



Cultivation of *Gigartina skottsbergii* (Gigartinales, Rhodophyta): Recent advances and challenges for the future**

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Abstract

This study integrates landings statistics and biological studies of the red alga *Gigartina skottsbergii* Setchell et Gardner. The analysis of the landings and carrageenan production in Chile suggests that this resource will suffer a strong harvesting pressure during the next years. Biological results on sporulation, germination, sporeling growth and survivorship in laboratory, indoor tanks and field conditions, indicate that cultivation of this species is technically feasible, as spores can be seeded on ropes and other substrata. Vegetative propagation of this species through tissue fragmentation is also possible. Vegetative fragments of this carrageenophyte have 20 to 30% higher growth rates than whole fronds in suspended culture systems. Protoplast production can be also explored for bypassing restrictions in spore availability. Major advantages that encourage the cultivation of *G. skottsbergii* include its gel quantity and quality, its pathogen-free condition, a high reproduction potential and its regeneration capacity. On the other hand, the major constraints are related to its relatively slow growth as compared to other carrageenophytes, limited availability of spores and high mortality during juvenile stages.

Introduction

In Chile, the demand for carrageenan-producing algae has increased steadily since 1996. *Gigartina skottsbergii* Setchell et Gardner is the most valuable carrageenophyte, with reported production of 32,438 t wet wt for 1996 (Buschmann et al., 1999a). This species shows a seasonal variation in biomass, with a maximum during summer on Atlantic and Pacific coasts

of southern South America (Piriz, 1996; Zamorano & Westermeier, 1996; Westermeier et al., 1999). Besides taxonomic (Kim, 1976), chemical studies (e.g. Piriz & Cerezo 1991), phenological and standing stock assessments (Zamorano & Westermeier, 1996; Piriz, 1996; Westermeier et al., 1999) and regeneration studies (Correa et al., 1999), there is a lack of information on the ecology, physiology, reproductive biology, among other aspects, that precludes the development of cultivation techniques. In this context it is important to consider that in the northern distribution limit of the Chilean coast, the available information on landings

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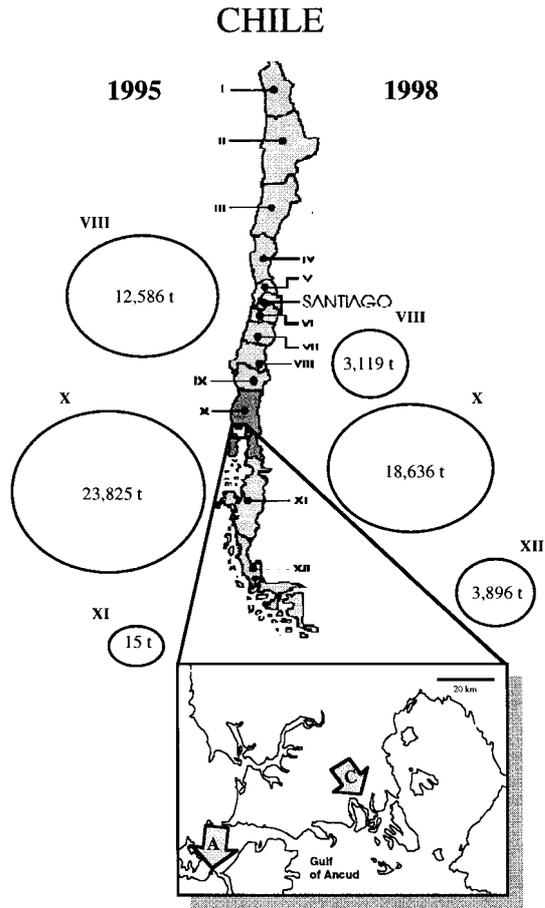


Figure 1. Harvesting sites (A = Ancud and C = Calbuco) for *Gigartina skottsbergii* in southern Chile and the administrative regions (Roman numbers). Biomass in the circles represent total wet weight harvested in the different regions in 1995 and 1998, respectively.

and stock assessments suggests that this species is being overexploited in some areas stressing the need for developing cultivation methods (Buschmann et al., 1999a; Buschmann et al., 2001).

This contribution analyzes the history of landings of this carrageenan-producing alga in Chile and includes recently obtained results on the biology of this resource to identify the technical and biological constraints that will have to be solved before farming becomes a reality. We concentrated on aspects related to sporulation, germination, germling growth and survivorship in laboratory, indoor tank and field cultivation and explored the potential use of alternative propagation methods. Finally, we critically assess the positive and negative aspects of an eventual farming of cultivating *Gigartina skottsbergii*.

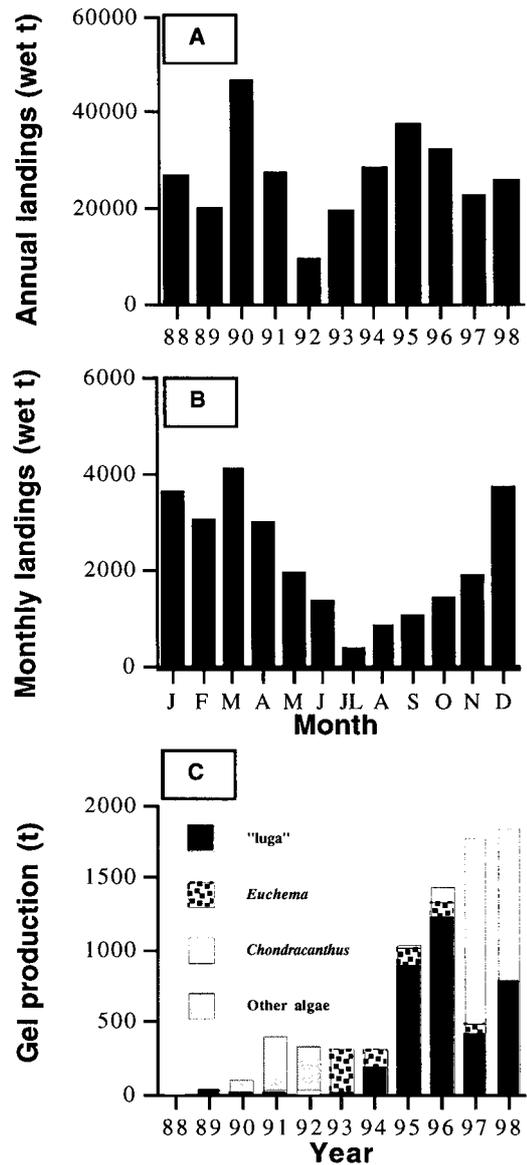


Figure 2. Carrageenophytes and carrageenan production in Chile. A: total annual landings of 'lugas' (common name given by fishermen to *Sarcothalia crispata* + *Mazzaella laminarioides* + *Gigartina skottsbergii*). B: monthly variations in landings during 1998. C: carrageenan production from different species.

Materials and methods

Seaweed landings, study sites and carrageenan content

Data on total and monthly landings during past years, as well as carrageenan production, were obtained

from the annual reports of the Chilean Fisheries Service (Anonymous, 1999). All *Gigartina skottsbergii* samples for the experiments were obtained from Ancud and Calbuco, southern Chile (Figure 1), where population studies have been conducted since 1996 (Zamorano & Westermeier, 1996; Westermeier et al., 1999). Commercial information on carrageenan was obtained by interviewing businessmen associated with its production in Chile. Additionally, during 1998, carrageenan content was determined in samples collected from Ancud ($n = 6$) and Calbuco ($n = 6$), following the methodology of Klein et al. (1984). Viscosity and gel strength of the carrageenan were determined following the protocols utilized by the Extractos Naturales Gelymar S.A. company (Zamorano, pers. comm.).

