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Early juvenile growth of the abalone *Haliotis australis* in culture

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Abstract

Experimentally cultured juvenile *Haliotis australis* were fed different diets and their growth was measured. For juveniles between 3 and 6 mm in length the best growth was achieved on diatoms but macroalgal and artificial diets were also suitable for sustaining growth. The size at which the diet should be changed from microalgae to macroalgae was determined as 7 mm. Two batches of juveniles grown in a V-shaped tank rearing system averaged 15.9 ± 0.53 and 17.5 ± 0.69 mm in length after 13 and 12 months respectively. Methods to enhance the early juvenile growth are discussed.

Keywords: *Haliotis australis*; weaning; growth; abalone.

Introduction

In New Zealand, research into abalone culture began in the early 1980's with work on *Haliotis iris*, the black-footed paua, (Tong 1982) and the first commercial farming of this species began in 1987 (Henriques *et al.* 1989). Commercial operators would prefer to farm an abalone with a lighter coloured foot and *H. australis*, the yellow-foot paua, has been considered a suitable species. The feeding (Poore 1972a), growth (Poore 1972b), and reproduction (Poore 1973; Wilson & Schiel 1994) of adult *H. australis* have been investigated but little work has been done on juveniles because of their low abundance in the field (McShane *et al.* 1994). Research on *H. australis* at the NIWA Aquaculture Research Centre in Wellington has led to the development of techniques to bring the broodstock into ripe condition, to spawn them, and to rear the larvae (Moss submitted).

Newly settled abalone can ingest diatoms as early as 2 days after settlement (Norman-Boudreau *et al.* 1986), a food which can be part or all of their diet up to and into adulthood (Paul *et al.* 1976). In the wild adult *H. australis* feed preferentially on the rhodophytes *Pterocladia lucida* and *Hymenocladia lanceolata* (Poore 1972a) but nothing is known about the juvenile diet. It is not feasible to grow or harvest sufficient quantities of these seaweeds for culturing *H. australis* but the red seaweed *Gracilaria chilensis* is considered suitable as it can be readily cultured and is eaten by juvenile abalone (Tong & Moss 1992). Manufactured diets (Hahn 1989) may also be suitable. For a farmed abalone it is important to enhance the growth at all stages and therefore information is required on the dietary requirements of each size group. This study was carried out to investigate the growth of early juvenile *H. australis* in culture and to determine the diets most conducive to fast growth for different sized juveniles.

Materials and methods

Growth in V-shaped tanks

Adult *H. australis* collected from the Wellington south coast between March and May in 1991 and 1992 were held at the NIWA Aquaculture Research Centre, and brought into spawning condition

using elevated temperatures and *ad libitum* feeding of mixed seaweeds (Moss submitted). The broodstock were stimulated to spawn with hydrogen peroxide (Tong *et al.* 1992) in October 1991 (batch 1) and September 1992 (batch 2). The larvae were reared in 500 l flow-through tanks (Tong & Moss 1992) and at 10 days post-hatch, were settled onto diatom-coated (*Navicula* sp.) acrylic plates in 2m long V-shaped tanks (Tong & Moss 1992) at ambient water temperature (10°–18°C). When the juveniles reached about 5 mm, the rhodophyte *G. chilensis* was added to the tanks to supplement the diatoms.

Growth was monitored for 12 months following settlement, by measuring *in situ* the length of 100–250 juveniles, sampled at random and at irregular intervals. All measurements are presented as mean lengths ± 1 se. During the 12 months some of the juveniles (<1%) were removed for short-term growth and feeding experiments (see below) and not replaced. These removals reduced the density of juveniles in the tanks from about 1 juvenile per 28 cm^2 of tank surface to about 1 juvenile per 29 cm^2 but this should not have greatly biased calculations of growth in the tanks.

