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Studies on technology for seaweed forest construction and transplanted *Ecklonia cava* growth for an artificial seaweed reef

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Abstract

We installed seaweed reef for restoration of barron ground coast. We hollowed out a U-shaped groove in a cross-shaped artificial seaweed reef and covered it with a zinc sheet (U-bar) to transplant *Ecklonia cava* growing on *Dellenia* wood by hand, installing the U-bar on the artificial seaweed reef, fixing it with concrete. Thus seaweed can be attached easily, with pre-installed stainless bolts and nuts. The length of *Ecklonia cava* leaf transplanted to the cross-shaped reef was 7.2 cm in February 2005 reached its maximum size, 35.9 cm (n=30) by July. Thereafter, it decreased to 18.9 cm in October due to shedding. The leaf weight after the experiment was 24.8 from the initial 0.4 cm (n=30). Regression analysis showed Y=0.7875X-4.6488 (R^2=0.7225) for blade length and Y=0.0025$X^{2.6733}$ (R^2=0.8711) for leaf weight. The high values of the R^2 values for the two measurements were highly reliable, with the reliability of the linear regression function higher than that of the functions of 2 variables. The artificial seaweed forest constructed in the barren ground was highly comparable with natural seaweed forest in terms of growth, indicating that the artificial seaweed construction can be done in an easy, efficient and economically viable way. This further indicates that the technology developed by the present study can be extensively used for the project for artificial seaweed forest construction.

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Introduction

Though seaweeds in the east coast are known to be distributed from the shore to 30 m in depth (Shin *et al.,* 2008; Choi *et al.,* 2009; Kim *et al.,* 2010), they are restricted to within 10m in the southern part of the east coast and distributed to 30 m in the northern part. The difference in vertical seaweed distribution is determined by light intensity given transparency and regional water temperature (Lobban and Harrison, 1997). Various weeds can grow in the east sea as its sediment quality is mainly rock beds and it has a relatively higher transparency than the south and west seas. These seaweeds reduce carbon dioxide and produce oxygen through carbon fixation processes in marine ecosystems, providing feed for useful marine resources such as fish and invertebrates, spawning grounds, shelters and nursery grounds (Lindstrom, 2009). However, recently seaweed flora has become stressed and its biomass has been sharply reduced. It is a serious problem as the output of useful marine resources is decreasing as well.

Barren ground reducing primary productivity is gradually spreading and destroying the coastal ecosystem in Korea (NFRDI, 2005). Barren ground is the area where seaweed forests disappear and cannot grow anymore. It is called Getnoguem (in Korean) that means burning rocks, indicating the desertification of coastal rock bases. *Crustose* coralline algae living on the surface of rock beds are pink and the color turns white after death. Thus, fishermen mention this as to 'whitening phenomenon' (Kim *et al.,* 2006, 2007). The causes of barren ground are the destruction of seaweed communities resulted from overharvesting of commercial seaweeds, overgrazing by marine animals (i.e. urchins, sea slugs and gastropods), and destroying seaweed habitat destruction due to coastal industrialization and pollution (NFRDI, 2005, 2006). In 2004, barren areas were approximately 7.0% in Gangneung, Gangwon Province (mid coast of East Sea), and ca. 31.0% in the southern coast of east sea (Gyeongbuk). In the southern coast of Korea, the barren ground areas around Yeosu were more than 70 ha in 2006 and 655 ha in 2009.

To restore barren ground areas, the National Fisheries Research and Development Institute constructs a large seaweed forest in order to enhance seaweed resources (i.e. bio-energy) and coastal productivity, and to remove nutrients from seawater (NERDI, 2009).. The perennial brown alga, *Ecklonia cava* grows naturally from Jeju Island to Samcheok, Gangwon Province and is utilized frequently for constructing seaweed forest. It is also very important commercial species not only for feeding abalone and but also for fostering marine animals in the marine ecosystem. Seaweed forests should be constructed after considering environmental condition where the forest is created. So far, marine seaweed forests are created by using the technologies of spore transplantation, underwater bottom long lines, and artificial sac facilities (NERDI ESFRI, 2007). It is well known that spore transplantation is the most effective method for contributing seaweed forests because seaweeds transplanted grow and then release zoospores which make natural marine forests. Here, we introduced the spore transplantation method and monitored the growth of *Ecklonia cava* transplanted via this method.

