# Biology and Status of Aquaculture for Giant Clams (Tridacnidae) in the Ryukyu Islands, Southern Japan

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#### Introduction

The Ryukyu Islands consist of many islands located between Kyushu in mainland Japan and Taiwan. The islands in the southwestern area of the Ryukyu Islands belong to the Okinawa Prefecture (Fig. 1). The Ryukyu Islands are strongly affected by the Kuroshio Current and are renowned for their coral reefs with high diversity of tropical and subtropical species.

Giant clams traditionally have been utilized as fisheries resources for a long time in this area. According to fisheries statistics,

catches of *Tridacna crocea* in Okinawa have decreased drastically during the last 30 years and currently are less than one tenth of previous catches (Fig. 2). Fishing can easily deplete stocks of giant clams because the clams inhabit shallow waters and take at least three years to attain sexual maturity.

Techniques for the mass seed production and aquaculture of three species (*T. crocea*, *T. squamosa*, and *T. derasa*) were established in Okinawa. Four hundred thousand seeds of giant clams of 8 mm shell length (SL) are supplied to fishermen for use in aquaculture or stock enhancement every year.

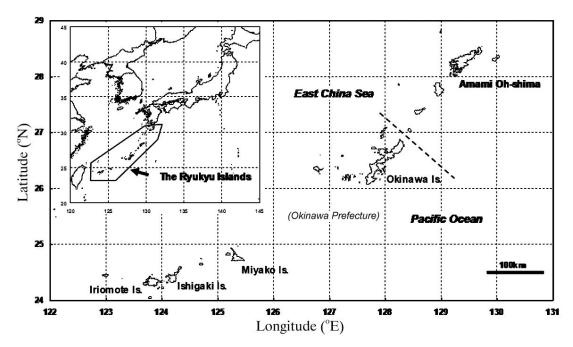


Fig. 1. Map showing location of the Ryukyu Islands in southern Japan.

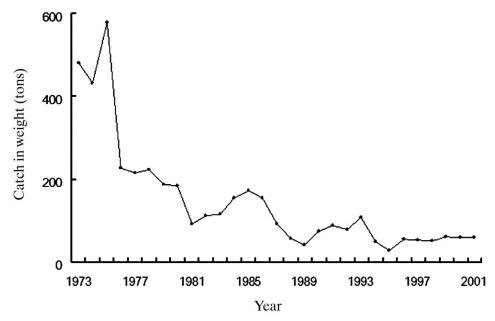


Fig. 2. Annual catches (tons) of *Tridacna crocea* in Okinawa, Japan.

This paper will review the (1) biology of giant clams, (2) present status of aquaculture of giant clams in Okinawa, and (3) other studies on giant clams in southern Japan.

## **Biology of Giant Clams**

#### Classification of giant clams

Classification of giant clams is shown as follows:

> Phylum: Mollusca Class: Bivalvia Order: Veneroidea Superfamily: Cardioidea

Family: Tridacnidae (Giant clams) Genera: Tridacna and Hippopus

Giant clams are distributed on coral reefs and areas adjacent to reefs in the Western Pacific and Indian Ocean. Nine species of giant clams belonging to two genera in the family Tridacnidae are currently known to science (Knop 1996). Three species (T. tevoroa, T. rosewateri, and Hippopus porcellanus) have been discovered in the 1980s and 1990s, but the areas where they are distributed are limited.

## Giant clam species in the Ryukyu Islands and morphological characters

In the Ryukyu Islands, six species of giant clams have been recorded including five extant species, T. crocea, T. squamosa, T. derasa, T. maxima, H. hippopus (Fig. 3) and T. gigas, although the latter species has rarely been found in this area recently.

Murakoshi (1988) described the morphological characters of the five extant species as follows:

T. crocea is the smallest of the five species and grows to 15 cm SL. There are short scutes on the radial ribs and unequal anterior and posterior sides of the shell margin. This species shows marked variations in mantle colors. It has the best taste among the five species and it is the most important species of giant clam as a fisheries resource in the Ryukyu area.

