CULTIVATION OF THE AUTOCTONOUS SEAWEED *Laminaria saccharina* OFF THE GALICIAN COAST (NW SPAIN): PRODUCTION AND FEATURES OF THE SPOROPHYTES FOR AN ANNUAL AND BIENNIAL HARVEST.

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**ABSTRACT**

The production and characteristics of the sporophytes for an annual and biennial harvest were studied in a cultivation of *Laminaria saccharina* ('sugar kombu') carried out off the Galician coast (NW Spain). For the indoor production of seedlings attached to string the European technique developed by the French Fisheries Institute (IFREMER) was used. This technique has been adapted by the Spanish Oceanographic Institute (IEO) for this species. The string with the *L. saccharina* seedlings was outplanted in cultivation ropes which were deployed horizontally ('longline') in the sea, in February 2002. Productivity was about 6 wet kg m⁻¹ culture rope for annual-harvest and 4 wet kg m⁻¹ for biennial-harvest, after 4 and 13 months respectively. The decrease in the harvestable biomass (about 35 %) came as a result of a striking reduction in the number of plants on culture rope between both crops, probably because of high mortality of sporophytes throughout the summer. The results on blade features of *L. saccharina* showed important differences between annual and biennial harvests which affected the "substantiality value" rate that expressed the quality of blade for food for human consumption used in Asia. Substantiality value of *L. saccharina* was 36 mg cm⁻² wet weight of blade for annual-harvest and 61 mg cm⁻² for biennial-harvest. Another aspect which needs to be highlighted was the high cost of the biennial harvest due to the necessary maintenance practice of this cultivation.

**INTRODUCTION**

The brown algal genus *Laminaria* (*Phaeophyceae, Laminariales*) encompasses several economically important species that are commonly known by the Japanese name of kombu. These marine seaweeds are used mainly as direct human food or as a source for the industrial extraction of the alginates, colloidal cell wall polysaccharides (McHugh, 2003). Recently, laminarian species have been considered a food source for mariculture of herbivorous animals of commercial
value such as abalone and urchin (e.g. Shepherd & Steinberg, 1992; Viana et al., 1993; Mai et al., 1994; Spirlet et al., 2001; Takami, et al., 2003; Daume & Ryan, 2004; Chang et al., 2005). In addition, Laminariales may be of interest for the feeding of bivalve molluscs (e.g. clam) as single cell detritus (SCD) (e.g. Pérez Camacho et al., 2002, 2004).

The worldwide production of Laminaria comes mainly from mariculture in Asia or from wild populations in Europe but about 90% of the total harvest is obtained through cultivation (Zemke-White & Ohno, 1999). The farming of Laminaria japonica Areschoug in 2000 produced about 916,011 tons dry weight worth $2.8 billion (Tacon, 2003).

Since the mid-20th century, several species of Laminaria have been collected from natural stocks off the European Atlantic coast for the extraction of alginates (Kain & Dawes, 1987; Briand, 1991). This yield of Laminaria spp. is approximately 50,000 tons dry weight and this production is important for the alginophyte industry, representing 24% of the total alginophyte resources (Mchugh, 1991).

Laminaria saccharina (Linnaeus) Lamouroux is the native European species that has more potential for future commercial cultivation due to its high added value, its high growth rate and the fact that its cultivation in the open sea is both biologically and financially feasible.

'Sugar kombu', common name for L. saccharina, is only found in Spain along the Asturian and Galician coast (Pérez-Ruzafa et al., 2003) and natural population resources are very rare.

At present, Laminaria saccharina is harvested and marketed for human consumption in Galicia. Moreover, abalone hatcheries (Haliotis spp.) will be soon developed and this seaweed can be used as fodder. On a mid-term basis, it is unlikely that the commercial abalone culture or edible seaweed industry can be maintained from natural seaweed populations so that it will become necessary to guarantee sustained quantity and quality of algal production with a view to ensure the success of these activities. Therefore, there will be a well-established need for seaweed production in Galicia in the near future. One way of meeting the demand could be the development of intensive culture of Laminaria saccharina in the open sea.

The life cycle of species belonging to the order Laminariales consists of an alternation between a microscopic filamentous gametophyte phase and a macroscopic laminar sporophyte phase (Fritsch, 1945). Laminaria saccharina is a perennial algae and the life span of the sporophyte is biennial; its longevity does not exceed 3 years (Parker, 1948).

