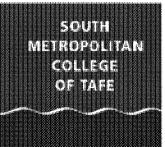
# *The Development of Aquaculture Techniques for Production of WA Dhufish (Glaucosoma hebraicum)*

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FISHERIES RESEARCH & DEVELOPMENT CORPORATION



Project 95/095

### 95/095 'The Development of Aquaculture Techniques for Production of WA Dhufish ( <u>Glaucosoma hebraicum</u> )'

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OBJECTIVES		

- The production of fertilised eggs from wild fish 1.
- 2. Production of fertilised eggs from captive fish
- 3. Larval rearing

### NON TECHNICAL SUMMARY:

### Need

By world standards Australia has not developed a significant marine finfish farming industry. One of the principal constraints has been the absence of suitable technology for Australian species. This technology is currently being developed in a number of research facilities in temperate regions of Australia.

There are currently several companies intending to farm marine finfish in Western Australia. The species intended for culture (snapper and black bream) have medium level prospects for price and markets. An urgent need exists for the development of technology suitable to culture a high priced market driven species, such as the WA dhufish reported here, to support the endeavours of this fledgling industry.

Information was obtained during the course of this project for WA dhufish for fish capture, growth rates, fish health, egg production and larval requirements.

### Fish capture

A total of 70 WA dhufish were captured, transported to the Fremantle Maritime Centre in Western Australia and acclimatised to captivity during 1995/6.

### Growth rate indicator

An indicator of the growth rate potential of this species was obtained when a 1 kg male captured early in December 1995 had grown to 1.6 kg by the end of March 1996.

### Fish health issues

As with all new aquaculture species the WA dhufish has parasites and other health concerns that can cause serious problems if not controlled in captivity. Two such problems have been identified with this species and these are exophthalmia, or 'pop-eye', and a gill parasite.

### Exophthalmia

A considerable number of fish held for this project developed unilateral exophthalmia. Staff at the Fremantle Maritime Centre, the Fisheries Department and Agriculture WA are continuing investigations into causal agents and preventative measures. It is important to note that this problem did not delay the research into the development of culture technology for this fish.

### Gill Parasite

A monogenean trematode, or fluke, which has been tentatively identified as from the Genus <u>Gyrodactylus</u>, is a common gill parasite on the WA dhufish. An effective treatment was discovered which consists of a one and half hour freshwater bath. This resulted in a near 100 % kill of the flukes with no adverse effects on the fish.

### Production of fertilised eggs

#### Natural Spawning

The majority of the WA dhufish females held in the Fremantle Maritime Centre tanks underwent gonadal maturation as the season progressed. One of these fish spawned spontaneously from December to February. The eggs collected were not fertilised but this was a significant achievement considering the term of this project.

#### Hormonal Inducement

Approximately 150 000 fertilised eggs were obtained by hormonal inducement using a) HCG saline injection; and b) LHRH slow releasing pellet. Fertilised eggs were obtained from broodstock held in captivity for one to six months prior to inducement. Eggs and sperm were hand stripped and fertilisation occurred artificially. Rates of fertilisation ranged from 0.3% to 95%. Hatching rates ranged from 4 to 37%. The total number of larvae hatched was 31 000.

### Larviculture

At a rearing temperature of 22° C, the mouth of the larvae opened by day 4 (by day 3 at 23.5° C). The first feeding organisms (rotifers, *Brachionus plicatilis*) were captured by larvae during the 5th day of age. Swim bladder inflation occurred at day 5. Nauplii of *Artemia salina* were accepted on day 17 and enriched metanauplii by 22 days of age. The larvae completed metamorphosis between days 30 and 40 at 22° C average rearing temperature. Approximately 500 larvae underwent metamorphosis. 24 juveniles survived and are still alive and well at 180 days of age.

