NEW ZEALAND FRESHWATER FISHERIES MISCELLANEOUS REPORT NO. 31

SILVER CARP SPAWNING

by

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Report to: Northland Regional Council

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Servicing freshwater fisheries and aquaculture

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NEW ZEALAND FRESHWATER FISHERIES MISCELLANEOUS REPORTS

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REPORT TO NORTHLAND REGIONAL COUNCIL ON SILVER CARP SPAWNING

SUMMARY

Silver carp were induced to spawn on three occasions between 1988 and 1989. About 10,000 fry were produced and transferred to Lake Omapere.

The number of brood stock is limited and climate and feeding conditions at Rotorua probably limit egg and fry production.

This report discusses spawning cycle, breeding methods and results, the problems and risks of breeding silver carp at Rotorua, and possible options for 1990-91.

The options are :-

- 1. Make further attempts to breed silver carp and produce sufficient fry in 1990-91
- 2. Seek to overcome climatic limitation and develop a hatchery in or near Kaikohe.
- 3. Abandon silver carp production.

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1. BACKGROUND

1.1 NUMBERS OF STOCK

When this project started, the main stock of silver carp was present in Lake Orakai and a smaller number had been stocked in Elands pond, Hawkes Bay. Nine silver carp were captured from Lake Orakai in June 1987, but it was apparent the population was smaller than we had thought.

On a further expedition in November 1988, we recovered 16 fish from Elands pond. The landowner informed MAFFish staff that most of this population was killed by flooding during Cyclone Bola. There are an unknown number of broodstock remaining in this pond.

Disease, the stress of transfer from Hawkes Bay to Rotorua, and induced spawning all took their toll. Anoxic conditions in one pond at Rotorua killed eight mature silver carp at the end of December 1989. There are five broodstock left in the Rotorua Ponds.

Last years fry will take at least four years to grow to maturity. We will try to recover further mature fish from Elands Pond this winter.

1.2 SPAWNING CYCLE

Like grass carp, silver carp are warm water fish which originate from large rivers in northern China. The optimum temperature for silver carp is about 25-27 °C. In nature, the fish move up river during the monsoon season (early spring) and spawn downstream of obstructions, in turbulent water.

The eggs float downstream for about 36 hours, before the larvae hatch out. Larvae continue to develop for a further 48 hours or so, until the gut and mouth parts develop. For the first week to fortnight, the fry feed on small zooplankton, but when they reach 12-14 mm, the gut elongates, body depth increases, and the fry start to feed on phytoplankton.

Growth to maturity can take two to five years depending on temperature and feeding conditions. Males usually mature before females.

In the females, eggs develop through several phases which increase the size and food reserves of the cell. Occytes grow from around 6 $\mu \rm m$ to over 1000 $\mu \rm m$. The pace of oogenesis is determined primarily by the mean and seasonal water temperatures and probably by the

condition of the female in the year prior to spawning. Occytes in all stages can be found in the gonads, but only a few ripen in any one year. The proportion and number that are ripe, probably depends on temperature and condition of the females in the previous summer. Those occytes which are sufficiently developed remain in the penultimate stage over winter.

With spring, environmental cues signal to the fish's brain to enter the final maturation in readiness for spawning. Hormones released by the pituitary gland control the final development of oocytes.

Maturation is a physiological process and is controlled by temperature. The temperature history is measured in physiological degree days (PD). Overseas reports and experience at Rotorua indicate that 1200 to 1500 PD are necessary for egg maturation. This is equivalent to 4-6 weeks in inside tanks at mean temperature of 24 °C, or the middle of December in outside ponds.

Ripe oocytes are held in readiness prior to ovulation (release of mature ova into the body cavity prior to fertilisation). Ovulation is triggered by specific environmental cues such as temperature drop, turbulent water, presence of other fish, pheromones and so on. These conditions stimulate a surge of hormones from the pituitary gland, which cause ripe oocytes to be released from the ovary into the body cavity. The eggs remain viable for a limited period, probably only about one hour. If mating occurs, ova are expelled through the vent and fertilised in water outside the body cavity.

We have no data for fecundity of silver carp in New Zealand conditions, but I expect it to be 20,000 - 50,000 eggs/kg each female. Fertilisation rates should be about 80%, and hatch out rates 40-60%. With perfect conditions and good females, a 5 kg fish may produce 100,000 fry from 250,000 eggs. If the induced females are not in good condition, more females would be needed to produce 100,000 fry.