Materials and Methods

Overall test sea area and artificial reef facility conditions: The test coast area is a test fishery belonging to a director of the East Sea Regional Fisheries Research Institute in Samcheok, Gangwon-do (province) (Fig. 1). Its depth is 6~10m, with well developed even rock beds, and smooth current flow, as it is connected to the open sea, in a gulf shape, with breakwaters on the coast.

In the director's test fishery in Samcheok, we established 12 cross-shaped artificial reefs on a rock bed at a dept of 7~9 m. We left 2 m of space between the artificial reefs, as it is a sea area where barren ground has deepened. The artificial reef specifications were

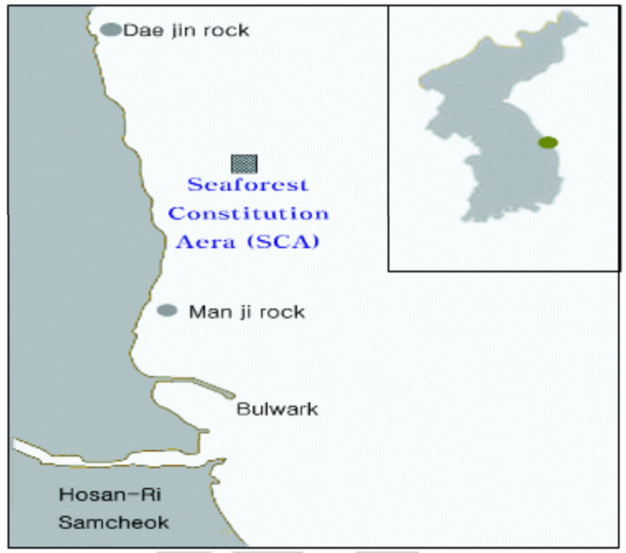

Fig. 1: Location of the seaweed forest construction site at Imwon, Samcheok.

3.5×3.5×0.5 m, 4.596 tons, and 1.915 m^3 volume, and they are designed for 15 m of spores to be transplanted. It is designed low so that it can be installed near coasts and the effects of current were minimized by making holes in the artificial reefs.

Seaweeds transplantation technique: For the cross-shaped artificial reefs used in this study, we hollowed out a U-shaped groove in a cross-shaped artificial seaweed reef and covered it with a zinc sheet (U-bar) to transplant *Ecklonia cava* seaweed growing on *Dellenia* wood by hand, installing the U-bar on the artificial seaweed reef, fixing it with concrete. The spore house where seaweed attaches is resistant to current flow, thus seaweed can be attached easily, with pre-installed stainless bolts and nuts. The spore house was then fixed on seawater-resistant *Dellenia* wood using tacks, inserted into the prefabricated U-bar groove and tightened the nuts. Since these artificial reefs are designed to be easily attached and detached in transplanting seaweed spores, it is easy to establish them again if seaweed dies naturally or withers after seaweed forest has been created.

There are two methods of spore planting. One is to load artificial reefs on a barge and transplant them onboard, and the other is to transplant them under water. For transplantation onboard, spores can be transplanted quickly and efficiently, but the seaweed dries out easily. To address this, the time spent onboard should be minimized. To prevent transplanted spores from drying after transplantation but before submergence, they should be covered with nonwoven fabric soaked in seawater, and constant re-wetting. Underwater transplantation takes a long time to establish a facility and costs a lot, as professional divers transplant the spores underwater. Seaweed planted in this way, has high survivability however, as seaweed planted this way are never dried and there is no damage to the leaves due to exposure to light or high temperatures. This study used underwater spore transplantation, temporarily transplanting *Ecklonia cava* spores produced and attached to a cremo screw for 5 days. These were then fixed to *Dellenia* wood after winding them around 22 mm of rope and transplanted to the artificial reefs. The leaf length was 7.2 mm and the weight was 0.4 g at the time of transplantation.