### Conclusion

The performance indicators for project 95/095 were achieved, ie:

- 1. Successful production of eggs
- 2. Successful production of larvae
- 3. Successful production of fingerlings

Further research is being conducted on the development of culture technology for this species through a three year project supported by the South Metropolitan College of TAFE, the Fisheries Department of WA and the FRDC.

## Key words - fish culture; aquaculture development; aquaculture techniques; egg production; larval development; <u>Glaucosoma hebraicum</u>; WA dhufish; jewfish.

### BACKGROUND

The WA dhufish is the most valuable commercial finfish species in the Western Australian market. It is a premium quality, high value table fish with a wholesale value of \$12.50 per kg (whole fish) in WA. It is at least equivalent in quality and greater in value in the WA wholesale market to premium fish species at the Sydney Fish Markets such as the pearl perch, estuary cod, red emperor and pink snapper. (It should be noted that the wholesale value of the pink snapper in the WA market is approximately half of that obtained in the Sydney market).

Wild stocks of WA dhufish cater to the local seafood market in Perth and to a large recreational fishing fraternity. The wild stocks are not large enough to support an export industry, so the only avenue open for large scale exploitation of this species is by aquaculture.

The WA dhufish is reported by the WA Fisheries Department in their 'Biological Synopsis of Westralian Jewfish (*Glaucosoma hebraicum*)' (Sudmeyer et al, 1990) to grow to 30 cms in one year. As fish produced by aquaculture grow more rapidly than those in the wild, this reported rapid growth rate may allow one harvest per year, a significant advantage over many other marine finfish aquaculture research candidates.

Another factor which makes the WA dhufish a favourable candidate for aquaculture research is its low inherent stress level. WA dhufish caught from the wild are calm and can be handled and moved about in tubs with ease. They do not exhibit the typical agitated capture/escape response of the pink snapper and many other species of fish. They readily settle into culture tanks, resume feeding within a week of capture and can be handled relatively easily at any time. Low stress in broodstock is an advantage for an aquaculture candidate as stress level is related to natural spawning ie. low stress is a precursor of gonadal development.

### NEED

By world standards Australia has not developed a significant marine finfish farming industry. One of the principal constraints has been the absence of suitable culture technology for Australian species. This knowledge is currently being developed in a number of research facilities in temperate regions of Australia.

There are currently several companies intending to farm marine finfish in WA. The species intended for culture (snapper and black bream) have medium level prospects for price and markets. An urgent need exists for the development of technology suitable to culture a high priced market driven species, such as the WA dhufish reported here, to support the endeavours of this fledgling industry.

It is probable that the cost of WA dhufish culture would be similar to that of other marine finfish species. Therefore the high economic value of the WA dhufish would result in more flexibility in the way the industry can develop and grow. This may be particularly important to establish an economically viable marine finfish farming industry in WA.

Concerns have been expressed in various forums regarding uncertainty over the grow-out phase of marine finfish farming along the WA coastline. Such concerns include:

\* the lack of protected sea-pen sites along the high energy coastline of WA;

\* environmental implications of the location of sea-pens over or near seagrass beds. These include degradation from anchors, shading, sedimentation or eutrophication; and

\* the detrimental effects of high nutrient loading on areas immediately surrounding sheltered (and therefore low energy) potential sea-pen sites, where reduced wave and current action may limit the effective flushing and dilution of effluents.

These problems can be minimised by the establishment of land based facilities. The location of land based growout facilities adjacent to high energy coastal areas would avoid the present uncertainty surrounding the concept of sea based growout and result in a controlled growing environment, rapid dilution of effluents and subsequent reduction in environmental impacts. Land based growout facilities are however, more expensive to establish than sea based growout and economics will play a vital role in deciding which option to take.

The availability of a species such as the WA dhufish, with its high economic value and good marketability, may present the opportunity to carry out the grow-out phase of the culture process in land-based facilities.