Indoor cultures

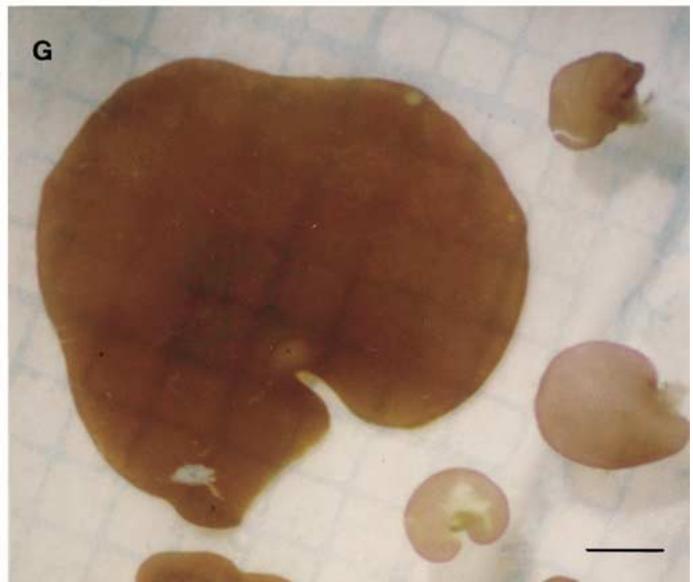
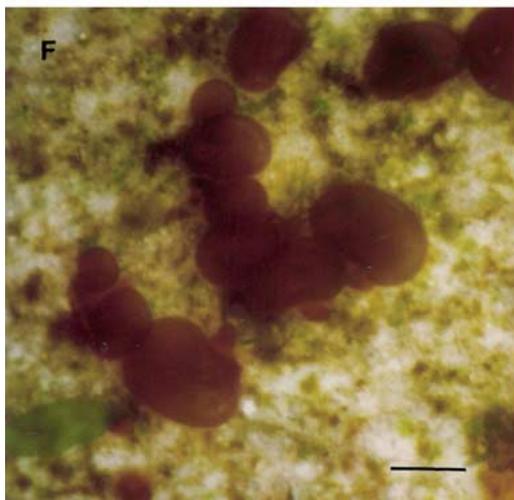
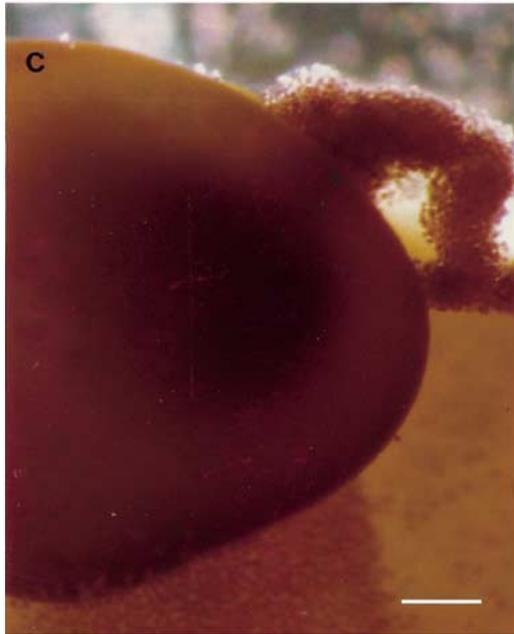
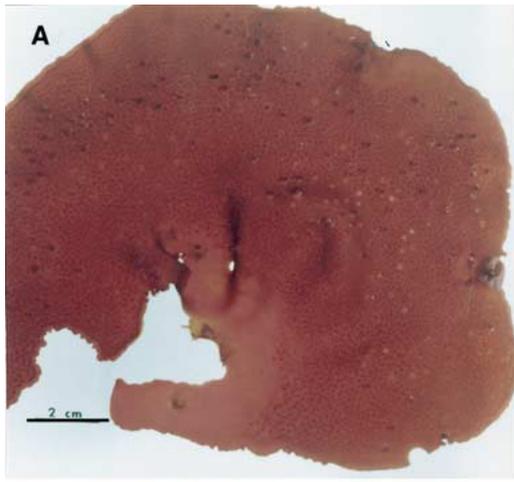
Reproductive fronds were collected by SCUBA diving and transferred on ice to the laboratory at the Pontificia Universidad Católica de Chile in Santiago within 12 to 24 h of collection, in order to initiate spore cultures and inoculations of artificial substrata. Wild fronds were induced to sporulate by air drying at room temperature and the released spores were used in laboratory experiments. Aliquots of ca. $360,000$ spores mL^{-1} were seeded on 20-cm^2 ceramic plates and grown indoor in 3-L glass tanks. Twenty plates were sent to the Metri Marine Station of Universidad de Los Lagos. The seeded ceramic plates were set up under two light conditions (5 and $10 \mu\text{mol m}^{-2} \text{s}^{-1}$) in 50-L tanks with $1.2 \mu\text{m}$ filtered seawater. During this experiment, water temperature fluctuated between 12 and 15°C , salinity ranged from $30\text{--}32\text{‰}$, pH from $8\text{--}8.2$, and the photoperiod was maintained at 12:12 (L:D). Survival and growth of the juvenile stages was photographically recorded using a stereomicroscope. The number of individual discs in the photographs were counted and each disc mapped on an acrylic sheet and numbered. Changes in the area of 50 randomly selected juveniles were recorded monthly, and changes in density (number of individual discs per cm^{-2}) and specific growth rates [$\text{SGR} = (\ln \text{final area} - \ln \text{initial area}) / \text{time (days)} \cdot 100$] were determined. The percent cover of fouling algae was also recorded photographically and quantified by using 100-random-dots acrylic quadrants. Correlation analyses were done between fouling algae and *Gigartina skottsbergii* sporeling density and changes in frond area of the juveniles.

Open sea cultivation

Replicates of the seeded ceramic plates incubated in the indoor tanks were taken to Ancud and Calbuco, fastened to concrete blocks and installed at $9\text{--}12$ m depth, within the native populations of *Gigartina skottsbergii*. Measurements of germling density and size were recorded monthly since October 1998. Comparisons between growth responses of field- and laboratory-raised juveniles were done using 250 haphazardly selected individuals exposed to their respective environments from October to November.