T. squamosa reaches about 40 cm SL. The anterior and posterior sides of the

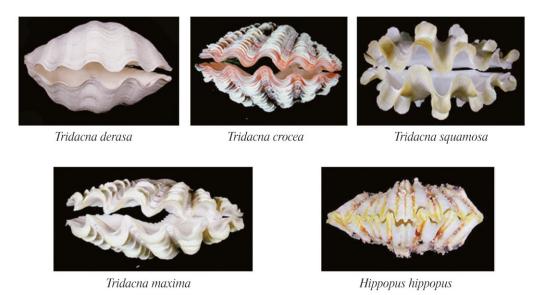


Fig. 3. The extant species of giant clams in the Ryukyu Islands, Japan.

lower shell margin are almost equal and large regular scutes exist on the radial ribs.

T. derasa is the largest of the five species and grows to 50-60 cm SL. The anterior and posterior parts of the lower shell margin are unequal. There are no scutes and the shell surface is smooth.

The byssus disappears as the clam grows to a large size.

T. maxima reaches 30-35 cm SL. Shell morphology of this species is similar to *T. crocea*. However, it is possible to distinguish between the two species by the size of the adductor muscle impression.

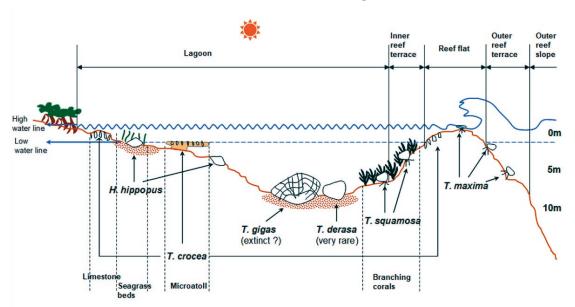


Fig. 4. Schematic illustration of major habitats of giant clams (*Tridacna crocea*, *T. squamosa*, T. derasa, T. maxima, Hippopus hippopus) in the Ryukyu Islands, Japan.

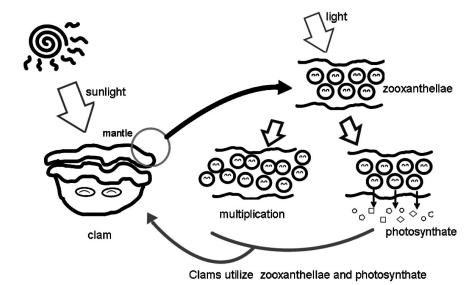


Fig. 5. Schematic illustration of the relationship between giant clams and zooxanthellae.

H. hippopus grows to 30-40 cm SL. The valves are diamond-shaped in adults and are quite different from those of other species. Their byssus orifices are nearly closed.

## Habitats of giant clams in the Ryukyu **Islands**

Habitats of giant clams in the Ryukyu Islands are illustrated in Fig. 4. *T. crocea* bore into limestone substrates near the shore. on reef flats or dead parts of massive coral colonies, and most of the valves are hidden in the hollows. T. squamosa are found among branches of coral colonies in lagoons. T. derasa inhabit lagoons but this species is very rare in the Ryukyu Islands as this area is the northern limit of its distribution. T. maxima bury part of their shells in substrates at the outer reef edges where waves are rough and currents are strong. *H. hippopus* are generally found on sand or algal beds in lagoons.

#### **Biological characteristics**

Giant clams show the following biological characteristics during their life cycle.

The most distinctive character of giant clams compared with other mollusks is the

symbiotic relationship with zooxanthellae. Zooxanthellae (Symbiodinium spp., Dinophyta) are 7-10  $\mu$ m in diameter. The clam utilizes the photosynthetic products formed by the algae and consumes any excess algae in the mantle tissue directly (Fig. 5). Because the symbiotic alga is not transmitted through their eggs, the symbiotic relationship is established during larval development. Giant clams die if the symbiotic relationship with the zooxanthellae is not established.

