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Farming of the giant kelp *Macrocystis pyrifera* in southern Chile for development of novel food products

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Key words: Macrocystis pyrifera, farming, food products, Chile

Abstract

This study explores the potential cultivation of the giant kelp *Macrocystis pyrifera* (L.) C.A. Agardh in southern Chile, for the development of novel food products. The study demonstrates the importance of considering the collection site of the parent sporophytes for successful cultivation. This study also shows that the ropes must be seeded with 10,000 to 40,000 spores ml^{-1}, depending on the culture method used. We also demonstrated that under environmental conditions in southern Chile, the seeded ropes must be put at sea at the latest during autumn (April) in order to reach the harvesting season in December. However, several other management aspects must be considered to improve the quality of the product. Our final estimation indicates that over 14.4 kg m^{-1} of rope (fresh weight) can be produced and from this total production, over 70% can reach the quality to produce different food products that are already being introduced in oriental countries. The remaining 30% can be used for abalone feeding and is also available for the organic fertilizer industry located in Chile.

Introduction

California kelp beds started to be harvested as a source of potash during the first decade of the 20th century and commercial interest in the giant kelp *Macrocystis pyrifera* expanded significantly between the 1970s and 1980s (Neushul, 1987; Druehl et al., 1988). This interest was primarily for the production of alginates, but also to produce biomass as a feedstock for methane production as a consequence of the energy crisis at that time (North et al., 1982; Gerard, 1987; Neushul & Harger, 1987). Nevertheless, *M. pyrifera* commercial cultivation for methane production was never a reality. At present, the supply of *M. pyrifera* biomass for the alginate industry relies exclusively on restoration practices and management of natural beds to obtain a sustainable production (North, 1979; McPeak & Barilotti, 1993; Vásquez & McPeak, 1999). After the energy crisis and because of the low price of alginates, farming research on *Macrocystis* declined sharply.

On the other hand, other brown algal species began to be commercially cultivated in Japan, China and Korea, mainly for human consumption (Tseng, 1987; Kaneko, 1999; Hanisak, 1998) while kelp-farming attempts for this purpose have also proved technically feasible in other regions (e.g. Druehl et al., 1988; Kain, 1991; Merril & Gillingham, 1991). Interestingly, the demand for brown algae is also increasing due to the introduction of new uses such as fertilizers, cultivation for bioremediation purposes, and abalone as well as sea urchin feeding among others (Petrell et al., 1993; Vásquez & Vega, 1999; Buschmann et al., 2001c; Ugarte & Sharp, 2001; Chopin et al., 2001). In Chile, despite the commercial importance of various algal species, aquaculture is still limited to the red alga *Gracilaria chilensis* (Buschmann et al., 2001b).

In Chile, Abalone and sea urchin cultures, organic fertilizer production and novel seafoods have created a new niche market for the giant kelp *Macrocystis pyrifera*. Increased harvesting is already causing some deterioration of different kelp populations (Vásquez & Vega, 1999). Considerable information on *Macrocystis* cultivation has been published in the past (North, 1979). However, some basic knowledge necessary to run a successful commercial activity is still lacking, especially with regard to the different environmental conditions and complex morphological and reproduction variability between populations, that can have important commercial consequences.

Considering this new market scenario, the potential impact on natural populations and the lack of biological knowledge necessary to produce a high quality product, this paper deals with the cultivation of *M. pyrifera* in southern Chile. Specifically, the effect of the origin of the parental plants on the survival and growth of young sporophytes cultivated on ropes was tested, in both hatchery and field conditions. Finally, a pilot cultivation was established to determine the potential yields of *M. pyrifera* in southern Chile and we describe some of the food products developed.

Materials and methods

Study sites

Fertile sporophylls of *Macrocystis pyrifera* were collected at six localities in southern Chile: Metri (41°35′S; 72°42′W), Pargua (41°47′S; 73°25′W), Calbuco (41°46′S; 73°08′W), Pucatrihue (40°33′S; 73°43′W), Bahía Mansa (40°34′S; 73°44′W) and Curaco de Velez (42°26′S; 73°35′W) (Figure 1). Site selection was based on the presence of abundant kelp populations and different water movement conditions. The plants were collected by scuba divers and transported, within 6 h, on ice to the seaweed culture laboratory in Metri. All field cultivation experiments were carried out in Metri and the pilot culture in Calbuco (Figure 1).

