



Patterns of growth of juvenile pink abalone *Haliotis corrugata* fed re-hydrated natural feeds at a laboratory and a hatchery

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Abstract

The feasibility of obtaining a similar growth response from juvenile pink abalone *Haliotis corrugata* at a research laboratory and a hatchery, when using natural feeds was evaluated. Four macroalgae, *Egrelia menziesii*, *Eisenia arborea*, *Macrocystis pyrifera*, *Gracilaria* sp., and the surfgrass *Phyllospadix torreyi* were used as feeds. Response patterns of abalone were very similar at both facilities in terms of final length, weight, and survival, varying from 13.0 to 15.50 mm, 0.31 to 0.52 g, and 60.0% to 78.6%, respectively. Better growth was obtained when *E. menziesii*, *M. pyrifera*, and *Gracilaria* sp. were offered. Poor results were obtained with surfgrass. The feed conversion ratio was determined at the laboratory and did not vary significantly, ranging from 42.3 to 199.0; although a significant inverse correlation was observed with growth rate. Growth in length and weight and survival rates varied within 1.6–20.2 $\mu\text{m day}^{-1}$, 0.123–1.664 mg day^{-1} , 0.4–0.64% day^{-1} , respectively. Mean growth rate in length (14.7 $\mu\text{m day}^{-1}$) and weight (1.18 mg day^{-1}) at the hatchery were significantly higher than that obtained at the laboratory (9.4 $\mu\text{m day}^{-1}$ and 0.77 mg day^{-1}), which is most likely a consequence of more suitable water temperature at the hatchery. Mean survival rate was significantly higher at the laboratory (53.1% day^{-1}) than at the hatchery (46.1% day^{-1}).

KEY WORDS: algae, *Haliotis corrugata*, natural feeds, surfgrass

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Introduction

Pink abalone *Haliotis corrugata* and green abalone *Haliotis fulgens* represent nearly 97% of the commercial capture of the Mexican abalone fishery (Ramade-Villanueva *et al.* 1998). Local fishing cooperatives produce larvae and juvenile in hatcheries to enhance natural stocks of both species (McBride 1998).

Microalgae and macroalgae are used to feed abalone in those hatcheries. Several algal species have been evaluated as constituents in single-species diets for abalone. Day & Fleming (1992) stated that, although a single-species diet does not support sustained growth of abalone, it may be of great value when part of a mixed diet. Abalone along the Pacific coast of the Baja California Peninsula graze on kelp *Macrocystis pyrifera* and regional hatcheries use kelp as the main source of natural feed (McBride 1998). However, many algae of the subtidal zone along the Pacific coast may be used as alternative potential feed. Studies evaluating common macroalgae along the Baja California coast as feed for abalone are important for better understanding of the biology of local abalone species and improving hatchery practices (Serviere-Zaragoza *et al.* 1998, 2001).

Research on natural feeds for abalone that inhabit coastal waters of Baja California is scarce. To date, studies have been limited to juvenile *H. fulgens*. Viana *et al.* (1993, 1996) used *M. pyrifera* as a control in a feeding trial and Serviere-Zaragoza *et al.* (2001) evaluated the growth of juveniles fed *M. pyrifera*, *Eisenia arborea*, *Gelidium robustum*, and *Phyllospadix torreyi*. There are no antecedents dealing with natural feed for juvenile *H. corrugata*. Evaluation of such feed is of major relevance because pink abalone is one of the most important species for the local abalone fishery and aquaculture.

Scaling-up results obtained in research laboratories to commercial aquaculture production is rarely addressed, despite its major practical relevance. Rigorous tests that simultaneously evaluate feeding at hatchery and laboratory facilities are needed to have confidence in experimental results. This study assessed the possibility of reproducing growth response at a research laboratory and a hatchery when juvenile pink abalone were fed common algae and a surfgrass from benthic communities along the western coast of Baja California.