Metamorphism occurs in the young stage so the symbiotic algae can obtain sunlight efficiently (Knop 1996). The larvae of giant clams do not differ markedly from other bivalves before metamorphosis. The shells are connected on their upper side by a ligament. The respiratory siphons stick out from one side of the shell. The gills are located in the central part of the larva. The larvae have two adductor muscles - anterior and posterior. After metamorphosis, the ligamentous joints move to the lower side and thus open upwards. The two siphons become separate and increase in length. The gills move to the upper side. The posterior adductor muscle moves to the center and the other one shrinks.

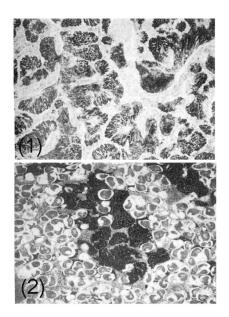


Fig. 6. Photomicrographs of histological sections of gonads in T. crocea (adapted from Murakoshi 1988): 1) Male-phase, 5 cm SL testes are observed; 2) Hermaphrodite condition, 8 cm SL - ova and testes are observed.

Giant clams are protandric hermaphrodites (Wada 1952). T. crocea up to 5 cm SL are all males with testes. In clams over 5.5 cm SL, the testes are filled with sperm and ovaries are scattered among them (Fig. 6), the latter

develop as the clams grow. T. squamosa up to 15 cm SL are all males with testes, while ovaries are found in shells over 17 cm SL (Leonid 1991).

#### Life cycle

The life cycle of giant clams is reported by Murakoshi (1988), Lucas (1988), Knop (1996), and Heslinga et al (1990).

As reported by Murakoshi (1988), the life cycle of *T. crocea* is shown in Fig. 7. Twenty hours after fertilization, eggs of T. crocea develop to D-shaped larvae of 0.14 mm SL. After a week of planktonic life, the larvae become pediveligers (0.19 mm SL) with both velum and foot, and gradually become benthic. After one more week, the velum gradually degenerates and the pediveligers develop into larval clams of 0.20-0.24 mm SL which begin to acquire symbiotic algae. The zooxanthellae which are ingested from seawater by the pediveligers move out through the wall of the gut, and the algae multiply. A row of algae reaching the ventral margin can be recognized as the establishment of symbiosis between larval clam and algae (Fig. 8).

Larval clams (0.25-0.50 mm SL) have conspicuous umbones and clear growth

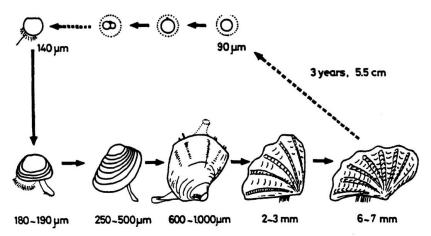


Fig. 7. Schematic illustration of the life cycle of Tridacna crocea (from Murakoshi 1988).

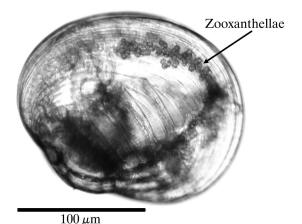


Fig. 8. Photomicrograph of larval clam of Tridacna crocea (0.2 mm SL) when the symbiotic relationship is first established.

lines on the shell. As the hinge area of the shell becomes large at 0.60-1.00 mm SL, remarkably long siphons develop. The radial ribs develop strongly and scutes appear on the shells as they grow thicker and become opaque. The mantle lobe is extended and the orifice of the byssus opens. The juvenile clams attach to substrates more firmly by the byssus. The shape of valves gradually changes from almost triangular at 2-3 mm SL juveniles to the adult shape in 6-7 mm SL juveniles. In about 3 years, they grow to 5.5 cm SL and start to reproduce.

## **Status of Giant Clam Aquaculture** in Okinawa

## **Induction of spawning and egg** collection

The water temperature around Ishigaki Island shows seasonal variation ranging from 20 to 30°C (Fig. 9). Mature clams appear from March to August and are brought from the sea to laboratory tanks where spawning is induced by artificial stimuli. One of the most effective methods is pouring gonad tissue dilution into the tank. Rapid changes of sunlight intensity are also

used to stimulate spawning in T. squamosa and T. derasa. A light shielding net is used to cover the tank until spawning induction is performed on a sunny day.

