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# PRELIMINARY STUDY ON THE BREEDING TECHNIQUES OF *Neolissochilus soroides* (Duncker,1904)

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**Abstract**

Tengas or Copper Mahseer is highly demanded and commercially sought after angler. It has significant aquacultural and conservational values but still no study has been done on the reproduction and breeding technique for this species. To protect the species from extinction, few attempts were undertaken to breed and produce seed of Tengas in a captive rearing system. Fish were acclimatized, artificially fed and prepared to breed in captive conditions. Two breeding methodology with the use of hormones is discussed in this paper. However, this species was not successfully bred due to several factors. Induced breeding for this species is possible if the broodstock is matured enough to spawn and the provision of better climatic conditions in spawning tank may help to induce the maturity in this species.

*Keywords*: *Neolissochilus soroides, breeding technique, aquaculture production, conservation*

## 1 INTRODUCTION

*Neolissochilus soroides (Dunker,1904),* locally known as "Tengas" or "Copper Mahseer" are widely distributed in freshwater habitats throughout tropical and subtropical areas of the southern and southeastern Asia (Rainboth, 1991). 2 species of the genus *Neolissochilus*, *Neolissochilus soroides* (Duncker, 1904) and *Neolissochilus hendersoni* (Herre,1940) are reported to occur in peninsular river systems (Khaironizam, 2013). In peninsular Malaysia, *N. hendersoni* is endemic to northern part of Peninsular Malaysia (Khaironizam, 2010) while *N. soroides* has a widely distribution range through Cambodia (Rainboth, 1996), Thailand (Vidthayanon and Kottelat, 2003). This species reported has high market demand in the aquarium trade and sought after by anglers in Malaysia (Khaironizam, 2010).

According to Bagenal (1968) most of tropical fish reproduce throughout the year. Ingram (2007) reported that spawning in Mahseer takes place after rainy season which normally occurred from December to March. *N.soroides* shows high percentage of ripe oocytes during heavy rainfall, suggests 2 peaks of spawning activity of this species take place in a year and may precede the heavy rains. (Khaironizam and Zakaria-ismail, 2013).

Captive spawning and technology is used in order to supplement populations of many species that are declining in the wild (Hitoshi et al., 2007). However, the captive production for most species in aquaculture has proven challenging mainly due to the complex reproductive biology, the small size of the newly hatched larvae, the long larval phase and the lack of suitable feed which eventually leads to poor survival of larvae (Madhu and Rema, 2014). In order to manage any species, knowledge on their early life, breeding biology and feeding are the most importance (Rahman et al,. 2011).

Although some studies on the reproductive biology, condition factors and feeding habits of Neolissochilus soroides have been reported by Khaironizam and Zakaria-Ismail,2013, none information on the breeding technology and induce spawning of this species have been reported. Considering the enormous important of *Neolissochilus soroides,* information on the early life stage is an important requirement for optimization large scale seed production. Studies on embryonic and larval development are useful

for fish farmers to enhance their seed production by promoting larval growth and survival (Bilal *et al*., 2014). Hence, this study was attempted to provide information on the suitable breeding techniques and embryonic development as well as larval development of the induced bred Tengas, *Neolissochilus soroides* under captive conditions.

## 2 MATERIALS AND METHDOS

### 2.1 Broodstock selection

Brood fishes of *Neolissochilus soroides* were collected from Sungai Chiling, Kuala Kubu Baru, in collaboration with Department of fishery, Selangor. Sampling was conducted three times started from December 2015 followed by second and third times in January and February 2016 respectively. A total of 15 *N.soroides* broodstocks were sampled during the first sampling and 6 broodstocks were collected in January and February 2016. Sampling and spawning experiment were conducted during the natural breeding season of N.soroides from November 2015 to April 2016 (Khaironizam & Ismail, 2013). In situ water quality parameters recorded during sampling were as follows: water temperature ($23.94\pm0.73^0$C), pH (7.5 ± 0.15) and dissolved oxygen (7.5± 0.15 mg/L). All broodstocks were transported to Aquatic Lab, Faculty of Science and Biotechnology, Universiti Selangor with least disturbance and reared in fibre tank (2m x 1m x 1m). All brooders were acclimatized to laboratory conditions and fed by high protein pellet for a month before been used for first breeding experiment. The lengths of brooders were ranged from 20 – 40 cm and 200 – 540 gm weight.

### 2.2 Induced Breeding

Induced breeding experiments were conducted during the natural breeding season (November – April) of *Neolissochilus soroides* in fibre tank size (2m x 1m x 1m) capacity. Three trials were performed by using two different breeding techniques throughout the experiment. Two techniques that have been used in this experiment were semi-natural breeding and artificial propagation. Healthy males and females broodstock used in both techniques were selected by sexual dimorphism (Bilal et al. 2014). In both methods, ovaprim hormones (0.5 ml/kg) were used as it was the most successful hormone treatment and dosage for an artificial propagation for Thai Mahseer (Kunlapapuk & Kulabtong, 2011, Ingram et al., 2005).

