Competition between Dunaliella species at high salinity

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Introduction

The coexistence of species of phytoplankton has been of interest since Hutchinson (1961) raised the “paradox” of many species sharing the same resources. Many investigations have considered competition between microalgae of different phyla (Goldman et al., 1982; Richmond et al., 1982) or genera (Tilman, 1981). Relatively little research has concerned closely related species.

In the high-salinity waters of Hutt Lagoon, Western Australia, the main interspecific interaction appears to be between two congeneric species, Dunaliella salina Teodoresco and Dunaliella viridis Teodoresco (Chlorophyta, Volvocales). These algae coexist in the water column of the ephemeral salt lake, Hutt Lagoon, and in the man-made ponds of a research facility and pilot plant. No other algae are observed in the water column of the lake or ponds when the salinity of the water is above ca 200 g·L⁻¹. At lower salinities certain diatoms, blue-green algae (Aphanathece and filamentous forms) and dinoflagellates appear in the water column. Some of these algae occur in the bottom sediments at higher salinities, but appear to be inactive or not to interact with the water column. Various protozoa occur in the high-salinity waters at Hutt Lagoon (Post et al., 1983), but major predators of Dunaliella appear to be restricted to salinities below 200 g·L⁻¹. Some flagellate protozoa (Bodo spp.) and ciliates (Euplotes spp.) are active up to 310 g·L⁻¹, but appear to feed on bacteria and organic debris. We have observed the flagellate stage of Heteramoeba sp. and Euplotes sp. to prey on Dunaliella in laboratory cultures at a salinity of 250 g·L⁻¹, but they did not greatly affect the population dynamics of the cultures; in the experiments reported below, such predation was not observed except as noted (one case).

Interaction between D. salina and D. viridis is potentially important in the commercial production of β-carotene being developed at Hutt Lagoon (Borowitzka et al., 1984, 1985; Moulton et al., 1987). D. salina produces massive amounts of β-carotene in the high-salinity conditions of the salt lake and ponds (50–150×10⁻¹² g·cell⁻¹ β-carotene, or ca 10% dry wt). D. viridis produces 0.4–0.8×10⁻¹² g·cell⁻¹ of mixed carotenoids, or up to 0.02% dry wt. Therefore to maximize the commercial production of β-carotene, D. salina should predominate.

D. salina appears to have a higher salinity optimum for growth than does D. viridis. The two species also differ in their behavioral response to high salinity; D. viridis tends to remain at the bottom of a pond at 250 g·L⁻¹, whereas D. salina remains active in the water column at salinities up to 310 g·L⁻¹, the saturation point for sodium chloride in Hutt Lagoon.

The intensity of solar radiation is also expected to influence the competitive interaction between D. salina and D. viridis, because the accumulation of β-carotene by D. salina appears both to protect the alga from intense radiation and to increase the point of compensation between photosynthesis and respiration (Borowitzka et al., 1984; Loeblich, 1982). The behavior of the two species reflects this, D. salina migrating towards intense light and D. viridis seeking lower light intensities. Relative initial concentrations of the two species (“inoculum
size”) also influences the outcome of interspecific competition.

Taxonomic note

The taxonomic positions of the various recognizable forms of *Dunaliella* in this study are not precise (Borowitzka & Borowitzka, 1987). We consider all forms that contain relatively high amounts of β-carotene to be *D. salina* Teodoresco. The forms range from oval and almost rectangular cells ca 22 × 18 μm, to pear-shaped cells 12 × 8 μm. Other forms with swollen anterior ends and protuberances may result from different culture conditions. Under the natural conditions of the salt lake at Hutt Lagoon and our culture conditions, *D. salina* remained red with ca 50–150 × 10⁻¹² g·cell⁻¹ β-carotene. The species appears to be identical to *Dunaliella bardawil* of Ben-Amotz & Avron (1983).

The predominant forms of *Dunaliella* that do not contain high amounts of β-carotene at high salinity at Hutt Lagoon appear to be a single species which alters form according to the conditions and age of the culture. The cells vary in content of granular material and chlorophyll, and range from 14 × 10 to 8 × 6 μm. We consider these forms to be *D. viridis* Teodoresco following Masyuk (1973), on the basis of their relatively large, red-brown, protuberant eyespot, size range and color. In earlier work with Hutt Lagoon cultures Borowitzka et al. (1985) described this species as *Dunaliella parva*. The species probably matches the *D. salina* of Ben-Amotz & Avron (1983). *D. viridis* from Hutt Lagoon contains 0.4 × 10⁻¹² g·cell⁻¹ of carotenoid in young cultures and 0.8 × 10⁻¹² g·cell⁻¹ in old, stationary-phase cultures. At this level of carotenoid they do not appear “yellowish”, hence do not comply with that criterion for *D. parva*.

We have observed other species of *Dunaliella* in pond cultures below ca 150 g·L⁻¹ salinity, including *Dunaliella jacobae* and *Dunaliella bioculata*; they appear to be of minor importance.

Material and methods

All cultures were derived from the ephemeral salt lake and experimental ponds at Hutt Lagoon, W.A. The salt lake is dry in summer, fills with water in winter and is rarely diluted to salinities <200 g·L⁻¹ (Arakel, 1981; Arakel & Moulton, 1986). The experimental ponds are constructed from earthen walls on the lake bed and are maintained at >250 g·L⁻¹ salinity and 200 mm depth (Borowitzka et al., 1984, 1985; Moulton et al., 1987), and are fertilized with nitrate and phosphate. All organisms in the ponds derive from the salt lake.

Laboratory experiments were carried out in growth cabinets under controlled light and temperature. In the experiment in which light intensity was varied, light was provided from an upper and lower row of ten Sylvania cool white 20-W fluorescent tubes. Light intensity was measured using a Li-Cor LI-1935B underwater spherical quantum sensor with Li-Cor LI-185B meter. Culture medium was either derived from the ponds or chemically constituted to approximate the major anions and cations of the ponds.

A unialgal stock of *D. viridis* was produced by plating out material from Hutt Lagoon, and was used in laboratory experiments. The stocks of *D. salina* were obtained by selecting from Hutt Lagoon cultures containing <1% *D. viridis*.

Growth rate was obtained either by fitting a straight line to the log-transformed first few points of the growth curve (“initial” growth rate), or by a least-squares fit of the (not log) growth data to the logistic relationship. In the latter case, the maximum concentration of cells (“carrying capacity”) was either estimated subjectively or obtained by further iteration, and the growth rate was termed “intrinsic” growth rate. The theoretical maximum production of cells was calculated from the logistic relationship; it is the intrinsic growth rate multiplied by the carrying capacity divided by four.

Salinity was determined by temperature-compensated refractometry calibrated to Hutt Lagoon brine sampled in winter; small variations may arise from variation in the constituents of the brine.

Occurrence in the salt lake

In summer, *D. salina* and *D. viridis* exist in and be-