In Okinawa, the spawning season falls in March-August for T. squamosa, March-May for T. derasa, and April-September for T. crocea. It is possible to check gonad development in *T. crocea* through the byssus orifice on the underside of the shell in order to select mature individuals.

Spawning usually releases sperm first, followed by eggs. The eggs are very small, about 90  $\mu$ m in diameter. The range in number of spawned eggs is 5-20 million for T. crocea, and 10-100 million for T. squamosa and T. derasa. When the clams begin to spawn, they are immediately moved to another tank (500-l polycarbonate container) to avoid the effects of polyspermy. Spawning continues intermittently for 20-40 min. About 300 ml water with sperm is poured into the tank to fertilize the eggs. Fertilized eggs are stocked in the tank at a density of 6 eggs/ml. Strong aeration is necessary so the eggs do not settle on the tank bottom.

### **Seed production**

Seed production procedures are outlined in Fig. 9. The day after fertilization, D-shaped larvae are transferred to another tank at a density of 0.3 larvae/ml. The indoor tanks are 10 tons (2  $\times$  5  $\times$  1 m) and 5 tons (1.2  $\times$  4.0  $\times$ 1.0 m) in capacity. Moderate aeration is maintained to ensure water circulation in the tank (Fig. 10). It is necessary to provide zooxanthellae for the larvae, because the algae are not incorporated in the eggs of giant clams. Some of the ingested algae are digested in the gut of larval clams. The algae are provided to the larvae from Day-3 (3 days after hatching) at a density of 30-100 cells/ml.

There are two methods of obtaining zooxanthellae for use in larval culture. One

	Egg Larvae	(Establishment of symbiosis)	mbiosis)	Juvenile		Seed (utility size)
Shell length	0.08mm	0.2mm	0.6~0.8mm	1mm	3mm	8mm
Day	Day 0	Day 10∼30	Day 40~60	Day 60∼80	Day 100~200	Day 150~300
Density (Maximum)	300,000/m³ (~500,000)	$25,000/\text{m}^2$ $(\sim 50,000)$	$15,000/\text{m}^2$ (~30,000)	$10,000/\text{m}^2$ (~20,000)	4,000/m <sup>2</sup> (~6,000)	$2,000/\text{m}^2$ (~4,000)
Light intensity	$\sim$ 500 $\mu$ mol/m <sup>2</sup>		$\sim 1000  \mu  \mathrm{mol/m^2}$	No light shielding	ielding	
Culture water	No running water		running water, running water (1~2 times per day) (ca. 1 times per h	running water (2~3 times per h)	vater es per h)	
Exchange of water	one half 2 to 3 days	ys complete 10 to 20 days	0 days	complete 3 to 4 weeks	t weeks	
Aeration	moderate weak	No aeration				
Feed zooxanthellae	Fed (Day 3~)	/ Not fed				
Co-rearing with herbivorous gastropods				Batillaria spp.	Troe	Trochus niloticus

Fig. 9. Procedure for seed production of giant clams at Yaeyama Branch, Okinawa Prefectural Fisheries Experimental Station, Japan.

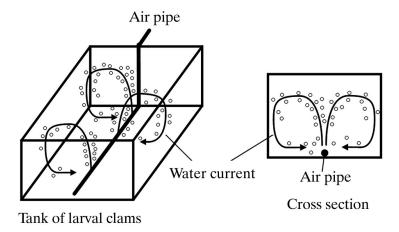


Fig. 10. Design of the 10-ton  $(2 \times 5 \times 1 \text{ m})$  and 5-ton tanks  $(1.2 \times 4.0 \times 1.0 \text{ m})$  showing water current direction to prevent dead spots in the tank.

is to take tissue directly from the mantle of live clams, mash it and separate the algae from tissue debris. Another method is to use cultivated algae maintained in artificial medium without aeration under white fluorescent light (about  $60 \, \mu \text{mol/m}^2/\text{s}$ ) with a 12: 12-h light-dark cycle at 28°C.

