

Mesocosm technology advances grouper culture

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Juvenile flowery cod *Epinephelus fuscoguttatus* at 92 days post-hatching (average weight = 16g, TL = 9.4cm)

Researchers in Northern Australia are using this protocol to produce fingerlings of two grouper species with difficult early life stages.

At the Queensland Department of Primary Industries and Fisheries (DPI&F) in Cairns, Northern Australia, researchers have succeeded in producing juvenile flowery cod (*Epinephelus fuscoguttatus*) for the first time

in Australia. The fish, known as 'tiger grouper' in Asia, is in great demand in the live reef fish trade.

Demand for flowery cod, and other reef fish species such as gold-spot cod (*Epinephelus coioides*), is focussed on the live fish markets of Hong Kong and China. Flowery cod retails for AUD55 to 75/kg (USD 43-58) in these markets. While flowery cod is farmed in many Asian countries, many of the fingerlings used are caught as undersize juveniles in the wild. Hatchery production of fingerlings, as developed by DPI&F researchers, is essential to ensure the long-term sustainability of the aquaculture of the flowery cod.

The Reef Fish Aquaculture Project is based at Northern Fisheries Centre (NFC), Cairns. The recent upgrading of NFC, through the construction of a new aquaculture and stock enhancement facility, provides a purpose-built, marine aquaculture research facility that supports the development of tropical aquaculture in Queensland.

Finfish research at this centre is currently focussed on the production of high-value reef fish species including barramundi cod (*Cromileptes altivelis*), flowery cod and gold-spot cod. Research focus is on the development of methodologies for rearing larvae of these species, including the production of novel prey species such as copepods. The project is funded by the Queensland Government. Associated projects, funded by the Australian

Centre for International Agricultural Research (ACIAR) have been valuable in providing opportunities for collaborative research in the Asia-Pacific region, particularly Indonesia and the Philippines.

Larval rearing

Rearing methodologies were initially developed from small-scale replicated experiments used to address issues during the early larval stages. However, larval survival in small scale recirculation systems has been poor with 100% mortality commonly occurring by day 10. Physical parameters identified during small-scale trials were then transferred to a larger pilot scale rearing trial using mesocosm technology. The same protocol is being used to produce fingerlings of two grouper species with difficult early life stages.

Mesocosm is a semi-intensive technology integrating both intensive and extensive aquaculture principles. It applies semi-intensive rearing principles which provide more diverse conditions during the early development phase, when larvae are highly sensitive and easily stressed. As larvae mature and become more robust, intensive rearing methodology can be introduced.



Development of flowery cod larvae cultured at Northern Fisheries Centre, Cairns. Top: Day 16 after hatching and bottom, Day 35 after hatching.

Tank management

Fertilised eggs were stocked (30/l, day 0 = hatch) into a 6m² fibreglass mesocosm system. Water was exchanged during days 1-2 at 5% tank volume per hour during the day. No water was exchanged on days 3 to 4 to prevent the removal of prey, particularly copepod nauplii, from the tank during the critical first feeding period. From day 5, water was exchanged overnight starting at 5%/hr increasing to 11%/hr and a continuous flow from day 17 post-hatching.

Squid oil was added to the water surface twice daily from day 1 to 6 post-hatching to prevent larvae from becoming caught in the water surface tension. A photoperiod of 12L:12D was supplied by two overhead daylight fluorescent tubes supplemented by a low level of natural light. Light intensity ranged between 300 and 700 lux across the tank water surface.

Feeding Schedule

Four microalgal species (*Tetraselmis sp.*, *Cryptomonad sp.*, *Isochrysis sp.* (T.ISO) and *Nannochloropsis oculata*) were added daily from day 0 to 22. They were added on an equal ration (organic weight) basis to maintain an algal concentration equivalent to 2.2 x 10⁶ *N. oculata* cells/ml. (Organic weight is an ash free dry weight calculation which the team has profiled for each algal species.)