A mature female in good condition has a slightly distended vent, the belly is soft to the touch and, occasionally, eggs can be expressed from the vent. Males develop small tubercules on the pectoral fins (fins feel like sandpaper), and milt can be squeezed from the abdomen.

Mature females are susceptible to stress and can spontaneously resorb the ova if they are badly handled or stressed in other ways.

1.3 BREEDING METHOD

1. Conditioning

The aim of induced breeding is to trigger ovulation in mature females carrying ripe eggs. The drugs used cause the same surge of hormones as natural environmental conditions, but enable us to predict ovulation time and strip out viable eggs.

However, it is self-evident that no amount of drugs can create ripe, viable eggs if they are not already in the penultimate stage. As outlined above, the quantity of mature ova is probably influenced by the condition of the female the year <u>before</u> spawning and the immediate temperature history in the two months prior to spawning.

At Rotorua, we try to create the best possible conditions for induction by bringing silver and grass carp into tanks inside a hot house for at least one month at 23-27 °C before induction. There are two main advantages in this procedure. Firstly, pond temperatures at Rotorua rarely exceed 23 °C which is too low for reliable spawning. Secondly keeping the fish indoors reduces the possibility that a sudden cold snap will cause oocyte resorbtion (for example, 27-30 November 1989). The fish are examined after 1200 PD and if the females appear in good condition, induction is attempted.

2. Hormones and induction

Until this year, whole carp pituitaries (Carp Pituitary Extract - CPE) collected from wild carp, and Human Chorionic Gonadotrophin (HCG) were used to induce ovulation. This method is labour-intensive and expensive, as wild carp have to be caught and the pituitaries extracted, dehydrated and prepared before injection. Apart from the cost (\$500 per induction) the method is unreliable because:-

- (a) there is no means of establishing the potency of carp pituitary before the induction attempt;
- (b) repeated inductions may induce an immune response in older broodstock which reduces the sensitivity to induction in future years; and

(c) at least two and sometimes three injections are given to induce the fish. Repeat injections cause stress which may cause resorbtion of the ova.

Recent overseas developments have demonstrated the efficacy of synthetic analogue hormones. Lutenizing Hormone-Releasing Hormone (LHRH-a) is a synthetic peptide which causes ovulation in several vertebrates. Two years ago salmon gonadotrophin (SGnH-a) was synthesised. Published reports indicate that SGnH-a is more powerful than LHRH. Both hormones are used in combination with a 'dopamine antagonist', either Pimozide or Domperidone.

Analogue hormones have several advantages over the CPE protocol.

- (a) It should be feasible to standardise the potency of the drugs.
- (b) There is no auto-immune response to the analogues so the efficacy does not decrease with repeat inductions
- (c) the drugs are cheaper (\$100 per induction).
- (d) the fish are less stressed since only one injection is given.

However, synthetic hormones do have disadvantages. In particular, the drugs are so powerful, that ovulation of unripe eggs that are not viable may be induced. This ovulation may take place several days after injection, and the additional stress may kill the female if the eggs are not stripped out. I believe that this may have occurred on two occasions with silver carp.

In both CPE and analogue hormone protocols, there is no external sign that ovulation has occurred. Fertilisation can only occur during a 'window' of about 1 hour 8-14 hours after the final injection. If fertilisation is too soon or too late, the eggs begin development then die 12 to 14 hours after fertilisation.

With grass carp, the usual way of assessing that ovulation has occurred is to examine the outlet water for eggs every half-hour and to periodically lift the females. If the females orgasm, or expel any eggs, we assume that ovulation has occurred.

Silver carp are not so helpful. They require a higher hormone dose for induction and appear to respond differently from grass carp to synthetic hormones. We have never observed eggs in the outlet water prior to stripping silver carp, and never observed orgasm when the females are lifted. In consequence we have

developed a protocol where the females are gently lifted, and the belly stroked to strip any ovulated eggs.

However the continual handling of the silver carp may cause partial resorbtion or blocking of ovulation.

2 BREEDING RESULTS.

We made two attempts to spawn silver carp in 1988 using the CPE + HCG protocol.

2.1 In the first attempt, sixteen fish were transported from Hawkes Bay on 3rd November and three females and two males were spawned with partial success on 24 November. The quality of the eggs (judged subjectively) was variable and the milt was poor and watery. About 20,000 fry were eventually produced from this batch, of which about 5,000 fingerlings survived.