U-BAR: A thin iron plate covered with thin zinc film, with a much higher chemical resistance than tin plate. Its specifications are 3,000 mm in length, 30×40×30 mm and 1.2 mm in thickness. Its fastener is coated with zinc, 140 mm long, 40 mm wide, and 3 œ3 thick. It fixes seaweed rope tied to *Dellenia* wood for transplantation.

Stainless anchor (sus set-anchor; SUS304): SUS304 grade stainless steel. The body and bolt are integrated, with a wedge drawn out and fixed, adhering more closely to the cover when tensile force is applied to the bolt. A Set Anchor is particularly advantageous when high intensity resistance is required and is often used in Elevators, and Guide Rails. Anchor bolt specifications are W1/2" bolt size, 17.3 mm external diameter, 120 mm full length, 70 mm screw length, 55 mm boring depth, 3,200 maximum concrete strength at 200 kg cm^{-2} and 3,400 kg maximum shear resistance.

Dellenia wood: To prevent ropes for seaweed growth from moving, *Dellenia* wood which is used in ships and enclosing nets, was used. *Dellenia* is appropriate as it is highly salt-resistant (about 7 years) and lasts in seawater while the seaweed is taking root. Its specifications are 30 mm × 30 mm × 30 mm and both 1 m and 3 m lengths were used.

Seaweed floras and community structure: Seaweed floras and community analysis was carried out in the Imwon, Samcheok sea area in the East Sea, in February 2005, before the seaweed forest was constructed, and in February 2006 and 2007, 1 year and 2 years after creation. For a quantitative survey measuring biomass, we laid out two quadrates (50 cm × 50 cm) partitioned into 10 cm × 10 cm to a depth of 5, 10, and 15m each in the subtidal area and collected all seaweeds that appeared there, using chisels. The collected seaweeds were fixed with a 5-10% formalin-seawater solution on the spot, carried to the laboratory, identified by microscope and a list of appearing species and National names was made following Lee and Kang (2002).

We removed sand and impurities from quantitatively collected seaweed by washing them many times with fresh water, then identified them, measured their wet weight in 0.1 g, and converted the value into biomass per unit area (g wet wt. m^{-2}).

We calculated richness, diversity, evenness and dominance indices using PRIMER version 6 on the basis of average biomass by species for every quadrat according to study period and quantitative data.

Regression analysis: The growth (blade length, blade width) of transplanted *Ecklonia cava* was calculated every month from February to November using the skin scubar method. Only length was calculated by non-destructive methods in this sea area, and biomass was estimated for the measured data by carrying out regression analysis on the basis of data acquired every month using destructive methods for an *Ecklonia cava* seaweed forest being cultivated in the same way in the Hosan sea area about 20 km away from test sea area (Ko *et al.,* 2008). In addition, for monthly distribution according to transplanted *Ecklonia cava* blade length and width growth, we used Hosan data which used a destructive method.

Results and Discussion

The transplanted *Ecklonia cava* blade length in Imwon, Samcheok was 7.2 in February 2005, at the early stage of transplantation, and increased to 35.9 in July, showing maximum growth. After that, it decreased to 18.9 in October due to shedding (Fig. 2). The blade weight increased by 0.4 in February along with blade length, and showed maximum weight decrease (24.8) in July. For blade length, there was a sharp increase from May to June and a sharp decrease in July and a slow decreased from July to August, showing a large difference between blade length and weight (Fig. 3). Transplanted *Ecklonia cava* did not grow during the study period.

