P. yezoensis*, *G. incurvata*, and *U. compressa*). In this study, the V_{max} of the green and red algae was higher than that of the brown algae (Fig.2). Of all species, *U. compressa* had the highest V_{max} (140.3 μmol g^{-1} DW h^{-1}) and thus was assigned a rating of 2 in the PSASI II (Table 4 and Fig.2). Whereas the V_{max} of *P. yezoensis* (111.5 μmol g^{-1} DW h^{-1}) was assigned a rating of 2 in the PSASI II, *G. incurvata* showed a low V_{max} value (50.6 μmol g^{-1} DW h^{-1}) and received a rating of 1 (Table 4 and Fig.2). In particular, all brown algae (*U. pinnatifida* and *E. cava*) showed very low V_{max} values (less than 30 μmol g^{-1} DW h^{-1}) and, accordingly, were assigned ratings of 1 in the PSASI II (Table 4 and Fig.2).

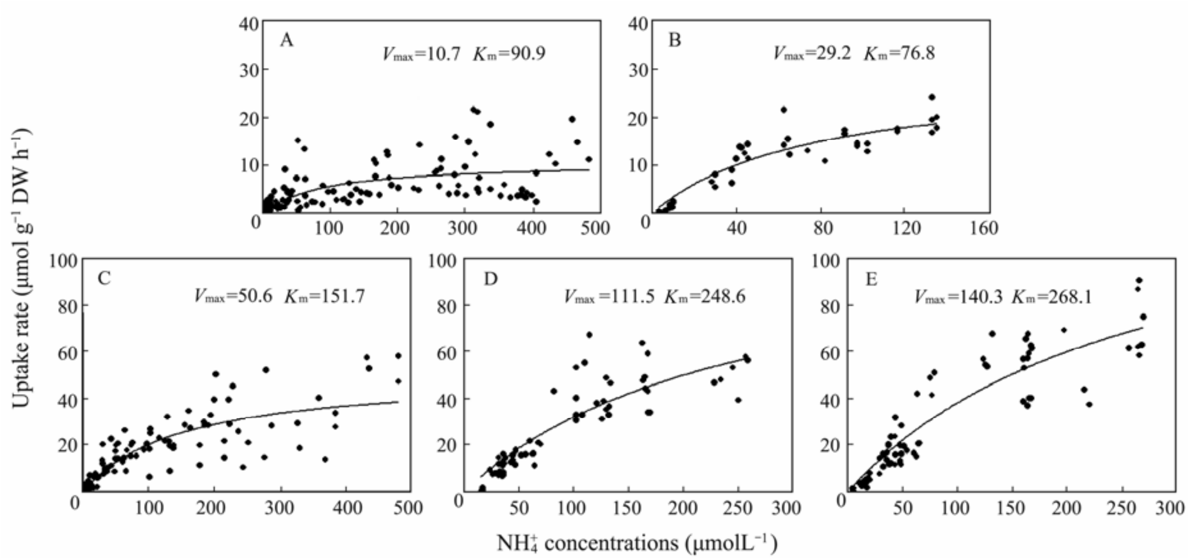

Fig.2 NH_4^+ uptake rates of seaweeds as a function of NH_4^+ concentration. Curves represent the best fits of the Michaelis-Menten equation; A, *Undaria pinnatifida*; B, *Ecklonia cava*; C, *Gracilaria incurvata*; D, *Porphyra yezoensis*; and E, *Ulva compressa*.

In IMTA, the NH_4^+ levels in fish farm effluent are relatively high, approximately 150 μmol L^{-1} (Neori *et al*., 1996; Carmona *et al*., 2006). Therefore, seaweeds exhibiting high K_m should be utilized as biofilter material for NH_4^+ effluent in IMTA, as the low-affinity system is operative and exhibits very high values of K_m at high concentrations (Lobban and Harrison, 1994). Our results showed that the K_m values of green and red algae were highest, whereas those of brown algae were relatively low. The K_m of *P. yezoensis* and *U. compressa* were 268.1 and 246.9 μmol L^{-1}, respectively (Fig.2). *G. incurvata* also showed a relatively high K_m (151.7 μmol L^{-1}, Fig.2). Therefore, these species were assigned ratings of 2. In contrast, the brown algae had K_m values below 100 μmol L^{-1} and thus were assigned ratings of 1 (Fig.2).