Alternative propagation methods

Propagation results considered in the present study are based on the regeneration capacity of vegetative frond fragments and rhizoids under laboratory conditions (Correa et al., 1999). In this study, the potential of regenerated tissues to grow was also assessed by installing fronds and rhizoid fragments in the field. The responses of these fragments in the field were photographically monitored. Cultivation in the field was done by fastening 20 tagged fronds to ropes. The surface of each frond was drawn on an acrylic plate at the beginning of the experiment and during each of the monthly controls done by scuba-divers. The feasibility of producing protoplasts was evaluated using female gametophytes of *Gigartina skottsbergii* collected in Ancud, following the basic protocol described by Le Gall et al. (1990). Algae were carefully cleaned and pre-incubated in autoclaved seawater containing 0.1% penicillin (5000 U) and streptomycin (5 mg L^{-1}) for $20\text{--}30$ min. A gram of material was chopped into $0.1\text{--}0.3$ mm pieces and incubated for 30 min in the presence of 450 mM NaCl, 120 mM MgCl_2 , 100 mM Tris (pH 7.2) and 0.2 mM PMSF (phenyl methyl sulfonyl fluoride). The incubation medium was then discarded and the remaining material was incubated in a giratory shaker ($60\text{--}70$ rpm). The enzymatic medium used for tissue digestion consisted of 1% (w/v) cellulase CELF (6500 U) and a mixture of kappa- and lambda-carrageenases (200 U/ 180 U) from *Cytophaga drobechiensis* (Potin et al., 1991), which was dissolved in 10 mL of 0.5 M NaCl, 40 mM MgCl_2 , 5 mM KCl, 50 mM Tris (pH 7.2), 0.2 mM PSMF and 0.35 m sorbitol. The concentration of kappa- and lambda-carrageenases was adjusted to match the higher polysaccharide content of *G. skottsbergii*. Undigested tissue debris was removed by filtration with a $20 \mu\text{m}$ nylon mesh and protoplast yield was recorded at various incubation times with



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Figure 3. Various developmental stages of *Gigartina skottsbergii*. A: wild tetrasporic frond, bar = 2 cm. B: wild cystocarpic frond, bar = 2 cm. C: sporulating cystocarp with a thread-like mass of released spores, bar = 500 μm . D: germinating tetraspores seeded on artificial substrata, bar = 18 μm . E: juvenile, umbrella-like gametophytes in culture, bar = 3 mm. F: juvenile, laboratory-borne gametophytes on artificial plates, cultivated in the open sea; bar = 500 μm . G: gametophytes harvested after four months in the field, bar = 1 mm.

a Newbauer haemocytometer. Cell viability was assessed by autofluorescence, while the removal of the cell wall was confirmed by staining with 0.01% w/v calcofluor white (Maeda & Ishida, 1967; Zablackis et al., 1993).

Results and discussion

Analysis of seaweed landings

Commercial exploitation of *Gracilaria chilensis* in the 1970s (Santelices, 1996; Alveal, 1998) was accompanied by limited exploitation of other red algae. However, during the last few years seaweed exploitation in Chile has entered a diversification phase (Buschmann et al., 2001). In recent years, and partially as the result of the establishment of carrageenan processing plants such as Gelymar S.A. and Danisco Ingredients in the southern part of the country, exploitation has expanded rapidly, with *Sarcothalia crispata*, *Mazzaella laminarioides* and specially *Gigartina skottsbergii*, becoming the most valuable target species. The main harvesting activity of Chilean carrageenophytes occurred during 1995 and affected Region VIII and X (Figure 1). Operations are now consistently moving further south, looking for new unexploited stands, specially those of *G. skottsbergii* (Figure 1). Current exploitation of this algal resource has reached Punta Arenas, in Region XII (Figure 1) and stock assessments have already been done by private companies even in the Falkland Islands (Westermeier, unpublished data).

Even though carrageenophyte landings have fluctuated during the last decade, total landings reached ca. 30,000 t in 1998 (Figure 2A). Harvesting of these resources is a seasonal activity with a maximum recorded in spring and summer (Figure 2B). As *Gigartina skottsbergii* is the most commercially valuable carrageenophyte, it is expected that harvesting pressure will increase in the near future. Indeed, carrageenan production has increased steadily during the past years (Figure 2C), in spite of the economic crisis in east Asia. To cope with this demand, Chile is additionally importing *Eucheuma* spp. (Figure 2C).

Table 1. Range (n = 6) of carrageenan yield, viscosity and gel strength in extracts of *Gigartina skottsbergii* in winter (A) and summer (B)

	Yield (%)	Viscosity (cPs)	Strength (g cm ⁻²)
A. Autumn – Winter			
Gametophytic fronds	45–46	62–378	104–126
Tetrasporic fronds	38–59	1000–1360	No Data
B. Spring – Summer			
Gametophytic fronds	72–75	15–30	18–26
Tetrasporic fronds	53–60	90–312	No Data

Carrageenan content

Cystocarpic fronds of *Gigartina skottsbergii* had consistently higher carrageenan yield than tetrasporic fronds, which is in agreement with information available for plants from Argentina (Piriz & Cerezo, 1991). Cystocarpic fronds produce carrageenans with χ/ι and μ/ν -characteristics and the tetrasporic fronds produce only λ -carrageenans (Piriz & Cerezo, 1991), in agreement with reports for other members of the Gigartinales (Craigie, 1990). Because of the 20–25% higher carrageenan yield, and the presence of kappa-2 carrageenan, there is a higher demand for *G. skottsbergii* over other local species. Gametophytic populations of this species showed carrageenan yields that varied between 45–46% in autumn-winter to 72–75% in spring-summer (Table 1). In contrast, carrageenan yields from tetrasporic fronds were lower in spring (60%) and summer (53%; Table 1). Carrageenan yields of both cystocarpic and tetrasporic fronds were consistently higher than for populations from Argentina (Piriz & Cerezo, 1991). Viscosity of carrageenan in tetrasporophytes was higher in winter than summer and gel strength from gametophytes was higher during winter. Carrageenans from tetrasporic fronds did not show gelling capacity (Table 1). Furthermore, the chemical characteristics of the *G. skottsbergii* gels did not show seasonal variations related to environmental conditions (Piriz & Cerezo, 1991), contrasting to what has been shown in other species of *Gigartina* (McCandless & Craigie, 1974; Pickmere et al.,

1975; Mathieson & Tveter, 1976). The effect of the culture conditions, specially nutrients, influence the carrageenan yield and quality (Zinoun et al., 1993; Chopin et al., 1999), a phenomenon that remains to be demonstrated in *G. skottsbergii*.

Indoor cultures

Flat thalli, like those of *Gigartina skottsbergii*, are generally cultivated in multi-step culture systems, manipulating the microscopic stages in hatcheries and transferring the juveniles to the open sea (Santelices, 1999). Our research has concentrated on elucidating the factors that control spore performance, including the processes of release, settlement and germination (Buschmann et al., 1999b). *G. skottsbergii* shows a 'Polysiphonia-type' life history with tetrasporangia embedded in the frond (Figure 3A) and cystocarps inside papillae (Figure 3B). The two types of reproductive structures are scattered over the whole frond (Avila et al., 1999a). Spores are usually discharged massively (Figure 3C), to then settle and germinate by forming a septum perpendicular to the substratum (Figure 3D) and described as 'Dumontia-type' germination (Guiry, 1990). The germling develops into an 'umbrella-like' plantlet (Figure 3E). In general, temperatures from 10 to 13 °C and dim light conditions are required for adequate settlement and germination, while nutrients are needed only during later stages of development (Correa et al., unpubl.). These results differ from those reported for Antarctic populations of *G. skottsbergii* by Bischoff-Bässman & Wienecke (1996), who found higher growth and survival responses at 5 °C or lower. It seems that genetic differences among the various populations have developed as a consequence of different selection pressure existing in the regions located at the distributional limits of *G. skottsbergii* (Martínez et al., unpubl.).