There is a strong feeling among commercial and recreational fishing groups that in certain areas, the WA dhufish is under heavy pressure and its numbers are dwindling. Fisheries management of the WA dhufish includes size and bag limits but little is known of the biology of the species in the wild. A 'Biological Synopsis of Westralian Jewfish (Glaucosoma hebraicum)' (Sudmeyer et al, 1990) compiled by the WA Fisheries points out that "..there is a paucity of information regarding jewfish biology". This situation is currently being addressed with a three year FRDC supported Murdoch University investigation into the biology of the species.

Work on the current TAFE/Fisheries project has indicated that WA dhufish caught in depths of over 20 metres stand little chance of survival because of decompression effects rupturing capillaries within the fish leading to necrosis, infection and death. If substantiated with further research, this has major implications for fisheries management in that current practice is to impose a size limit and return undersize fish to the sea.

### **OBJECTIVES**

The objectives of the project were as follows:

- 1. The production of fertilised eggs from wild fish
- 2. Production of fertilised eggs from captive fish
- 3. Larval rearing

Due to the difficulty of capturing wild fish during the months of December 1995 to March 1996 we were unable to produce eggs from wild fish. We were successful however in obtaining fertilised eggs from captive fish. Larval rearing experience was gained from larvae that hatched from some of these eggs in March 1996.

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The performance indicators for the project were:

- 1. Successful production of eggs
- 2. Successful production of larvae
- 3. Successful production of fingerlings

These performance indicators were achieved.

### **METHODS**

### Broodstock collection

A total of 70 WA dhufish were captured, transported to the Fremantle Maritime Centre in Western Australia and acclimatised to captivity during 1995/6.

Fish were captured by hand line fishing in depths of less than 20 metres from waters off Ledge Point, approximately 200 km north of Perth. After fish were caught, they were placed directly into a 250 litre tub of seawater onboard the capture vessel where they were vented (the swim bladder punctured with a sterile 18 gauge hypodermic needle to remove excess gas) and held until the completion of fishing. They were transported ashore in a 50 litre tub and transferred into a 900 litre tank on a truck where they were provided with air and oxygen. The transport by truck from Ledge Point to the Fremantle Maritime Centre lasted two hours. On arrival, fish were anaesthetized with 2-phenoxyethanol, cannulated to check the maturation stage of the gonads, tagged intraperitoneally and allocated to a tank.

Broodstock WA dhufish were kept at the Fremantle Maritime Centre in the following tanks: two round fibreglass tanks of 10m<sup>3</sup> capacity, two oval shaped fibreglass tanks of 25m<sup>3</sup>, one round concrete tank of 40m<sup>3</sup> and one round fibreglass tank of 50m<sup>3</sup>. Most tanks were exposed to natural conditions of temperature and photoperiod. One 10m<sup>3</sup> tank was exposed to a photoperiod and temperature regime retarded by two months compared to natural conditions.

All tanks operated on a flow through system with a water renewal of at least 10% per hour.

The size of captured broodstock fish varied between 2 and 12 kg. Fish biomass in tanks ranged from 0.7 to  $3.0 \text{ kg/m}^3$ . The sex ratio of the males to females was 1:1 in all the tanks.

Feeding of broodstock WA dhufish commenced within the first two days after capture. Fish were fed a set diet of coral trout, squid, prawns and pilchards in an attempt to meet their presumed natural diet preferences. A vitamin premix and a product stimulating the immune response of the fish were added to the feed from February 1996 onwards.

### Natural spawning

To encourage natural spawning under culture conditions, the broodstock tanks were set up in such a manner that stress endured by the fish was reduced to a minimum. For instance, the tanks were covered with black plastic to protect the fish from excessive illumination.

Under the assumption that the eggs had a positive buoyancy, a 500  $\mu$ m mesh size egg net was installed on each tank overflow.

The conditions of photoperiod and temperature selected in the controlled environment tank closely matched the day length and temperature variation observed during 1994 at the WA Marine Research Laboratories (Marmion, WA). These data were the closest representation available of the conditions of photoperiod and temperature that are experienced by fish in the wild around the metropolitan area of Perth.

