MORPHOLOGICAL DEVELOPMENT OF NAPOLEON WRASSE, *Cheilinus undulatus* LARVAE

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ABSTRACT

To improve the survival rate, napoleon wrasse larval rearing trial was conducted at Research Institute for Mariculture, Gondol-Bali in 2003. The trial aims at assessing initial feed for larvae, food habit, and morphological development from early larval stage to juvenile. The results showed that chicken egg yolk could be applied as initial feed and followed by rotifer, *Artemia* and mysid (*Mesophodopsis* sp.). Three swimming behavior of larvae were observed, drifting, free swimming and hiding on the substrate as larvae develop. Digestive system development, simple tube like, transition stage and coiled where digestive system could be distinguished between stomach, intestine and rectum.

KEYWORDS: napoleon wrasse, larvae, feed, morphology

INTRODUCTION

Napoleon wrasse, *Cheilinus undulates,* locally called Siomae, Lemak, Lambe is a humphead fish distributes around coral reef area in Indonesia, Malaysia, Philippine, and Western Pacific Islands. Lives around caves near coral reef, they feed on mollusks, crustaceans, echinoderms, etc. (Anonymous, 2004). Naturally, first maturation occurs after total length reaching 40—50 cm (Sadovy, 2005) or after 5 years old; its live span was at least 3 decades and spawns in a group mating (Anonymous, 2004).

Napoleon wrasse is one of the most economical fish in Asia with the major market destinations are Hong Kong, Taiwan, and Singapore (Anonymous, 2004). The high price is due to its good taste and the scarcity of this fish in the nature. Napoleon wrasse population is nearly extinct caused by the high fishing intensity, non-selective fishing and the low rate of stock recruitment or probably has been over fished. Size composition of captured were ranged from 0.25 to 10 kg/fish meaning that non-selective fishing has been undergoing (*personal comm - fishermen.*).

Napoleon fish has become an international issue because this fish is nearly extinct and now is listed in Appendix II of the CITES (Sadovy, 2005). In 1995, Indonesia announced the regulation HK.330/S3.6631/96, to prohibit to catch and export but no implementation and there is still widespread use of mariculture exemption to take juveniles from the wild and hold them until sale (Anonymous, 2004).

Realizing the problem, Research Institute for Mariculture, Gondol-Bali started napoleon wrasse breeding program in 1998. Development of broodstock acclimation technique, gonadal maturation and spawning technique by water management, food, vitamin, and hormonal inducement have been implemented successfully in captivity (Slamet & Sutarmat, 2001; Slamet & Sutarmat, 2001; Sutarmat & Slamet, 2002; Slamet *et al.*, 2002; Slamet *et al.*, 2003; Slamet *et al.*, 2003; Slamet & Sutarmat, 2003).

Embryology study was conducted by Yunus *et al.* (1999) and embryonic development faster than other marine finfish and in 12—13 hours after spawning the eggs hatched-out. Compared to Marble grouper (*Epinephelus microdon*) and humpback grouper *Cromileptes altivelis* which need about 18 hours after fertilization (HAF) to hatch (Tridjoko *et al.*, 1996; Slamet & Tridjoko, 1997), and Orange spotted grouper *E. malabaricus*, 17-19 HAF (Ruangpanit *et al.*, 1993).

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Development of hatch-out technique was also conducted by Imanto *et al.*, 1999; showed that incubation time for Napoleon wrasse eggs was about 17—24 hours after spawning under water salinity of 33 ppt and temperature of 29°C—31°C. However, larval rearing development technique is still left behind. Suitable feed for initial feeding period is the main problem in larval rearing of this fish.

Newly-hatched napoleon wrasse larval size is very small and the mouth opening size at the initial feeding is much smaller. Actually, based on measurement of mouth opening size, the size of super small (SS-type) rotifer is suitable for initial feed. Unfortunately, from several trials conducted, it was never found rotifers in the digestive tract of the larvae (Ismi *et al.*, 2001). This means that rotifer was not suitable for initial feed for napoleon wrasse larvae. Probably, at the initial feeding period, napoleon larvae filter smaller particles. It is frequently found unidentified small particles in the digestive tract of larvae.