Growth on diatoms and seaweed

Growth of juvenile *H. australis* on two diets, microalgae (diatoms) and macroalgae (*G. chilensis*) was compared. Juvenile *H. australis* of 3 mm, 5 mm, 7 mm, and 9 mm length (with a range of ± 0.5 mm), were taken from 1 tank (batch 2) five months after settlement, tagged and assigned to 24 x 2 l flat-bottomed plastic containers. The tags were small pieces of coloured plastic attached to the shell with cyanoacrylate "super" glue (brand name "Loctite Prism 401"). The containers, which had a surface area of about 750 cm^2, were supplied with 18° ± 1°C seawater, filtered to the 1-5 micron range, at a flow rate of about 0.1 l min^{-1}. All treatments were maintained under a 12:12 photoperiod.

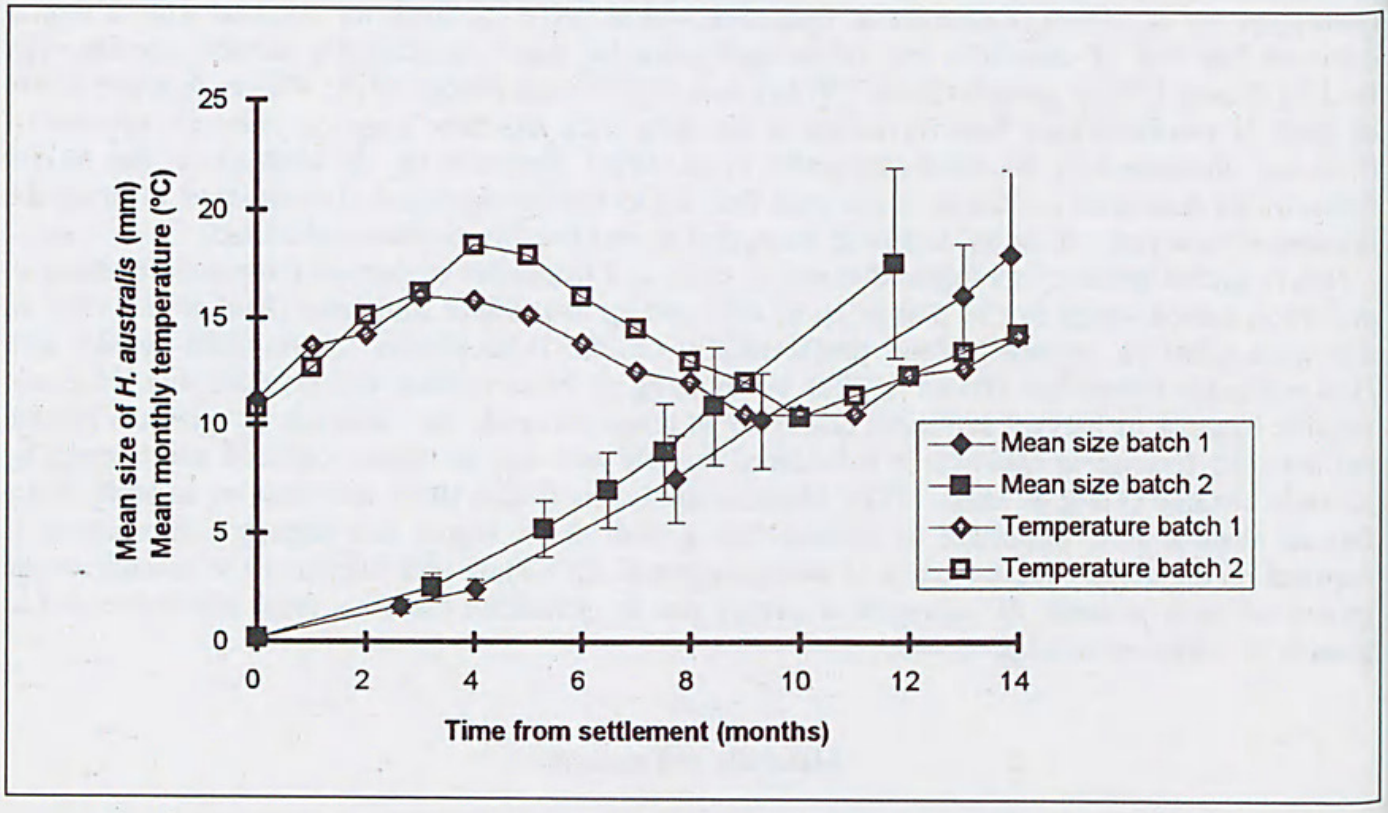

Figure 1. First year growth of juvenile *Haliotis australis* in semi-commercial V-shaped tanks at ambient temperatures at Mahanga Bay, Wellington. Batch 1 settled (September 1991) in one tank and batch 2 settled (October 1992) in two tanks. (All size measurements ± 1 S.D.).