Spore densities for the initial inoculation yielded an average germling density of 1013 discs cm^{-2} on the plates at the end of the laboratory stage. Although the density dropped drastically in the 1000-L tanks, more than 100 juveniles cm^{-2} (Figure 4A) survived after three months of culture. The two photon flux densities tested did not significantly affect germling survivorship ($F = 3.522$, $p = 0.077$) and resulted in an average growth rate of 2% day^{-1} (Figure 4B). Frond surface of the germlings reached 0.15 mm^2 , with no significant differences ($F = 0.458$, $p = 0.499$) induced by the two photon fluxes (Figure 4C).

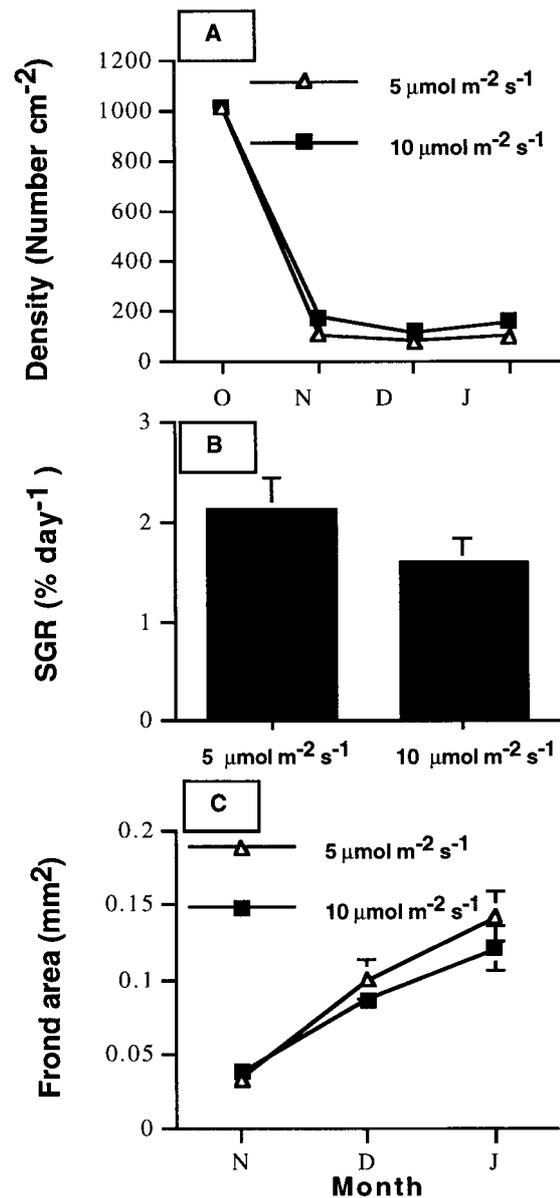


Figure 4. *Gigartina skottsbergii* cultivated in nursery tanks. A: density evolution. B: specific growth rate (S.G.R.) displayed by sporelings. C: frond size of juveniles.

At the beginning of the tank experiments, a low cover of filamentous algae, mainly Ectocarpaceae, fouled the ceramic plates. During the next two months, fouling algae increased up to 80% cover, showing significantly ($F = 4.659$, $p = 0.035$) higher fouling levels at the higher photon flux treatment (Figure 5A). Numerous studies have demonstrated that fouling organism can produce adverse effects on growth and sur-

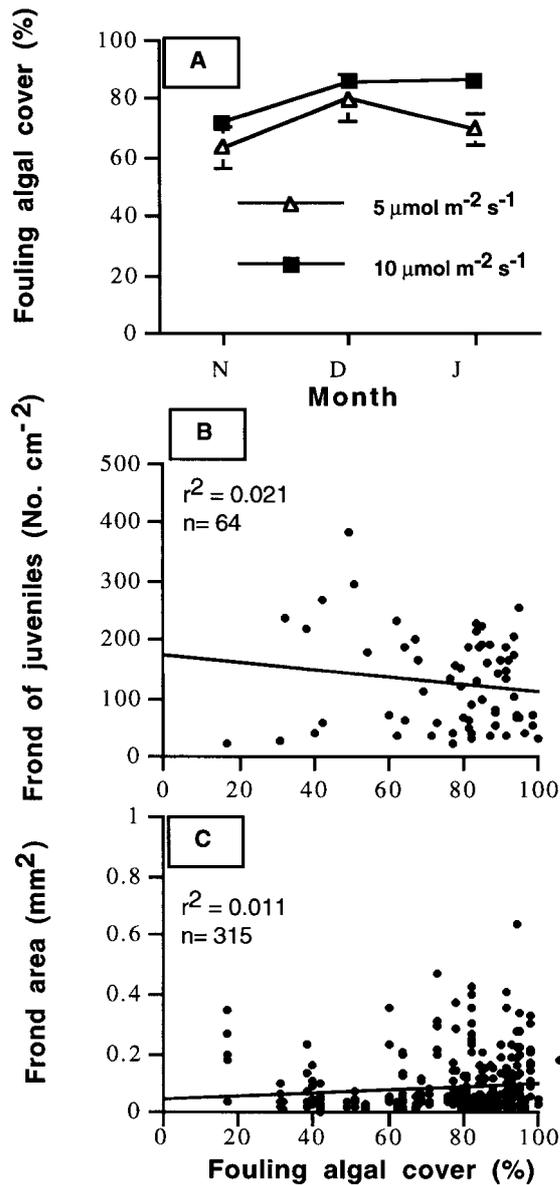


Figure 5. Effect of fouling algae density and size of *Gigartina skottsbergii* juveniles in tank cultures. A: cover of fouling algae on ceramic plates. B: correlation between cover of algal fouling and density of *G. skottsbergii* germlings. C: correlation between cover of algal fouling and size of *G. skottsbergii* germlings.

vivorship of farmed and wild algae (Fletcher, 1995). Nevertheless, we observed no significant effect of fouling on the density and size of *Gigartina skottsbergii* (Figure 5B, C), and some control of fouling algae in tank cultures could be achieved by reducing photon flux density below $10 \mu\text{mol m}^{-2} \text{s}^{-1}$.

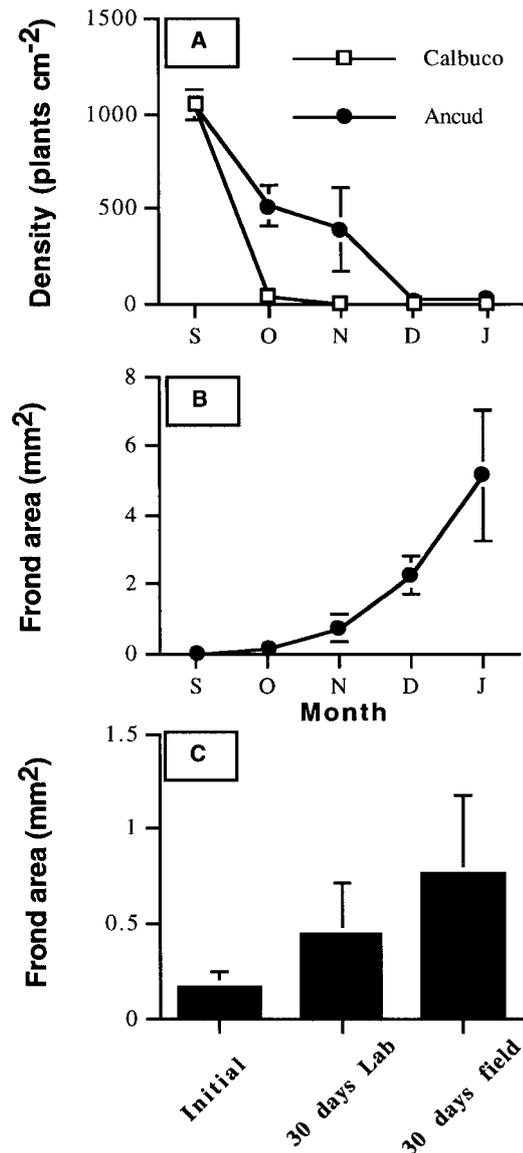


Figure 6. Growth of *Gigartina skottsbergii* germlings seeded on ceramic plates and transplanted to the field. A: density in Calbuco and Ancud. B: frond size increments in Ancud. C: size comparison of 30-d old germlings from the laboratory and from the field.

