Induced Spawning of the Common Carp and Aischgrund Carp (*Cyprinus carpio*) and the Largemouth Yellowfish (*Barbus kimberleyensis*)

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Abstract

Spawning of the common carp (*Cyprinus carpio*), the Dinkelsbühl variety of the Aischgrund carp (*C. carpio*) and the largemouth yellowfish (*Barbus kimberleyensis*) was induced by means of pituitary gland extracts obtained from Aischgrund carp donors. The ova were artificially fertilized and kept in Zuger jars up to the hatching stage whereafter they were transferred to trays suspended in 1 m^3 indoor tanks into which the larvae were released after hatching.

Introduction

Induced spawning and the artificial fertilization of fish ova are valuable techniques which facilitate mass production of fry for fish farming purposes and the breeding of rare and endangered species for restocking purposes. This paper deals with experiments conducted during 1979 at the Hartbeespoort Dam hatchery of the Transvaal Nature Conservation Division which resulted in the first ever successful artificial fertilisation of the largemouth yellowfish (*Barbus kimberleyensis*) ova.

This extremely popular angling species is one of the few endemic predators in the Orange River system. Due to the fact that females attain sexual maturity at an age of eight years (Mulder, 1973) and to the ever increasing turbidity of the Vaal and Orange Rivers as a result of erosion and to the tremendous angling pressure it is subjected to, it was deemed necessary to investigate the culture of large numbers of the species for restocking purposes. The water supply to the hatchery is drawn from far below the surface of Hartbeespoort Dam and due to the relatively low temperature, natural breeding in the ponds normally commences in early summer. To condition brood fish to spawn at an earlier stage they were transferred to heated tanks within the hatchery building. This not only facilitated the handling of the fish during induced spawning experiments but also resulted in having fry available much earlier in the season. Induced spawning was based on the methods described by Rothbard (1978) and two varieties of carp were used *viz.* the common carp (*Cyprinus carpio*) and the Dinkelsbühl variety of the Aischgrund common carp.

Methods

Pituitary glands were obtained from mature male and female Aischgrund carp donors varying in mass from 800 to 1 000 g and preserved in absolute ethyl alcohol. A few hours before use the glands were removed from the alcohol, dried on blotting paper and ground to a fine powder by means of two spoons. The powder was diluted with a 0,9% saline solution.

Prior to treatment fish were transferred to the hatchery and the sexes kept apart in 1,0 m^3 plastic containers. For two weeks the temperature of the water was gradually raised to 23°C and kept constant for a week before pituitary dosages were administered. Throughout the experiment the water temperature was regulated between 23°C and 25°C and a waterflow of 6 ℓ/min was maintained through the tanks. For one week the fish were treated regularly with 40 mg/ℓ formalin and 0,15 mg/ℓ malachite green in the tanks to prevent infections on wounds sustained during handling and also to remove external parasites. During treatment the waterflow was cut off for a period of 30 min after which the normal waterflow was resumed. Prior to spawn taking all fish were anaesthetised with 1 g of M.S. 222 dissolved in 10 ℓ water. Care was taken to use the same water as the holding tanks to prevent shock, and within 2 min the fish were sufficiently anaesthetised to be handled.

To prevent coagulation of ova by water, the fish were dried carefully prior to the fertilization process and the ova and sperm were stripped into a container while continuously stirring with a feather for 2 min. Rinsing commenced immediately to remove albuminoid substances. The rinsing process was done according to the technique originally described by Wyonarovich (1962) and modified by Rothbard (1978). It consists of two solutions which are made up as follows:

Solution A
- 3 g carbamid (urea)
- 4 g table salt
- 1 ℓ water

Solution B
- 1 g tannic acid
- 1 ℓ of water

Solution A was used to rinse the eggs for a minimum period of 45 min and the solution was replaced every 5 min. The ova were then rinsed in solution B for 10 s and then again for the same period using solution B diluted by half with tap water. The rinsing process was concluded using water to remove all remaining particles. The ova were then transferred to Zuger jars (Schäperclaus, 1949) where the water temperature was kept fairly constant at approximately 24°C and a flow of 1,5 ℓ/min was maintained through the jars.

Hormonal Treatment

Common carp

On 27 August 1979 at 22h00 three females and four males of approximately 6 kg body mass were injected each with one pituitary gland made up to 1 mℓ with a physiological saline solu-

tion. At 07h00 the following morning the females received a further dosage of pituitary gland extract. The dosage was calculated on a basis of 1 gland per kg body mass. Six pituitary glands were therefore homogenized, made up to 1 mℓ with 0,9% saline and injected intramuscularly posterior to the dorsal fin.