Four individually tagged *H. australis* from 1 size group were assigned to each container with 3 replicate containers for each size group. The 24 containers (4 size groups x 2 diets x 3 replicates) were randomly positioned in an 8 x 3 grid.

In the microalgal diet, one 225 cm^2 acrylic plate, heavily coated with cultured diatoms (*Navicula* sp.) (Tong *et al.* 1992) was placed in each container. As the plates were grazed, further cultured diatoms were settled onto the plates in the containers to ensure a constant supply of food. Where the diatoms were grazed faster than they could be supplemented, the plates were substituted with others on which diatoms had been cultured. Care was taken to ensure that the *H. australis* transferred naturally from old plates to new. In the macroalgal treatment clean plates were placed in the containers along with whole fronds of the red seaweed *G. chilensis*. More *G. chilensis* was added during the experiment to ensure that the juvenile *H. australis* were fed *ad libitum*. The experiment was run for 30 days before the juveniles were remeasured and growth increments calculated.

Growth on diatoms, chopped seaweed and a manufactured diet

Growth of juvenile *H. australis* fed three diets was compared. The diets were: diatoms (*Navicula* sp.); *G. chilensis* which had been finely chopped in a domestic blender; and a powdered manufactured diet, brand name Makara (Walker 1993). Thirty-six 3 mm, 6 mm, and 9 mm (with a range of ± 0.5 mm) *H. australis*, were taken from the second tank (batch 2), tagged (as above). Four individually tagged juveniles of one size group were randomly assigned to each 2 l plastic container and there were three replicates of each treatment. The 27 containers (3 size groups x 3 diets x 3 replicates) were randomly positioned in a 9 x 3 grid and conditions were maintained as described above.

The microalgal (diatom) diet was fed as described above. The containers with *G. chilensis* were cleaned by siphoning every 3–4 days and the freshly chopped seaweed added and allowed to settle. Likewise, the manufactured diet was added every 3–4 days and allowed to settle on plates after the containers had been siphoned.

The experiment was terminated after 29 days and the juveniles remeasured and growth increments calculated and compared. ANOVA's were used to test treatment effects with Student-Newman-Keul (SNK) tests used to test differences between specific treatment means.

Results

Growth in V-shaped tanks

The juveniles settled in the V-shaped tanks in 1991 (batch 1) were first measured after 11 weeks when they had mean size of 1.6 ± 0.03 mm and again after 17 weeks when they were 2.6 ± 0.05 mm (Fig. 1). Those settled in 1992 (batch 2) reached a mean size of 2.6 ± 0.08 mm after 14 weeks. Growth rates over the first 3–4 months were between 20 and 30 microns per day for both batches. In both years high mortality was observed at about 4–5 weeks post-settlement, when there was a major decrease in the numbers of post-larvae in the tanks. This was confirmed when empty shells were collected from the bottom of the V-shaped tanks and measured. Their lengths ranged between 0.24 and 1.28 mm in length (mean 0.80 ± 0.05 mm). The first respiratory hole formed when the juveniles were 1.7 ± 0.01 mm in length (c. 8–10 weeks).

Growth rates between measurements for the first 6 months of monitoring ranged from 18 to 45 microns per day (batch 1) and from 24 to 49 microns per day (batch 2). Only after 6 months of age when the *H. australis* were had reached a mean length of about 7–8 mm did growth rates exceed 50 microns per day. Growth rates between measures for the remainder of the year ranged between 51–60 microns per day (batch 1) and 56–75 microns per day (batch 2). Batch 1 averaged 15.9 ± 0.53 in length (40 microns per day) after 13 months, but batch 2 grew faster over the first year and averaged 17.5 ± 0.69 mm (47 microns per day) after 12 months.