Open sea cultivation

Following an initial sporeling density of ca. $1054 \text{ discs cm}^{-2}$, the number of plants under field conditions decreased sharply between September and October at Calbuco and to December at Ancud (Figure 6A). All juveniles were lost by November at Calbuco, while the January densities remained at 1 plant per cm^2 at

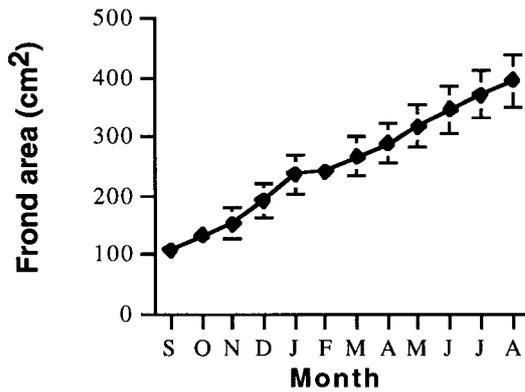


Figure 7. Growth of whole juvenile fronds of *Gigartina skottsbergii* fastened to ropes in the open sea, expressed as mean (± 1 SE) values of frond area increase.

Ancud. The size of the surviving fronds at Calbuco increased from 0.16 mm² to 5 mm² in 4 months (Figure 6B), and growth was consistently higher in the field than in the laboratory (Figure 6C).

Grazing can be of great importance for explaining subtidal abundances of macroalgae (e.g. Schiel & Foster, 1986). This phenomenon appears to operate at quite different intensities in Calbuco (high herbivory pressure) and in Ancud (low herbivory pressure) and thus explains contrasting differences in abundance and recruitment of *Gigartina skottsbergii* in the two populations (Westermeier et al., 1999). In Calbuco, the sea urchin *Pseudoechinus magallanicus* may reach densities over 100 individuals m⁻², whereas in Ancud urchins are absent and gastropod herbivores occur at much lower densities, with presumably low effects on the abundance of understory species (Moreno & Sutherland, 1982). Canopy also modulates the abundances of subtidal algae (e.g. Dayton et al., 1984; Santelices & Ojeda, 1984; Kennelly, 1989). In natural stands of *G. skottsbergii*, however, canopy-forming seaweeds are absent, making unsustainable the hypothesis that the Calbuco – Ancud differences in abundance and recruitment is due to that factor. Water movement, on the other hand, has different intensities in Ancud and Calbuco (Westermeier et al., 1999), although its ecological effect on *G. skottsbergii* distribution and abundance remains to be elucidated.

Juveniles of *Gigartina skottsbergii* were successfully grown when fastened to ropes in the open sea. In the field, these juveniles increased from 100 to 390 cm² in 8 months (Figure 7), with a monthly mean growth rate of 10.3%. These values are relatively low as compared to other temperate subtidal

carrageenophytes present in the same study area such as *Sarcothalia crispata* (1.2 to 1.4% d⁻¹; Avila et al., 1999b), *Chondrus crispus* on the Atlantic coast of Canada (2 to 4% d⁻¹; Chopin et al., 1999) or *Gigartina exasperata* (8% d⁻¹; Waaland, 1977). Growth of the latter could be significantly increased through a selection program (Waaland, 1979) and it has been shown by Merrill & Waaland (1979) that higher growth rates of selected strains were associated with the absence of reproduction. Similarly, *G. skottsbergii* shows a high vegetative propagation capacity, a feature required to develop a strain selection program.

Alternative propagation techniques

Considering the restricted seasonal spore availability, low spore survivorship in hatchery and the eventual need to propagate specific individuals for strain selection purposes, alternative vegetative propagation techniques for *Gigartina skottsbergii* are needed. Under laboratory conditions it is possible to propagate *G. skottsbergii* through regeneration of excised tissue fragments (Correa et al., 1999). *G. skottsbergii* can also regenerate from rhizoids, forming new fronds under laboratory (Figure 8A, B) and field conditions (Figure 8C, D). Thus, it is important not to remove the rhizoids from the substratum during harvesting, as shown for another economically important red seaweed (*Mazzaella laminarioides*: Santelices & Norambuena, 1987; Westermeier et al., 1987; Gómez & Westermeier, 1991). Further, healing and regeneration were shown to be regulated by nutrients, temperature and photon flux density. Frond fragments have been also successfully propagated and cultivated in the sea (Buschmann et al., 1999a). Vegetative fragmentation and subsequent regeneration resulted in plants with an elongated habit and a growth potential higher than non-fragmented fronds (Westermeier et al., unpublished data).

Protoplasts can be used for biotechnological purposes and as alternative means for propagation (Polne-Fuller & Gibor, 1986; Saga et al., 1986; Kloareg et al., 1991). Protoplasts are highly suitable for gene transfer, are capable of growing into whole plants at high frequency (Minocha, 1999), and have been used to produce new genetic variants in algae (Cheney, 1999). Protoplasts have been successfully isolated from a number of seaweeds, four of which are carrageenan producers, including: *Chondrus crispus* (Smith & Bidwell, 1989; Le Gall et al., 1990), *Gigartina corymbifera* (Gross, 1990), *Soliera filiformis* (Gomez-