On day 2 post-hatching, copepods (*Acartia sinjiensis*) and SS-rotifers (*B. rotundiformis*) were added at densities of 1.25/ml and 10/ml, respectively. Enriched rotifers (Algamac 2000) were added from day 6 until day 16 to maintain a density of 15 to 20/ml. Artemia nauplii were introduced from day 9 – 13 and enriched (Algamac 3050) meta-nauplii from day 13 to 28. Artificial diets were introduced from day 9 onwards as detailed in Table 1.

Metamorphosis/cannibalism

The first metamorphosis of larvae to juveniles was noted on day 29/30 for both species and the majority had metamorphosed by day 40. Growth rates were similar for both species during the larval phase with a slight increase in gold-spot cod growth rates compared to

Figure 1. Total length (mean value and standard error) of flowery cod (open circles) and gold-spot cod (closed squares).



Table 1. Succession of feed types used to rear larval flowery cod and gold-spot cod.

Food regime for mesocosm rearing of grouper	Flowery cod		Gold-spot cod	
	Month post hatch	Day post hatch	Month post hatch	Day post hatch
Month post hatch 40'	20µm	50µm	100µm	200µm
Month post hatch 90'	40µm	100µm	200µm	400µm

Day post hatch	0	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31	32	33	34	35	36
Algae (mix)	[Green bar from day 0 to 22]																																				
Copepods	[Green bar from day 2 to 16]																																				
SS-rotifers	[Green bar from day 6 to 16]																																				
Artemia AF	[Green bar from day 9 to 13]																																				
Artemia EG	[Green bar from day 13 to 28]																																				
Enriched rotifers (every hour)	[Orange bar from day 9 to 28]																																				
30-300µm	[Orange bar from day 9 to 16]																																				
150-500µm	[Orange bar from day 13 to 20]																																				
300 - 500µm	[Orange bar from day 16 to 23]																																				
400-600µm	[Orange bar from day 20 to 27]																																				
500-800µm	[Orange bar from day 23 to 30]																																				
800-1200µm	[Orange bar from day 27 to 34]																																				
1200-2000µm	[Orange bar from day 30 to 36]																																				

that of flowery cod during the juvenile phase (Figure 1). Cannibalism coincided with the start of metamorphosis and was the major cause of mortality.

Grouper larvae are very sensitive to handling stress prior to metamorphosis and grading is not possible. This results in very high levels of cannibalism of larvae during the metamorphosis window. On day 43, surviving juveniles were transferred into a raceway tank where they were graded into three size classes (<5.5, 5.5 - 6.5 and >6.5 mm bar width). Grading reduced cannibalism from 26.9 to 13.7% and cannibalism ceased altogether from day 65 onwards when the total length of juveniles averaged 5.7 to 6.3cm.

Behaviour

During metamorphosis (around day 30-35), larval behaviour changed from an active swimming, surface-oriented feeding pattern to a less active and benthic feeding behaviour. Artificial hides were introduced to correspond with this behavioural change and juveniles rather than larvae that had not yet metamorphosed, used them predominantly.

Juveniles were increasingly prone to startling in response to stimuli such as activity around the tank. They began to display behaviour similar to that of the adult fish around day 95 – 100. Territorial and dominant behaviour involved individuals displaying a pale underside to other fish or head to tail pushing and mouthing. The provision of adequate hides remained an important factor in reducing this aggressive behaviour.

Future application

This initial success in rearing of two grouper species indicates the application of mesocosm technology to rear multiple marine finfish species that have sensitive early life phases. Mesocosm systems provide a broader physical parameter range for the sensitive early larval phases reducing the need for strict control over conditions essential in small-scale intensive systems. The system can be changed to more intensive management once this early

phase has passed.

Research into production of juvenile grouper continues to focus strongly on resolving problems occurring during the early larval feeding stages. The current success represents a breakthrough that will allow the concurrent development of techniques for production of juveniles. The recent progress in this area forms the basis of the next project phase, where the more active transfer of outcomes to industry proponents can commence.



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