#### 2.2.1 Semi-natural propagation

First attempt was conducted on December 2015, by using semi-natural propagation. In this experiment, two sets of broodstocks consisted of two females and one male each set with range size (150 – 240 gm) and (20 – 25 cm). Male and female of broodstocks prepared for spawning were anaesthetized to set the fish in deep narcosis in 2-3 minutes and ready to be manipulated with less injury (Adam Nemeth, 2013).

Injections were administered intramuscularly in the dorsal lateral region of the body. Injections were given during late night. Immediately after administration, the breeding sets were released together into the spawning tank for natural spawning activity. During the early hours, the brooders were closely observed for their responses to hormone and breeding behavior. The water quality parameters recorded during the study were as follows: water temperature ($29.7^o$c ± $0.46^o$c), pH (6.53 ± 0.25), and dissolved oxygen (5.5 ± 0.52 mg/L).

#### 2.2.2 Artificial propagation

Second and third attempt were conducted on February and March 2016, by using artificial propagation method. In these experiments, three females and four males with body weight 350-540gm with 32-40cm in length were used. All broodstocks were anaesthetized to set the fish in deep narcosis prior to hormone injection with less injury.

All fish were covered with wet towel during hormone injection. Ovaprim hormone with 0.5 ml/kg were used for both trials, hormone were administrated intramuscularly in the dorsal region of the body. After injection, both females and males were placed together in a fibre tank with aeration.

After 12 hours, the stripping procedures were applied to both male and female by gently

pressing on the abdomen after anesthetization in order to get the milt and eggs from the broodstock.

## 3 RESULT

The present study used two methods of induced breeding (semi natural propagation and artificial propagation) through hormonal administration (0.5 ml/kg ovaprim).

The first attempt was using a semi natural propagation, with hypophysation breeding set were released back immediately to the spawning tank for natural spawning activity. Broodstock in spawning tank were observed for breeding behaviour and 8 hours after hormonal administration, courtship was noted whereby 2 males were chasing a single female. After 12 hours, still no eggs and milt were obtained from the natural spawning activity. Semi natural spawning method was used during the first attempt due to the small size of broodstock, and gentle press on the abdomen will harm the fish.

The second and third attempts were conducted two and three months after the first attempt with bigger size of broodstock. Artificial propagation was conducted by using hormonal administration and stripping procedure was applied on the male and female 12 hours after hormonal administration. During the both attempts, milt were successfully stripped from the males, however there are still no eggs were successfully stripped out from the females.

Physicochemical characteristics of the water from natural environment and during experiment are given in Table 1.

Table1: Physiochemical of water quality parameter in natural environment and during the experiments.

| Parameters | Natural | Captivity |
|---|---|---|
| Dissolved Oxygen (mg/l) | 7.5 ± 0.15 mg/l | 5.5 ± 0.52 mg/l |
| pH | 6.91 ± 0.90 | 6.53 ± 0.25 |
| Water Temperature | 23.94 ± 0.73$^0$C | 29.7 ± 0.46$^0$C |

## 4 DISCUSSION

Till date, there are no reports on the breeding technology of *Neolissochilus soroides*, previous study on this species was focused on its biology aspects and its distribution. No reference is available on its breeding technique with or without hormonal administration for this species. This study was the first attempt made to breed this species in captivity but could not succeed due to several factors. Even hypophysation failed to induce the spawning of *Neolissochilus soroides*.

The main factor which leads to the unsuccessful breeding of *Neo.soroides* is none of the females were found ready to spawn and could not be sexually matured in captivitiy though males were sexually matured and always ready to spawn. Sampling of Neolissochilus soroides were conducted during the rainy season in December 2015 for the first sampling followed by second and third sampling during the extreme dry periods in January and February 2016. Extreme dry season affects the sampling process of wild matured broodfish.

Extrinsic factors such as temperatures, annual rainfall distribution and photoperiod may had little effect on fish breeding activities (Gillet and Quetin, 2006; Khaironizam and Ismail, 2013).
Other factor that leads to the unsuccessful breeding of *Neo.soroides* is that the difference between natural and captive conditions i.e dissolved oxygen, water temperature and water current may cause stress or may not provide the cues needed in reproductive process.

## 4 CONCLUSION

Knowledge on the breeding techniques and early development of Neolissochilus soroides is important in order to protect this species as well as to develop their culture techniques for aquaculture purposes. Induce spawning technique for this species is possible if the broodstock is matured and ready enough to spawn. The provision of better climatic conditions in spawning tank may help to induce the maturity in this species.

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