Laminaria japonica is also a biennial plant that has been commercially cultivated in Asia on an annual or biennial life spans. Both of them are referred to as the one-year cultivation or the two-year cultivation methods, respectively. In the annual method the sporophyte is left to grow for only one winter, whereas in the biennial method this process takes two winters (Kawashima, 1984, 1993; Druehl, 1988).

The quality of Laminaria for human food is judged mainly by the thickness of the blade, which is given by the length of cultivation time in the sea. The two-year cultivation is the method which produces the highest quality Asian kombu. (Kawashima, 1984, 1993).

So far, there have been several trials on open-water Laminaria saccharina cultivation in New York, U.S.A (Brinkhuis et al., 1983), British Columbia, Canada (Druehl et al., 1988) and in the Isle of Man, United Kingdom (Kain et al., 1990). However, there are no comparative studies on productivity and features of L. saccharina for annual and biennial cultivation. In addition, no studies have been published on this subject.

In this paper we examined, on cultivation of Laminaria saccharina, the productivity and features of sporophytes for annual and biennial harvest off the Galician coast. We also discuss the industrial use of annual and biennial yield of L. saccharina. This study provides valuable information for future commercial cultivation of this species in Galicia.

MATERIALS AND METHODS

Laboratory culture

The laboratory cultivation involves the indoor production of artificial seedlings of Laminaria saccharina attached to string under carefully monitored environmental conditions. This stage was conducted by the Spanish Oceanographic Institute (IEO) in Santander, which has developed the technology to make seedlings of this species on a semi-industrial scale.

For the production of seedlings the technique developed by the French Fisheries Institute
Annual and biennial harvest for a cultivation of Laminaria saccharina off Galicia

(IFREMER) was used. This technique is known as the European method. The steps of this technique are described in detail by Pérez et al. (1984, 1991). In contrast to the Asian method, based on the direct sowing of spores on collectors (lines of string wound on a frame), the European method uses gametophytes in free-living. These gametophytes are kept as vegetative stock by active planktonic growth in the culture medium, which is known as free-living culture. When the seedlings are required, there is a suspension of gametophytes which are projected onto the collectors and these become attached to string. Subsequently, the collectors are placed in tanks under adequate environmental conditions until the growth of the seedlings (i.e. young sporophytes) on the string, the so-called 'seedling string'.

The stock gametophytes strain of Laminaria saccharina used in the present study comes from the same gametophyte bank of the IEO collection so that any heritable differences in morphology would be minimal. At the time of planting the seedlings which were obtained were 2 to 4 mm long.

Field culture

The field cultivation of Laminaria saccharina occurred in the waters of Ría de Ares y Betanzos (Galicia, Spain) into the northwest Atlantic Ocean. Figure 1 shows the culture ground site where it was carried out. This location is in a fixed coastal area specifically devoted to marine culture, mainly to intensive commercial cultivation of mussel.

The cultivation apparatus consists of horizontal ropes (i.e. 'longline' type) held in place at a given depth by a series of buoys, each of which is linked at its ends to a concrete block. The so-called horizontal rope is the culture rope with attached seedling string. The structure and design of the floating raft for cultivation used in the present study were similar to the ones described by Pérez-Cirera et al. (1997) for farming of Undaria pinnatifida (Harvey) Suringar on Galician coasts.

The young sporophytes of L. saccharina were planted in the open sea in February 2002. The method to outplant the seedling string on the cultivation ropes was as follows: the string was wound helicoidally around the rope, fixing it with a plastic tape at 25-cm intervals. The cultivation ropes were placed horizontally ('longline') at a 2-metre depth. The depth of the cultivation remained the same during the growth process, except in summer, when the cultivation ropes were placed at a 7-metre depth so that the L. saccharina could survive more easily under the higher temperatures of the summer season.

Measurement of productivity and features of sporophytes

After outplanting in February 2002, harvesting took place in May for the annual cultivation and in March of the following year for the biennial cultivation; that is to say, the cultivation of Laminaria

Figure 1.
Map showing the location of cultivation ground (·) in Ría de Ares y Betanzos (Galicia, N.W. Spain).
saccharina took about 4 and 13 months of culture in the sea respectively. At harvest time, 4 random samples of 50-cm culture rope were collected and transported to the laboratory for the measurement of yield and the characteristics of rope-cultured sporophytes.

The biomass production for annual and biennial cultivation of L. saccharina was calculated considering the 4 stretches of the culture rope collected and expressed as wet weight of plants per metre of cultivation rope. The total number of plants grown on the unit of rope was also measured.