#### Hormonal inducement

The following inducing hormones were used in this study:

a) human chorionic gonadotropin (HCG) at a dose rate of +/- 1000 IU/kg body weight

b) luteinising hormone-releasing hormone analogue (LHRHa) in both saline and cholesterol-base slow release pellet forms at a dose rate of  $+/-100 \mu g/kg$  body weight

All hormones were injected intraperitoneally. Pellets were inserted using a 2mm inner bore needle on a tag inserter.

The stage of development of the female gonad was determined after removal of a sample by cannulation while the fish was under anaesthetic. The sample was examined microscopically and the size of the largest oocytes measured. Saline hormones (HCG or LHRH) were injected when egg size was 600  $\mu$ m in diameter or above. LHRH pellets were inserted if egg size was greater than 200  $\mu$ m and less than 600  $\mu$ m in diameter.

Males were also anaesthetized before verifying the stage of maturity of their gonads by stripping. Males were induced regardless of the stage of spermiation.

#### Fertilisation

Sperm was collected by stripping. The male fish were removed from the anaesthetic bath, rinsed with clean seawater and the abdomen dried with a towel. Pressure was exerted along the abdomen to release sperm which was collected in a sterile syringe. The sperm was kept on ice and the motility was checked under a microscope.

Eggs were expressed from a mature female after rinsing and drying the abdominal area. Eggs were collected in a measuring cylinder and transferred into one litre of filtered seawater. Sperm from at least two males was added directly to the suspension of eggs and both were mixed gently.

After 5 minutes, eggs were counted and the floating fraction was transferred into an egg incubator.

#### Larviculture

Larval trials were undertaken in three tanks of 300 litres and two tanks of 2 m<sup>3</sup> volume. These tanks utilised flow-through seawater filtered to 5  $\mu$ m. Rearing water temperature was maintained between 22 and 24° C.

The first feed organism offered to larvae was the rotifer *Brachionus plicatilis*. From 17 days onwards, the larvae were offered brine shrimp *Artemia salina* nauplii. Enriched brine shrimp metanauplii were provided from day 22.

### Photomicrography

An Olympus zoom stereo microscope (SZ6045TR-F) was matched to a SZ-STS illumination base and Olympus SC35 mm Type 12 photomicrography camera to record egg and larval development.

### DETAILED RESULTS

#### Size at maturity

The smallest female WA dhufish in spawning condition at the Fremantle Maritime Centre weighed 1.6 kg and measured 43 cm; the smallest spermiating male observed weighed 2.4 kg and measured 52 cm.

#### Natural maturation and spawning

The majority of the WA dhufish females held at the Fremantle Maritime Centre underwent gonad development in enclosed tanks after their capture and oocytes reached at least 300 µm in diameter.

One of the females of a weight of 5.5 kg, matured and ovulated naturally in a tank. This fish spawned spontaneously late in December 1995 after spending only three and a half month in captivity. The eggs were positively buoyant and were collected in an egg net on the overflow spillway of the tank. The collected eggs however, were not fertilised. Another female of 6 kg in weight matured naturally in a tank after 3.5 months in captivity but died prior to spawning due to a gill parasite infestation. The fish was stripped after death and 350 ml of ovulated eggs were obtained.

Four males of various sizes (2.6 to 7.5 kg) underwent natural gonad development and produced sperm of good quality after spending less than a year in enclosed tanks.

Male WA dhufish were observed to have low sperm volumes. The size of the male testes were relatively small (maximum observed gonadosomatic index of 0.17%) and did not substantially increase in size during the spawning season.

#### Induced maturation and spawning

Successful hormonal inducement of the WA dhufish first occurred in December 1995. A female was injected with 870 units HCG/kg body weight. Two males were injected with 780 and 1000 units HCG per kg body weight respectively. A total number of 60 000 eggs were stripped from the female; 500 eggs were subsequently fertilised and 100 hatched into larvae. This represents a fertilisation rate of less than 0.3% and a hatching rate of the fertilised eggs of 20%.