Materials and methods

Natural feeds

The brown algae *Egrecia menziesii*, *Eis. arborea*, and *M. pyrifera*, the red algae *Gracilaria* sp., and the surfgrass *P. torreyi* were used as feeds. At the beginning of the trials, *E. menziesii* and *Gracilaria* sp. were collected from the northern Pacific coast of the Baja California Peninsula and the other species from the central coast of the peninsula. For the brown algae, pneumatocysts were removed from the blades after harvest. Specimens were air-dried and stored in cardboard boxes at 20 °C to prevent deterioration. Feed were re-hydrated before administration. The proximal composition of the feed was performed using the methods of AOAC (1995) displayed in Table 1.

Feeding trials

Two feeding trials were carried out simultaneously. One trial was conducted for 138 days in a marine research laboratory at Bahía de La Paz and the other for 155 days in a hatchery at Bahía Tortugas (Fig. 1). The conditions of cultivation at the two facilities are listed in Table 2.

Hatchery-produced juvenile *H. corrugata* [$12.7 \text{ mm} \pm 1.84$ (SD), $0.30 \text{ g} \pm 0.10$ (SD) g] were held in 16-L fiberglass rearing tanks ($50 \times 30 \times 35 \text{ cm}$) with a concave bottom placed within large rectangular containers. Tanks were covered with 500- μm -pore-size Nyltex mesh to prevent escape. Before

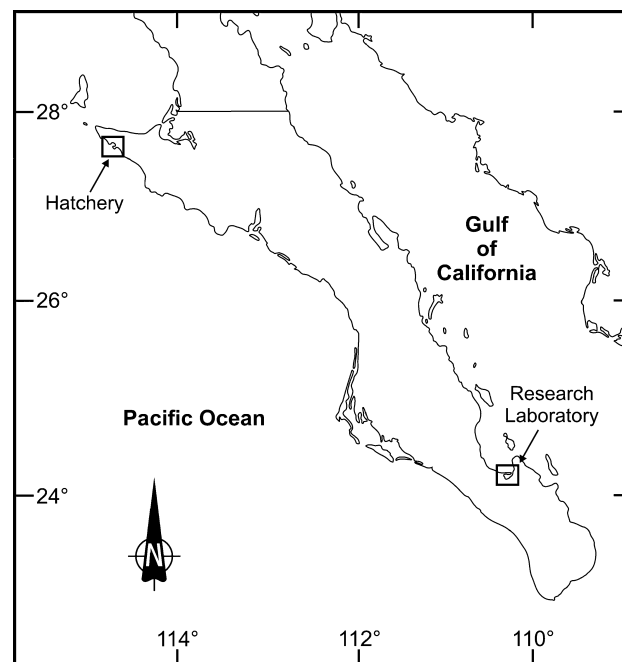


Figure 1 Locations of the research laboratory and the hatchery used for the investigation.

beginning the trials, ANOVA was conducted to ensure homogeneity of initial size and weight among treatments. Three experimental tanks per feed treatment with 40 juveniles per tank were used. Treatments were randomized among tanks ($n = 15$). Rehydrated feeds were given *ad libitum* in the afternoon at 2-day intervals after uneaten food and faeces were removed. Microalgae growing on the inner walls of the tanks were removed twice a week with a brush.

Data collection

Shell length was measured with a Vernier caliper and wet body weight with an electronic balance (nearest 0.001 g) approximately once each month during the trials. Dead specimens were counted and replaced with similar-sized marked abalone to maintain a standard density. Replacements were excluded from sampling. Absolute growth rate on

Feed	Crude protein	Ether extract	Crude fibre	Ash	NFE	Energy (J g^{-1})
EM	101.6 ± 1.4	3.7 ± 0.3	51.0 ± 1.7	315.6 ± 0.4	528	11579.9 ± 57.1
EA	124.5 ± 0.5	7.4 ± 0.5	33.1 ± 2.2	303.3 ± 1.4	531	10689.3 ± 238.2
MP	110.6 ± 2.6	3.7 ± 0.3	32.1 ± 1.2	411.3 ± 0.9	442	10571.3 ± 270.0
GR	194.0 ± 2.4	0.6 ± 0.1	17.6 ± 1.2	283.5 ± 1.9	504	13584.9 ± 147.8
PT	145.7 ± 1.5	1.5 ± 0.1	93.5 ± 2.9	399.1 ± 1.4	360	10590.9 ± 196.4

NFE, nitrogen-free extract.