Cultivation of different populations

The sporophylls collected in Metri, Pargua, Calbuco, Pucatrihue and Bahía Mansa, were washed under tap water and UV treated filtered seawater (0.2 μm) containing commercial iodine (0.5% for 10 s), packed in filter paper, covered with aluminum foil and stored at 15 °C (Figure 2A). After 12 h, 10 to 15 sporophylls were placed in 20 L sterile plastic containers filled with filtered (0.2 μm) and autoclaved seawater (Figure 2B). Sporulation started in all cases after 25 to 35 min and, after 1 h the sporophylls were removed and the water was filtered with a 100 μm mesh. Eight PVC cylinders covered with a 1.5 mm nylon rope were introduced into each of the 20 L containers to allow for spore settlement (Figure 2B). After 12 h, the eight PVC cylinders were removed and placed in a 30 L glass tank filled with autoclaved, filtered, and Provasoli enriched seawater (McLachlan, 1973; Figure 2B). Culture was carried out at a photon flux density of 30–40 μmol m^{-2} s^{-1}; a temperature of 9–10°C; a salinity of 30% and a pH of 7.8–7.9. Photoperiod was 16:8 (L:D) during the first week; 14:10 (L:D) during the second week; 12:12 (L:D) during the third week and 10:14 (L:D) thereafter (following a previously determined protocol; Buschmann unpublished results). After 44 days, 3 cm pieces of rope were randomly cut off and plant density (number of sporophytes fronds per cm of rope fragment) estimated under a stereomicroscope. Furthermore, the maximum lengths of the juvenile sporophytic fronds were determined using an ocular micrometer. The data were statistically analyzed by a one-way ANOVA after logarithmic transformation to ensure the normality and homocedasticity of the data. If significant differences were detected between treatments, a Tukey *a posteriori*-test (according to Steel and Torrie, 1985) was performed. As the data were obtained from independent tanks, seeded from independent plants and comparisons between times were not considered, no pseudoreplication exists (*sensu* Hurlbert, 1984).

After 60 days in the hatchery (September), seeded ropes were attached to a 3 m long horizontal supporting rope (18 mm diameter) in groups of three placed at 2 m depth (Figure 3) in Metri (Figure 1). Three seeded ropes were used for each one of the five original populations. Plant density and length of the different *Macrocystis pyrifera* populations were estimated, after one and two months in the field, by random sampling of 5 cm seeded rope sections under a stereomicroscope. All data were analyzed as above.

Pilot study

Parent sporophytes were collected in Curaco de Velez (Figure 1) and seeded on ropes following the same methods and culture conditions described above. The ropes were seeded in mid January and were brought to the Calbuco culture site (Figure 1) in March. The initial culture conditions of the sporophytes in the field were

