MANUAL FOR INTENSIVE HATCHERY PRODUCTION OF ABALONE

Theory and practice for year-round, high density seed production of blacklip abalone (Haliotis rubra)

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Introduction

This manual was commissioned by the Indigenous Fisheries Initiative of the NSW Government as a practical guide to commercial scale seed production of blacklip abalone (*Haliotis rubra*) in NSW. Methods and equipment are based on those described by Hone et al. (1997) with more recent refinements developed by Dr Arthur Ritar and Mark Grubert in Tasmania during the course of the Fisheries Research and Development Corporation (FRDC) Project 2000/204 and in NSW during the course of additional projects supported by the FRDC, namely projects 1998/219 and 2001/033.

The manual provides specialized instruction on:
- how to collect and reproductively condition blacklip abalone broodstock
- how to induce them to spawn
- how to hatchery-rear their young through the larval and early juvenile stages to an age and size suitable for on-farming or for seeding depleted fisheries.

With minor adjustments to methods and equipment, the manual should also serve as a useful production guide for most temperate species of abalone. There is an emphasis on year-round production of spat as opposed to the seasonal production practised to date by commercial abalone farms in Australia. In view of the scarcity and high cost of coastal sites with access to marine waters in NSW, the techniques described are geared to efficient and intensive production of abalone spat using small areas of land. These techniques in some respects are quite different to those generally practised to date by commercial abalone farms in Australia.

This manual is not intended to cover all aspects of blacklip abalone biology, nor hatchery production technology. There is a large amount of such information published both in Australia and overseas in relation to blacklip and other abalone species. The manual can be used by people with a range of backgrounds. It is primarily intended for abalone farmers and technicians with a knowledge of aquaculture fundamentals; however, the information is also expected to be useful to scientists and researchers.
How to use this Manual

This manual consists of a logical sequence of chapters that provide background theory as well as practical guides of how to:

- collect wild adult blacklip abalone, domesticate them and bring them into spawning condition
- induce them to spawn
- fertilise and incubate their eggs
- produce larvae, post-larvae and juveniles of a size suitable for seeding depleted fisheries or for on-farming, in accordance with the systematic scheduling of operations illustrated in Appendix 1.

To help you apply these procedures, we have also provided step-by-step summaries that itemize key actions and essential equipment and materials. Supplementary information on the design and operation of facilities and equipment and on sub-procedures is provided as a series of appendixes. Specialist technical terms are presented in bold font and are defined the first time they appear in the text. A reminder of the meaning of these specialized words can be readily accessed in the glossary at the back of this manual.

We intend this manual not as a stand-alone text but as a companion to other publications, especially the excellent array of manuals and reports listed in ‘References and companion reading’. We strongly encourage readers to acquire and consult these and additional references as recommended from time to time through the text.
References and companion reading


Chapter 1  Abalone Biology
Chapter 1 Abalone Biology

A balone are marine snails that belong to a group of invertebrates (animals lacking a backbone or other forms of internal skeleton) called molluscs. Molluscs also include common bivalves such as scallops, oysters, mussels, pippies and cockles as well as octopus, squid and cuttlefish. Most species of abalone have a moderate to heavily calcified snail-like shell that is flattened (Figure 1). This flat body shape reduces drag forces generated by powerful waves and currents typical of their shallow (1 to 10 m deep) exposed coastal reef habitat. The shell protects the soft body tissues including a very large and undulating muscular foot that allows the abalone to remain firmly attached to rocky substrates even while moving and feeding.

Figure 1 Abalone: a marine snail distinguished by a flattened shell with a set of respiratory pores.

A series of holes (Figures 1 and 2) along the left upper margin of the flattened shell defines abalone as a particular group (genus) of snails, called Haliotis. These holes (or respiratory pores) are outlets through which seawater, partially stripped of oxygen by the gills, is exhaled together with urine, faeces, and either sperm or eggs. The head and mouth of abalone are flanked by two pairs of sensory tentacles (Figure 2); the shorter outer pair are stalked eyes. The mouth lacks teeth but instead is equipped with a long rasping tongue called a radula that is used to tear, dislodge and ingest food, which comprises mainly plant materials.

Food includes seaweed that is either still attached to rock surfaces or has already been detached by the actions of waves and currents. Detached seaweed—
principle food source of larger juvenile and adult abalone—is commonly referred to as 'drift'.

As already implied, abalone have separate sexes. The gonads (egg or sperm-producing organs) lie adjacent to the margin of the shell at the rear of the body on the side closest to the spire of the shell (Figure 2). The gonad can be clearly seen only by deflecting the foot out of the way (Figure 3). Male abalone can be identified by their white to cream gonads (testes) (Figure 4). Female abalone (Figure 4) have gonads (ovaries) that reflect the colour of their eggs, which varies widely from green to olive and from maroon to brown. In practice, determining the sex of blacklip abalone can be difficult because the overlying skin on the outside of the foot can be heavily pigmented or banded, ranging from pale grey to black, with the latter totally obscuring the gonad colour.

