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	Journal Name	Manuscript No.		Author Received: No. of pages: 6	PE: HR/Suresh

Aquaculture Research, 2005, 1–6 doi: 10.1111/j.1365-2109.2005.01321.x

First attempt to cultivate the carrageenan-producing seaweed *Chondracanthus chamissoi* (C. Agardh) Kützing (Rhodophyta; Gigartinales) in Northern Chile

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Abstract

Chondracanthus chamissoi is an important source of carrageenan in Chile. Presently, all the production is harvested from wild populations. This study reports the first attempt to cultivate *C. chamissoi*. Experiments were conducted with female gametophytic and sporophytic thalli by inserting them among braids of a 7 mm polypropylene rope. Inoculated ropes were placed at 1, 3 and 5 m depths in two sheltered bays in northern Chile. The fronds adapted well to cultivation and grew at all the depths tested, although the greatest increase in biomass was observed at 1 m depth. On a yearly cycle, higher biomass increases were observed in autumn and winter months compared with the spring and summer. We believe that by adapting the cultivation methodology, this species could be cultivated year round.

Keywords: carrageenophyte, Chile, *Chondracanthus chamissoi*, mariculture, seaweed, vegetative propagation

Introduction

Production of seaweeds in Chile has been based mainly on the harvest of wild populations, with commercial cultivation being restricted to a single species, *Gracilaria chilensis* Bird, McLachlan & Oliveira. The success of *Gracilaria* cultivation stimulated the establishment of new seaweed-processing plants in Chile in the last 10 years, which resulted in an increase in the demand for raw material (Ávila, Ask, Rudolph, Nuñez & Norambuena 1999; Buschmann, Correa, Westermeier, Hernández-González & Norambuena 2001a). This is also partly due to the development of carrageenan industries and an increase in the demand of seaweeds for human consumption (Buschmann *et al*. 2001a). This demand motivated efforts to provide a scientific and practical basis for the management and cultivation of carrageenophytes such as *Sarcothalia crispata* (Bory) Leister (Ávila *et al*. 1999; Romo, Alveal & Werlinger 2001) and *Gigartina skottsbergii* Setchell & Gardner (Ávila *et al*. 1999; Correa, Beltrán, Buschmann & Westermeier 1999; Westermeier, Aguilar, Sigel, Quintanilla & Morales 1999; Buschmann, Correa, Westermeier, Paredes, Aedo, Potin, Aroca & Hernandez-González 2001b).

Chondracanthus chamissoi is a benthic marine red algae that may reach up to 50 cm in length. This species is found from the lower intertidal zone to 15 m depth (Hoffmann & Santelices 1997). It is distributed from Paita, Peru (5°S) to Ancud, Chile (42°S) (Ramirez & Santelices 1991). In northern Chile, the main harvesting areas are Caldera (27° 4′S), La Herradura (29° 58′S) and Puerto Aldea (30° 58′S).

Chondracanthus chamissoi is recognized as an important source of income in northern Chile coastal settlements (González & Meneses 1996). Buschmann and colleagues (2001a) suggested that the demand for *C. chamissoi* will increase in the next years. In addition to its commercialization for the extraction of carrageenan, there is an increasing demand for human consumption in Asia (González, Meneses & Vasquez 1997; Hoffmann & Santelices 1997; Anonymous 2000). As a consequence of this demand, overharvesting of *C. chamissoi* has already occurred in Puerto Aldea, diminishing the annual historical production from 500 to 60 tons in recent years (Anonymous 2000). Consequently, it is expected that the pressure on wild beds will extend to other localities, as has occurred in other seaweed populations of commercial interest in Chile (Norambuena 1996; Correa *et al*. 1999).

Ecological and physiological studies of *C. chamissoi* aiming to establish the basis for culture and management techniques have been carried out (González & Meneses 1996; González *et al*. 1997; Bulboa & Macchiavello 2001; Vásquez & Vega 2001; Macchiavello, Bulboa & Edding 2003). However, so far, no cultivation techniques have been developed to grow this species.

In this paper, we test an experimental approach to cultivate *C. chamissoi* that could lead to the development of a commercial system.

Materials and methods

The experiments were performed at the laboratory of Marine Botany and Ranching at Universidad Católica del Norte, Herradura Bay, Coquimbo, Chile (29°58′S) from April 2002 until May 2003. Parallel experiments were conduced in Calderilla Bay, Caldera (27° 4′ S), between September and December 2002.