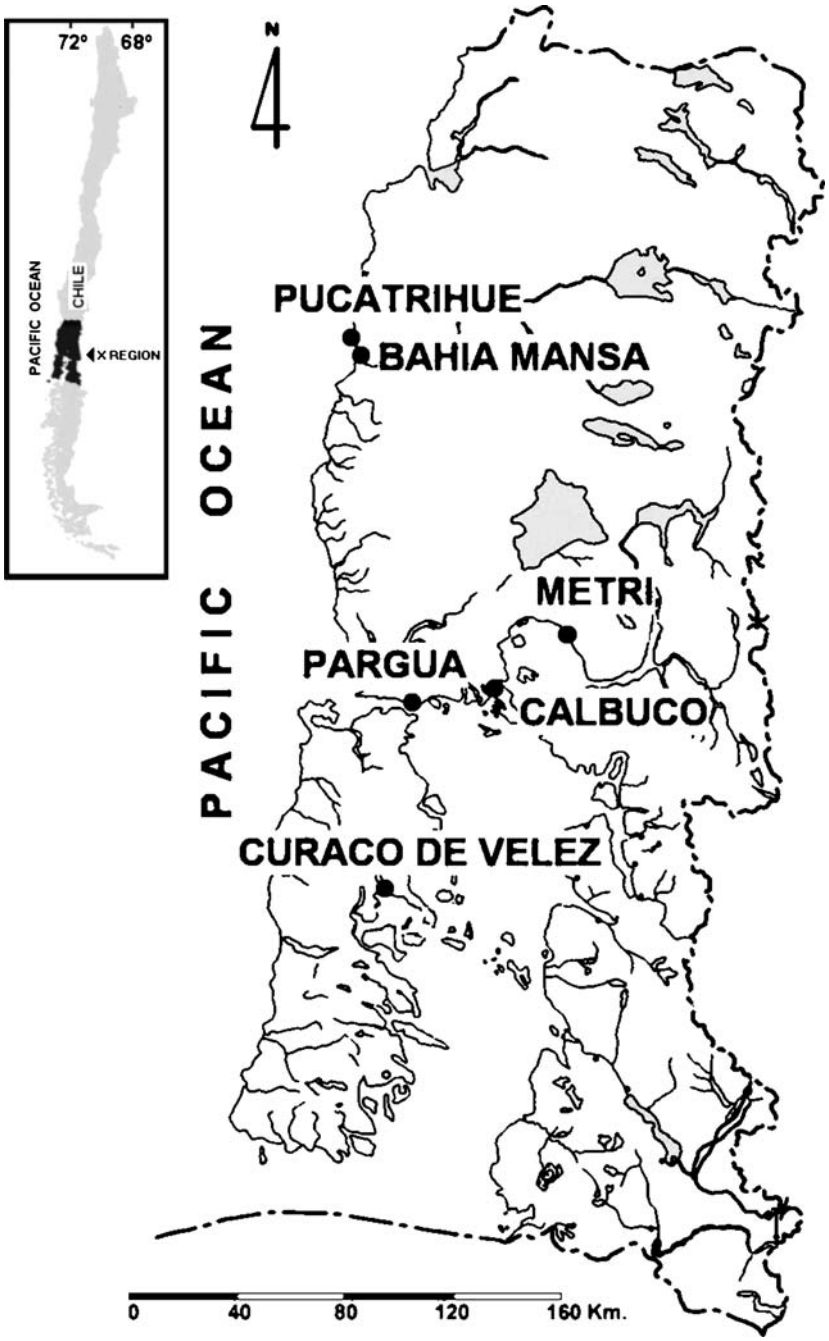

Figure 1. Map indicating the five collection sites of fertile tissues of *Macrocystis pyrifera* in southern Chile (Calbuco, Pargua, Metri, Pucatrihue and Bahía Mansa) and the experimental and pilot cultivation sites in Metri and Calbuco respectively.

1 mm plant length, and a mean density of 51 plants mm^{-1}. The experiment was initiated in March using the horizontal culture system at 1 m depth (Figure 3), based on previous results (Buschmann, unpublished data). The *Macrocystis pyrifera* fresh weight produced per m of long-line was evaluated by taking 10 random 1.5 m rope samples. Total fresh weight was determined and then the blades, stipes and pneumatocyst