Table 1. Mean growth increments (mm ± 1 s.e.) and percent survival for 4 size groups of juvenile *H. australis* fed on either diatoms or *Gracilaria chilensis* over a 30 day trial period at 18°C. < or > indicates a significant difference between diets, = indicates no significant difference using the Student-Newman Keul (SNK) test

Size group:	Treatment: Diatoms	*Gracilaria chilensis*	SNK test
3 mm	1.20 ± 0.094 (75%)	1.54 ± 0.089 (92%)	D=G
5 mm	2.36 ± 0.101 (100%)	1.93 ± 0.134 (100%)	D=G
7 mm	2.68 ± 0.113 (100%)	3.27 ± 0.125 (100%)	D<G
9 mm	1.70 ± 0.107 (92%)	2.90 ± 0. 251 (92%)	D<G

Growth on diatoms and seaweed

The juvenile *H. australis* grew between 1.0 and 4.2 mm over the 30 days of the trial, with treatment means ranging from 1.20 ± 0.094 to 3.27 ± 0.125 mm (Table 1). Some mortalities occurred with both diets (Table 1) so the analyses of growth were done using data for only three *H. australis* per replicate. In those where all four had survived, data for one individual selected at random was removed to ensure the analyses remained balanced.

There was a significant difference in growth increment with size ($df = 3$; $F = 17.68$; $p = 0.001$) and there was a significant diet*size interaction ($df = 3$; $F = 12.99$; $p = 0.001$). The SNK tests showed there were no significant differences in growth of the 3 mm or 5 mm groups of *H. australis* fed the two diets (Table 1). However the 7 mm and 9 mm *H. australis* fed *G. chilensis* grew significantly faster than those fed diatoms.

Growth on diatoms, chopped seaweed and a manufactured diet

The mean growth of juvenile *H. australis* in all treatments was between 0.60 mm and 2.81 mm over the 29 days of the trial (Table 2).

Some mortalities did occur, particularly in the treatments which were being cleaned and fed every 3–4 days (Table 2). Analyses of growth increments used only three *H. australis* per replicate as explained above.

There were significant differences with diet ($df = 2$; $F = 15.15$; $p = 0.000$) and size ($df = 2$; $F = 16.27$; $p = 0.012$) and a significant diet*size interaction ($df = 4$; $F = 3.05$; $p = 0.044$). The SNK tests showed that the 3 mm and 6 mm *H. australis* fed on diatoms had significantly greater growth increments than the same sized juveniles fed either the finely chopped *G. chilensis* or the manufactured diet (Table 2). However there were no significant differences in the growth increments of the 9 mm *H. australis* fed any of the three diets.

Table 2. Mean growth increments (mm ± 1 s.e.) and percent survival for 3 size groups of juvenile *Haliotis australis* fed on either diatoms, finely chopped *Gracilaria chilensis*, or manufactured diet (brand name "Makara") over a 29 day trial period at 18°C. < or > indicates a significant difference between diets, = indicates no significant difference using the Student-Newman Keul (SNK) test

Size group:	Treatment: Diatoms	Chopped *G. chilensis*	Manufactured diet	SNK test
3 mm	1.83 ± 0.124 (100%)	0.78 ± 0.206 (83%)	0.60 ± 0.231 (67%)	D>G=M
6 mm	2.81 ± 0.111 (100%)	1.38 ± 0.238 (75%)	1.78 ± 0.213 (100%)	D>G=M
9 mm	2.40 ± 0.148 (100%)	2.20 ± 0.212 (100%)	1.93 ± 0.198 (100%)	D=G=M

Discussion

In culture systems, diatoms are the principle food source for post-larval and early juvenile abalone up to a size of about 5–10 mm (Ebert & Houk 1984; Hahn 1989; Hooker & Morse 1985; Seki 1980). This study showed there was no difference in growth and survival of 3 mm or 5 mm juvenile *H. australis* fed either diatoms or whole *G. chilensis*. However, growth on an artificial diet and on finely chopped *G. chilensis* was significantly slower than on the diatoms. The small (3–6 mm) *H. australis* fed whole *G. chilensis* grazed over the surface of the seaweed feeding on the epiphytes and diatoms associated with the surface of the seaweed and ate little of the seaweed itself. This type of feeding behaviour has previously been observed for juvenile *H. iris* fed epiphytised *G. chilensis* (Pickering 1990).