In NSW, wild blacklip abalone become sexually mature by the time they reach a shell length of about 90 mm at 3 to 5 years of age. However, their farmed counterparts become sexually mature 1 to 2 years earlier at a smaller size of 50 to 70 mm. A abalone are broadcast spawners that release sperm and eggs (Figure 5) into the surrounding seawater, where fertilisation takes place. Some populations of wild abalone form aggregations and spawn in unison to increase the chance of successful fertilisation.

**Figure 2** Drawing of dorsal view of an abalone with shell removed to expose soft tissues (adapted from Shepherd, 1975).
**Figure 3** Simple method of deflecting the foot muscle of an abalone to determine whether it is sexually mature and, if so, its gender and breeding stage.

**Figure 4** View of soft tissues of blacklip abalone from above after removal of the shell. Note the ripe gonads of these adult abalones, characterized in the case of males (left specimen) by a large cream-coloured testis and in females (specimen on the right), by a maroon-coloured ovary. Reproduced from Liu (2005).
In established well-run hatcheries, 60% to 80% of female abalone and 80% to 90% of males held at temperatures of 16 to 18°C—conducive to ripening of the gonads—can be expected to spawn in response to the induction protocols discussed in Chapter 4. However lower rates of success are to be expected during the first year of operation of new hatcheries because of incomplete domestication of recently captured stock; this problem is commonly compounded by staff inexperience and teething problems with the newly commissioned equipment.
Figure 6  Schematic life cycle of abalone (source Hone et al., 1997).
Twin attachment muscles

Figure 7  Fully shelled second stage veliger larvae

Figure 8  Photomicrograph of recently settled and metamorphosed post-larvae grazing on surface of crustose coralline algae coated rock (courtesy of Sabine Daume) Note: Smooth larval shell (red arrow) and sculptured post larval shell growth (yellow arrow) Black bar = 500 micron
Fertilised eggs hatch into first-stage swimming larvae called **trochophores** *(Figure 6)* after 18 to 24 h at an optimum temperature of about 18°C. The time spent as **planktonic** larvae (living in the water column) differs among and within species and depends on temperature. Trochophores develop further into shelled **veliger** larvae *(Figures 6 and 7)* after 20 h at 18°C. The larvae of abalone do not feed, but live on the egg yolk and by directly absorbing soluble nutrients in seawater through the skin. Blacklip abalone larvae first become competent to settle and **metamorphose** (transform) into reef-dwelling **post-larvae**. Post-larvae *(Figure 8)* are simply early-stage juveniles that range from 0.3 to 3 mm in shell length.

Metamorphosis of larvae into post-larvae occurs after a minimum of 6 days at an optimum temperature of 18°C. Larvae can however, delay settlement and metamorphosis until they find a suitable habitat *(Figure 9)*, which consists of rocky surfaces coated by a thin skin of tough, limestone-depositing **crustose coralline algae**. This is commonly known as ‘pink rock’ *(Figure 10)*. These predominately shallow (1 to 6 m) rock habitats include wave-rambled ‘boulder fields’ and other deeper bare rock areas called **barrens**. In NSW, barrens are commonly the product of grazing by dense aggregations of the black (or purple) sea urchin *(Centrostephanus rodgersii)* *(Figure 11)*. Urchin barrens are in fact the most common type of reef habitat in New South Wales, accounting for almost half of the 5000 ha of reef located from Port Stephens (120 km north of Sydney) south to the Victorian border.

Ready-to-settle blacklip abalone larvae ‘home in’ on these rocky habitats, being attracted by chemical cues released by the surface layer of crustose coralline algae and by recent grazing trails of juveniles and adults of their species. During and immediately following settlement, larvae and early post-larvae suffer heavy mortalities that exceed 95% during the first week alone. The principal causes are predation or accidental ingestion by a diverse array of other common and prolific reef surface grazers, especially urchins, turban shells and other marine snails, in combination with predatory free-ranging sea worms.

With progressive growth from 2 mm to about 10 mm, and further development of the rasping tongue, the main source of food for juvenile blacklip abalone progressively shifts from microscopic algae *(Figure 12)* towards the very abundant but tough cells of the encrusting coralline algae. Surface grazing on crustose coralline algae tissue remains an important way of obtaining food for juvenile blacklip abalone up to a shell length of about 40 mm and 18 months of age. Thereafter it is progressively replaced by a diet of seaweed that consists mainly of broken-off pieces *(drift)*, rather than whole plants, which need to remain firmly attached to rock substrates via holdfasts in order to survive.