Collection and maintenance of plants

Gametophytic (bearing cystocarps) and sporophytic (bearing tetrasporangial sori) fronds of *C. chamissoi* were collected at Herradura Bay, Coquimbo, Chile. The plants were transported wet in plastic bags, washed with seawater and cleaned of visible epiphytes. Gametophytic and sporophytic fronds were maintained separated in a 2000 L outdoor tank, with continuous flow filtered seawater (1 μm) until use in the experiments.

Inoculation of ropes and experimental design

Gametophytic and sporophytic fronds (5–10 cm long) were closely inserted among braids of 2 m polypropylene (7 mm diameter) ropes (ca. 10–15 g per rope). The main axis of the fronds was inserted in the rope, leaving the lateral branches free to grow.

Ropes with both gametophytic and sporophytic plants, in triplicate, were suspended horizontally, from a long-line system, at 1, 3 and 5 m depth.

During a 3-month period, the ropes were returned to the laboratory every 30 days and cleaned of fouling. The ropes were drained to eliminate excess water, weighed and returned to the sea. Biomass was expressed as the mean fresh weight per lineal metre of rope (g m^{-1}).

In Herradura Bay, the experiments were repeated in autumn (April), winter (July), spring (September) and summer (December). For the experiments in Calderilla Bay, the ropes were inoculated in the laboratory and transported wet, in plastic bags. In this case, the ropes were inoculated only in spring (September).

An analysis of variance (ANOVA) was used to evaluate the effects of depth and reproductive state on the biomass variation. Homogeneity of the variances and normality were reviewed for all results. A Tukey test was used when treatments showed significant differences (Sokal & Rohlf 1981). All data were analysed using SYSTAT 8.0 Software (SPSS, Chicago, IL, USA).

Results

The gametophytic as well as the sporophytic fronds grew well in all the depths tested (1, 3 and 5 m), producing ropes densely covered by mature thalli (Fig. 1). Secondary attachments of the fronds were observed after 2 months in the sea (Fig. 2).

Herradura Bay experiments

Autumn

Biomass accumulation at 1 m depth was significantly greater than at 3 and 5 m, for both reproductive phases ($P < 0.05$). Largest biomass accumulation occurred at 1 m during the second month of cultivation

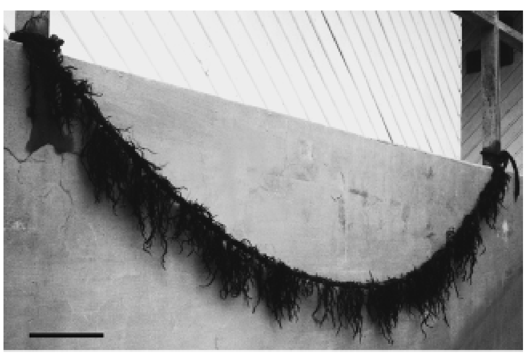

Figure 1 Spring inoculated rope (Calderilla Bay) with female gametophytic fronds of *Chondracanthus chamissoi* grown after 3 months in the field at 3 m of depth. Scale bar = 10 cm.

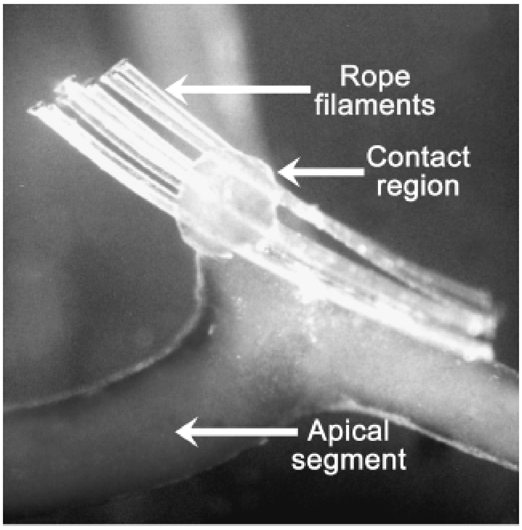

Figure 2 Regions of adhesion between the fronds of *Chondracanthus chamissoi* and the rope filaments. Scale bar = 2 mm.