Further experimentation through January to March 1996 resulted in obtaining approximately 210 000 eggs in total, of which 150 000 eggs were fertilised and 31 000 hatched into larvae. The hatched larvae resulted from four different strippings during March of 1996. The fertilisation rates of the batches of eggs collected increased from 30% to 95% during this time, and hatching rates were successively 4, 21, 17 and 37%. These results were obtained with two females: the first was induced with 1200 units HCG/kg body weight by injection and the second with 110 $\mu$ g LHRH/ kg body weight by pellet implant.

A preliminary indication of the latency period between injection of the hormones and ovulation was obtained. These were approximately 44 hours after saline injection and approximately 6 days after implants. These results need to be verified during the 1996/7 spawning season.

### Larviculture

The fertilised eggs underwent development and hatched after 32 hours at 23° C. The mouth of the larvae opened at day 4 and the first feeding organisms (rotifers) were captured by the larvae from day 5 onwards. Swim bladder inflation occurred at day 5. There was 100% of larvae with positive swim bladder inflation. Artemia nauplii were offered from day 15 but were only captured at day 17 by the largest larvae. Enriched Artemia metanauplii were captured by larvae at 22 days old.

The larvae completed metamorphosis between days 30 to 40 at an average temperature of 22°C. Metamorphosis was characterised by the completion of adult features. At about 45 days of age, the larvae became benthic in preference to their previously planktonic orientation in the water column. The horizontal darkly pigmented band formation appeared approximately at this time and the fish became lighter in colour. Pelvic fins seemed very large in proportion with the others fins.

Nearly 200 larvae reached metamorphosis and 24 were still growing at Day 180. The largest of these fish were approximately 95 mm in length and 19 grams in weight.

A parasitic organism of the genus *Cryptosporidium* first infested the gut of the larvae prior to completing metamorphosis. This organism is 1 to 2  $\mu$ m in size and the genus is known to be pathogenic to fish. Symptoms of the genus include anorexia, regurgitation and digestion disturbances resulting in severe emaciation (Dr Brian Jones, Fisheries Department WA, Fish Pathologist, pers. com). The symptom observed was poor digestion from day 25 onwards with mortalities from day 37 to 48. There is no known treatment for this organism and the surviving juveniles were weak for some time. It is probable that this infestation caused a period of slow growth as the juveniles recovered.

Photographs of selected stages of stripping, egg, larval and juvenile development of the WA dhufish can be seen in Appendix 1.

### BENEFITS

The Western Australian marine finfish farming industry will directly benefit from the successful development of culture technology for the WA dhufish.

The increased knowledge that has been developed concerning the reproduction and early life history of the species may also assist in the management of the wild WA dhufish fishery. An article concerning the identification of eggs and larvae of the WA dhufish is currently being prepared for publication.

There are insufficient data at this stage to provide information about export prices, costs of production and non-market benefits etc.

### INTELLECTUAL PROPERTY

There is no intellectual property concerning this project.

### FURTHER DEVELOPMENT

Further research is being conducted on the development of culture technology for this species through a three year project (commenced July 1996) supported by the Fremantle Maritime Centre (FMC) of the South Metropolitan College of TAFE, the Fisheries Department of WA (FDWA) and the Fisheries Research and Development Corporation (FRDC).