Trials using pytoplankton such as *Nannochloropsis* sp., *Tetraselmis* sp. (*personal com.*), and fish egg powder (*personal comm.* - Suko Ismi) as initial feed showed that high percentage of larvae successfully ingest those particles but after several days no larvae survive. Conclusion from those trials was napoleon wrasse larvae may filter smaller particles but those phytoplankton and egg powder were not able to be digested.

Based on trials above, study on larval development was necessary to conduct to understand feeding habit and morphological development from newly hatch to juveniles of napoleon wrasse.

MATERIAL AND METHOD

Trial was conducted at Research Institute for Mariculture at Gondol-Bali in October 2003. One-m^3 cylindrical fiberglass tank filled with 0.7 m^3 filtered sea water was used for this trial. Inner wall of the tank was yellow and the tank was equipped with continuous aeration system and cartridge filter to supply fresh sea water if necessary.

Napoleon wrasse eggs were provided from natural spawning of broodstock in 100 m^3 concrete tank. Eggs were collected from broodstock tank into an egg collector by water flow-through system. One hour after spawning, eggs were collected and placed in 30-l fiberglass to separate between fertilized and unfertilized egg. Fertilized and developing eggs (morula stage) were transferred into larval rearing tank. Number of eggs transferred was 35.000 (50 eggs/L).

Larval rearing method applied was modification of milkfish and grouper larval rearing method (Ahmad *et al.*, 1993; Sugama *et al.*, 2001).

Boiled chicken egg yolk was applied as initial feed for larvae. Prior to feeding, yolk was sieved through 30 ìm mesh size of plankton net. Size compositions of egg yolk particles were:

0.5 - 1.0 µm	:	60%
1.0 - 5.0 µm	:	20%
5.0 - 20.0 µm	:	10%
> 20 µm	:	10%

Observed and measured parameters were feeding habit, digestive development, the presence of feed in digestive system, and morphological development of larvae.

RESULT AND DISCUSSION

Hatching rate of egg was 90%, this value was high compared to those of the same spawning period (0%—90% hatching rate). Total length of newly hatched larvae was 1.97 mm (Figure 1). The survival activity index (SAI) of the larvae was 2.0 (SAI in October 2003 spawning period ranged from 0.4—3.0) meaning that high hatching rate was not related with high quality eggs. This finding agreed with Mushiake & Sekiya (1993), that high SAI showed less initial floating death and no relationship among floating rate, fertilization rate and hatching rate or egg and oil globule diameter of egg and survival activity index. Hence, egg quality still needed to be improved. On stripped jack (*Pseudocaranx dentex*), larval production was only possible when the SAI exceeded 6.0 (Mushiake & Sekiya, 1993). But with low SAI value, it was still possible to produce some napoleon juveniles.

Boiled chicken yolk as initial feed was applied from D-3 (3 days after hatch-DAH) 3 times a day. Observation under microscope clearly showed that larvae ingested the yolk particles and improved survival and growth of larvae (Figure 2).

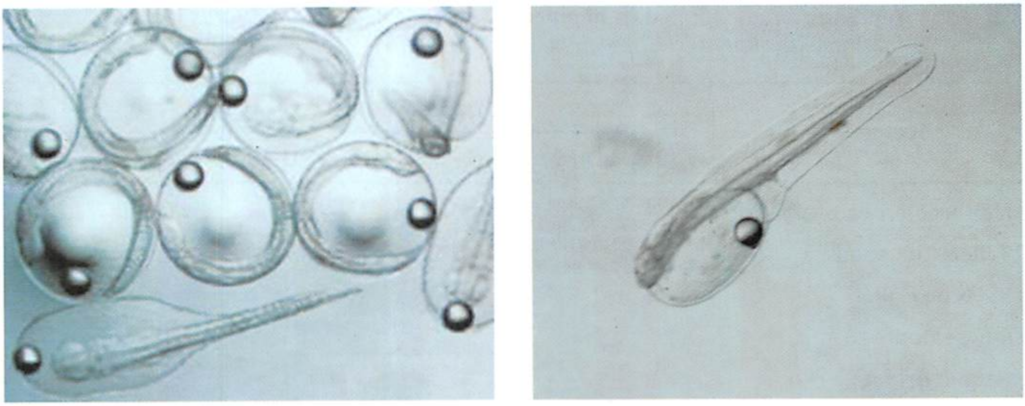


Figure 1. Egg and newly hatched napoleon wrasse, *Cheilinus undulatus* larvae (20x)