Larger juveniles are fed seaweeds because they require more diatoms than can easily be produced (Uki *et al.* 1981; Ebert & Houk 1984). The 7 and 9 mm juvenile *H. australis* fed *G. chilensis* ate the seaweed as well as the epiphytes growing on it. The 9 mm juveniles in the second trial showed no significant differences in growth between the three diets fed, however the 9 mm juveniles in the first trial fed diatoms grew significantly slower than those fed *G. chilensis*. A possible explanation for the slower growth is that there were insufficient diatoms for the number and size of juveniles in the first trial. Additional diatoms that were added to plates and allowed to settle were rapidly consumed. In the second trial sufficient diatoms were available so they were not limiting.

The calculated growth rates of *H. australis* in the V-shaped tanks were generally lower (18–75 microns per day) than in the experimental trials fed either diatoms (40–100 microns per day) or *G. chilensis* (50–110 microns per day). Higher overall water temperature and better availability of suitable food probably contributed to the faster growth of the juveniles in the small scale trials. Temperature may also have accounted for the differences in growth seen between the two batches of juveniles in the V-shaped tanks as the ambient temperatures in 1992 (batch 2) were higher than those in 1991 (batch 1).

In the V-shaped tanks the small pennate diatoms, most suitable as food for post-larval abalone (Norman-Boudreau *et al.* 1986; Hahn 1989), were rapidly grazed and replaced by overstory diatoms, which are less suitable as food (Matthews & Cook 1995). The addition of *G. chilensis* when the juveniles reached about 5 mm did not immediately increase their growth rate. Turbulence created by aeration in the V-shaped tank caused the *G. chilensis* to circulate and the 5 mm juveniles were unable to climb onto the seaweed to feed on the epiphytes as they did in the experimental containers. The macroalga may also have inhibited diatom growth in the V-shaped tanks by stripping the nutrients from the water (Pickering 1990). Once the juveniles reached 7–8 mm in the V-shaped tanks they were able to catch and ingest the *G. chilensis* and growth rates increased to more than 50 microns per day. This is still lower than the growth rates of similar sized juveniles from the experiments (60 –110 microns per day) indicating that the feeding systems in the V-shaped tanks can be improved. In a commercial system best growth will be achieved by weaning the 7–9 mm juveniles on to either macroalgal or manufactured diets because of the difficulties associated with maintaining a diatom coat.

The most appropriate diet for *H. australis* culture will depend on the availability and cost of various foods, the size and number of juveniles to be grown and the tank design to be used. Tanks that are suitable for feeding and growing diatoms may not be suitable for feeding out seaweeds or artificial foods. The V-shaped tanks we use have a high surface area (11.5 m^2) for growing diatoms and for the juveniles to graze over (Tong & Moss 1992). They are also suitable for feeding out seaweeds once the juveniles are large enough to catch the seaweed (Tong *et al.* 1992). Flat-bottomed settlement tanks of a similar volume to the V-shaped tanks have a much smaller surface area (3.8 m^2) and are less productive at growing diatoms and juvenile abalone (Ebert & Houk 1984; Tong & Moss 1992) but may be more suitable for feeding manufactured powdered foods (Ewing pers comm.) which settle out on the horizontal surface.

Careful management of diatom supply, and changing the diet of juveniles from diatoms to seaweeds or manufactured diets at the appropriate size will enhance the early growth of *H. australis* in culture.

Other factors such as water temperature (Leighton 1974; Leighton *et al.* 1981; Uki *et al.* 1981; Hahn 1989) and species (Uki & Kikuchi 1979) and nutritive value of the diatoms (Austin *et al* 1990) have also been shown to affect the growth rates of abalone. These factors, and the interactions between them, will also affect the early growth of juvenile *H. australis* and need to be investigated.

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