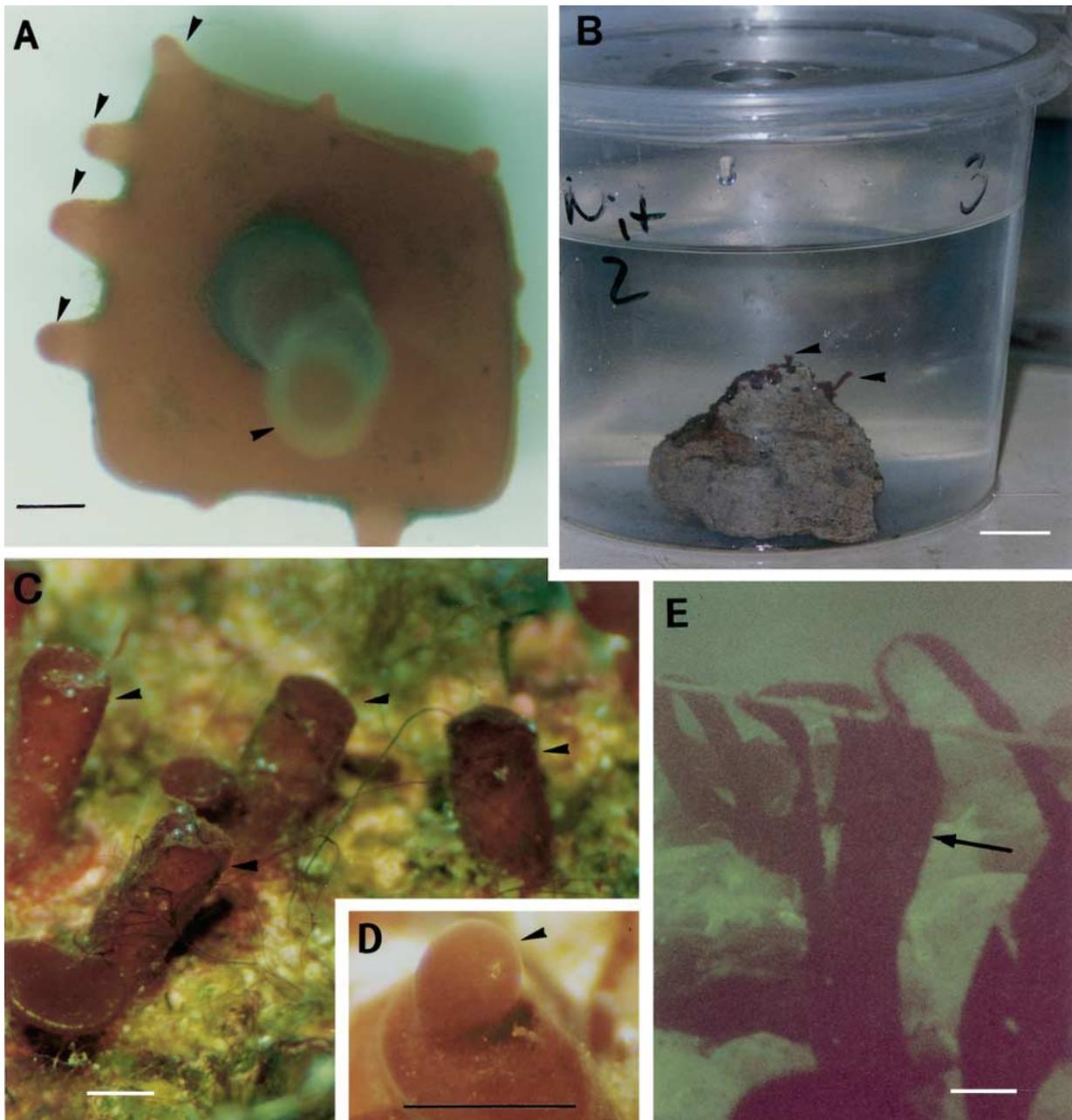


Figure 8. Regeneration of *Gigartina skottsbergii*. **A:** explants with rhizoid and frond tissues showing regeneration processes along the margins (arrows), scale bar = 0.2 cm. **B:** rock chips with rhizoids cultivated in 1-L plastic containers, scale bar = 1.5 cm. **C:** rhizoid remains (after experimental cuttings) attached to rocks with clear signals of healing and regeneration, scale bar = 1.5 mm. **D:** close-up of regenerated (arrow) rhizoid, scale bar = 1.5 mm. **E:** fronds obtained through vegetative propagation (arrow) and rope cultivation in Calbuco, scale bar = 10 cm.

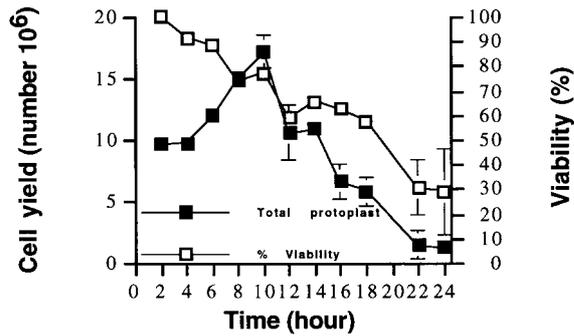


Figure 9. Cell yield and cell viability of protoplasts of *Gigartina skottsbergii* after different incubation times with a mixture of cellulase and kappa- and lambda-carrageenases. Error bars represent ± 1 SE, $n = 4$.

Pinchetti et al., 1993) and *Kappaphycus alvarezii* (Zablackis et al., 1993). Protoplasts of *Gigartina skottsbergii* were consistently obtained in the laboratory with yields of 17×10^6 cells g^{-1} after 10 h of incubation in the enzyme mixture (Figure 9). This yield was 226% and 230% higher than the reported values for *Kappaphycus alvarezii* and *Soliera filiformis* respectively (Zablackis et al., 1993; Gómez-Pinchetti et al., 1993), but 2.3% lower than for *Chondrus crispus* (Le Gall et al., 1990). A clear decline in cell yield was recorded after 10 h of incubation. Viability of the protoplasts was 100% after 2 h, but declined to 29% after 24 h (Figure 9). *G. skottsbergii* has elongated medullary cells, which are morphologically distinct from the cortical and subcortical cells (Figure 10A). Survival of medullary cells initially was 88%, and then declined after 10 h to 8% (Figure 10B). The calcofluor trials indicated the presence of cell walls in the medullary filaments. Protoplasts size ranged from 3.7 to 12.5 μm in diameter, probably reflecting the types of cells in the digested tissues. The protoplasts appear vacuolated, and cells are spherical with a parietal chloroplast (Figure 10C). Using TEM (Figure 10D), the cells show healthy plastids filling most of the intracellular space, and a central nucleus. To date, plants have not been regenerated from protoplasts, but the importance of this approach to open new technological alternatives, including genetic engineering (Minocha, 1999) and genetic manipulation (Cheney, 1999), encourages further studies.