Overall, *U. compressa* and *P. yezoensis* had the highest PSASI II scores (score = 4), followed by *G. incurvata* (score = 2, Table 4). *U. pinnatifida*, and *E. cava* showed the lowest values (with PSASI II scores of 1) due to the low NH_4^+ uptake rate.

Table 4 Ratings of the PSASI II parameters and final PSASI II scores for each species

Species	V_{max}	K_m	PSASII score
Undaria pinnatifida	1	1	1
Ecklonia cava	1	1	1
Porphyra yezoensis	2	2	4
Gracilaria incurvata	1	2	2
Ulva compressa	2	2	4

Note: maximum possible PSASI Ⅰ = 4.

3.3 SASI Data: Physiological Parameters of Seaweeds

To examine the physiological characteristics of seaweed, tests of the NH_4^+ uptake efficiency, tissue N content, and chlorophyll *a* content were conducted on five species (*U. pinnatifida*, *E. cava*, *P. yezoensis*, *G. incurveta*, and *U. compressa*). Four species, excluding *E. cava*, showed NH_4^+ uptake efficiencies higher than 75%. In particular, *U. compressa* absorbed over 99% of the NH_4^+ in the water column during 7 d. Generally, *Ulva* showed high biofiltering effciency in IMTA system regardless of culture condition (Copertino *et al*., 2009). Thus, four of the tested species were assigned ratings of 2, whereas *E. cava* received a rating of 1 due to its relatively low uptake rate (Table 5 and Fig.3).

Table 5 Ratings for each parameter are multiplied to determine the SASI score for seaweed species

Species	PSASI I	PSASI II	Removal efficiency	Tissue N content	Chl *a*	SASI score
Undaria pinnatifida	2	1	2	2	2	16
Ecklonia cava	1	1	1	2	1	2
Porphyra yezoensis	2	2	2	2	1	16
Gracilaria incurvata	2	1	2	2	1	8
Ulva compressa	2	2	2	2	1	16

Notes: Maximum possible SASI score = 32. Optimal species for IMTA are those with SASI ≥ 8; species with a SASI < 8 would be rejected.

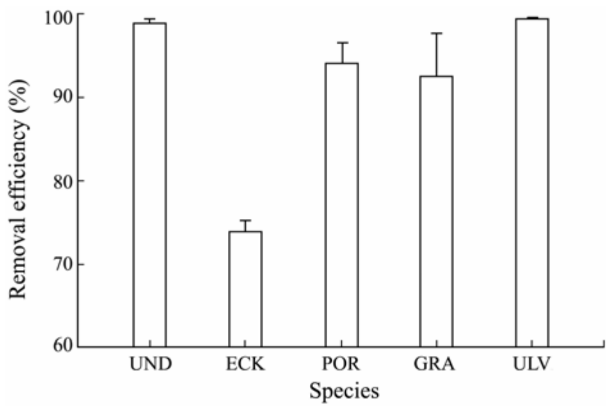

Fig.3 NH_4^+ uptake efficiency of seaweeds during the 7-d experiment. Data are expressed as mean ± SE (n = 6); UND, *Undaria pinnatifida*; ECK, *Ecklonia cava*; POR, *Porphyra yezoensis*; GRA, *Gracilaria incurvata*; ULV, *Ulva compressa*.

The tissue N content of seaweed is an optional parameter because it is not easy to generalize on the nutrient sequestration of seaweed (Neori *et al*., 2004). Nevertheless, the tissue N content of seaweed increases with N enrichment in seawater until saturation (Neori *et al*., 1991; Chopin *et al*., 1999; Davison *et al*., 2007; Kang *et al*., 2011). Martinez and Rico (2002) reported that some N can be sequestered as photosynthetic pigments, free amino acids, and proteins. Additionally, N content of *Garcilaria* steadily increases in various culture scale system for 1–2 months (Abreu *et al*., 2009, 2011). Therefore, higher tissue N content or the ability of N accumulation in tissue could be an important factor in the selection of seaweed species for IMTA. In the present study, the mean tissue N contents of *U. pinnatifida*, *E. cava*, *P. yezoensis*, and *G. incurvata* at the end of the experiment exceeded their initial values, whereas the mean tissue N content of *U. compressa* decreased slightly compared with the initial value. This was despite lack of significant differences in tissue N content between the start and the end of the experiments. Because the tissue N contents (mean ± SD) of all species were equal to or higher than (mean – SD) of the initial pre-experiment samples, they were assigned ratings of 2 (Table 5 and Fig.4).