Table 1 Proximal composition ($\text{g kg}^{-1} \pm \text{SD}$ of dry matter) of the natural feeds: *Egrecia menziesii* (EM), *Eisenia arborea* (EA), *Macrocystis pyrifera* (MP), *Gracilaria* sp. (GR), and *Phyllospadix torreyi* (PT)

Table 2 Conditions of culturing at the research laboratory and the hatchery during the assays

Features	Research laboratory	Hatchery
Water treatment	Filters: sand, ultraviolet (UV), 1- μ m cartridge. Aeration: vigorous with air stones	Filters: sand, UV, 10- μ m cartridge. Aeration: vigorous with air stones
Daily water exchange	90 mL min ⁻¹ (800%)	90 mL min ⁻¹ (800%)
Dissolved oxygen (mg L ⁻¹)	>6.9	>7.1
Salinity (g L ⁻¹)	38.3 \pm 0.5	35.0 \pm 1.1
Temperature (°C)	20 \pm 1.0 (controlled)	13.4–26.4 (uncontrolled)

a per day basis was calculated for shell length and wet body weight at the end of the trials.

Feed conversion ratio (FCR) was determined for the trial at the laboratory and calculated as: dry feed consumed/wet weight gain per animal per day (Britz 1996). Control tanks without abalone were used to estimate feed loss in seawater. Consumption was calculated on a dry weight basis following Uki & Watanabe (1992), where $FC = (GS/100) - R$, where G is the weight of feed provided each day, S is the correction factor (as a percentage) from the control tanks, and R is the remaining food (in grams) after feeding the abalone. For calculation of weight gain, the correction by Kitabayashi *et al.* (1971) was used to compensate for abalone mortality during the trial.

Statistical analysis

For each trial, ANOVA followed by a Tukey post-hoc mean comparison test was performed at the end of the experiments to determine if significant differences in shell length, weight, FCR, and survival occurred. Correlation analysis was also carried out between FCR and shell length.

Differences in growth and survival between the facilities were tested using multi-factor ANOVA. Daily growth and survival rate were used for comparison, as they constitute an adequate, standardized measurement to compensate for the difference in duration between the experimental trials. The arc-sine transformation was used for survival values previous to analysis. Significance level was set to $P < 0.05$. Procedures available in STATISTICA 6.0 were used to perform the statistical analyses.

Results

The growth pattern of abalone was very similar at the two facilities (Fig. 2). At the end of the experiments, significant differences in final length (laboratory: $F = 19.23$, $P = 0.000$; hatchery: $F = 4.3$, $P = 0.027$) and weight (laboratory: $F = 19.56$, $P = 0.000$; hatchery: $F = 6.68$, $P = 0.007$) were observed, where *E. menziesii*, *M. pyrifera*, and *Gracilaria* sp.