The characteristics of L. saccharina for annual and biennial harvest were evaluated by measuring the 5 longest sporophytes in each of the 4 samples: plant wet weight and length, blade length, greatest blade width and blade thickness 25 cm above the blade-stipe junction. To determine the quality of blade for human consumption (the so-called 'substantiality value') the following equation by Kawashima (1984, 1993) was used: [blade wet weight (mg)] × [blade length (cm)] × blade greatest width]-1. In Asia, this equation is applied to commercial cultivation of Laminaria japonica.

### Statistical analysis

The differences in production and features of sporophytes between an annual and biennial harvest were analyzed using analysis of variance (one way ANOVA). The assumptions of normality and homocedasticity for the ANOVA were contrasted with the Kolmogorov-Smirnov test for normality, and the Levene for the homogeneity of variances. In some instances, there was no homogeneity of variances but the ANOVA was nonetheless applied because its Fisher test was robust to departures of variance from homogeneity when sample sizes are balanced. (Zar, 1996; Underwood, 1997). The ANOVA was performed with the SPSS® v. 11 statistical package for Microsoft Windows.
RESULTS

The cultivation of Laminaria saccharina in the waters of Galicia (N.W. Spain) was outplanted in February 2002 and harvested after about 4 and 13 months growing in the sea. The harvestable biomass was about 6 kg per metre of rope in May of the same year for the one-year cultivation whereas for the two-year cultivation about 4 kg were obtained in March of the following year (table 1). These production values differ significantly, but only slightly, according to $a=0.05$ (table 3: ANOVA; $p = 0.045$). This decrease in biomass production in the biennial harvest as compared to the annual harvest is in line with the strong reduction in the number of plants on the cultivation rope between both harvests (table 1), which are statistically different (table 3: ANOVA; $p = 0.000$). However, biennially-harvested plants showed greater length and weight than annually-harvested plants (table 1) and these differences are significant (table 3: ANOVA; $p = 0.000$).

The blade of plants from both harvests was morphologically different. Thus, the blade from the biennial cultivation had greater wet weight, as well as greater width and thickness than the blade from the annual cultivation (table 2). For these characteristics there are significant differences between the plants from the annual and biennial harvests (table 3: ANOVA; all with $p = 0.001$).

Using blade weight, length and width, the sporophytes of the biennial harvest turned out to have a higher quality value (i.e. 'substantiality value') than the sporophytes from the annual harvest (table 2). More specifically, the average 'substantiality value' obtained for the two-year cultivation was about twice as much as that for the one-year cultivation (table 2). This variation was very significant (table 3: ANOVA; $p = 0.000$).

Another aspect worth mentioning was that, unlike in the annual-cultivation, in the biennial-cultivation there were many epiphytic organisms on the culture.

<table>
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Significantly differ shown in bold and different letters (super indices) indicate the significant level (á) where a is 0.01 and b is 0.05.

Lack of normality or homogeneity of variances at á = 0.05 shown with asterisk (*).
rope and the structure of the floating raft. The main epiphytic found was the mussel and its presence made it difficult to maintain the cultivation ropes at a given depth. In addition, for the biennial cultivation, the strong reduction of density on the culture ropes allowed for the establishment of other algal, namely the opportunistic seaweed Saccorhiza polyschides (Lightfoot) Batters.

**DISCUSSION**

The average productivity of Laminaria saccharina cultivation started in Galician waters in February was about 6 wet kg m\(^{-1}\) rope for the annual harvest which took place in May the same year and about 4 wet kg m\(^{-1}\) rope for the biennial harvest, after about 4 and 13 months of cultivation respectively. The difference in biomass production was slightly significant, but a 35% of the production obtained in the biennial harvest was lost on average when compared to the annual harvest.

The decrease in the production between the annual and biennial harvests came as a result of the strong reduction in the number of plants on a rope-length basis, probably because of high mortality of sporophytes throughout the summer season. In this respect, Brinkhuis et al. (1983; 1984) and Lee & Brinkhuis (1986) reported similar observations for L. saccharina at its southern limit of distribution in the northwestern Atlantic Ocean. Our study takes place in the north west of the Iberian Peninsula, where L. saccharina has its southern limit of distribution in the northeastern Atlantic Ocean (Yarish, 1988; Lüning, 1990). For that reason, it is very likely that most of the L. saccharina sporophytes found in the Galician waters will disappear during the summer, unlike what happens within its finest distribution range where no noticeable mortality occurs during the summer (Parker, 1948; Kain 1979).