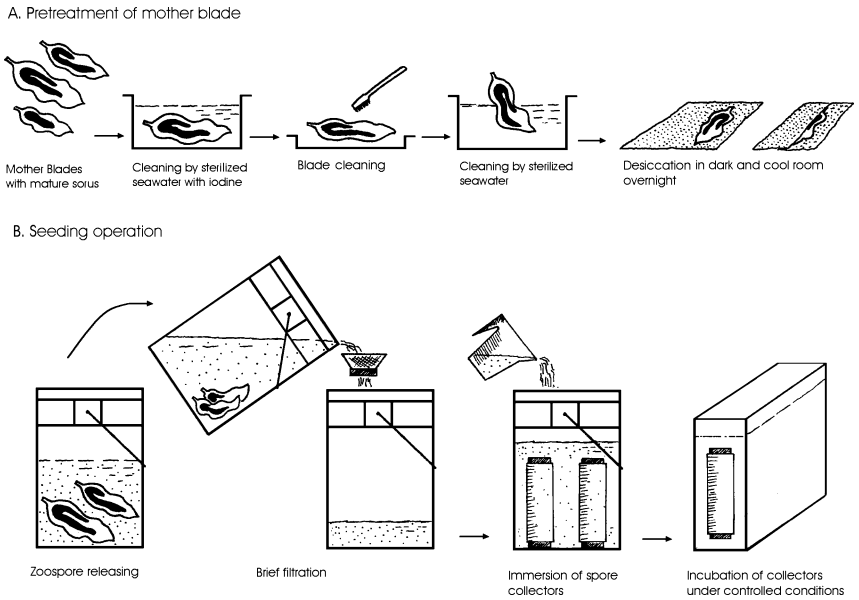

Figure 2. Scheme representing the pretreatment and sporulation induction procedure (A), and the seeding operation (B) for *Macrocystis pyrifera* mass production. The procedure was modified from Merrill and Gillingham (1991).

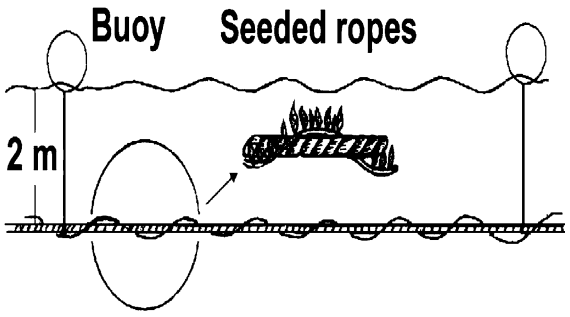

Figure 3. Suspended culture method used in this study: horizontal culture method adapted from Kawashima (1993) and Merrill and Gillingham (1991).

and disposable parts (holdfast, necrotic, perforated, and epiphyted tissues) of 22 plants obtained at random, were separated and weighed individually. Finally, "substantiality" (the weight per frond area; *sensu* Kawashima, 1993) of 30 randomly taken fronds was determined by weighing 1 cm^2 frond discs on a digital balance ($\pm$ 1 mg). Statistical analysis was performed following the above-described protocol.

Results

Cultivation of different populations

In the hatchery, the mean number of sporophytes produced at 9–10°C, 40 μmol photons m^{-2} s^{-1} with a variable photoperiod, varied between 1.0 and 12 individuals mm^{-1} of seeded rope (Figure 4A). The number of sporophytes produced was significantly higher ($p < 0.05$) from fertile sporophytic tissues, which were collected from Metri and Pucatrihue. Pargua and Bahía Mansa showed the lowest seeding success under the same culture conditions, whereas Metri and Bahía Mansa showed the highest size increment, with mean values of 2.85 mm in 44 days (4B).

Under field conditions, all the individuals produced from plants collected in Bahía Mansa and Pucatrihue (wave exposed coast) showed a mortality of 100% after only one month of cultivation (Figure 5). No significant size difference ($p > 0.1$) was found between plants originating from Calbuco, Pargua and Metri during the first month of culture. After two months in culture, no significant differences ($p > 0.08$) were found between the studied populations (Figure 5), although Calbuco plants showed a trend towards increased growth.

Pilot study

The pilot study carried out in Calbuco produced a biomass of 14.4 kg m^{-1} ($\pm$4.8 kg m^{-1}; S.D.) after 7 months of culture, and showed a substantiality value of the harvested fronds of 68.5 mg cm^{-2} ($\pm$1.6 mg cm^{-2}; S.D.). The different tissue types of *Macrocystis pyrifera* produced yields that varied from 1.3 kg m^{-2} (pneumatocyst) up to 4.1 kg m^{-2} (stipe) and 4.8 kg m^{-2}