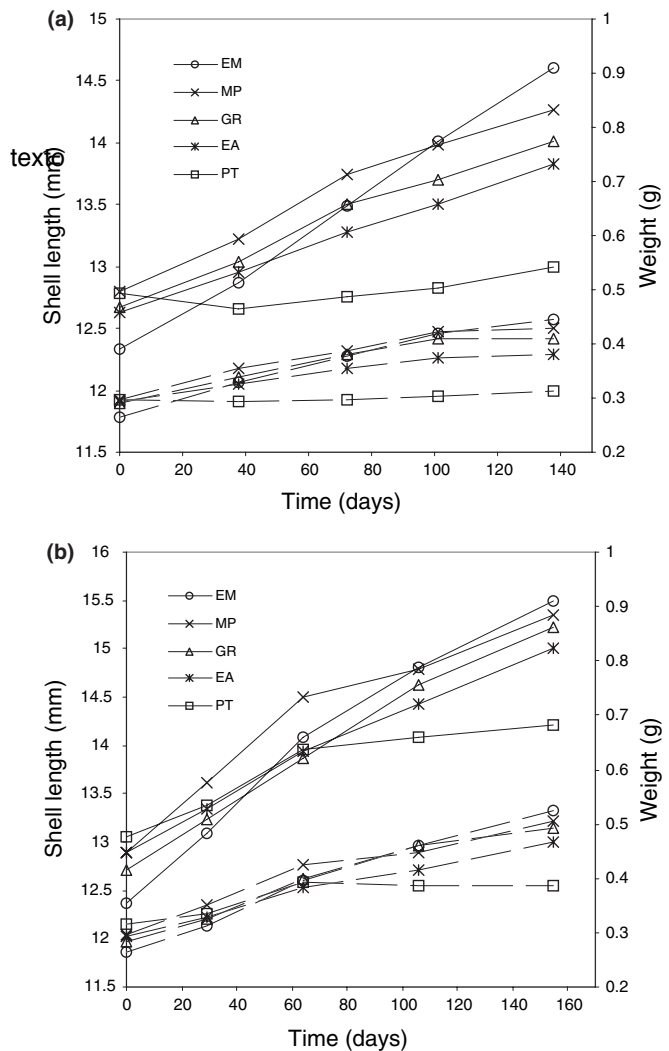


Figure 2 Growth response in shell length (solid line) and weight (dashed line) of juvenile *Haliotis corrugata* fed *Egrecia menziesii* (EM), *Macrocystis pyrifera* (MP), and *Gracilaria* sp. (GR), *Eisenia arborea* (EA) and *Phyllospadix torreyi* (PT) raised in a research laboratory (a) and a hatchery (b).

feeds produced better growth in both trials (Table 3). Poor results were consistently obtained at both facilities when surfgrass (*P. torreyi*) was provided. *Eisenia arborea* produced

Table 3 Mean values (\pm SE) of final shell length, weight, feed conversion ratio (FCR), and survival of juvenile *Haliotis corrugata* at the laboratory and the hatchery

Feed	Length (mm)		Weight (g)		FCR	Survival (%)	
	Laboratory	Hatchery	Laboratory	Hatchery	Laboratory	Laboratory	Hatchery
EM	14.61 \pm 0.14 ^a	15.50 \pm 0.43 ^a	0.45 \pm 0.003 ^a	0.52 \pm 0.034 ^a	42.3 \pm 10.4 ^a	77.0 \pm 2.08 ^{ab}	67.6 \pm 8.8 ^a
MP	14.26 \pm 0.20 ^{ab}	15.35 \pm 0.26 ^a	0.43 \pm 0.020 ^{ab}	0.47 \pm 0.009 ^{ab}	56.0 \pm 14.2 ^a	60.0 \pm 2.88 ^a	74.3 \pm 3.4 ^a
GR	14.02 \pm 0.14 ^{ab}	15.22 \pm 0.15 ^{ab}	0.41 \pm 0.010 ^{ab}	0.50 \pm 0.023 ^a	119.5 \pm 72.1 ^a	88.3 \pm 1.66 ^b	78.6 \pm 0.6 ^a
EA	13.83 \pm 0.05 ^b	15.01 \pm 0.10 ^{ab}	0.38 \pm 0.006 ^b	0.49 \pm 0.018 ^a	72.1 \pm 36.7 ^a	72.0 \pm 5.56 ^{ab}	63.3 \pm 4.9 ^a
PT	13.00 \pm 0.08 ^c	14.21 \pm 0.06 ^b	0.31 \pm 0.009 ^c	0.39 \pm 0.001 ^b	199.9 \pm 140.3 ^a	69.3 \pm 8.29 ^{ab}	73.3 \pm 2.7 ^a