Post-larvae and other small juvenile blacklip abalone live on the tops and sides of submerged rocks. As they grow beyond about 5 mm, they start seeking shelter under the rocks by day, emerging only at night to feed in the absence of their most important predators. These predators include carnivorous fish and especially species such as wrasse, morwong *(Figure 13)* and wirra cod. Coincident with the shift in diet to seaweed, the distribution of juvenile blacklip abalone becomes more ‘patchy’. This patchiness reflects the uneven distribution
of drift, including common red seaweeds such as *Gracilaria* and green seaweeds such as sea lettuce (*Ulva* spp.). Drift is generally more abundant and accessible on the outer margins of reefs directly exposed to prevailing currents.

Red seaweeds are generally more palatable to blacklip abalone than the common brown seaweeds such as giant kelp (*Macrocystis* spp.), and this probably also accounts for the large variation in the carrying capacities of individual reefs for abalone—even of those within a few hundred metres of one another. Highly productive reefs in NSW are commonly characterised by fast-growing blacklip abalone with thin, oval shells. By contrast, low-productivity reefs commonly carry slow-growing abalone with thicker, heavier shells that are more circular. Many abalone within these low-productivity reefs fail to reach the minimum legal shell length, which in NSW is 115 mm. Such populations are commonly referred to as ‘stunted’.

*Figure 9* Typical shallow exposed rocky coastal habitat of blacklip abalone
**Figure 10** Typical ‘pink rock’ shallow crustose coralline algae coated boulder habitat of settlement and early juvenile stages of blacklip abalone. In the centre is an early prototype mass seed-deployment device that disperses button-sized juvenile abalone. Photo taken 24 h after release. From: Heasman et al., 2004.

**Figure 11** Urchin barrens - the most common type of reef habitat in NSW, accounting for about half of the State’s 5000 ha of commercially fished abalone-bearing reef
Figure 12  Examples of benthic microalgae that comprise the principal diet for early post-larvae and to juvenile abalone. These include algae eaten by abalone, especially diatoms such as species of Navicula (a & b), Nitzchia (c), Cylindrotheca (d), Ulvella (e) & crustose coralline algae (f). Reproduced from Daume, 2004. A-E scale bar indicates 10 micron; F scale bar indicates 1 millimetre.
Figure 13 Above: Holding tank with red & banded morwong. Morwong together with common species of wrasse and other reef and demersal fish like wirra cod, are major predators of juvenile abalone 5 to 50 mm long.

Right: A collection of shells of juvenile abalone regurgitated by the morwong.
Chapter 2  Broodstock Collection and Husbandry
Chapter 2 Broodstock Collection and Husbandry

To run a hatchery cost-effectively it is a great advantage to be able to produce multiple batches of larvae and juveniles year-round. Unfortunately, in NSW wild blacklip abalone are not reliable sources of ripe, ready-to-spawn broodstock, even during the natural late spring/early summer breeding season. Wild adult blacklip abalone collected in apparently ripe condition (i.e. with large, swollen gonads) generally prove over-ripe or are inhibited from spawning by the stress of capture and confinement. As a consequence, abalone hatcheries located in NSW must include provision for long-term holding and year-round reproductive conditioning and spawning of captive broodstock. Fortunately, reproductive conditioning of blacklip abalone is a simple matter of feeding them a good quality manufactured diet while maintaining them in excellent (near-oceanic) quality seawater and at favourable temperatures within the range 16 to 18°C. These temperatures mimic those of sea temperatures in coastal NSW in winter, a time when blacklip abalone develop gonads at the expense of growth.

Before broodstock may be collected from the wild, a compulsory permit must be acquired from the relevant authority. In NSW this is the Fisheries Division of the Department of Primary Industries. Permits restrict size and number and the methods of collection and areas where abalone can be legally collected. All such restrictions must be strictly observed. When collecting broodstock, choose young adults in the range 90 to 120 mm that have clean, undamaged shells free of heavy biofouling and obvious infestation by shell damaging mud-worms and boring sponges. The most vigorous and healthy individuals are those with thinner and elongated shells that can rapidly right themselves when upturned and rapidly seek shade and shelter when exposed to bright light. Be aware that numbers of males and females in natural populations might not be equal and that the sex of selected stock is best checked (see methods of sexing below) as soon as possible after collection. This will ensure that sufficient numbers and the best examples of each sex are retained and that the remainder can be returned to the wild unharmed.

Special care must be taken not to injure stock when removing them off rocky surfaces. Do not use abalone knives or other sharp implements that may cut the foot of the abalone, resulting in major loss of blood, infections and eventual death. Instead, use a metal paint scraper with its corners ground off (Figure 14) or a plasterer’s plastic spatula and place the blade under the back of the foot in a rapid but smooth sliding (not levering) action to break the seal between the foot and rock surface.