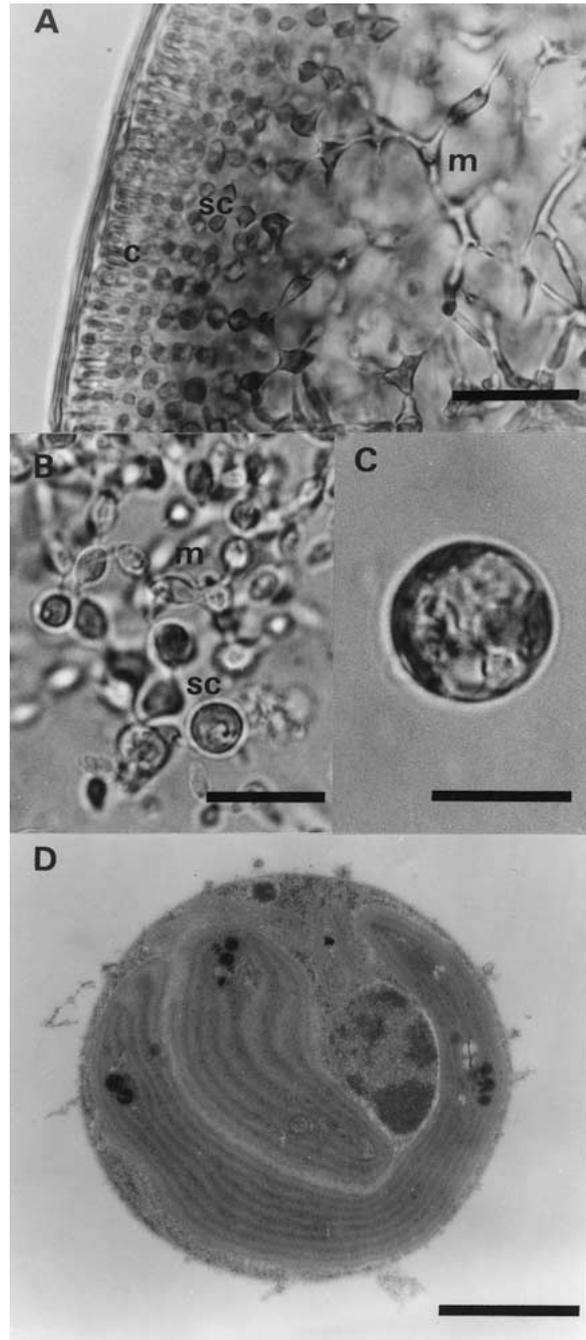


Figure 10. Protoplasts from *Gigartina skottsbergii*. A: section through a vegetative frond with (c) cortical, (sc) sub-cortical and (m) medullary cells, scale bar = 50 μm . B: different cellular types; (sc) sub-cortical and (m) medullary cells released during enzymatic digestion, scale bar = 12 μm . C: isolated protoplast, scale bar = 7 μm . D: TEM of a protoplast from the cortical cell layer, scale bar = 1 μm .

Conclusions

This analysis indicates that the demand for *Gigartina skottsbergii* will probably increase steadily and consequently the need for raw material will pose an unsustainable harvesting pressure on natural stands in southern Chile. Cultivation is, however, feasible. Spores can be seeded on ropes or other substrata, and thalli can be grown via fragmentation. The possibility of using protoplasts to by-pass temporal restrictions in spore availability also remains as a challenge for future research.

Cultivation of *Gigartina skottsbergii* requires special attention to ameliorate high mortalities of new recruits in the field and to select cultivation areas, where temperature cannot exceed 14 °C. Because of the low growth rates of *G. skottsbergii*, it is essential to select fast-growing strains, as it has been done in other red algae (Cheney, 1999) including species of *Gigartina* (Waaland, 1979). Farming of *G. skottsbergii* in Chile is currently limited by its slow growth and market values. As with other gel-producing seaweeds, prices are rather low, and in the case of *G. skottsbergii* the price paid to the producers has fluctuated between US\$ 0.7 and US\$ 0.9 kg⁻¹ dry wt in the past (1998–1999).

The characteristics of the gel produced by *Gigartina skottsbergii* will likely induce a sustained harvesting pressure on this resource, which will not be altered with the introduction of algae such as *Eucheuma* spp. Thus, it is our opinion that the establishment of commercial farming of *G. skottsbergii* will require an integrated effort of private investors and research centers to move beyond the basic understanding of the biology of the resource. An integrated private and academic effort should aim to stabilize the production of this alga, including biomass available mainly from cultivated stands rather than from wild stocks.

Alternatively, land-based tank cultivation could be another opportunity that requires to be studied in the near future. Experimental tank cultivation of other species of *Gigartina* (Waaland, 1977), *Chondrus crispus* (Bidwell et al., 1985; McLachlan, 1991;) and different *Gracilaria* species (Hanisak, 1987; Ugarte & Santelices, 1992; Buschmann et al., 1994; Friedlander & Levy, 1995) has been successful. However, carrageenophytic species such as *Chondrus crispus* can develop severe disease in tank cultures (Craigie, 1999) and so high tolerance to epiphytes and fouling organisms seems an important advantage of *G. skottsbergii* (Buschmann et al., 2001). These studies

suggest that, although it seems possible, tank cultivation of *G. skottsbergii* still requires to be demonstrated.

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References

- Alveal K (1998) The seaweed resources of Chile. In Critchley AT, Ohno M (eds), *Seaweed Resources of the World*. Japan International Cooperation Agency, Yokosuka, pp. 347–365.
- Anonymous (1999). Anuario Estadístico de Pesca 1998. Subsecretaría de Pesca, Valparaíso, Chile, 283 pp.
- Avila M, Ask E, Rudolph B, Nuñez M, Norambuena R (1999a) Economic feasibility of *Sarcothalia crispata* (Gigartinales, Rhodophyta) cultivation. *Hydrobiologia* 398/399: 435–442.
- Avila M, Candia A, Nuñez M, Romo H (1999b) Reproductive biology of *Gigartina skottsbergii* (Gigartinales, Rhodophyta) from Chile. *Hydrobiologia* 398/399: 149–157.
- Bidwell RGS, McLachlan J, Lloyd NDH (1985) Tank cultivation of Irish moss, *Chondrus crispus* Stackh. *Bot. mar.* 28: 87–97.
- Bischoff-Bäsmann B, Wienecke C (1996) Temperature requirements for growth and survival of Antarctic Rhodophyta. *J. Phycol.* 32: 525–535.
- Buschmann AH, Correa JA, Westermeier R (1999a) Recent advances in the understanding of the biological basis for *Gigartina skottsbergii* (Rhodophyta) cultivation in Chile. *Hydrobiologia* 398/399: 427–434.
- Buschmann AH, Correa JA, Westermeier R, Hernández-González MC, Norambuena R (1999b) Mariculture of red algae in Chile. *World Aquaculture* 30: 41–45.
- Buschmann AH, Correa JA, Westermeier R, Hernández-González MC, Norambuena R (2001) Red algal farming: a review. *Aquaculture* 194: 203–220.
- Buschmann AH, Mora OA, Gómez P, Böttger M, Buitano S, Retamales C, Vergara PA, Gutierrez A (1994) *Gracilaria chilensis* outdoor tank cultivation in Chile: use of land-based salmon culture effluents. *Aquacult. Eng.* 13: 283–300.
- Cheney DP (1999) Strain improvement of seaweeds through genetic manipulation: current status. *World Aquaculture*: 30: 55–56 + 66–67.
- Chopin T, Sharp G, Belyea E, Semple R, Jones, D (1999) Open-seawater aquaculture of the red alga *Chondrus crispus* in Prince Edward Island, Canada. *Hydrobiologia* 398/399: 417–425.
- Correa J, Beltrán J, Buschmann AH, Westermeier R (1999). Healing and regeneration responses in *Gigartina skottsbergii* (Rhodophyta, Gigartinales): optimization of vegetative propagation for cultivation. *J. appl. Phycol.* 11: 315–327.
- Craigie J (1990) Cell walls In Cole MC, Sheath (eds), *Biology of the Red Seaweeds*. Cambridge Press, Cambridge, pp. 221–257.