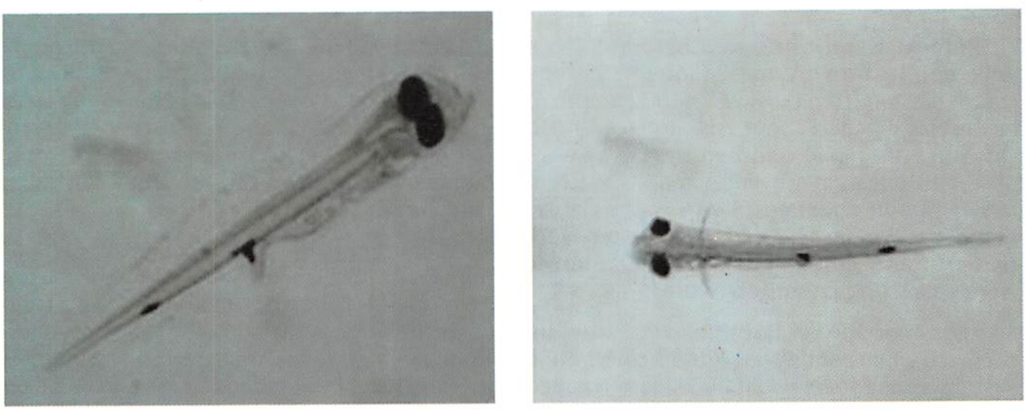


Figure 2. Napoleon wrasse, *Cheilinus undulatus* larvae at initial feeding (D-3), TL: 2.0—2.3 mm (20x)

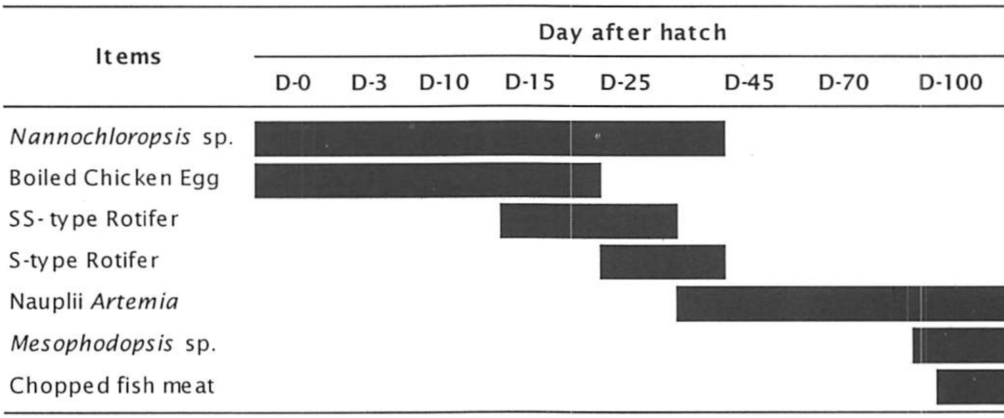
Feeding using yolk was not without problems, since the water quality tended to deteriorate. To solve this problem, 3 efforts, i.e. siphoning the uneaten feed, stocking *Nanochloropsis* sp. at a density of 0.2—0.6 $\times 10^6$ cell/mL to stabilize water quality as well as feed supply for rotifer, and exchanging water about 10% per day from D-3 to D-12 and increased gradually to about 50%—100% per day from D-20 to juvenile.