Different superscripts indicate significant differences from ANOVA between feeds tested: *Egrecia menziesii* (EM), *Macrocystis pyrifera* (MP), *Gracilaria* sp. (GR), *Eisenia arborea* (EA), and *Phyllospadix torreyi* (PT).

slightly better results than surfgrass (Table 3). There were no significant differences in FCR between feeds at the laboratory ($F = 1.2$, $P = 0.361$). However, FCR values were significantly correlated with growth ($F = 4.9$, $P = 0.047$). Lower FCR values were related to high growth rates.

There was a general trend for the effect of feeds on growth to be more clearly distinguishable in the trial conducted at the laboratory. This was particularly evident for final survival, as no significant differences were determined between feeds at the hatchery ($F = 1.46$, $P = 0.283$), but significant differences between extreme survival rates occurred at the laboratory ($F = 4.70$, $P = 0.021$), which corresponded to the *Gracilaria* sp. and *M. pyrifera* treatments (Table 3).

Results from the multi-factor ANOVA indicated that there was no significant interaction between the facilities and feeds affecting growth rate in length ($F = 0.54$, $P = 0.704$), weight ($F = 0.29$, $P = 0.879$) and survival rate ($F = 2.83$, $P = 0.052$). Growth rate (length and weight) and survival rate were significantly related to the facility where the trials were conducted ($F = 67.47$, $P = 0.000$; $F = 35.65$, $P = 0.000$; and $F = 11.07$, $P = 0.003$, respectively), and the feed used ($F = 48.70$, $P = 0.000$; $F = 34.03$, $P = 0.000$; and $F = 3.61$, $P = 0.022$, respectively).

Mean growth rate of abalone (length 14.7 $\mu\text{m day}^{-1}$, weight 1.18 mg day^{-1}) at the hatchery was higher than at the laboratory (9.4 $\mu\text{m day}^{-1}$ and 0.77 mg day^{-1}) (Table 4). There were no significant differences in growth rates between the *E. menziessi* feed used at the laboratory and *E. menziessi*, *Gracilaria* sp., *M. pyrifera*, and *Eis. arborea* feed used at the hatchery. Surfgrass at the hatchery resulted in growth rate similar to the lower ones obtained at the laboratory. Mean survival rate was higher at the laboratory (53.1% day^{-1}) when compared with what occurred at the hatchery (46.1% day^{-1}). The lower survival rates were more discernible, corresponding to *E. menziessi* and *Eis. arborea* at the hatchery and *M. pyrifera* at the laboratory.

Table 4 Mean values (\pm SE) of growth and survival rates of juvenile *Haliotis corrugata* obtained at the hatchery (H) and laboratory (L)

Facility	Diet	Growth rate ($\mu\text{m day}^{-1}$)	Growth rate (mg day^{-1})	Survival (% day^{-1})
H	EM	20.2 \pm 0.9 ^a	1.664 \pm 0.11 ^a	0.43 \pm 0.057 ^b
L	EM	16.5 \pm 0.1 ^{ab}	1.304 \pm 0.01 ^{ab}	0.52 \pm 0.040 ^{ab}
H	GR	16.3 \pm 1.4 ^{ab}	1.344 \pm 0.14 ^{ab}	0.50 \pm 0.004 ^{ab}
H	MP	15.9 \pm 1.5 ^{ab}	1.336 \pm 0.13 ^{ab}	0.47 \pm 0.022 ^{ab}
H	EA	13.7 \pm 0.6 ^{bc}	1.118 \pm 0.09 ^{bc}	0.40 \pm 0.031 ^b
L	MP	10.6 \pm 0.9 ^{cd}	0.967 \pm 0.01 ^{bcd}	0.43 \pm 0.020 ^b
L	GR	9.7 \pm 0.9 ^{cd}	0.848 \pm 0.07 ^{bcd}	0.64 \pm 0.012 ^a
L	EA	8.7 \pm 0.6 ^{cd}	0.621 \pm 0.03 ^{cd}	0.55 \pm 0.015 ^{ab}
H	PT	7.4 \pm 0.3 ^d	0.442 \pm 0.02 ^{cd}	0.47 \pm 0.017 ^{ab}
L	PT	1.6 \pm 1.3 ^e	0.123 \pm 0.15 ^e	0.50 \pm 0.060 ^{ab}