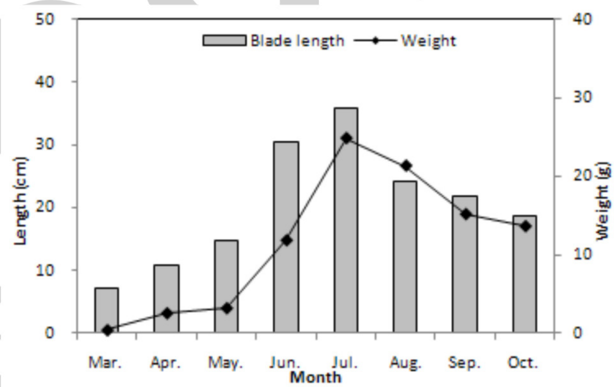

Fig. 2: Monthly variations in mean plant length and wet weight of *Ecklonia cava* in the study period at Imwon. Values are mean of 30 replicates ± SD

We identified the blade length and weight of *Ecklonia cava* in Imwon by substituting the blade length of transplanted *Ecklonia cava* in Imwon for the results of regression analysis of *Ecklonia cava* transplanted in Hosan. For blade length of the *Ecklonia cava* population, whole individuals were distributed within the 0-10 cm range in March 2005, at the early stage of transplantation, individuals in the 10-20 cm range were dominant in April and May, accounting for 53.3% and 70.0%, respectively. The 20-30 cm range was dominant accounting for 53.3%, the 30-40 cm range also showed high frequency accounting for 43.3%, and the 40-50cm range also accounted for 3.3% in June. In July, individuals in the 30-40 cm range accounted for 86.7% of all individuals, those of the 40-50cm range amounted to 10.0% and moved to the range where grown blades were bigger by July. On the other hand, shedding started to appear in August, so individuals of 40-50 cm didn't appear and those of 30-40 cm also sharply decreased, showing only 6.7%. Those of 20-30 cm accounted for 83.3% of the total. In September and October, shedding continued and individuals in the 20-30 cm range and the 10-20 cm range were dominant, respectively (Fig. 3A).