(June), resulting in 158 g m^{-1} fresh weight (SD ± 29) and 160 g m^{-1} (SD ± 40) of gametophytic and sporophytic thalli respectively (Fig. 3a and b). No significant differences were found between reproductive phases ($P > 0.05$). Throughout the experiments, plants remained free of epiphytes, with their natural reddish tone and abundant new branches reaching as long as 15 cm. However, at 5 m, plants developed thinner fronds with sparse branching. Detachments of the heavier plants occurred after 3 months at 1 and 3 m depths.

Winter

Figure 3c and d show a slight increase in biomass during the first month of culture (August) for both reproductive phases. Weight increase was significant ($P < 0.05$) in September and October, respectively, at 1 and 3 m. Gametophytes showed a biomass peak of 120 g m^{-1} (SD ± 36) in October at 1 m, whereas sporophytes reached a biomass peak of 102 g m^{-1} (SD ± 21) in September. These values were significantly larger than those registered at 3 and 5 m depths ($P < 0.05$).

Similar to the autumn experiment, no significant differences in the biomass were observed between the two reproductive phases ($P > 0.05$). In September, at 1 and 3 m depths, *Polysiphonia* spp. epiphytized *C. chamissoi*. The plants at 5 m produced branches up to 15 cm long, which were free of epiphytes until the end of the experiment. However, the new branches were thinner and sparsely branched. Heavier fronds started to detach from the rope after 3 months.

Spring

The plants developed robust and abundant branches in this season. However, detachment of the heavier specimens took place from the first month in the sea. No significant differences between the tested depths were found ($P > 0.05$). The gametophytic fronds registered maximum biomass at 1 m in November (62 g m^{-1} SD ± 19), and the sporophytic fronds in December at 2 m depth (78 g m^{-1} SD ± 24) (Fig 3e and f). No significant differences were observed between the two reproductive phases ($P > 0.05$).

From October, *Polysiphonia* spp. appeared as epiphytes on *C. chamissoi* at 1 and 3 m depths. Although bleached fronds were observed at 1 m, plants remained clean and retained their natural colour, with branches reaching 20 cm in length at 5 m.

Summer

Figure 3g and h show a significant increase in the biomass during January at 1 m, in relation to the initial weight (December) for both reproductive phases ($P < 0.05$). Peak biomasses of 57 g m^{-1} (SD ± 7) and the 44 g m^{-1} (SD ± 13) were reached by gametophytic and sporophytic phases respectively. However, significant differences between gametophytic and sporophytic biomass were not observed ($P > 0.05$). Throughout the experiments, detachment of the heavier plants occured, mainly at 1 and 3 m depths. At 3 and 5 m, branching was sparse and branches were thinner, although free of epiphytes. Epiphytes *Polysiphonia* spp. became abundant from the beginning of the experiments, mainly at 1 m depth.

Calderilla Bay, spring experiment

A significant increase in biomass was registered during the first month of cultivation (October) at 1 and 3 m depths ($P < 0.05$), for both reproductive phases (Fig. 4). Maximum biomasses reached by gametophytic fronds were: 122 (SD ± 28), 84 (SD ± 5) and 85 g m^{-1} (SD ± 23) at 1, 3 and 5 m depths respectively (Fig. 4a). For the sporophytic fronds, the biomass peaks were 115 (SD ± 11) at 1 m; 79 (SD ± 7) at 3 m and 37 (SD ± 2) at 5 m depth (Fig. 4b). The accumulated biomass at 1 m was significantly greater than that noted at 5 m depth ($P < 0.05$) for both

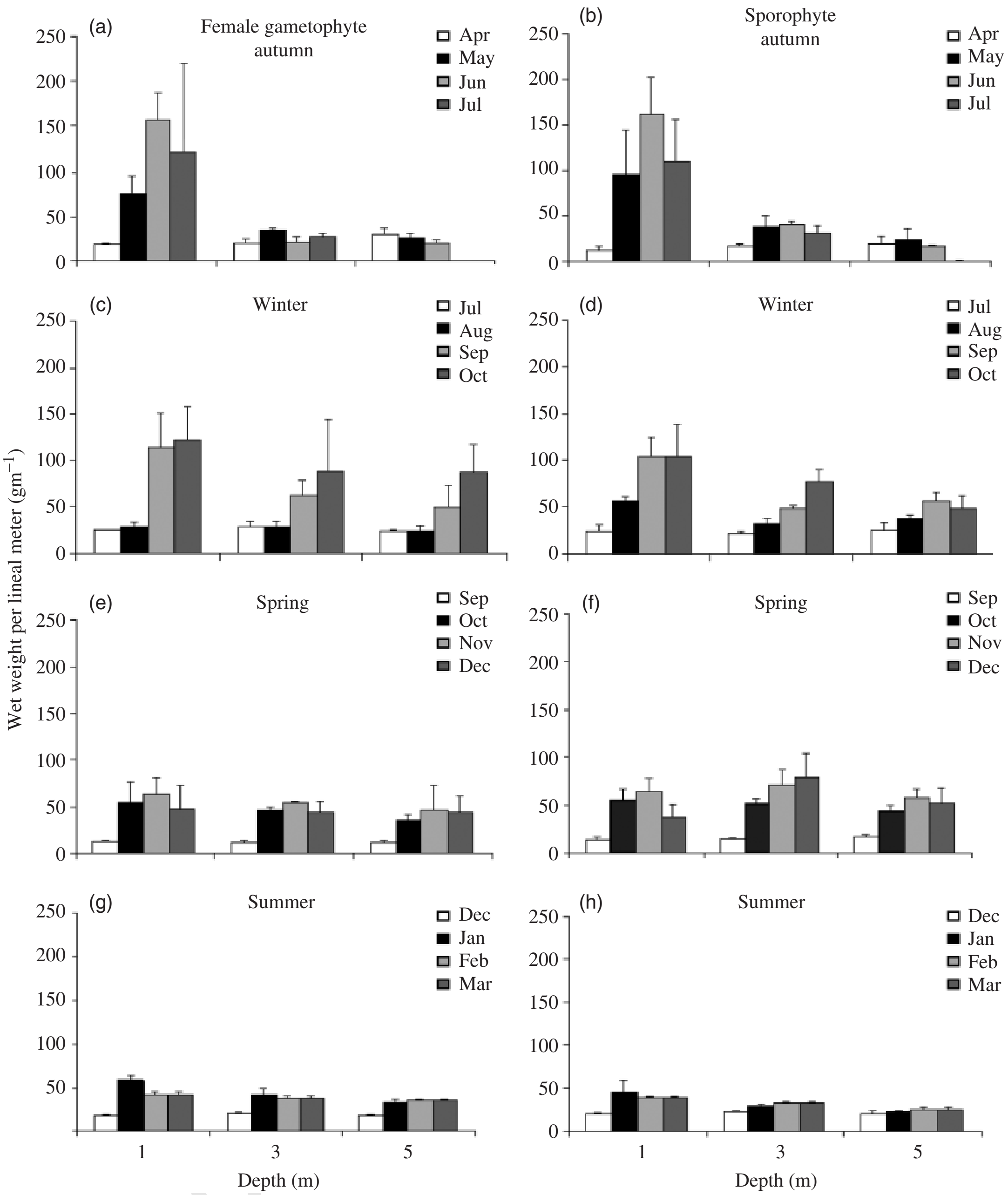

Figure 3 Biomass accumulation and standard deviation of female gametophytic and sporophytic fronds of *Chondracanthus chamissoi*, cultivated in the sea at 1, 3 and 5 m depths and different seasons, in the Herradura Bay. (a, b) Autumn inoculation, (c, d) winter inoculation, (e, f) spring inoculation and (g, h) summer inoculation.

reproductive phases. However, no significant differences were observed in biomass accumulation between gametophytes and sporophytes ($P > 0.05$).

Epiphytism by brown filamentous algae was observed mainly in the first month (September) at 1 m depth. Detachment of the heavier plants increased gradually until the end of the experiments.

Discussion

This paper reports the first attempt to cultivate *C. chamissoi* with the aim of decreasing exploitation pressure on natural populations of this valuable resource in Chile. This contribution is an addition to the efforts underway in Chile to establish a sustain-

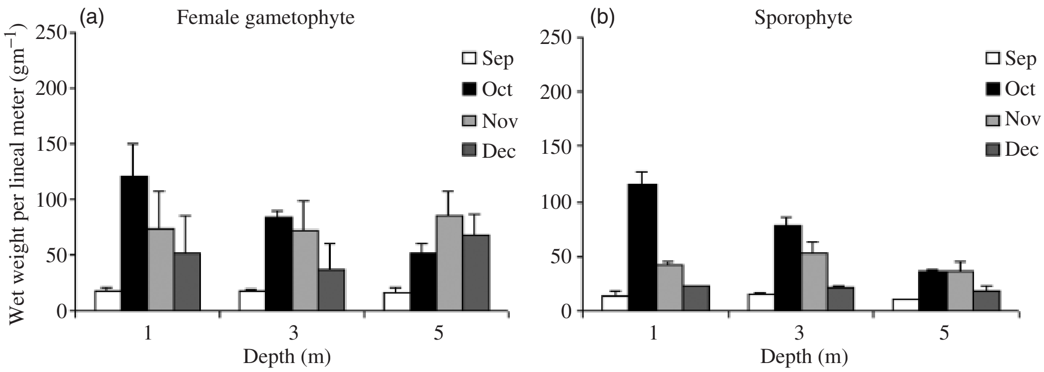

Figure 4 Biomass accumulation and standard deviation of female gametophytic (a) and sporophytic fronds (b) of *Chondracanthus chamissoi*, cultivated in the sea at different depths (1, 3 and 5 m) in Calderilla Bay. Spring inoculation.

able production of carrageenophytes, including propagation through spores (Buschmann *et al.* 2001b; Romo *et al.* 2001; Ávila, Pavez, Candia, San Martin & Caceres 2003), vegetative (Correa *et al.* 1999; Buschmann *et al.* 2001b) and protoplasm propagation (Buschmann *et al.* 2001b).

Our experiments show that cultivation of *C. chamissoi* in northern Chile is feasible in the colder months (autumn and winter), which was unexpected considering that the production of this species typically diminishes in the natural beds during these seasons (González & Meneses 1996; Vásquez & Vega 2001). On the other hand, in warmer months (spring and summer), the lower production, observed mainly at 1 and 3 m depths, was due to the detachment of the heavier plants, mainly associated with the higher weight and size of the fronds and epiphytism. This result agrees with Bulboa and Macchiavello (2001), who showed a better physiological performance at higher temperatures, allowing a fast growth in warmer months.

During autumn and winter experiments, harvests could be made after 2 and 3 months respectively. On the other hand, in spring and summer experiments, harvest frequency should be less than 30 days, in order to prevent the loss of the heavier plants.

A different situation was observed in Calderilla Bay where a lower presence of epiphytes during the first months of cultivation, and a smaller loss of plants, allowed a higher production of *C. chamissoi*. These results, added to the eurithermic behaviour of *C. chamissoi* (Bulboa & Macchiavello 2001), provide the possibilities to cultivate this species in other localities on the northern coast of Chile with potentially favourable abiotic and biotic conditions.

Epiphytism has been considered a major problem for the cultivation of seaweed (Lüning & Pang 2003), and the present case is not an exception. Vásquez and Vega (2001) mention massive appearance of epiphytes on *C. chamissoi* in Herradura Bay from January to April. In our case the epiphytism problem became worse mainly from September to March. On the other hand, epiphytism is not a problem during autumn and winter, and not at 5 m depth even during spring and summer. These results are encouraging and represent a possibility to access the human consumption market, with higher prices and where strict measures of quality are imposed.

González and colleagues (1997) suggested that drifting fronds of *C. chamissoi* would be able to re-attach. Recently, this was also confirmed by Macchiavello and colleagues (2003). Here, we provide evidence of the ability of *C. chamissoi* to develop secondary attachment to the supporting rope, which is an additional advantage for the cultivation of this species.

We also show that the suspended rope cultivation methodology is technically feasible to grow *C. chamissoi* in northern Chile. This technique allows control of the cultivation depth, which can be a strategy to grow the species all year round and to reduce the problem of epiphytism. The lower production at greater depth could be compensated by the production of a cleaner product for the food market, which yields much better money value. Besides, the rope cultivation system allows the selective use of gametophytes or sporophytes for the production of raw material for the κ- or λ-carrageenan market.

Cultivation of *C. chamissoi* is technically feasible in northern Chile, and yields can be improved through small adaptations in the methodology to avoid frond loss and epiphytism. However, it remains to be demonstrated that cultivation of this species is economically competitive with harvest of natural populations.

Acknowledgments

This study was supported by a DGI (UCN) Grant. We are grateful to Jaime Montenegro for assistance in the field. C. Bulboa is grateful to the Red Latinoamericana de Botánica (RLB – Tyler Prize 2004) for the PhD fellowship.

References

Anonymous (2000) *Estudio de situación base y plan de manejo y explotación de recursos bentónicos en el área de Puerto Aldea (bahía de Tongoy, Coquimbo, IV Región)*. Informe técnico, Universidad Católica del Norte, Coquimbo, Chile, 67 pp.

Ávila M., Ask E., Rudolph B., Nuñez M. & Norambuena R. (1999) Economic feasibility of *Sarcothalia* (Gigartinales, Rhodophyta) cultivation. *Hydrobiologia* **398/399**, 435–442.

Ávila M., Pavez H., Candia A., San Martin R. & Caceres J. (2003) Effects of temperature and irradiance on the germination and growth of *Sarcothalia crispata* in southern Chile. In: *Proceedings of the International Seaweed Symposium*, 17, Cape Town, South Africa, pp. 27–33. Q3

Bulboa C.R. & Macchiavello J.E. (2001) The effects of the light and temperature on different phases of the life history in the carrageenan producing alga *Chondracanthus chamissoi* (Rhodophyta, Gigartinales). *Botanica Marina* **44**, 371–374.

Buschmann A., Correa J.A., Westermeier R., Hernández-González M.C. & Norambuena R. (2001a) Red algal farming in Chile: a review. *Aquaculture* **194**, 203–220.

Buschmann A., Correa J.A., Westermeier R., Paredes M., Aedo D., Potin P., Aroca G. & Hernandez-González M. (2001b) Cultivation of *Gigartina skottsbergii* (Gigartinales, Rhodophyta): recent advances and challenges for the future. *Journal of Applied Phycology* **13**, 255–266.

Correa J., Beltrán J., Buschmann A. & Westermeier R. (1999) Healing and regenaration responses in *Gigartina skottsbergii* (Rhodophyta, Gigartinales): optimization of vegetative propagation for cultivation. *Journal of Applied Phycology* **11**, 315–327.

González J. & Meneses I. (1996) Differences in the early stages of development of gametophytes and tetrasporophytes of *Chondracanthus chamissoi* (C. Agardh) Kützing from Puerto Aldea, northern Chile. *Aquaculture* **143**, 91–107.

González J., Meneses I. & Vasquez J. (1997) Field studies in *Chondracanthus chamissoi* (C. Agardh) Kützing: seasonal and spatial variations in life-cycle phases. *Biología Pesquera* **26**, 3–12.

Hoffmann A. & Santelices B. (1997) *Flora Marina de Chile Central*. Ediciones de la Universidad Católica de Chile, Coquimbo, Chile, 434pp.

Lüning K. & Pang S. (2003) Mass cultivation of seaweeds: current aspects and approaches. *Journal of Applied Phycology* **15**, 115–119.

Macchiavello J., Bulboa C. & Edding M. (2003) Vegetative propagation and spore recruitment in the carrageenophyte *Chondracanthus chamissoi* (Rhodophyta, Gigartinales) in northern Chile. *Phycolgical Research* **51**, 45–50.

Norambuena R. (1996) Recent trends of seaweed production in Chile. *Hydrobiologia* **326/327**, 371–379.

Ramirez M.E. & Santelices B. (1991) Catálogo de las algas marinas bentónicas de la costa temperada del Pacífico Sudamericano. *Monografías Biológicas* **5**, 1–499.

Romo H., Alveal K. & Werlinger C. (2001) Growth of the commercial carrageenophyte *Sarcothalia crispata* (Rhodophyta, Gigartinales) on suspended culture in central Chile. *Journal of Applied Phycology* **13**, 229–234.

Sokal R.R. & Rohlf F.J. (1981) *Biometry*. W. H. Freeman, San Francisco, CA, USA, 895pp.

Vásquez J.A. & Vega J.M.A. (2001) *Chondracanthus chamissoi* (Rhodophyta, Gigartinales) in northern Chile. Ecological aspects for management of wild populations. *Journal of Applied Phycology* **13**, 267–277.

Westermeier R., Aguilar A., Sigel J., Quintanilla J. & Morales J. (1999) Biological basis for the management of *Gigartina skottbergii* (Gigartinales, Rhodophyta) in southern Chile. *Hydrobiologia* **398/399**, 137–147.

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