## Repopulation of intertidal areas with Lessonia nigrescens in northern Chile

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### Abstract

Intertidal rocky areas in northern Chile were repopulated experimentally with the brown alga *Lessonia nigrescens* using spore seeding and placement of reproductive fronds. The results were successful, and it is suggested that methods developed in the field can be done by people without special training.

### Introduction

Chilean brown seaweeds are exported as raw materials and to a lesser extent as condiment. The largest landings (Avila & Seguel, 1993; Vásquez & Fonck, 1994) have been species of *Lessonia* and *Macrocystis* which are harvested mainly in northern Chile. In addition to harvesting pressure, production of subtidal and intertidal populations has been adversely affected by the El Niño phenomenon of 1982–83 (Soto, 1985; Tomicic, 1985; Arntz, 1986).

We have explored the possibility of repopulating areas of the intertidal in order to increase production of these commercially important seaweeds. Here we report simple and inexpensive, but effective, techniques using *Lessonia nigrescens* Bory.

#### Materials and methods

#### Study area

The experimental localities were Punta Lagunillas  $(30 \circ 06' \text{S}-70 \circ 26' \text{W})$ , an exposed site, and La Herradura Bay  $(29 \circ 71' \text{S} -71 \circ 21' \text{W})$ , a protected area. The rocky intertidal at the former was dominated by *Lessonia nigrescens*, whereas at La Herradura Bay the density of *L. nigrescens* was low and the substratum dominated by crustose calcareous algae, with abundant black sea urchin.

#### Laboratory experiments

#### Spore seeding on different substrata

Maximum spore release from fronds of wild L. nigrescens was achieved by partial desiccation followed by re-immersion in seawater. The fronds were first washed and wiped with paper towelling and dried for about 12 h in darkness at 15 °C. In December, spores were released after re-immersion of desiccated reproductive fronds in 500 mL filtered (0.45  $\mu$ m) and pasteurized seawater contained in shallow plastic trays. Spores were seeded onto Velcro line (120 cm<sup>2</sup>), nylon cord (2 m long, 0.5 cm diam.), nylon mesh  $(100 \text{ cm}^2)$  and small stones  $(19-20 \text{ cm}^2)$ . Ten replica substrata for each type were inoculated with a spore suspension of about  $2 \times 10^6$  spores. The seeded substrata were incubated with aeration in small aquaria containing 500 mL enriched seawater (Provasoli, 1964), which was changed every five days. The temperature was maintained at 15 °C with an irradiance of 70  $\mu$ mol m<sup>-2</sup> s<sup>-1</sup> on a 12:12 light: dark period. Samples were scraped from substrata weekly and examined microscopically. In March one-half of these substrata were transferred to Punta Lagunillas and fixed to rocky intertidal areas by cement nails and plastic cable ties. The Kruskal-Wallis analysis of variance and multiple comparison test between treatment for ranks (Sokal & Rohlf, 1969) was used to compare sporophyte densities.

### **Field repopulation studies**

### Spore seeding experiments

One-hundred reproductive fronds were desiccated and transported to Punta Lagunillas and rehydrated in 40 L seawater for 2 h. The resulting spore suspension ( $8 \times 10^6$  spores) was introduced into the water at low tide over three 10 m<sup>2</sup> sites lacking *L. nigrescens* and sea urchins. Three control sites of comparable area without spore seeding were established about 15 m distance from the experimental area. This experiment was replicated at two sites in La Herradura Bay. Experimental and control areas were sampled monthly for recruitment.

## Frond seeding experiments

At Punta Lagunillas, 17 bundles of desiccated reproductive fronds (5 fronds) were fixed to hard substrata  $(10 \text{ m}^2, n=3)$  with concrete nails and plastic cable ties. These fronds were retained in the field for one week for spore release. Three similar areas were established as control plots, and in both control and experimental plots sea urchins were nominally absent. These areas were sampled monthly for new recruits. Thirty juveniles in a natural population were tagged in December and growth patterns and reproduction were followed at Punta Lagunillas. Holdfast diameter, number of stipes, maximum length of plants and occurrence of reproductive stipes were recorded at monthly intervals.

# Transplantation experiments with juvenile and adult plants

During February, plants were transplanted to Punta Lagunillas and La Herradura Bay. Juveniles (20-60 cm length, n = 20) were attached to natural substrate using non-toxic epoxy cement. Adult reproductive plants were tied to boulders with rubber bands and anchored to the bottom with cement nails. Individual plants were tagged, and holdfast diameters, number of stipes and maximal length of the plants were measured monthly.

### **Results and discussion**

# Laboratory experiments: spore growth on different substrata

Microscopic sporophytes were first observed following 24-d incubation, and after 44 d the density was greatest on gravel (Kruskall Wallis t=30.7, p<0.01) with  $3267\pm833$  plants cm<sup>-2</sup>. Densities were lowest on nylon cord ( $140\pm25$ ) and nylon mesh ( $24\pm3$ ) and intermediate on velcro line ( $1829\pm386$ ).

## Field work: spore seeding experiments

Two months after the experimental seeding, three plants, less than 10 cm in length, were detected at Punta Lagunillas, and these grew continuously during the following 10 months; holdfast diameter increase to 15 cm, number of stipes to 25 and maximum length to 250 cm. Five plants were recruited during the one-year observation, although none in the control plots. There was no development of plants in the seeded areas at La Herradura Bay.

## Frond seeding experiments

Following frond seeding, four sporophytes were observed in the experimental plots after the second month, and these were tagged for measurements. There was an apparent early decline in all measurements, followed by increased growth which peaked in the late fall-early winter (Fig. 1A). Within 6 mo, 30% of the plants had sori and this increased to 60% in the subsequent 6-mo period. Plants in the natural population showed a similar pattern of growth and reproduction (Fig. 1B). No recruits were observed in the control plots throughout the 12-mo observations.

## Transplantation experiments with juvenile and adult plants

Juvenile plants transferred to natural substrata did not survive beyond 15 d. There was a 50% mortality of transplanted adult plants during the first two weeks in the field. The surviving plants reattached to the substrate by their holdfasts and produced additional fronds. Three months after transplantation (September), 38% of the remaining plants were reproductive, although no recruits were found in adjoining areas during the following month.

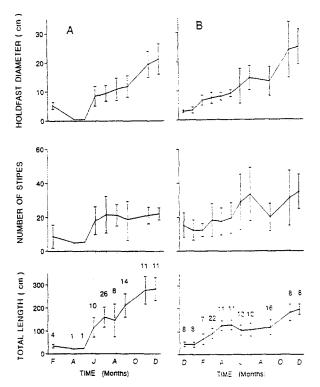


Fig. 1. (A) Frond seeding experiments and (B) natural repopulation. Temporal variation of holdfast diameter (cm), number of stipes, and maximum length (cm) of *Lessonia nigrescens*. Number of plants measured is indicated for each monthly evaluation ( $X \pm SD$ ).

It is feasible and practical to repopulate intertidal rocky areas with *Lessonia nigrescens*, and our initial results have been very encouraging. The most satisfactory results were obtained using reproductive fronds and spore seedings. The field methodology developed here is simple, an it can be done by personnel without special training. The physical and biological characteristics of the habitat proposed for repopulation must be taken into consideration. These include exposure, substrate stability and herbivory. For example, the high density of herbivores in La Herradura Bay is likely to have prevented recruitment in that area.

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