Failure in napoleon wrasse larval rearing occurred mostly due to disability of larvae to prey on rotifer or copepod nauplii introduced into the larval rearing tank. Based on the mouth opening size of about 133 um, theoretically some percentage of rotifer or copepod nauplii should be able to be ingested but from some trials conducted, it has never been found rotifer (Ismi *et al.*, 2001) or copepod nauplii (*personal com.*) in the digestive tract of larvae at the initial feeding stage. Another possible reason was movement speed of those feed organisms was too fast for the napoleon larvae to catch.

The results of the trial gave one suggestion for feeding regime on napoleon larval rearing (Table 1). Even so, it is necessary to find live organisms for initial feed to properly manage water quality in the larval rearing trial.

Feeding with chicken yolk continued until D-18. Larvae fed actively and the stomach part became yellowish. Based on this observation, it was confirmed that larvae fed on the chicken yolk and not from bacteria that growth in the yolk.

Table 1. Feeding regime applied in napoleon wrasse, *Cheilinus undulatus* larval rearing experiment



Rotifers were introduced before chicken yolk application period stopped. SS-type rotifer started from D-14 when total length of larvae was reached about 2.95 mm (Figure 3). One hour after giving rotifer, the larvae were observed under microscope and stomach of larvae samples contains 5—15 rotifer/larvae even the larvae still fed on chicken yolk particles and probably the Napoleon larvae were able to ingest rotifer before D-14.

High mortality of larvae on D-15 at night occurred when continuous water flow-through system applied. A lot of larvae were trapped on the tank wall and died. Probably, at night larvae did not swim actively and just drifted with the current.

Artemia nauplii were introduced from D-34. It seemed that larvae did not feed on *Artemia* when copepod available in the larval rearing tank. In this age, larvae tended to hide around tank wall where a lot of filamentous algae grew. Larvae used the algae for hiding and hunting copepod.

Total length of larvae reached about 9—11 mm on D-40 and some were metamorphosing to juvenile. Their habit, body shape and pattern were the same as the adult fish. Metamorphose to juvenile perhaps was started several days before. Juveniles tended to hide and came out when chasing the feed. Swimming pattern of juvenile changed, when chasing the feed, juvenile swam straightly and fast, but in passive

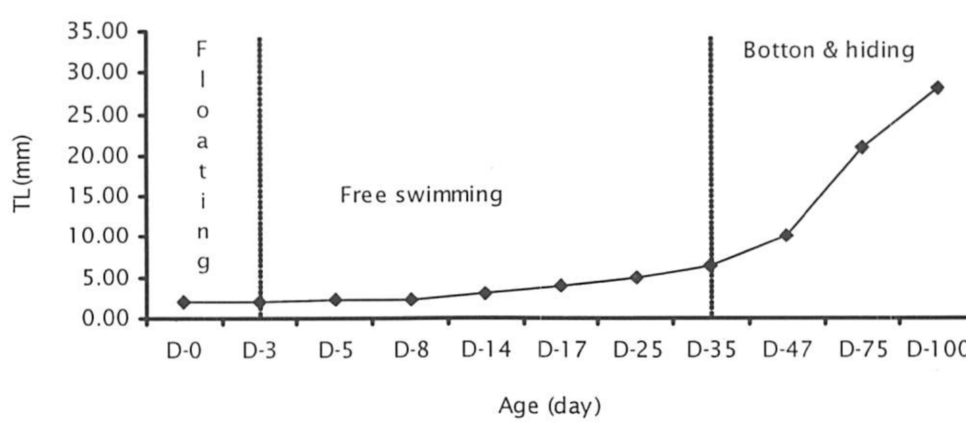


Figure 3. Growth pattern and swimming behavior of napoleon wrasse, *Cheilinus undulatus* larvae

swimming, they were just drifting and bending the body to control the swimming direction. This swimming pattern of juvenile might be one of the strategies to save energy.

Number of juveniles produced was 118 (0.37% survival rate from hatch-out). Total length of juveniles at D-100 were 30—32 mm. From this period, juveniles were fed with small shrimp (*Mesopodopsis* sp.) then followed by chopped fish.

Digestive system development from larvae to juvenile was clearly distinguished (Figure 4). Newly-hatched napoleon larvae just like other marine finfish larvae where the digestive tract was not well developed yet.