After the zooxanthellae are provided, larvae should be observed every day. If no algae are visible in the gut of larval clams,

they should be re-introduced. One half of the culture water is replaced at 2-3-day intervals. Many larvae settle and metamorphose from Day-7 and it is necessary to reduce aeration. Larval clams establish symbiotic relationship with algae between Day-10 and Day-20. It is possible to rear with no feeding once the symbiotic relationship is established. Many larvae die during this period. Average survival rate is 5% from D-shaped larvae to larvae with symbiotic relationship.

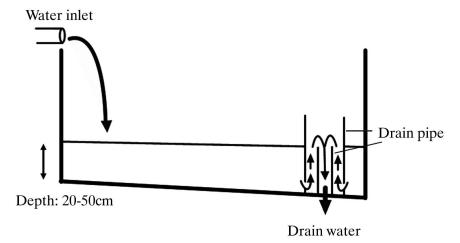


Fig. 11. Vertical section of the tank showing drainage of water from tank to prevent dead spots the bottom. The tank size is similar to that of Fig. 10.

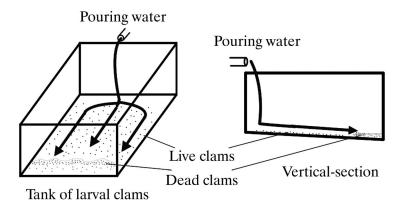


Fig. 12. Design of sweeping dead shells by pouring water in tank.

Flowthrough seawater (100-200% daily) is used as culture water and is kept at a shallow level (20-50 cm depth) after the establishment of symbiosis. Aeration is not used, to minimize the growth of weed in the tank. The bottom of the tank is drained using a large pipe sleeve with water inlets cut in its bottom then set over the drainage pipe (Fig. 11).

Growth of the larvae becomes stable with multiplication of the zooxanthellae in their mantle. Because the shells of larval clams are translucent, it is necessary to keep light intensity in the tank below 500  $\mu$ mol/ m<sup>2</sup>/s by light shielding. The larvae grow to juveniles of 1 mm SL 2-3 months after fertilization. The light shielding should be gradually removed before the clams grow bigger than 1 mm SL. Suitable clam density at this stage is 10,000 ind/m<sup>2</sup>.

If dead shells are found in large numbers at this period, they should be removed to avoid mass mortality. Dead shells can be swept by pouring seawater slowly over the bottom of the tank. After Day-50, live clams attach to the tank by means of their byssus (Fig. 13).

Juvenile clams bigger than 1 mm SL can be reared in direct sunlight in outdoor tanks. To clean weeds in the nursery tank, small

herbivorous gastropods (Batillaria spp.) are reared with the clams. Small trochus (Trochus niloticus) are used as cleaning guests after the clams reach 3 mm SL because they are easier to rear than Batillaria spp. The feces of trochoids should be swept away routinely. Juveniles should be moved to a new tank at least every four weeks and the density maintained at a suitable level. This way, it takes about six months to a year for larval clams to grow to a size that can be used for aquaculture (i.e., 8 mm SL). Average survival rate is 0.3% from larvae to the utility size of 8 mm SL in Okinawa.

#### Aquaculture

Trochus crocea is generally cultured by planting the seeds in pits prepared by drilling in limestone substrates. The following three points are important for good aquaculture: a) site selection areas with a cover of sand or seaweed are not suitable; b) vertical boring into the substrate to minimize the ability of seeds to move away from the pits; and c) protection from predators by covering planted sites with nets to effectively protect clams against fishes or crabs, the major cause of mortality.

Trochus squamosa and T. derasa are cultured in cages fixed on the sea bottom.