Different superscripts indicate significant differences between the facilities and the feeds tested: *Egrecia menziesii* (EM), *Gracilaria* sp. (GR), *Macrocystis pyrifera* (MP), *Eisenia arborea* (EA), and *Phyllospadix torreyi* (PT).

Discussion

Feeding local species of macroalgae and surfgrass to juvenile *H. corrugata* produced growth in very similar response patterns at a laboratory and a hatchery. This indicates the feasibility for reproducing growth response patterns of the pink abalone, regardless of differences in protocols used at either facility.

Higher growth rates at the hatchery were attributed to more suitable water temperature (Leighton 1974). Water temperature at the laboratory was maintained at 20–22 °C following Leighton (1974). However, temperature in the hatchery was not controlled, and progressively increased from 13.4 to 26.4 °C during the trial. Recently, Díaz *et al.* (2006) established that the preferred temperature range for juvenile *H. corrugata* is 22–26.3 °C.

A less restricted filtering system at the hatchery could also contribute to higher growth rates. Benthic microflora plays an important role in the nutrition requirements of abalone (Kawamura *et al.* 1998). Larger particles could promote

bacteria, benthic diatoms, blue-green algae or juvenile algae on macroalgae and surfgrass, which in turn, enrich the nutritional value of the natural feeds.

Species of abalone vary considerably in growth rate when fed macroalgae. For example, Simpson & Cook (1998), working with *Haliotis midae*, estimated growth rates at 15–53 $\mu\text{m day}^{-1}$ and Upatham *et al.* (1998), working with *Haliotis asinina*, estimated growth rates at 2–70 $\mu\text{m day}^{-1}$. This investigation provided the first estimates of growth rates and FCR for juvenile *H. corrugata* fed natural feeds. The growth rate varied from 1.6 to 20.2 $\mu\text{m day}^{-1}$. There was no evidence of the FCR being affected by the feeds, probably as a consequence of the high variance of FCR. Nevertheless, the macroalgae that produced better growth rates significantly tended to produce lower FCR values, which indicates better nutritional value and feed efficiency.

Estimates of growth for *H. corrugata* are lower than those reported for species of abalone that have been studied more extensively. Rates as high as 80 and 174 $\mu\text{m day}^{-1}$ were observed for juvenile *Haliotis discus* fed fresh algae of high nutrient value (Uki 1981; Uki *et al.* 1986). Juvenile *H. fulgens* grew at 120 and 140 $\mu\text{m day}^{-1}$ when provided with *Egrecia* sp. (Leighton & Peterson 1998). Growth rates of *H. midae* were generally higher when fed mixed or rotation diets (Simpson & Cook 1998). The lower growth rates observed for *H. corrugata* in this study could be partially a consequence of using rehydrated rather than fresh algae. Given the higher growth rates for related species when fed fresh natural diets, it is reasonable to expect that the growth rate of the pink abalone could significantly increase when fed fresh algae. Further investigation must be conducted to prove this hypothesis.