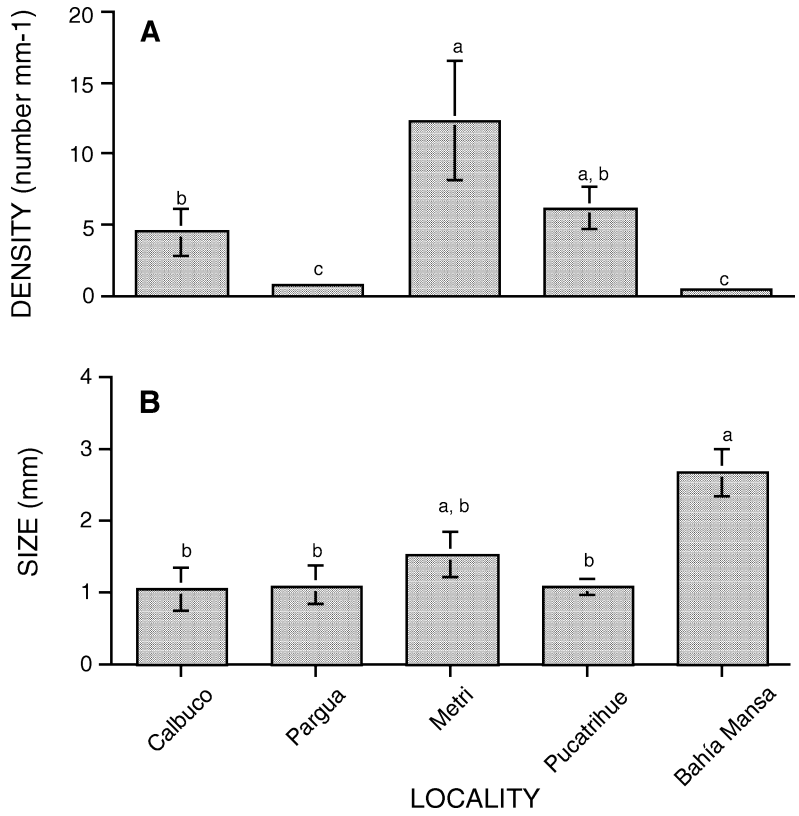

Figure 4. Cultivation of *Macrocystis pyrifera* under controlled conditions, initiated from fertile sporophytes collected in 5 sites of southern Chile: Calbuco, Pargua, Metri, Pucatrihue and Bahía Mansa. (A) Mean density (number mm^{-2}) of sporophytes on nylon ropes; and (C) mean length (mm) of sporophytes attached to nylon ropes after 44 days in the hatchery (9 replicates per location).

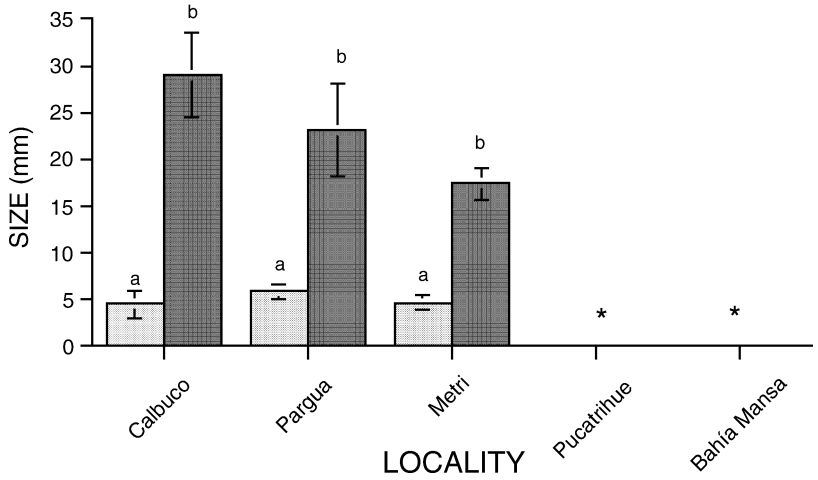

Figure 5. Cultivation of *Macrocystis pyrifera* at a depth of 2 m under field conditions, initiated from fertile sporophytes collected in 5 sites of southern Chile: Calbuco, Pargua, Metri, Pucatrihue and Bahía Mansa. Mean size (mm $\pm$ 1 SE) of sporophytes attached to nylon ropes after one month (dotted bars) and two months (grey bars). Letters above the bars indicate no significant differences after a Tukey *a posteriori*-test ($p < 0.05$). Asterisks indicate no survival of the germlings. Three samples were taken at each location.