The chlorophyll *a* content of seaweeds is dependent on the N concentration in the water column (Costanzo *et al*., 2000). It was found that macroalgae exposed to high N concentrations responded with increased chlorophyll *a* production within the 3-d incubation period. Our results showed that the chlorophyll *a* contents (mean ± SD) of four species (except *U. pinnatifida*) were lower than the (mean – SD) of the initial pre-experiment samples on day 7, and thus received ratings of 1 (Table 5 and Fig.5). However, *U. pinnatifida* received a rating of 2 because its chlorophyll *a* (mean ± SD) was within the range of the (mean – SD) (Table 5 and Fig.5).

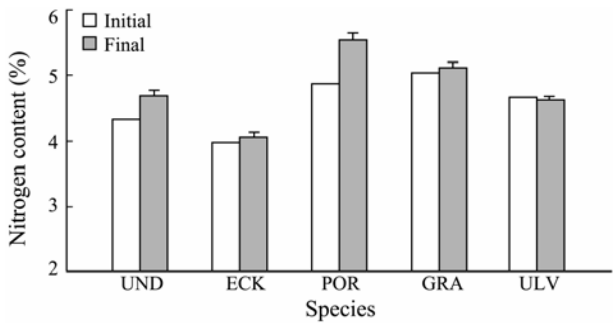

Fig.4 Tissue N contents of seaweeds at the beginning and the end of the experiment. Data are expressed as (mean – SD) and (mean ± SD) at the beginning and the end of the experiment, respectively (*n* = 6); UND, *Undaria pinnatifida*; ECK, *Ecklonia cava*; POR, *Porphyra yezoensis*; GRA, *Gracilaria incurvata*; and ULV, *Ulva compressa*.

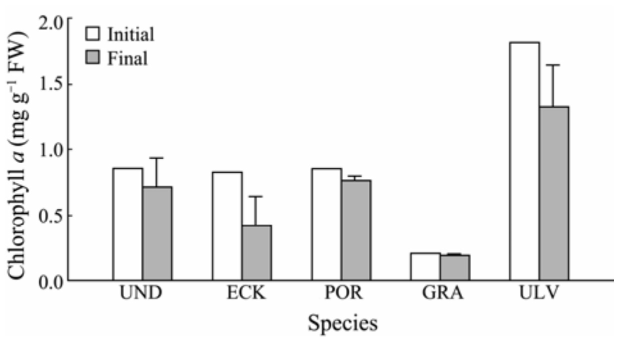

Fig.5 Chlorophyll *a* contents of seaweeds at the beginning and the end of the experiment. Data are expressed as (mean – SD) and (mean ± SD) at the beginning and the end of the experiment, respectively (*n* = 6); UND, *Undaria pinnatifida*; ECK, *Ecklonia cava*; POR, *Porphyra yezoensis*; GRA, *Gracilaria incurvata*; and ULV, *Ulva compressa*.

3.4 Calculation of SASI Ratings

The PSASI I based on PSASI I score is the first SASI ratings parameter. In the PSASI I, *U. pinnatifida*, *P. yezoensis*, *G. incurvata*, and *U. compressa* were as- signed a rating of 2 because their PSASI I scores were 4 or 8 (Tables 3 and 5). However, *E. cava* was assigned a rating of 1 for the PSASI I due to the low probability of its successful cultivation and application in seaweed- based aquaculture (Tables 3 and 5). For the PSASI II scores related to the NH_4^+ uptake rate of each species, two species (*P. yezoensis* and *U. compressa*) were rated as 2, and the other species received ratings of 1. The final SASI scores for each species, as calculated by multiplying together the SASI ratings (PSASI I × PSASI II × SASI data) are shown in Table 5. Because five SASI parameters were measured, the maximum achievable SASI score was 32. Seaweed species with SASI scores ≥ 8 were considered priority species for IMTA. In the present study, *U. pinnatifida*, *P. yezoensis*, and *U. compressa* received the highest SASI scores of 16. *G. incurvata* received a SASI score of 8. Accordingly, these four species were deemed suitable for IMTA. *E. cava* received the lowest SASI score and was considered unsuitable for IMTA due to its low removal efficiency and the difficulty in cultivation.