Aischgründ carp

On 17 September 1979 at 13h00 three females and four males of approximately 5 kg body mass were administered one gland each and the females were given a second dosage at 22h00 at a concentration of 1 gland per kg body mass.

Largemouth yellowfish

For this experiment only one female of 62,5 cm length and body mass of 4 kg, and two males of 2 kg each were available. Females of this species attain sexual maturity at approximately 55 cm and eight years of age (Mulder, 1973). As a result of the difficulty experienced in grinding one pituitary gland to a fine powder and the consequent loss of some of the substance, 4 glands were made up to 3 mℓ with physiological saline. One third thereof was administered at 22h00 on 23 October 1979 and the remaining dosage was given, as in the case of the carp, at 07h00 the following morning. Both males were given 2 glands each at 22h00 on 23 October. This was done to ensure that sperm would be freely available as was experienced with the two varieties of carp males. In both these cases it was found that stimulation by means of the pituitary glands resulted in a free flow of sperm when even the slightest pressure was applied to the male's abdomen, and this was also the case with the yellowfish males.

Results

Common carp

Within 9,5 h after the second dosage, one of the females exhibited breeding behaviour by chasing the others and pressing them into the corners of the tank. The ova were stripped, fertilized, rinsed and placed in the Zuger jars as described. Hatching commenced 52 h thereafter and eggs were immediately transferred to wooden trays with fine mesh bottoms and submerged 5 cm below the surface of the large tanks. Water was sprayed gently over the surface at a rate of 40 ℓ/h (Rothbard, 1978). Hatching was completed in 70 h.

Aischgründ carp

Two of the Aischgründ carp females reacted within 4 h of the last dosage. This was only discovered at 03h00 when they were immediately stripped of 1,5 ℓ of ova. The third female yielded 2,5 ℓ of ova at 06h00. The same procedures as mentioned above were employed in the rinsing and handling of the ova. Hatching commenced after 52 h and was completed in 72 h. Larvae of both carp varieties were motile within 19 h after hatching and feeding with hard boiled egg yolks was started.

Largemouth Yellowfish

The largemouth yellowfish female did not exhibit any breeding behaviour at all, yet when she was examined 19 h after the last dosage of pituitary gland extract she yielded approximately 6 000 ova which were fertilized. As this did not correspond to an average of 30 000 ova expected of a female of this size she was examined 24 h later and another batch of approximately 1 000 ova was obtained. After this she did not yield any more. Following the experience of Mulder and Franke (1973) the ova were not rinsed in the manner described by Rothbard (1978). Although the ova did cling together after they had been transferred to the Zuger jars they separated within 1 h after fertilization. The special rinsing process was used with the second batch of ova and although it resulted in the separation of the ova they all died within 3 h. This, however, cannot be ascribed to the rinsing process alone, as the ova might not have been fully mature.

As in the case of the carp varieties the ova started hatching within 53 h and was completed after 72 h. Upon hatching the larvae descended through the mesh to the bottom of the tanks where they congregated under submerged objects such as piping.

They tended to avoid light and eye pigments formed 30 h after hatching. The larvae remained immotile for approximately 86 h after which they rose to the surface and began to swim around in the tank. Ten hours later when most of the larvae were actively swimming about, feeding with finely sifted plankton and boiled egg yolk was started. These results compare favourably with the observations of Mulder and Franke (1973) and Groenewald (1961) who found that the motile stage of *Barbus holubi* larvae was reached between 4 and 6,5 days. In their experiments the average temperature of the water did not, however, exceed 21,5 °C.

Summary

Results obtained from the above-mentioned experiments clearly illustrate the validity of the techniques and pituitary extract dosages as prescribed by Rothbard (1978) for the three types of fish used here. As in the case of findings by Rothbard (1978) and Wyonarovich (1962) with carp, the process resulted in a more than 85% hatching success and 500,000 larvae could be grown and treated in 3 m^3 water for up to 3 weeks before being transferred to the nursery ponds. Similar to the findings of Mulder and Franke (1973) for *Barbus holubi* the hatching success of the largemouth yellowfish (*B. kimberleyensis*) was more than 95%. The advantages of this technique for the fish culturist are numerous when considering the tremendous mortalities amongst larvae due to coagulation and predation in nursery ponds. A much smaller breeding stock can be kept, selection is made easier and minimal losses are experienced due to the controlled environment for the first three weeks.

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