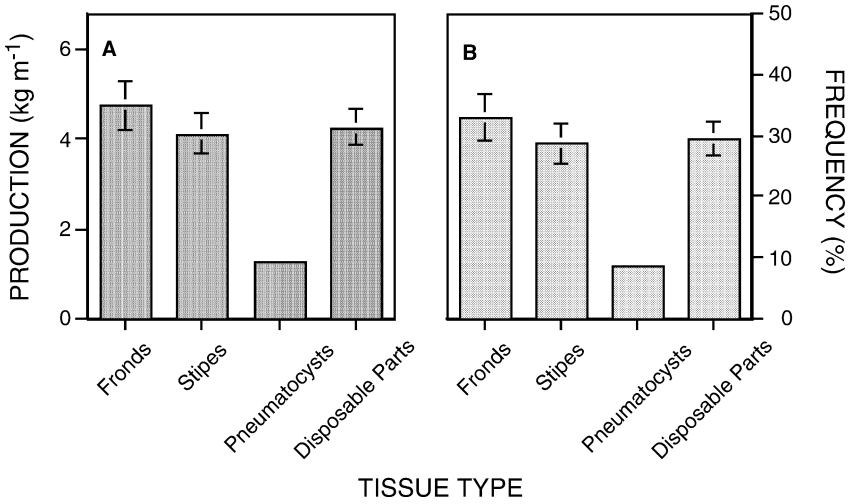

Figure 6. Mean (± 1 SE) biomass production (A) and percent yield (B) of different tissues and low quality tissues of *Macrocystis pyrifera* after seven months in a horizontal culture system installed in Calbuco. Twenty 1-m^2 samples were taken.

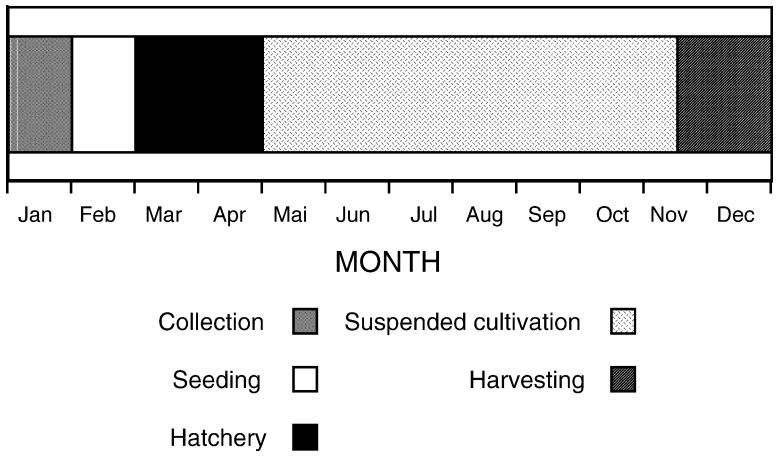

Figure 7. Production schedule of *Macrocystis pyrifera* in southern Chile.

(frond) (Figure 6A). Almost 30% of the total biomass was of lower quality and could not be used for food (Figure 6B), as it was covered with hydrozoan and bryozoan colonies, or necrotic tissues. Thus, *Macrocystis pyrifera* can be produced by collecting fertile tissues from the natural environment during January (summer), whereas rope seeding should be carried out at the latest during February (Figure 7). The hatchery phase takes at least 1.5 to 2 months, which means that the suspended culture at sea can be initiated in April or May (fall) and harvesting is possible in late November or December (Figure 7).

Discussion

This study demonstrates that the cultivation of *Macrocystis pyrifera* in southern Chile is technically feasible and that a productivity of over 14 kg m^{-1} during one production season can be obtained, which seems very promising (Figures 8A and B). The comparison of this value is not easy as most of the production estimates are based on extrapolations from physiological or growth measurements made on parts of kelp plants (Neushul & Harger, 1987). Estimations of 7 g ash free dry wt. m^{-2} day^{-1} (Neushul & Harger, 1987) and of

Table 1. Description of *Macrocystis pyrifera* food products developed.