3.5 Application of the Species-Selection Model

The seaweed species receiving high scores using this index were those genera that have been applied to IMTA in previous literature (*e.g.*, Troell *et al*., 2003). Both this index and previous identification of seaweeds for IMTA have primarily focused on the nutrient removal efficiency of seaweed. Based on the present index, *U. pinnatifida* can be selected as a good candidate seaweed species for IMTA. In fact, *U. pinnatifida* has dominated culture production, constituting 46% of the total wet weight production in Korea (Yoon, 2008), and has been co-cultured with fish in Korea for several decades. Few studies, however, have introduced *U. pinnatifida* as a biofilter in IMTA. Thus, this index reflects the cultural conditions of local areas and is a useful index for species selection in a seaweed-based integrated aquaculture system.

The experiments used in developing the SASI index were conducted on a laboratory (*i.e.*, relatively small) scale. As the nutrient uptake rates of seaweeds depend on areal nutrient loads (Troell *et al*., 2003), we should confirm whether these results are reproducible in tank-based or land-based systems. At present, the experiments are being conducted in a large tank-based system using the seaweeds selected by the proposed index. So far, the seaweeds have shown increased nutrient uptake efficiencies up to 50% and slight increases in tissue N content and growth rate (Kang, unpublished data).

3.6 Supplementation of the Species-Selection Model

Still, more components could be derived to complement the SASI index. It is necessary for a more powerful index to include supplementary physiological parameters of seaweeds such as biochemical composition (tissue phosphate content, C : N and N : P ratios), photosynthesis, growth rate, and phosphate and nitrite/nitrate uptakes. Vergara *et al*. (1993) demonstrated that the tissue C : N ratio is a powerful index for the physiological status of seaweed. Photosynthetic characteristics such as chlorophyll fluorescence are used as potential indicators of seaweed health status in the integrated cultivation of macroalgae (Figueroa *et al*., 2006). Seaweed growth rate will be measured for real nutrient removal rates by seaweeds in aquaculture systems during the culture period. As aquaculture effluents contain considerable inorganic phosphate and nitrate, the phosphate and nitrite/nitrate uptake rates of seaweed should be considered (Neori *et al*., 1998). The co-occurrence of different N forms can affect N uptake rate (Abreu *et al*., 2011). The application of multiple seaweeds to IMTA can increase N removal efficiency because the uptake rate of each species is dependent on the form of different N sources and different species are complementary in using nitrogen source (Bracken and Stachowicz, 2006; Kang *et al*., 2011). Furthermore, the effects of water temperature on the physiological characteristics of seaweeds should be considered due to a distinct seasonal trend in Korea. Wheeler and Srivastava (1984) reported that seaweeds have optimal temperature ranges over which nutrient uptake occurs. Kang *et al*. (2008) suggested that *Codium fragile* is a suitable species for use in IMTA in summer.

4 Conclusions

In summary, this study developed a species-selection index applicable to seaweed-based IMTA systems for identification of seaweed species best suited for improvement of water quality and reduction of necessary cost and effort for sustainable, environmentally friendly aquaculture. The proposed index takes into account the physiological characteristics indicative of seaweed quality, such as biochemical composition and pigment, as well as the economic value and nutrient removal efficiency of the seaweed species. The seaweed species that have been used in IMTA receive higher scores by this index, identifying species suitable for seaweed-based IMTA system. The species-selection index requires additional parameters and further testing of its application. Although the proposed index cannot account for every eventuality and may incur some errors, it provides valuable information for IMTA seaweed species selection.

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