Reliable production comparisons are difficult to make, but the average values of harvested biomass in this study are generally comparable to those observed for several experimental cultivations of Laminaria saccharina. Thus, in British Columbia, Canada, Druehl et al. (1988) reported a harvest of 8 wet kg m\(^{-1}\) rope, after 8 months of culture in the sea. The yield expressed per metre of rope has not been defined for the cultivation trials carried out in New York, U.S.A (Brinkhuis et al., 1983; 1984; 1987) and in the Isle of Man, United Kingdom (Holt & Kain, 1983; Kain & Dawes, 1987; Kain et al., 1990; Kain, 1991), but it can be estimated that crops over 5 kg m\(^{-1}\) would have been achieved within 6-8 months. In addition, biomass values that we have obtained in the current study are quite similar to the average value of 8 kg m\(^{-1}\) which occurs in commercial cultivation of Laminaria japonica by means of the forced-cultivation technique (i.e. biennial life span compressed into one year) (Mairh et al., 1991). Nevertheless, as regards harvestable biomass in cultivation of L. saccharina in Galicia, recent experiments with outplanting in December (in this paper the experiment took place in February) have shown an average yield of 11 wet kg m\(^{-1}\) rope within 4 months. (Peteiro et al., not yet published).

The results about the characteristics of Laminaria saccharina show important differences between the plants from the annual and the biennial harvests, which affects the blade quality value for human consumption. In order to assess blade quality, the so-called 'substantiality value', the equation applied to L. japonica ('kombu') in Asia was used, and this is expressed as wet weight per area of blade. The 'substantiality value' for the blade of L. saccharina was 36 mg cm\(^{-2}\) wet weight for the annual cultivation and 61 mg cm\(^{-2}\) for the biennial cultivation, after about 4 and 13 months of culture in the sea respectively. These 'substantiality values' show that the blade from the annual harvest is only half as good as that from the biennial harvest. Therefore, the quality of L. saccharina blade was determined by the time of culture in sea and it directly correlates to the thickness of its blade. Parker (1948) described that the thickness of L. saccharina blade increases mainly during the summer months, when there is a period of slow growth in length. This explains why biennial plants which grow during the summer had greater blade thickness, in contrast to annual plants. Up to now, there are no data about L. saccharina 'substantiality value', but biennially-harvested plants had average values of about 75-80 mg cm\(^{-2}\), similar to those reported for the cultivation of L. japonica by Mairh et al. (1991). However, the 'substantiality value' of 'kombu' can exceed 100 mg cm\(^{-2}\) (Kawashima, 1984, 1993).

Another aspect worth mentioning in the biennial-cultivation was the presence of a large number of mussels attached to culture ropes and the structure of the floating raft. Probably, this was favoured by the fact that our cultivation was carried out in a coastal
area where there is an intensive commercial cultivation of this species. The mussel caused mainly an increase in the weight that the buoys had to bear, so it became necessary to remove them manually. This maintenance practice of farming entails an expense which is not desirable from a financial viewpoint. A way to overcome this problem was the actual annual-cultivation, since a shorter cultivation time in the sea prevented any mussels from growing. On the other hand, in the course of the biennial-cultivation, the ropes of culture were also susceptible to the growth of other algae because of the reduction of plant-density during the summer. The opportunistic seaweed Saccorhiza polyschides was the fastest-growing algae in the biennial-cultivation ropes owing to its ability to quickly invade and colonise new areas. The occurrence of this alga, which can grow up to 4 metres, can interfere with the growth of Laminaria saccharina by competing for light and nutrients.

In conclusion, the results of this study point out that the moment in which the harvest of the annual or biannual cultivation takes place is important to the actual cultivation of Laminaria saccharina in Galician waters, as it affects the quantity and quality of the produce ('sugar kombu'). Thus, in a biennial harvest, a production loss (of about 35%) was reported with regard to that of the annual harvest. Nevertheless, its blade quality was much better (almost twice as good) with 'substantiality values' similar to those reported for the cultivation of Asian 'kombu'. At any rate, the biennial method is more costly than the annual method due to the maintenance practices required by this sort of cultivation, bringing about an increase in the production costs.

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REFERENCES


Kain (Jones) JM and Dawes CP (1987). Useful European seaweeds:


