

## Effects of stock loading and planting distance on the growth and production of *Gracilaria chilensis* in rope culture

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### Abstract

Through experimental manipulation of stock loading and planting distances in rope culture, this study evaluates the hypothesis that optimum density for growth and production of *Gracilaria chilensis* vary within a bed. Small packages of *Gracilaria* disposed at short distances exhibit higher growth and production rates in the landward ends of ropes than larger packages of seaweed disposed at longer distances. In these experiments, the landward end of ropes faces estuarine conditions, with reduced salinities and increased sediment loads during low tides. The interacting effects of stocks loading and position in the rope can affect growth and production rates so much as to nullify the 3.5-fold summer increments in production that occur in these estuarine beds of southern Chile. It is concluded that through manipulation of planting distances and stock loading, the production of this species could be increased up to 35% in some areas.

### Introduction

In contrast to land plants, the answer to how many individuals may be packed into an area without consequences is by no means obvious in seaweeds (Santelices, 1990). Some species increase mortality as density increases as it would be expected according to the  $-3/2$  thinning law (Cousens & Hutching, 1983). Other species grow faster and reach larger sizes with increasing densities (Schield & Choat, 1980; Schield, 1985; Martinez & Santelices, 1992). Therefore, the type of interaction to be found in high or low density stands seems to vary according to the species and habitat considered.

Ecological studies with *Gracilaria chilensis* suggest that the optimum density for growth may in fact vary within a given bed. Monitoring of experimentally harvested plots indicate (Santelices *et al.*, 1984) that the faster growing thalli in central parts of beds inhibit the growth of conspecific thalli occurring around them within a given distance. Therefore, in central parts of beds, density-dependent, negative intraspecific interactions are to be expected. By contrast, in the peripheric parts of beds, close inter-thallus distances result in fast growth of the inner thalli and in higher total production of experimental patches (Santelices & Fonck, 1979). In these parts of the bed there seems to be a protection effect of peripheric over

central thalli. Such an effect can be replaced lining the beds with artificial plants of polypropylene untwisted ropes (Silva & Poblete, 1991) and seems related to protection from sediment deposition.

Knowledge of the effects of planting distance on production is basic to field farming of *G. chilensis*, specially in southern Chile where most farms are developed over soft, sandy-muddy bottoms. Previous studies (Pizarro & Barrales, 1986; Westermeier *et al.*, 1991, 1993; Pizarro & Santelices, 1993) have shown that initial stocking loads affect production but those studies have not attempted combining various stock distribution patterns. Artificial protections, as those described by Silva and Poblete (1991) are uneconomical and cumbersome in the field. In this study we evaluate whether a variable planting strategy (e.g. closer planting distances in the periphery of patches combined with larger planting distances in central parts) could still provide the protective effect described for peripheric thalli, increasing production.

## Materials and methods

### a. The study area

All the experiments were done at the experimental site that the Universidad Austral de Chile has in the Maullin River (41° 35' S; 73° 42' W), close to Puerto Montt in southern Chile. A full description of the area has been provided already (Westermeier *et al.*, 1991). Therefore, only a summary account of the study site will be introduced here.

The experimental area is part of a soft-bottom estuary, located about 1000 m upstream of the mouth of Maullin River. The bottom is a mix of thin sand and sediments. Average water salinity is 25‰, with annual variations of 17 to 31‰ S. The lowest salinity values are found in low tides during winter months (June, July, August) when heavy rains occur in the area. The annual average water temperature is 10 °C, with an annual range of 6 to 13 °C. Light intensity varies seasonally, with an average value of 200  $\mu\text{E m}^{-2} \text{s}^{-1}$  in winter and 1580  $\mu\text{E m}^{-2} \text{s}^{-1}$  in summer. An impor-

tant part of this seasonal light variation arises from significant changes in day length, which ranges from 8 h in July (winter) to 14 h in January (summer). The estuary is normally protected from swells and waves from the open ocean, except when very heavy storms occur in the area. Water exchange in the area is by tidal currents.

Some rain occurs at all times in the study area, with a monthly maximum of 277.3 mm in winter and a monthly minimum of 81.6 mm in summer. Variable amounts of sediments therefore reach the estuary at all seasons, amounting to 4400  $\text{g m}^{-2} \text{month}^{-1}$  in the rainy season and to 75  $\text{g m}^{-2} \text{month}^{-1}$  in summer.

### b. Experimental testing

A two-factor experiment was designed to test the effects of planting distance (stock distribution pattern) and position on the growth and production of *Gracilaria chilensis*. Even though rope culture is uncommon in Chile, we used rope culture to tag in the field individual thalli or groups of thalli precisely calculating their growth rate. Three blocks were distinguished along a 4 m long plastic rope (Fig. 1). In one block ( $D_1$ ); each of 10 packages of 20 g (wet weight) of *G. chilensis* were attached to the rope every 10 cm. In the second block ( $D_2$ ), five packages of 40 g of algae were attached to the rope every 20 cm. In the third block ( $D_3$ ), each of 3, 100 g packages of *G. chilensis* were attached every 50 cm. Thus, the three blocks had a stocking load of 200  $\text{g m}^{-1}$  of rope, although the distance between packages along the rope (10, 20 or 50 cm) and the total number of replicate packages (10, 5 or 3) varied among blocks.

In the field, the ropes were placed with one end facing the seaward side and the other end facing the landward side of the estuary. The position of the rope closer to the landward end of the estuary was named Position 1 ( $P_1$ ), the central block  $P_2$  and the seaward end Position 3 (Fig. 1). Using random numbers, position and blocks were distributed in the field following a random block design. At each experimental run, a total of six ropes was used. They were placed 1 m apart one

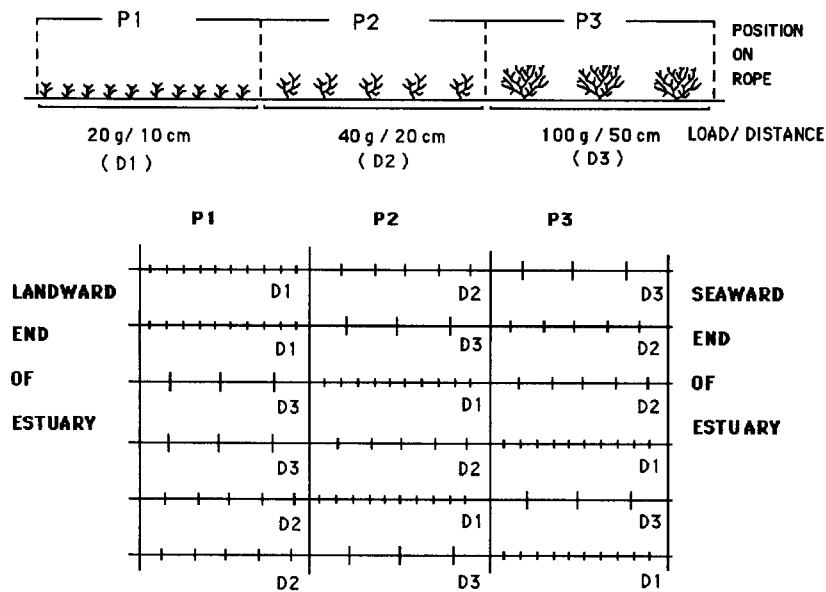


Fig. 1. Experimental set up showing the three stock loadings and planting distances used in this study ( $D_1$ ,  $D_2$  and  $D_3$ ) and the three possible positions for each treatment ( $P_1$ ,  $P_2$ ,  $P_3$ ) along a rope.

from the other, attached to wood sticks, at a vertical distance of 75 cm from the bottom. The study site had a depth of 1.25 m at low tide and 3.5 m during average high tide, so the ropes with the seaweeds were covered with water at all times during the experiments.

The experiment was repeated four times. Once during spring (October–November 1991), twice during summer (December 1991–January 1992; December 1992–January 1993) and once in late fall-winter (May–July 1992). The summer experiment lasted 30 days, the spring experiment 45 days and the winter experiment extended for 75 days. The very slow winter growth rates required longer experiments to obtain statistically significant responses.

At the end of each experiment, the ropes with algae were brought back to the laboratory and wet weighed. Specific growth rates were calculated using the formula

$$K = \frac{\log_2 X_2 - \log_2 X_1}{t_2 - t_1},$$

where  $K$  is growth rate and  $X_1$  and  $X_2$  are the values of wet weight at the beginning ( $t_1$ ) and at

the end ( $t_2$ ) of the experiment (Brinkhuis, 1985). Specific growth rates were calculated as daily growth rates and values were then transformed (arcsine) for further analysis. Since the summer and winter experiments are of different duration, seasonal comparisons of production were done on the basis of wet biomass produced per unit of rope per day. Data were then subjected to ANOVA (random block design) followed by Fisher PLSD test, whenever pertinent (Sokal & Rohlf, 1969).

Since the results gathered in the spring experiment are similar to those obtained in the two summer experiments, the following analysis contrasts the winter with one of the summer experiments only (December 1991–January 1992). These summer results are judged to be fully representative of the experimental runs done in spring and summer.

## Results and discussion

### a. Effects of season

Summer growth rates of *Gracilaria chilensis* cultivated on the ropes are about three times higher

than the winter growth rates ( $4.6 \pm 0.5\%$  versus  $1.8 \pm 0.4\%$ ); average biomass production was about 3.5 times higher in summer than in winter ( $36.6 \pm 6.1 \text{ g m}^{-1} \text{ d}^{-1}$  versus  $10.5 \pm 2.4 \text{ g m}^{-1} \text{ d}^{-1}$ ). Similar seasonal differences have been reported by Westermeier *et al.* (1993) in suspended cultures in the same experimental area. The very significant change in light availability and temperature are the most important abiotic factors correlated with this significant seasonal change in growth and production. As already mentioned, although other abiotic factors also vary in a seasonal way, light availability and temperature are those exhibiting maximum seasonal variation.

#### b. Effects of stocking distribution pattern

Small packages of *Gracilaria* disposed at short distances ( $D_1$ , Fig. 2) grew faster than large packages disposed at longer distances along the rope ( $D_3$ , Fig. 2). The growth differences between  $D_1$  and  $D_3$  are statistically significant in summer and winter ( $p = 0.05$  in both cases). In both seasons, the growth rates exhibited by  $D_2$  are intermediate between  $D_1$  and  $D_3$ . In summer the average  $D_2$

value is closer to  $D_1$  and significantly higher ( $p = 0.05$ ) than  $D_3$ . In winter there is statistically significant differences ( $p = 0.05$ ) between  $D_1$  and  $D_2$  but the difference between  $D_2$  and  $D_3$  is not significant.

Summer growth rates in all three stocking distribution patterns exhibit larger variability than the winter values. This is most pronounced in  $D_2$  and  $D_3$  (Fig. 2), suggesting that other factors, in addition to season and stocking distribution patterns are affecting growth.

Since stocking density per unit of rope length is similar in the three patterns of stock distribution, biomass production (Fig. 3) follows closely the results already described for specific growth rates. In summer, packages of 20 g of *Gracilaria* disposed every 10 cm ( $D_1$ ) produce, on average, 80% more than packages of 100 g of *Gracilaria* disposed every 50 cm ( $D_3$ ). Production differences between  $D_1$  and  $D_3$  are statistically significant in summer and winter ( $p = 0.05$  in both cases). Production in treatment  $D_2$  in summer is similar to  $D_1$ , but they differ significantly in winter ( $p = 0.05$ ).

Self-shading effects probably explain the increased growth and production rates of small

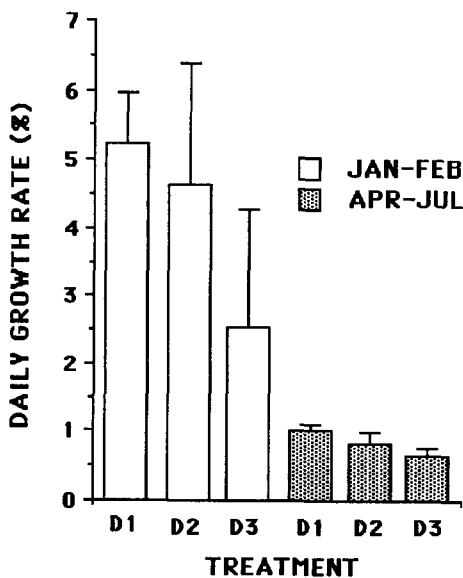


Fig. 2. Daily growth rate of *Gracilaria chilensis* on rope as a function of season and pattern of stock loading.

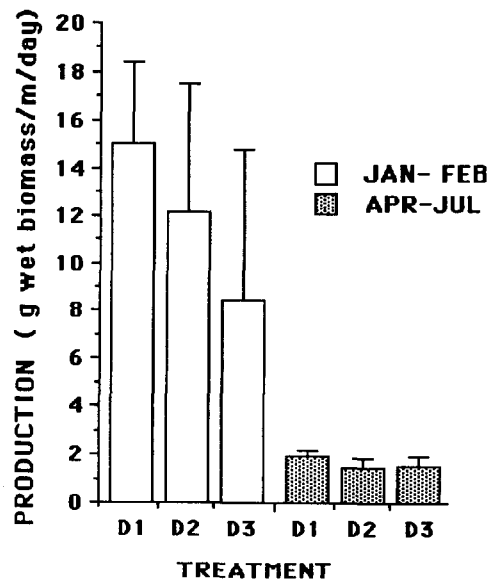


Fig. 3. Biomass production of *Gracilaria chilensis* in rope culture as a function of season and pattern of stock loading.

packages of *Gracilaria* disposed at short distances along the rope as compared with larger packages disposed at longer distances. Similar self-shading effects, which result in a reduction of growth and production, have been described in tank and laboratory cultivation of various seaweeds (e.g. Littler and Arnold, 1980; Macler and Zupan, 1991). The seasonal change in response of  $D_2$  is consistent with this interpretation. In winter, when light is limiting,  $D_2$  responds similarly to  $D_3$ . In summer, self-shading does not limit growth and  $D_2$  differs from  $D_3$ , approaching  $D_1$ .

### c. Position along the rope

The position of the packages of *Gracilaria* along the ropes also affects specific growth rates (Fig. 4). At both seasons, the end of the rope facing the landward end of the shore ( $P_1$ ) exhibited the lowest growth rate. The difference in the growth between  $P_1$  and  $P_2$  or  $P_3$  is statistically significant in summer ( $p = 0.05$  in both cases) but non-significant in winter. During summer,  $P_2$  showed average growth rate higher than  $P_3$ , but the opposite was true in winter. In neither cases the differences were statistically significant.

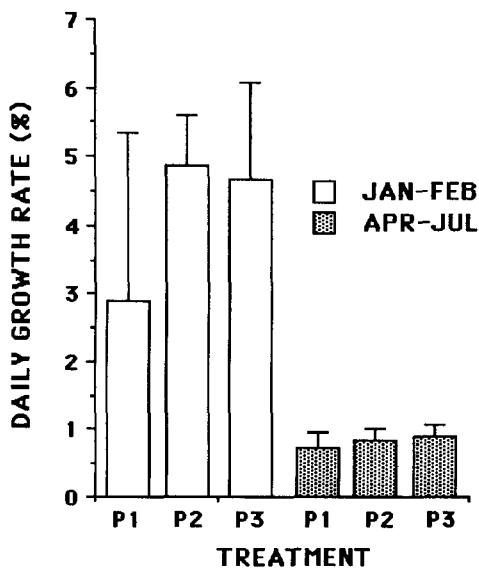


Fig. 4. Daily growth rate of *Gracilaria chilensis* in rope culture as a function of season and position along the rope.

Differences in biomass production due to position along the rope (Fig. 5) follow closely the pattern described for growth rates.  $P_1$  is less productive than  $P_2$  or  $P_3$  in summer and winter, but the differences are statistically significant only in summer ( $p = 0.05$ ). Similarly to growth rates,  $P_2$  is more productive than  $P_3$  in summer, but the opposite is true in winter. In neither cases the differences are statistically significant.

In this experiment, the plants located in  $P_1$  were facing the estuary, and therefore subjected to reduced salinities and increased sediment loads during low tides. Due to the geographic isolation of the study area, it was not possible to precisely evaluate the biological nature of the damage being suffered by the thalli in  $P_1$ . However, their reduced growth rates and low production values, as compared to the thalli in  $P_2$  and  $P_3$  are consistent with previous findings (Santelices & Fonck, 1979; Silva & Poblete, 1991) suggesting that in natural patches the peripheric thalli are affected more strongly by the abiotic environment than the inner thalli. Since in our experimental populations only the most external plants suffered this effect, it seems that its detrimental influence becomes reduced inside the patch. It is likely, however, that the amount of border affected by the abiotic en-

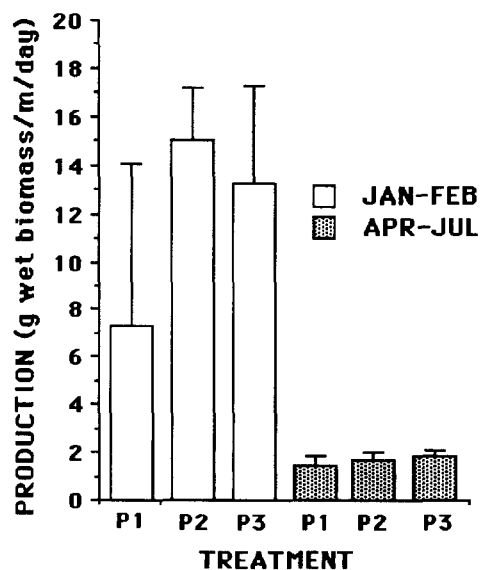


Fig. 5. Biomass production of *Gracilaria chilensis* as a function of season and position along the rope.

vironment may change seasonally and from one to other site in the estuary.

*d. The interacting effects of stocking distribution and position*

During summer, the interaction stock distribution/position affects very significantly the growth and production rates of *Gracilaria chilensis* (Fig. 6, 7). Small packages disposed at short distances ( $D_1$ ) exhibit a similar growth rate in a landward ( $P_1$ ), central ( $P_2$ ) or seaward ( $P_3$ ) position along the rope (Fig. 6; Table 1). However, for any one of the other two stock distribution patterns ( $D_2$  and  $D_3$ ), the landward position ( $P_1$ ) resulted in significantly less growth rate (Table 1) than the central ( $P_2$ ) or the seaward position ( $P_3$ ). In the case of the largest packages of *Gracilaria* disposed at the longest distances ( $D_3$ ), the central position yield the highest growth rates while for  $D_2$  the seaward position ( $P_3$ ) was the most productive (Fig. 6). In these last two cases however, the differences between  $P_2$  and  $P_3$  were not statistically significant.

The interaction stocks distribution/position also affects summer biomass production (Fig. 7).

Results follow closely those already described for growth rates.

During winter, some of the above tendencies are maintained, but others are modified (Fig. 6, 7). Small packages disposed at short distances along a rope ( $D_1$ ) grew equally well in a landward ( $P_1$ ), central ( $P_2$ ) or seaward position ( $P_3$ ). However, intermediate ( $D_2$ ) and large ( $D_3$ ) packages of *Gracilaria* grow better in the seaward position ( $D_3$ ) than in any of the two other positions, although these differences were not statistically significant (Table 1). Most of the significant differences found in winter arise from differences in stock distribution patterns ( $D_1$  versus  $D_2$  or  $D_3$ ).

The inhibition of growth exhibited by *Gracilaria* when placed in large packages in the  $P_1$  position could be related to light reduction through increased sediment trapping in the peripheric thalli. This would explain the significant effect in treatments  $D_2$  and  $D_3$  and the lack of effect on treatment  $D_1$ . In fact, the interacting effects of stock distribution and position can limit growth rates and production of *Gracilaria chilensis* in treatments  $D_2$  and  $D_3$  to the point of nullifying the seasonal production increments expected in summer (Fig. 6, 7). For example, the algae in treatment  $D_3$  and position 1 exhibit approximately

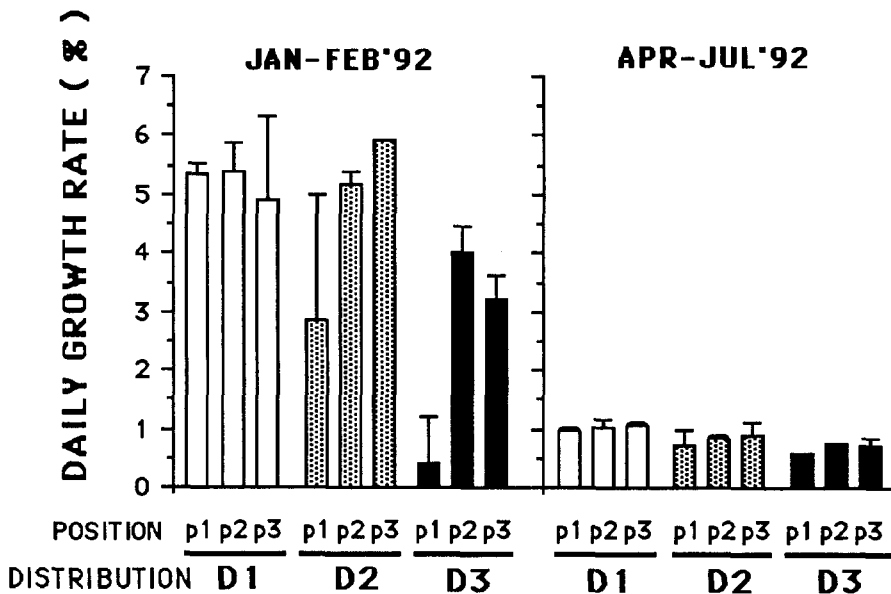


Fig. 6. Daily growth rate of *Gracilaria chilensis* as a function of season, position and stock loading along the rope.

Table 1. Significance of differences in specific growth rates among various treatments combining stock loading and position in rope culture of *Gracilaria chilensis* (NS = non significant; \* = significant at 5%; Fisher PLSD test).

Summer										
	D1P1	D1P2	D1P3	D2P1	D2P2	D2P3	D3P1	D3P2	D3P3	
D1P1	✓	N.S.	N.S.	N.S.	N.S.	N.S.	****	N.S.	N.S.	
D1P2		✓	N.S.	N.S.	N.S.	N.S.	****	N.S.	N.S.	
D1P3			✓	N.S.	N.S.	N.S.	****	N.S.	N.S.	
D2P1				✓	N.S.	****	****	N.S.	N.S.	
D2P2					✓	NS.S	****	N.S.	N.S.	
D2P3						✓	****	N.S.	N.S.	
D3P1							✓	****	****	
D3P2								✓	N.S.	
D3P3									✓	✓
Winter										
	D1P1	D1P2	D1P3	D2P1	D2P2	D2P3	D3P1	D3P2	D3P3	
D1P1	✓	N.S.	N.S.	N.S.	N.S.	N.S.	****	N.S.	N.S.	
D1P2		✓	N.S.	****	N.S.	N.S.	****	****	****	
D1P3			✓	****	N.S.	N.S.	****	****	****	
D2P1				✓	N.S.	N.S.	N.S.	N.S.	N.S.	
D2P2					✓	N.S.	****	N.S.	N.S.	
D2P3						✓	****	N.S.	N.S.	
D3P1							✓	N.S.	N.S.	
D3P2								✓	N.S.	
D3P3									✓	✓

similar growth and production rates in summer and winter. In the case of the algae in treatment  $D_2$  and  $P_1$ , their summer growth and production rates are higher than the winter values, but the differences are not statistically significant.

#### e. Effects of block distribution on production

Since the interaction stock distribution/position affects production, the planting regime of *Gracilaria* along the rope will have a most important effect on the biomass production in the farms. In the summer experiments, the ropes with treatments  $D_1$  in the landward end of the ropes produced, on average,  $41.76 \pm 7.74$  g wet weight  $m^{-1} d^{-1}$ . This value is 11% more than the ropes with the treatment  $D_3$  in such position ( $37.17 \pm 0.39$  g

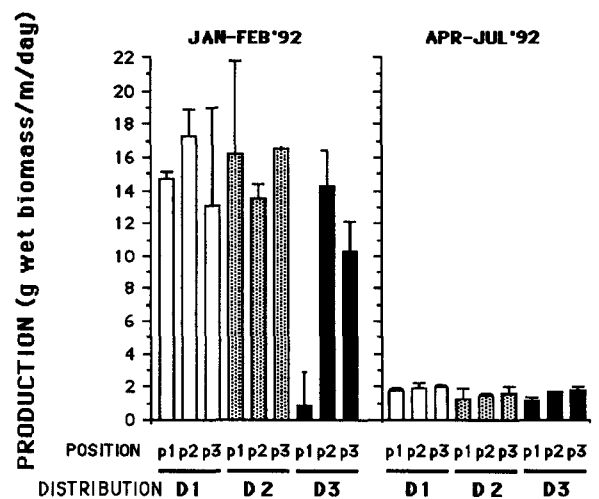


Fig. 7. Biomass production of *Gracilaria chilensis* as a function of season, position and stock loading along the rope.

wet weight  $\text{m}^{-1} \text{d}^{-1}$ ) and 26% more than those with  $D_2$  in the  $P_1$  position ( $30.81 \pm 1.31$  g of wet weight  $\text{m}^{-1} \text{d}^{-1}$ ). In winter the above differences can be as high as 30.72%, depending on the pattern of stock loading in the landward end of the rope.

## Conclusion

The overall conclusion emerging from this study is that the optimum planting distance for growth in *Gracilaria chilensis* varies according to the part of the bed being considered. Therefore, the production of this species could be increased to some extent (25–30% in our experiments) by manipulation of planting distance and stock loading in the field.

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