While the fish were in the tanks, four fish died from stress caused by transport, and several other fish were infected by fungus (Saprolegnia sp.) In other words, all the fish had been subjected to considerable unavoidable stress in the month prior to spawning. I believe that this reduced the probability of successful spawning.

It is notable that this year (1989) males have been maintained running ripe by additional injections of LHRH the week prior to inducing females.

- In the second attempt of 1988-89 season, two females and one male were taken from the Rotorua ponds on 7 December and injected the same night. Good quality milt was obtained from the male. Both females ovulated but neither produced viable eggs. In this case I suspect the females needed further temperature conditioning before producing viable eggs.
- 2.3 In 1989-90 season, five males and four females were kept in hot tanks for one month then induced with LHRH-a, SGnH-a and Domperidone protocols. The males were injected with 50 μ g/kg LHRH and all produced viable sperm.

Two females were injected with 50 μ g/kg LHRH + 5 mg/kg Domperidone. Two females were injected with 10 μ g/kg SnGh + 5 mg/kg domperidone. Three out four females produced eggs, but fecundity was low and only one batch of eggs proved viable. Eggs from this female developed normally and hatching rates were 40-50%.

- 2.4 There are four possible reasons that fecundity, viability and hatch out rates from fish kept at Rotorua are so low:
 - (a) Lack of condition in the female stock deriving from poor growing conditions in the year prior to induction.
 - (b) Lack of sufficient temperature conditioning in the hot house prior to induction.
 - (c) Females were in good condition, artificial hormones did induce ovulation but the eggs were stripped out either too early or too late.
 - (d) A combination of any or all of the above.

3 PROBLEMS AND RISKS

Some of the problems and risks of inducing ovulation are described above. It should be evident that spawning silver carp is not a straight forward affair that we can 'do' at the drop of a hat.

Firstly the broodstock must be well-conditioned from the previous year. All the broodstock induced in 1988 had been stressed or otherwise disturbed that year. Broodstock examined in 1989 were in better condition, but none of the females reached 'soft belly stage' before being placed in the hot house. There is little we can do to improve feeding conditions at Rotorua since these largely depend on summer temperatures.

Secondly, the temperature conditioning of the fish is not a precise business but a trade off between cooking the fish to create the physiological conditions for maturity, and blocking them by stressing the females. It is possible to sample the ovary in situ (by catheterisation) to establish the development of the eggs, but this is a risky procedure. This year I judged that with only four females available, I could not risk damaging one of them.

Thirdly, there is no way of establishing exactly when ovulation occurs. I consider, with hindsight, that analogue hormones may induce ovulation sooner rather than later. In other words, that we may have stripped the fish too late.

Overseas, fish are in better condition to begin with and many broodstock are spawned at one time. Unfortunately, we don't have enough broodstock to bracket the peak time and guarantee viable eggs.

4 OPTIONS FOR 1990-91

- Obtain further fish from Elands pond and induce further fish at the end of 1990. Though this is feasible, there are a limited number of broodstock available, and MAF Fisheries cannot guarantee to produce 100,000 plus fry. We would use the hot house and analogue hormone protocols and the experience gained in the last two years will perhaps increase the probability of success. However, an attempt in 1990 is largely a decision for the Northland Regional Council (NRC) as MAF Fisheries will not attempt to induce spawning without the support of NRC.
- 4.2 Climatic conditions in Lake Omapere are more favourable to silver carp than the conditions at Rotorua. It would be worthwhile transferring some broodstock to Omapere. Ideally the broodstock should be transferred to a site close to Omapere in a small secure pond. Given that we only have 8 broodstock remaining, MAF Fisheries would have be satisfied that the site was secure.

A separate proposal was made to the Foundation for Science and Technology for funding to develop a hatchery near Omapere but was not successful. The total cost would be about \$60,000 in the first year, and \$50,000 per year in subsequent years. Alternative sources of funding could be explored.

4.3 Northland Regional council may wish to abandon the proposal to stock Lake Omapere with silver carp.

5. COSTS OF SPAWNING SILVER CARP IN 1989.

Total cost of inducing spawning in silver carp from July 1st to November 26th, 1989 was :-

Personnel and overheads 4842 Consumables $\underline{642}$ Total $\underline{5484}$

The total cost included some further personnel costs involved with feeding fry and transporting fry to Kaikohe and the air freight.

Under the terms of the contract NRC paid only \$3700 plus the cost of air freight of fry to Kaikohe.

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