During collection, broodstock can be stockpiled either in live-wells of boats or in large tubs continuously flushed with new seawater. Alternatively, broodstock can be held in mesh cages (Figure 15) or net bags hung from the stern or sides of the dive vessel. Once collected, and during transportation to the hatchery, abalone must be either kept undisturbed in flowing seawater, or stored in damp, cool, dark conditions. For both methods the abalone are encouraged to attach to smooth inert surfaces, such as sheets of plastic (Figure 15) or to each other in clusters.
If transportation time to the hatchery is likely to be more than an hour or two, the abalone should be held in seawater at ambient temperature. If they are to be temporarily held in shore-based, flow-through or chilled recirculation seawater systems, such as those used by the live abalone export trade, then water quality must be maintained at near-oceanic quality.

**Figure 14**  A metal paint scraper with its corners and sharp edges ground off to minimise injury to broodstock being collected from the wild.

In the case of flow-through systems, the seawater flow rate per minute should equal or exceed the total abalone biomass. As stress experienced by recently collected wild abalone is a product of both time and intensity, periods of storage and transportation should be kept to a minimum. If holding and transportation periods are likely to exceed a few hours but no more than 24 h, damp cool storage at 12 to 16ºC is recommended. Preferably this should be within sealed plastic bags in an atmosphere of pure oxygen.

On arrival at the hatchery, newly collected broodstock should be immediately inspected and any stock with injuries or other serious afflictions culled. Healthy individuals should be placed in a quarantine tank supplied with ambient, 10 µm filtered seawater. The tank should be covered with 90% shade-cloth and the abalone provided with daytime shelters. Newly acquired broodstock should be retained in quarantine for at least 2 weeks to allow them to acclimatize to captivity and for the full effects of injury and stress to become evident.
Figure 15  Top: Plastic chicken crate for stock-piling and safe transportation (following collection from the wild) Bottom: Smooth plastic sheets that safeguard attached broodstock during transportation back to the hatchery.
The shells of broodstock retained on the basis of good appearance, health and vigour are cleaned under running seawater with a wire brush or an abalone knife to remove biofouling. Utmost care is needed not to damage soft tissues or the respiratory pores of the shell. In healthy blacklip abalone, cleaning will expose the prominent red/maroon layer of the outer shell surface (see Figure 1). Once cleaned, the shell may be treated with crushed sea salt for several hours to desiccate any undetected boring mud-worms that otherwise will multiply and spread to other broodstock within the conditioning units. An option to the use of salt is to suffocate the mud-worms by coating the shell with surfboard wax. Retain the abalone in quarantine for a further 1 or 2 days before peeling off the wax and rescrubbing the shell. (See Appendix 4 for further information).

Fully conditioned (ripe) blacklip abalone suitable for the induction of spawning are those that have been held at 16°C for 150 to 180 days (see Chapter 4 for details), either from the date of a previous spawning or from an initial state of little or no gonad development. Efficient reproductive conditioning cannot therefore be achieved without accurate and unambiguous records for individual broodstock. To do this, each broodstock must be individually marked before being measured, weighed, sexed and assessed for general health and breeding condition. Marking is done by threading commercially available tags (individually numbered and colour-coded cable ties) through the two respiratory pores closest to the front of the abalone (Figure 16). Care is needed to avoid damage to soft tissues.

Figure 16 Individually numbered or colour-coded cable-tie tags. These are inserted through the two respiratory pores closest to the front (head) of the abalone.
**Step By Step Summary and Check-List for Broodstock Collections**

1. Acquire all permits for collecting and holding broodstock while building and commissioning the hatchery (submit all applications at least 18 months ahead of intended start of operations).

2. Avoid injury of abalone by removing them from rocky surfaces with a rounded paint scraper or plasterer’s spatula, and transport them back to the hatchery at the prevailing sea temperature as soon as possible.

3. Provide smooth surfaces such as sheets of plastic for the abalone to adhere to during transportation to the hatchery.

4. Transport the abalone undisturbed in shaded or dark conditions in good quality seawater, or under damp, cool (less than 22 °C) conditions in air if the transportation period is less than an hour.

5. On arrival at the hatchery, allow the abalone to recover in clean, flowing seawater.

6. Remove biofouling from the shell with a wire brush or diver’s knife, taking great care not to damage the soft tissues.

7. Quarantine the newly collected abalone from existing stock for at least 2 weeks until they commence feeding (an indication of good health and habituation to captivity).

8. Double-tag, measure, and record the details of each individual on a database before introducing them into the broodstock conditioning system.