There was no apparent relation between the nutrient content of the macroalgae and surfgrass and the growth rate of juvenile *H. corrugata*. Mercer *et al.* (1993) did not find a relation between macronutrient content of macroalgae and growth rate in *Haliotis tuberculata* and *H. discus*; the authors suggested that differences in growth could be explained by the micronutrient contents (e.g. amino acids and fatty acids) in the algae. There is some evidence that high content of C₂₀ polyunsaturated fatty acids in *E. menziesii* stimulates growth of *H. fulgens* (Nelson *et al.* 2002).

The nutritional value of feed depends on many factors, such as nutrient composition, bioavailability, palatability, and digestibility (Mercer *et al.* 1993). In our study, the laminarian brown algae *E. menziesii* and *M. pyrifera* proved to be generally better feed for abalone in both trials. *Egrecia menziesii* was particularly superior in promoting growth. Algae of the order Laminariales have high nutritious value

for some abalone. In earlier growth studies of juvenile *H. discus*, approximately 60 species of brown, red, and green algae were evaluated, where 12 species of algae had high dietary value including order Laminariales (*Costaria*, *Undaria*, *Laminaria*, *Alaria*, *Eisenia*, and *Kjellmaniella*), *Desmarestia* spp., *Chondria* and *Enteromorpha* (Uki 1981; Uki *et al.* 1986).

Egrecia sp. produced the best growth when provided to *H. fulgens* (Leighton & Peterson 1998). Serviere-Zaragoza *et al.* (2001) provided native algae and surfgrass species of the coast of the Baja California Peninsula to *H. fulgens* and found that nutritional value of the brown algae *M. pyrifera* is higher than *E. arborea*, *G. robustum*, and *P. torreyi*. This is similar to results observed in the present study, suggesting that brown algae of the order Laminariales (*E. menziesii* and *M. pyrifera*) have high nutritional value for juvenile pink abalone.

Studies in Japan and North America indicate strong preferences of species of abalone for Laminariales macroalgae that form kelp forests (Leighton 1966, 1968; Uki *et al.* 1986). Along the coast of California, adult abalone accept a wide variety of brown algae, and in smaller proportion, red and green algae (Leighton 2000); e.g. *Haliotis rufescens* prefers *Macrocystis*, *H. fulgens* and *Haliotis cracherodii* prefer *Egrecia*, and *H. corrugata* prefers both brown macroalgae (Leighton & Boolootian 1963; Leighton 2000).

Our results indicate that feeding juveniles with red algae, *Gracilaria* sp., also produced good growth indicating that this species constitutes a good alternative for feeding during the nursery stage of *H. corrugata*. Simpson & Cook (1998) found that the red algae *Porphyra capensis* produced better growth of South African abalone *H. midae* compared with growth using the kelp *Ecklonia maxima* or *Laminaria pallida*. These results were obtained although the red algae are less abundant than kelp in the local coastal communities. The red algae *Gracilaria chilensis* is considered an excellent feed for *Haliotis iris* and is commercially available as a dry product (Clarke & Creese 1998). Along Australian and New Zealand coasts, abalone show marked preferences for red algae, forming the main ingredient in their natural diet (Poore 1972; Shepherd 1975; Shepherd & Steinberg 1992).

In this study, the surfgrass *P. torreyi* produced poor results. The role that surfgrass play in the nutrition of abalone species is unclear. When provided several choices, abalone does not consume surfgrass. Still, surfgrass has been occasionally found in the guts of California abalone species (Leighton 2000). As high nutrient content is normally associated with higher consumption of a feed, the uncommon occurrence of surfgrass in the gut content has been

interpreted as indicative of low nutrient content (Leighton 2000). Serviere-Zaragoza *et al.* (2001) also found that surfgrass is a low-nutrient feed for cultivated populations of abalone in Baja California. Yet, according to Guzmán del Prío *et al.* (2003), this surfgrass is frequently part of the natural diet of green abalone in Baja California. Further research must be conducted to fully understand the role of the surfgrass and macroalgae in the nutrition of *H. corrugata*.

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