The following points are important for good culture: a) selection of suitable site in shallow sea (2-5 m depth) and fixing the cage securely to prevent turning over during typhoons (Fig. 13-1); b) planting of bottom structure (e.g., coarse plastic mesh: Tamaki 2000) to prevent seed aggregation or clumping together that causes low survival; and c) maintenance of suitable culture conditions by cleaning the cage to secure sunlight and water supply (Fig. 13-2). The density of seeds in the cage should be maintained at 1000 ind/m² for 8 mm SL, and 300 ind/m² for 10 cm SL.

The market sizes in Okinawa are approximately 10-15 cm SL for *T. squamosa* and *T. derasa*, and 5-8 cm SL for *T. crocea*. Growth of the three species is shown in Fig. 14. At least two years are required to grow giant clams to market size. It is easy to rear giant clams because culture management does not require too much time as it needs only monthly or occasional monitoring. Survival rate is 40-50 % from utility seed size (8 mm SL) to market size. The utility size is based on the results of field tests (Tamaki 2000).

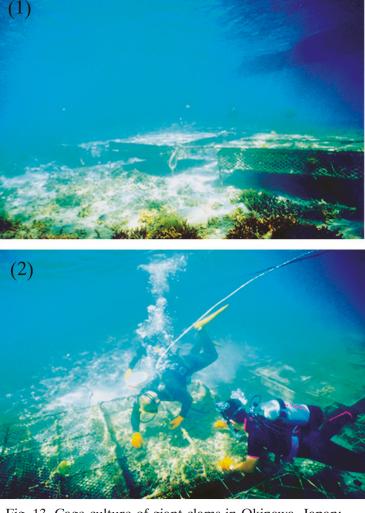


Fig. 13. Cage culture of giant clams in Okinawa, Japan: 1) set cages underwater, and 2) cleaning a cage.

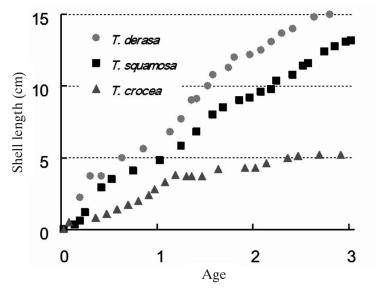


Fig. 14. Growth of three species of giant clams under culture conditions in Ishigaki Island, southern Japan.

Today, there is high demand in Okinawa for giant clams. Both the soft parts and the shells are utilized – the mantle and adductor muscles are mainly eaten raw while the shells are used for ornaments.

# Other Studies on Giant Clams in **Southern Japan**

Heterozygosity of allozyme loci and usefulness as artificial seeds of giant clams have been studied by Kobayashi (2003). The proportion of soft part weight (SW) per whole body weight (BW) giant clam was positively correlated with the number of heterozygotic loci. More heterozygous individuals also achieved a higher proportion of SW/BW (%). For seed production, it is therefore necessary to prepare a large number of mature clams to allow the production of heterozygous progenies.

In the Yaeyama Branch of the Okinawa Prefectural Fisheries Experimental Station, the following three studies are ongoing: 1) practical effectiveness of resource management for giant clams verified in a marine protected area (MPA) in Okinawa where

harvest of certain organisms is prohibited throughout the year; 2) seed production of T. maxima because the species is easy to rear in the winter season; and 3) artificial spawning for seed production in the winter season for seed distribution in summer when they are in high demand by fishermen.

The following two studies are planned at the Ishigaki Tropical Station, Seikai National Fisheries Research Institute, Fisheries Research Agency, Japan: 1) population genetic structure of clams in the islands in southern Japan to avoid genetic disturbance; and 2) factors in the establishment of symbiotic relationship for efficient seed production of giant clams.

The fishery regulation for stock management of giant clams in Okinawa bans harvesting of all species from June to August. Harvesting of T. crocea under 8 cm SL, T. squamosa under 20 cm SL, T. derasa under 30 cm SL, and H. hippopus under 15 cm SL is prohibited throughout the year. However, there is minimal enforcement of the rules by fishermen and locals. For sustainable utilization of giant clam resources, it is very important that the value of management and maintenance of fishing grounds is understood and recognized by fishermen and locals.

## **Acknowledgments**

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