Mouth and anus were opened on D-3, lower jaw was able to move up and down to open and close the mouth and digestive tract was in function. This stage is known as initial feeding period. Digestive tract was still very simple and just a tube-like without any clear separation. Digestive system of larvae on D-15 was a straight pattern but was clearly separated between anterior and posterior part. Probably at this time the enzyme gland was developed. Further development of digestion system was observed on larvae D-25. It was just started coiled. Fully coiled digestive system was observed on D-35, stomach, intestine and rectum part were clearly distinguished. Probably at the same time larvae were started metamorphose to juvenile.

CONCLUSION

Chicken egg yolk was suitable for initial feeding of napoleon wrasse larvae prior to be able to ingest rotifer, 14 days after hatch. Metamorphose from larvae to juvenile occurred at the same time with swimming pattern changed, tendency to hide, and fully coiled development of digestive system. Metamorphose to juvenile started 35—40 days after hatching when the total length reaching 9—11 mm.

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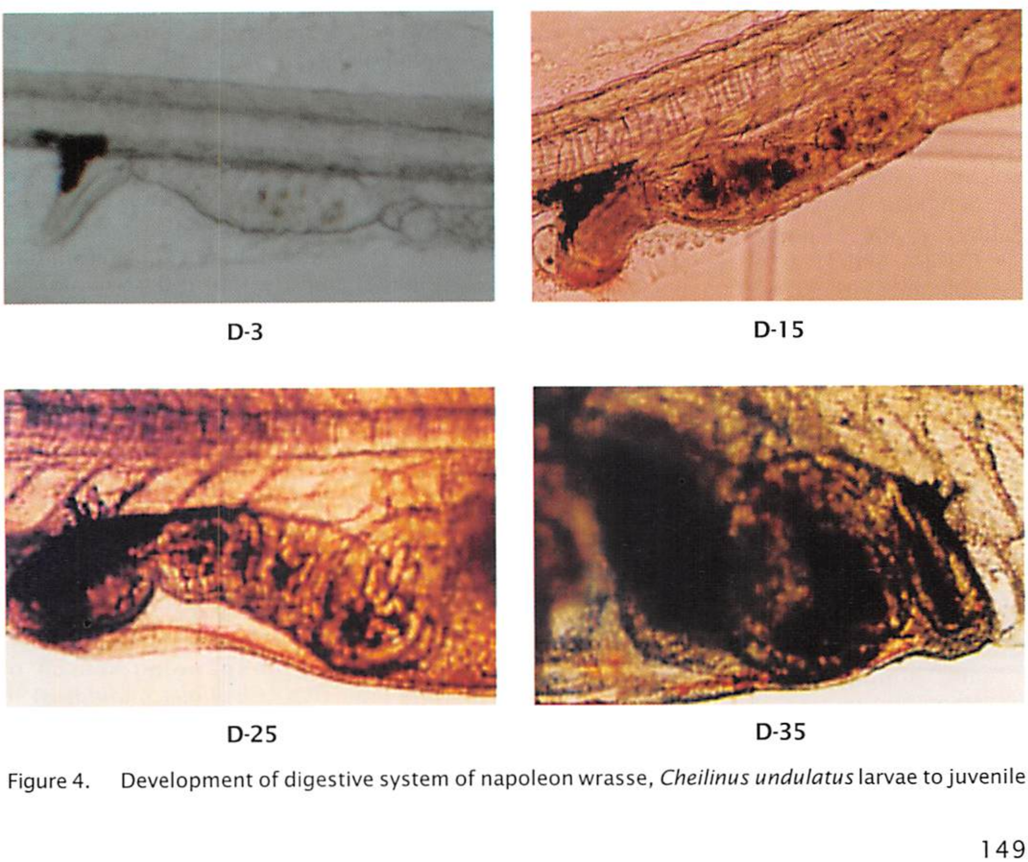

Figure 4. Development of digestive system of napoleon wrasse, *Cheilinus undulatus* larvae to juvenile

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