### STAFF

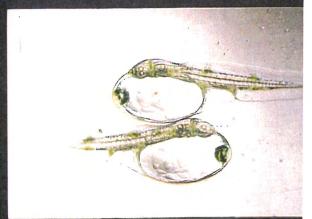
Personnel	Position	<i>Time commitment to project(%)</i>
Ms Francoise Pironet	Research Scientist (FMC)	100
Dr Noel Morrissy	Research Scientist (FDWA)	5
Mr Greg Jenkins	Project Manager (FMC)	20
Dr Brian Jones	Fish Pathologist (FDWA)	15
Mr Ken Frankish	Hatchery Manager (FMC)	20
Mr Bruce Ginby	Assistant Hatchery Manager (	(FMC) 25
Mr Kevin Smith	Technician (FMC)	30
Ms Tina Thorne	Technician (FDWA)	10
Mr Aaron Strawbridge	Technician (FMC)	5
Mr Craig Poller	Technician (FMC)	10
Mr Dean Kennerly	Technician (FMC)	50
Mr Anthony Arris	Technician (FMC)	5
Ms Rebekah Gilbey	Technician (FMC)	5

TAFE/ FISHERIES WA DHUFISH CULTURE PROGRAM 1995/6





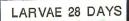
DEVELOPING EMBRYOS 22HRS



LARVAE AT HATCH

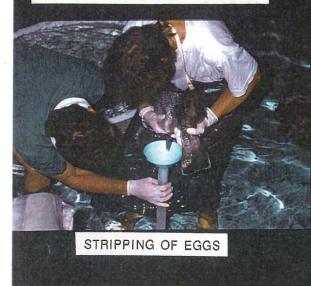








FEMALE IN SPAWNING CONDITION

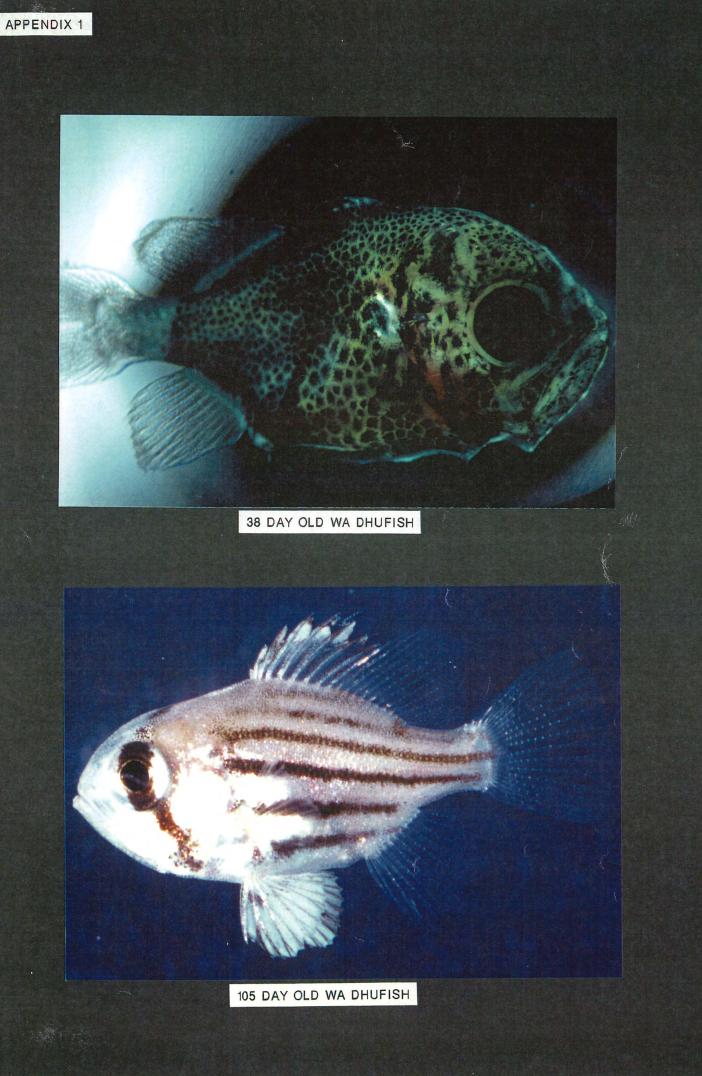




FERTILISED EGG - 5HRS

FERTILISED EGG - 12HRS





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