- Craigie J (1999) Rapid bioassay of a red food alga: accelerated growth rates of *Chondrus crispus*. World Aquaculture 30: 26–28.
- Dayton PK, Currie V, Gerrodette T, Keller BD, Rosenthal R, ven Tresca D (1984) Patch dynamics and stability of some California Kelp communities. Ecol. Monogr. 54: 253–289.
- Fletcher RL (1995) Epiphytism and fouling in *Gracilaria* cultivation: an overview. J. appl. Phycol. 7: 325–333.
- Friedlander M, Levy I (1995) Cultivation of *Gracilaria* in outdoor tanks and ponds. J. appl. Phycol. 7: 315–324.
- Gómez I, Westermeier R (1991) Frond regrowth from basal discs in *Iridaea laminarioides* (Rhodophyta, Gigartinales) at Mehuín, southern Chile. Mar. Ecol. Progr. Ser. 73: 83–91.
- Gómez-Pinchetti JL, Björk M, Pedersén M, García Reina G (1993) Factors affecting protoplasts yield of the carrageenophyte *Solieria filiformis* (Gigartinales, Rhodophyta). Plant Cell Rep. 12: 541–545.
- Gross W (1990) Preparation of protoplasts from the carrageenophyte *Gigartina corymbifera* (Kütz.) J. Ag. (Rhodophyta). J. microbiol. Methods 12: 217–23.
- Guiry MD (1990) Sporangia and spores. In Cole MD, Sheath RG (eds), Biology of the Red Algae. Cambridge University Press, Cambridge, pp. 347–376.
- Hanisak MD (1987) Cultivation of *Gracilaria* and other macroalgae in Florida for energy production. In Bird KT, Benson PH (eds), Seaweed Production for Renewable Resources, Elsevier Science Publishers, Amsterdam, pp. 191–218.
- Kennelly SJ (1989) Effects of kelp canopies on understory species due to shade and scour. Mar. Ecol. Progr. Ser. 50: 215–224.
- Kim DH (1976) A study of the development of cystocarps and tetrasporangial sori in Gigartinales (Rhodophyta, Gigartinales). Nova Hedwigia 27: 1–145.
- Klein E, Matsuhiro B, Utreras R, Zanlungo A (1984) Polysaccharides from Chilean seaweed. Part XIII. Studies on carrageenan from *Gigartina papillata* J. Ag. Rev. Latinoamer. Quim. 14: 128–131.
- Kloareg B, Benet H, LeGall Y (1991) Applications of protoplast isolation in basic and applied phycology. In García-Reina G, Pedersén M (eds), Seaweed Cellular Biotechnology, Physiology and Intensive cultivation, Universidad de Las Palmas de Gran Canaria, Canary Island, pp. 17–24.
- LeGall Y, Braud JP, Kloareg B (1990) Protoplast production in *Chondrus crispus* gametophytes (Gigartinales, Rhodophyta). Plant Cell Reports 8: 582–585.
- Maeda H, Ishida N (1967) Specificity of binding of hexopyranoxyl polysaccharides with a fluorescent brightener. J. Biochem. 62: 276–278.
- Mathieson AC, Tveter E (1976) Carrageenan ecology of *Gigartina stellata* (Stackhouse) Batters. Aquat. Bot. 2: 353–361.
- McCandless EL, Craigie JS (1974) Reevaluation of seasonal factors involved in carrageenan production of *Chondrus crispus*. Carrageenans of carposporic plants. Bot. Mar. 17: 125–129.
- McLachlan J (1991) General principles of on-shore cultivation of seaweeds: effects of light on production. Hydrobiologia 221: 125–135.
- Merrill JE, Waaland JR (1979) Photosynthesis and respiration in a fast growing strain of *Gigartina exasperata* (Harvey and Bailey). J. exp. mar. Biol. Ecol. 39: 281–290.
- Minocha SC (1999) Genetic engineering of marine macroalgae: current status and future perspectives. World Aquaculture 30: 29–30 + 57.
- Moreno CA, Sutherland JP (1982) Physical and biological processes in a *Macrocystis pyrifera* community near Valdivia, Chile. Oecologia 55: 1–6.
- Pickmere SE, Parsons MJ, Bailey RW (1975) Variation in carrageenan levels and composition in three New Zealand species of *Gigartina*. New Zealand J. Sci. 18: 585–590.
- Piriz ML (1996) Phenology of *Gigartina skottsbergii* Setchell et Gardner population in Chubut Province (Argentina). Bot. mar. 39: 311–316.
- Piriz ML, Cerezo AS (1991) Seasonal variation of carrageenan in tetrasporic, cystocarpic and 'sterile' stages of *Gigartina skottsbergii* S. et G. (Rhodophyta, Gigartinales). Hydrobiologia 225: 65–69.
- Polne-Fuller M, Gibor A (1986) Calluses, cells and protoplasts in studies towards genetic improvement of seaweeds. Aquaculture 57: 117–123.
- Potin P, Sanséau A, Le Gall Y, Rochas C, Kloareg B (1991) Purification and characterization of a new kappa-carrageenase from marine *Cytophaga*-like bacterium. Eur. J. Biochem. 201: 241–247.
- Saga N, Polne-Fuller M, Gibor A (1986) Protoplasts from seaweeds: production and fusion. Nova Hedwigia 83: 37–43.
- Santelices B (1996) Seaweed research and utilization in Chile: moving into a new phase. Hydrobiologia 326/327: 1–14.
- Santelices B (1999) A conceptual framework for marine agronomy. Hydrobiologia 398/399: 15–23.
- Santelices B, Ojeda FP (1984) Effects of canopy removal on the understory algal community structure of coastal forests of *Macrocystis pyrifera* from southern South America. Mar. Ecol. Progr. Ser. 14: 165–173.
- Santelices B, Norambuena R (1987) A harvesting strategy for *Iridaea laminarioides* in central Chile. Hydrobiologia 151/152: 329–333.
- Schiel DR, Foster MS (1986) The structure of subtidal algal stands in temperate waters. Ocean. Mar. Biol. Annu. Rev. 24: 265–307.
- Smith RG, Bidwell RGS (1989) Inorganic carbon uptake by photosynthetically active protoplasts of the red macroalga *Chondrus crispus*. Mar. Biol. 102: 1–4.
- Ugarte R, Santelices B (1992) Experimental tank cultivation of *Gracilaria chilensis* in central Chile. Aquaculture 101: 7–16.
- Waaland JR (1977) Growth of Pacific Northwest marine algae in semi-closed culture. In Krauss, R (ed.), The Marine Plant-Biomass of the Pacific Northwest Coast, Oregon University Press, pp. 117–137.
- Waaland JR (1979) Growth and strain selection in the marine alga *Gigartina exasperata* (Florideophyceae). Int. Seaweed Symp. 9: 241–247.
- Westermeier R, Rivera PJ, Chacana M, Gómez, I (1987) Biological bases for management of *Iridaea laminarioides* Bory in southern Chile. Hydrobiologia 151/152: 313–328.
- Westermeier R, Aguilar A, Sigel J, Quintanilla J, Morales J, (1999) Biological basis for the management of *Gigartina skottsbergii* (Gigartinales, Rhodophyta) in southern Chile. Hydrobiologia 398/399: 137–147.
- Zablackis E, Vreeland V & Kloareg B (1993) Isolation of protoplasts from *Kappaphycus alvarezii* var: *tambalang* (Rhodophyta) and secretion of *t*-carrageenan fragments by cultured cells. J. exp. Bot. 44: 1515–1522.
- Zamorano J, Westermeier R (1996) Phenology of *Gigartina skottsbergii* (Gigartinales, Rhodophyta) in Ancud Bay, southern Chile. Hydrobiologia 326/327: 253–258.
- Zinoun M, Cosson J, Deslandes E (1993) Influence of culture conditions on growth and physicochemical properties of carrageenans in *Gigartina teedii* (Rhodophyceae – Gigartinales). Bot. mar. 36: 131–136.