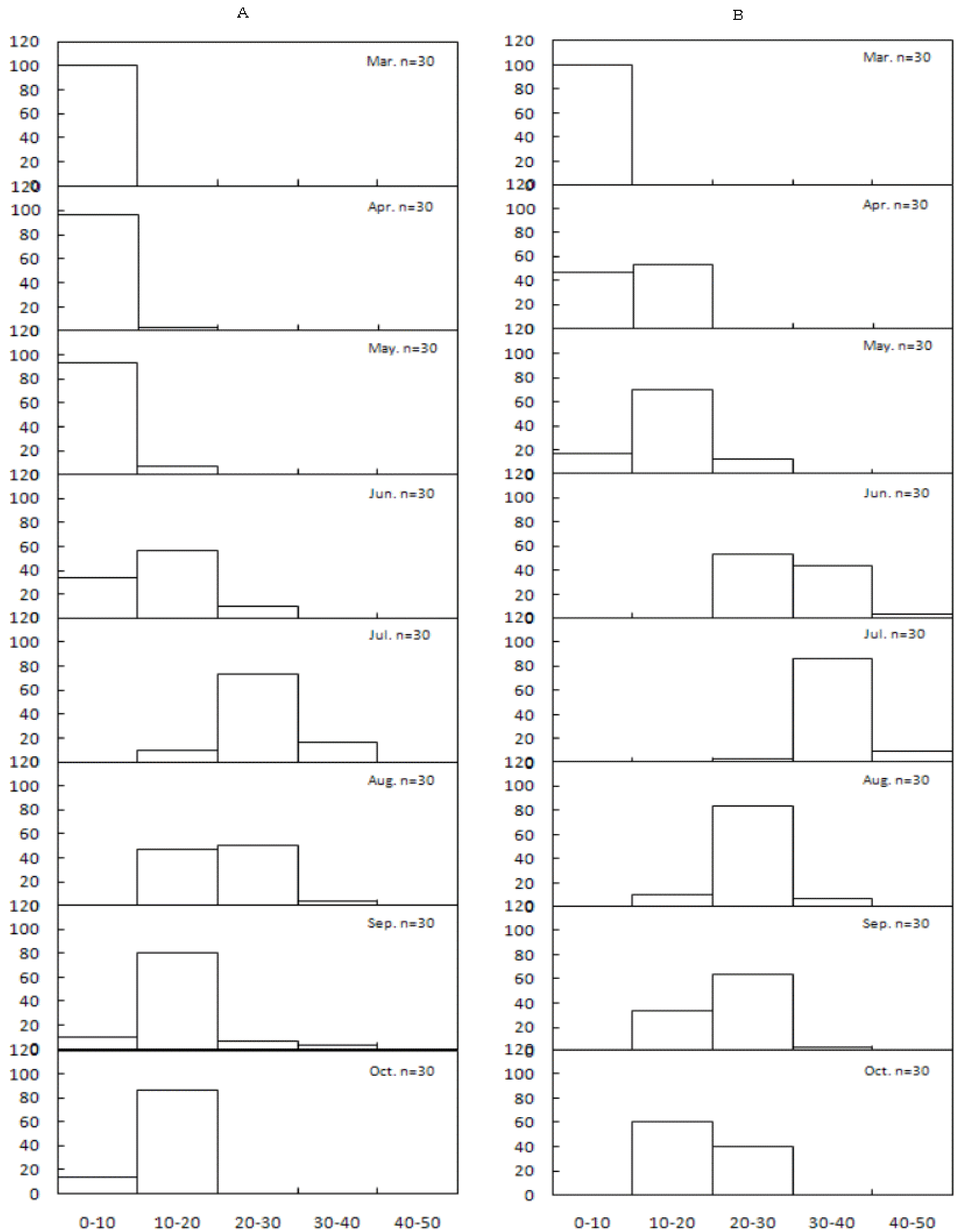

Fig. 3: Monthly blade weight change (A) and length (B) composition for *Ecklonia cava.* spore transplantation on artificial seaweed reef at Hosan.

For transplanted *Ecklonia cava* weight, 93.3-100.0% were below 10 g from February to May, 56.7% of in the 10-20 g range in June. In July, at maximum growth, 20-30 g accounted for 73.3%, individuals and 30-40 g accounted for another 16.7%. In August, when shedding appeared, individuals at 30-40 g sharply decreased, for 3.3% and those at 20-30 g were dominant, accounting for 50.0% as in July. Shedding lasted through September and October and individuals of 10-20 g were dominant with those above 20 g disappearing in October (Fig. 3 B).

Linear regression analysis was defined as a function of Y=0.7875X-4.6488 (R^2=0.7225) and the function of 2 variables were defined as a function of Y=0.0025$X^{2.6733}$ (R^2=0.8711). The R^2 value of both showed high reliability, but the reliability of the function of 2 variables appeared higher than that of the linear regression function (Fig. 4).

There were 24 seaweed species in the Imwon sea area from 2005, before the seaweed forest was created, to February 2007, 2 years after creation, including 4 species of green algae, 8 of

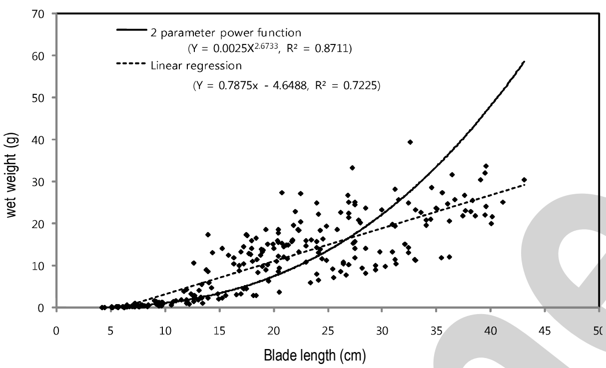

Fig. 4: Linear and nonlinear regression models for estimating *Ecklonia cava* biomass at Hosan. The solid line stands for standard linear regression and the dashed line for 2 parameter power regression.

brown algae, 21 of red algae and one species of phanerogamous plant (Table 1). With respect to classification, red algae appeared most frequently, with green algae, brown algae, and red algae comprising 11.8%, 23.5% and 61.8%, respectively. By study period, in 2005 before the seaweed forest was created, 21 total species appeared (2 of green algae, 7 of brown algae, and 9 of red algae), 20 species (2 of green algae, 7 of brown algae, and 11 of red algae) appeared in 2006, a year after creation, and in 2007, two years after creation, 13 species (species of green algae, 3 species of brown algae, and 9 species of red algae) appeared, showing that species decreased since the seaweed forest was created (Table 1).

Community indices calculated using biomass and species of seaweed are shown in Table 3. The abundance index was within a range of 2.58-4.18, highest in 2006, a year after seaweed forest creation, with a sharp decline identified in 2007. On the other hand, the evenness index was within a range of 0.40-0.69 and was highest

Table- 1: Community indices for biomass each year in Imwon.

Community index	Year		
	2005	2006	2007
Richness index, R	3.89	4.18	2.58
Evenness index, J'	0.53	0.40	0.69
Diversity index, H'	1.61	1.19	1.76
Number of species	21	20	13
Total biomass (g/m2)	171.43	94.00	103.97

in 2007, meaning that biomass was evenly distributed through the species in 2007. The species diversity index was within a range of 1.19-1.76, at a minimum value in 2006, a year after creation, and maximum in 2007, two years after creation. Species and species diversity appeared to decrease due to human interference and physical effects after seaweed forest creation, but it is thought that the seaweed community is gradually stabilizing.

Biomass by study period was within a range of 171.43-94.00 g (average 123.13 g) and showed maximum value in 2005 when the seaweed forest was created, sharply decreased to the minimum in 2006, then increased a little in 2007. With respect to main species by biomass, 89.37 g for *Gracilaria* sp. made it the dominant species followed by *Acrosorium polyneurum* and *Plocamium telfairiae* in 2005. In 2006, 67.57 g for *Desmarestia ligulata* made it the dominant species with no other species exceeding 5g. At 48.43 g *Sargassum honeri* was the dominant species and at 15.77 g *Gelidium amansii* was the second (Table 2).

A large quantity of catch is possible in a short period along the Korean coastline, as fishery technology has been highly developed. This is causing rapid decrease in costal resources (An *et al.,* 2007). Faced with this situation, a total allowable catch (TAC) system for catch limits to protect and manage fishery resources, as well as acceptable biological catch (ABC) are being carried out. However, there is no system for costal seaweed or method for efficient resource use, such as planned harvests or fallow years. resulting barren ground is thus sharply decreasing seaweed resources and ecosystem destruction which rapidly reduces fishery productivity is occurring. Seaweed performs a range of functions including photosynthesis which creates oxygen from carbon dioxide, sea water purification by absorbing excessive nutrient salt, space for epifauna to live and feed, spawning grounds and habitat for marine animals. Fishery production is thus influenced by the amount of seaweed resources (Ohno, 1993). The results of this study indicate 34 seaweed species were in the Imwon sea area from 2005, before the seaweed forest was created, to February 2007, 2 years after creation, including 4 species of green algae, 8 of brown algae, 21 of red algae and one species of phanerogamous plant. The composition ratio by classification was 11.8% green algae, 23.5% brown algae, and 61.8% red algae. Species diversity has thus been restored as a result of seaweed forest creation.

Ecklonia cava which is most commonly used in seaweed forests, grows from September to October and forms apothecium.

Table- 2: Mean biomass of dominant seaweed around the seaweed forest area in Imwon

2005	2006	2007
Gracilaria sp. (89.37)	*Desmarestia ligulata* (67.57)	Sargassum honeri (48.43)
Acrosorium polyneurum (19.10)	other species (26.43)	Gelidium amansii (15.77)
Plocamium telfairiae (17.63)		Colpomenia sinuosa (9.80)
Acrosorium yendoi (14.73)		Symphyocladia latiuscula (7.90)
Codium arabicum (10.93)		Bossoella cretacea (6.63)
Prionitis divaricata (8.40)		Dictyota dichotoma (5.30)
Agarum clathratum (7.23)		other species (10.13)
other species (4.03)		

Zoospores released develop into female and male gametophytes. When female and male gametophytes grow, diplophase sporophyte are formed through pollination and develops into young leaves. It lives mainly below 5-15 m depth but its habitat tends lower as water temperature in the East Sea rises (NFRDI, 2007). In Japan, *Ecklonia cava* is distributed up to a depth of 30 m in the temperate zones of the southern part of Japan (Kawashima, 1993).

In Japan, as barren ground referred to as Isoyake has occurred in many sea areas, fishery productivity sharply decreased, and natural restoration of seaweeds has been very difficult (Terawaki, 2001). Thus, Japan started a study on increasing barren ground sea area productivity, led by the Fishery Agency, and a study to crate seaweed forests in development areas was attempted by the Resource Energy Agency in the 1980's. Various studies began in the 1990's and the Ministry of Transportation created a developed port, the Ministry of Construction built breakwaters and the Fisheries Agency was in charge of new fishing grounds. In the recent 2000's, the creation of seaweed forests has being investigated for environmental conservation and ecosystem maintenance. A study on technology for seaweed forest creation, carried out by Terawaki (2001) is similar to the concept of artificial reefs for seashells and seaweed in Korea, in that it installs implanted natural spores living in artificial reefs in test sea areas. However, as it is difficult for natural spores to be formed or implanted and live in sea areas where barren ground has been aggravated (NFRDI, 2007), young spores are directly transported into artificial reefs in Korea. When creating seaweed forests in the early stage, ropes where spores are attached were wound around artificial reefs (Kim *et al.,* 2006, 2007). When using this method however, such ropes constantly move due to rainstorms, typhoons, and waves, causing transplanted seaweed to fall off. Spore transplantation used in this study however, made up for this weakness and prevented spores from falling off. Spore transplantation was used mainly in Pohang and Gangneung in the East Sea area, Namhae in the South Sea area, and Seoqwipo, Jejudo in 2009.

Ecklonia cava showed its longest leaf length in July and leaf length was verified as relatively long from June to August. This result is similar to that of *Ecklonia stolonifera*, a related species (Kim and Yoo, 2003). According to a study by Serisawa *et al.,* (2004), a population with a 20-30 cm leaf length occurred in July then the *Ecklonia cava* colony disappeared due to barren ground. Abalone production in this fishing ground was 1723.5 kg in 1996 but none was produced in 2000 when the *Ecklonia cava* colony disappeared. According to Kang *et al*., (2001), the *Ecklonia cava* around Dokdo thrived even at a depth of 2 m and its leaf length was longest in summer. *Ecklonia cava* growing around Dokdo had an average leaf length of 26.0, 678.7 g weight and a maximum value of 131.3, 678.7 g respectively. Considering that it is a perennial seaweed, it is estimated to be the result of 5 year's growth.

The natural habitat of *Ecklonia cava* is a depth of 5~25 m around Dokdo but 15~20 m in key habitats such as the Hongdo area (Kang *et al.,* 1993) and Tosa Bay in Japan (Tominaga *et al.,* 1999). For the East Sea, the Samcheok area water depth, in the northern part of the East Sea, was 5~12 m and 3~9 m in the southern part of the East Sea, exhibiting different distributions (NFRDI, 2007).

Recently, as barren ground along the coastlines of Korea become more aggravated (Kim et al. 2010), the creation of seaweed forests is being carried out as a countermeasure. In the 2000s, natural spores implanted to inhabit artificial reefs were investigated regarding environmental conservation and ecosystem maintenance (Terawaki, 2001). However, for regions where barren ground is extreme, there were no spores released from grown seaweed, as there were few extant seaweeds. Therefore, young spores were directly transplanted into artificial reefs in Korea. When creating seaweed forests in the early stage, ropes where spores are attached were wound around artificial reefs (Kim *et al.,* 2006, 2007). When using this method however, such ropes constantly move due to rainstorms, typhoons, and waves, causing transplanted seaweed to fall off. Spore transplantation used in this study however, made up for this weakness and prevented spores from falling off. Spore transplantation was mainly used in Pohang and Gangneung in the East Sea area, Namhae in the South Sea area, and Seoqwipo, Jejudo in 2009.

The water seaweeds habitation depth varies according to regional water temperature and transparency (Lobban and Harrison, 1997). *Ecklonia cava* lives at a depth of 5~25 m around Dokdo and 15~20 m in key habitats (Kang *et al.,* 2001). In Samcheok, in the northern part of the East Sea, the water depth was 5~12 m and in the southern part of the East Sea, it was 3~9 m (NFRDI, 2007). In this study, artificial reefs were installed at a depth of 10 m and the growth of *Ecklonia cava* was identified as very good as its leaf length was 7.2-35.9 cm. According to Kang et al (2001), *Ecklonia cava* around Dokdo grew best in summer, with an average leaf length and weight of 26.0 cm, and 64.0 g respectively. Maximum leaf length and weight was 131.3 cm, and 678.7 g, respectively, exhibiting a large difference compared to the 35 cm maximum growth in this study. The difference might be caused by the fact that we transplanted young leaves and measured the growth of only 1-year-old seaweed in this study while in case of Dokdo, the natural *Ecklonia cava* colony was measured and all individuals from 1 to 5 years old were included.

The primary blade length of *Ecklonia cava* was decreased from summer to fall (Serisawa, 2001). This is the result of shedding, as reported for *Ecklonia kurome*. In this study, *Ecklonia cava* leaf length and weight increased from March to July, but constantly decreased from July to October with. In addition, *Ecklonia kurome* showed maximum growth in April and May while *Ecklonia cava* in this study showed maximum growth in July. However, it is similar to the results of *Ecklonia stolonifera*, a related species, and the change in monthly leaf length proved similar (Kim and Yoo, 2003). According to the study of Serisawa *et al.,* (2004), a population of *Ecklonia cava* with 20-30cm leaf length appeared in July and then the colony disappeared due to shedding.

For a regression analysis for *Ecklonia cava* growth we used the linear regression function known to be appropriate for *Ecklonia cava* growth analysis and the function of 2 variables, with the nonlinear regression function defined as an exponential function (Ko et al. 2008). As a result, the regression formulas were defined as $Y=0.7875X-4.6488$ ($R^2=0.7225$) and $Y=0.0025X^{2.6733}$ ($R^2=0.8711$), showing very high reliability. According to Ko *et al.,* (2008), the R^2 value of the linear regression function was 0.7408, higher than this result but the value of the function of 2 variables proved most appropriate for *Ecklonia cava* growth analysis at 0.7496, which showed lower reliability than this result. The difference in reliability might be caused by the fact that the length of each individual was reflected in this study while regression analysis was implemented after classifying size classes of 10 cm in the study by Ko et al., (2008). Another reason is that this study measured more individuals and reflected those results.

The artificial seaweed forest constructed in the barren ground was highly comparable with natural seaweed forest in terms of growth, indicating that the artificial seaweed construction can be done in an easy, efficient and economically viable way. This further indicates that the technology developed by the present study can be extensively used for the project for artificial seaweed forest construction in Korea.

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