Food product	Product specification
Fresh Salted Frond (Figure 8 D)	1 × 3 cm salted blade pieces are washed in filtered seawater and blanched at 100°C for 1 min and then mixed with salt (23%). The product has a green color and is placed in 15 or 10 kg plastic bags
Pneumatocyst Rings and Stipe Pieces (Figure 8C and E)	Slices of pneumatocysts and 2-cm pieces of stipes are washed and heated in vinegar with alcohol content not higher than 1% and an acetic acid value not lower than 5%. The products are packed in glass or packs as required. The product must be maintained in cool conditions (2–6°C)

Figure 8. (A) Pilot culture of *Macrocystis* in Calbuco, (B) harvesting *Macrocystis* and products developed for human consumption in Chile: (C) stipe fragments; (D) blade strips; (E) pneumatocyst rings and (F) a general presentation plate of these *M. pyrifera* products.

4 wet kg per production period per m^2 (Coon, 1982) exist, suggesting that our results are encouraging. It is important to mention that over 70% of the harvested biomass can be used to produce high quality food products (Figures 8C, D, E and F; Table 1), which represents a significantly higher success rate for the use proposed in this study when compared to other uses. In addition, the introduction of massive kelp culture in Chile may have other associated benefits such as providing a means of removing nitrogen and phosphorus produced by salmon farming (Buschmann et al., 2001a; Chopin et al., 2001; Troell et al., 2003). It is important to remember that this area of Chile produces over 300,000 tonnes of salmon, thus creating significant environmental impacts and conflicts (Buschmann, 2001).

To date, *Macrocystis pyrifera* has not been used to produce food products in Japan. Traditionally the species used as food in oriental countries are *Laminaria japonica* (Kombu) and *Undaria pinnatifida* (Wakame) (Kawashima, 1993; Ohno & Matsuoka, 1993). In Chile, the bull kelp *Durvillaea antarctica* has been traditionally used as a food source, but has a very low price. Because *Laminaria* and *Undaria* are not present in Chile, the idea was to develop alternative products that could be exported to oriental countries with a tradition in seaweed consumption (Abbott, 1996). However,

as the characteristics of color, texture, substantiality, and mucilage content of *Macrocystis* cannot be compared to those of *Laminaria* or *Undaria* we pursued the commercial strategy of creating novel products (Figure 8C to F), which received a positive market response in Asia after a first dispatch and market tour. Thus, because this is a developing market, it is believed that the commercial cultivation of *Macrocystis* is possible in Chile. In addition, alternative use of this kelp as abalone and sea urchin feed, or as organic fertilizer, strengthens its economic feasibility. Lower grade parts of *Macrocystis* can also be used for the other purposes indicated above.

Recently, it has been demonstrated that Chilean stocks of *M. pyrifera* show small genetic differences from other Southern Hemisphere species, but stronger differences from Californian stocks (Coyer et al., 2001). Despite this low genetic diversity, plants collected at various sites in southern Chile showed different potential for use in aquaculture practices. The plants from the most exposed areas cannot be used for farming in the inner seas, of southern Chile, although they presented higher growth under hatchery conditions. In contrast, marginal differences were observed between *M. pyrifera* populations from the inner seas. In contrast to an earlier report that identifies *M. laevis* in this region (Aguilar-Rosas et al., 2003) and indicates that some morphologically distinct plants exist, our six years of observations and experimentation strongly suggest that this smooth bladed plants correspond to *M. pyrifera*. Nevertheless, these results recognize that to some extent each farmer must consider site-specific characteristics of his own licensed location and the morphological characteristics of the parent sporophytes, before starting commercial activities.

Despite the promising production results obtained here, *Macrocystis* production in Chile still needs more research. Several factors remain to be studied, but two aspects are especially important, as has been demonstrated for other algal resources (Wikfors & Ohno, 2001). Firstly, strain selection cannot be overlooked. The manipulation of the growth capabilities of this resource is important, but given that it is required to produce food products, it is important to study aspects that can be used to produce plants with specific morphological characteristics. Unpublished results indicate that morphological variation and life-history variability of *M. pyrifera* in southern Chile is high (Buschmann et al., 2004), but we still do not know how to manipulate morphological variation or how to maintain certain morphological characteristics under culture conditions. Secondly, we wish to highlight management requirements during the field phase that will allow a successful commercial operation. During this phase aspects such as plant density, harvesting strategies, and disease control are extremely relevant, as can be seen for example, during the intensive cultivation of *Laminaria japonica* (Kawashima, 1993). All these aspects remain unknown and will undoubtedly provide excellent material for future studies in southern Chile.

Acknowledgments

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