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Photoperiod Induced Larval Growth of Anemonefish *Amphiprion percula*

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Abstract: Rearing of anemonefish is relatively routine nowadays. To reduce time and cost of rearing anemonefishes, experiments were undertaken to improve the methods for rearing *Amphiprion percula.* This experiment was conducted to determine the effect of the length of photoperiod on larval duration, growth to metamorphosis and early juvenile phase. Growth of larvae was significantly faster under a photoperiod of 16 hours light/8 hours dark, compared to the photoperiod of 24 hours light/0 hours dark and 12 hours light/12 hours dark.

Key words: Photoperiod · Larval growth · Anemonefish · *Amphiprion percula*

INTRODUCTION

The culture of marine tropical fish conserves natural reef resources by offering alternatives to wild capture and develops a new source of organisms for the aquarium trade. The aquarium hobby draws 10-20 million enthusiasts who keep more than 90 million tropical fish [1]. Increasing pressures on natural populations of coral reef animals due to their expanding popularity in the aquarium trade has stimulated interest in developing culture techniques for marine tropicals. Controlled spawning through temperature and photoperiod manipulations to simulate seasonal changes and bring about gonadal maturation has been successful with several temperate species [2, 3]. The tropical marine anemone fishes (Pomacentridae) are important in ornamental fish culture [4] and are a popular subject of research [5]. Only two studies have experimented with ways of enhancing the efficiency of larval rearing of anemone fishes. Studied on the effect of high nitrate-N on the growth and survival of juvenile and larval anemonefish.

Amphiprion ocellaris [6]. Manipulation of light intensity has been shown to influence growth, survival, or feeding success of many larval teleosts [7-9]. This study examines the growth rates of larvae and juveniles under different light regimes. This variable is important as anemonefish larvae are visual feeders [10, 11]. In two separate experiments, haddock (*Melanogrammus aeglefinus*) larvae were raised under different photoperiods (24L:0D or 15L:9D), or different combinations of tank colour (black or white) and light intensity [12]. The Physical and biological factors that act on developing larvae are essential to create protocols that maximize survival and growth encountered under culture conditions. Light has been shown to be an important ecological factor for fish, influencing development from egg to sexually mature adult [13-16].

In nature, larval foraging may be limited by available daylight hours. Aquaculturists can mitigate this limitation by providing artificially extended daylengths [17]. Manipulation of photoperiod can have a major impact on larval growth and survival [18-21, 12]. Estimated the optimum growth conditions for anemonefish larvae to be a 16-hour daily light period, recommends a 24/h light regime. Full scale experiments in tanks were conducted to elucidate the effect of photoperiod regime, dietary fat level and stocking density on growth in spotted wolffish [22, 23, 4].

Most of these experiments suggest a growth enhancing effect of extended photoperiod or continuous light. This is in contrast to the earlier findings on photoperiod manipulated spotted wolffish [24]. The experiments were conducted to determine the effect of the length of photoperiod on larval duration, growth to metamorphosis and early juvenile phase. Growth of larvae

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was significantly faster and the duration of the larval phase was significantly shorter, under a photoperiod of 16/ h light/8/ h dark, compared to the photoperiods of 12/h light 12/h dark and 24/h light 0/h dark [25]. The objective of the present study is to investigate the effect of different photoperiods on the duration of larval growth of anemone fish *A. percula.*

MATERIALS AND METHODS

Collection and Acclimatization of Fish: Breeding pairs of *Amphiprion percula* were collected from the Ornamental Fish Trade, Chennai and acclimated to the laboratory condition for three months at the Institute of Center for Advanced Study in Marine Biology Annamalai University, Parangipettai. They were kept in 750-L tanks with UV treated estuarine water (26-30 ppt, 28-30°C temperature and 7.6-8.1 pH). Egg laying material comprised of bark rock, bivalve shells and ceramic plates in the tank as lining for home surrounding and sea anemone *Stichodactyla gigantean* were also kept to mimic the natural environment of this fish. Fishes were fed with brine shrimp, squid, fishes, clams and oyster. Spawning period is approximately every 10 days and produced 400 - 800 eggs per clutch. Embryos are hatched in nine days. The water in the hatching tank was gently aerated but not filtered, since the fish larvae are sensitive to currents.

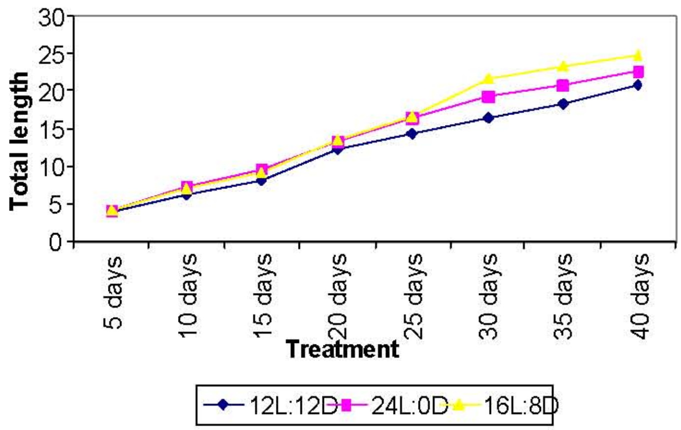
Effect of Photoperiod on Larval Growth: Larvae and juveniles were transferred to 21-L ash tank (tank height - 30 cm, width - 38 cm, length – 45 cm) connected to an 800-L recirculation system with biological filtration. The chemical and physical conditions were maintained as in the breading tank. The parental larvae are divided into three batches for the experiments. The fish larvae were measured to the nearest 0.3 cm total length (TL) and preserved in 5% formalin. Three batches of larvae from the same breeding pair were reared under three different light sources. The light regimes were: (I) 12/h light 12/h dark (12L: 12D); (II) 16/h light /8h dark (16L: 8D); and (III) 24/h light 0/h dark (24L: 0D). The light was provided by one 40 watts fluorescent light bulb. Feeding was made available for 24/h day in all three treatments. First day feed was mixed with algae and rotifers in the ratio 3:1, rotifer density is 6 - 8 per/ml and maintained up to 2nd day. The third day feed was enriched and the rotifers density was 8-10 per/ml and maintained up to 4th day. The 5th day feed rotifer density is 8-10 per/ml and maintained up to 9th day. 10th day feed was mixed with rotifer and *Artemia* in the ratio of 3:1 and maintained up to 12th day. The 13th day feed was mixed with rotifer and *Artemia* in the ratio of 1:1 and maintained up to 14th day. The 15th day feed was mixed with rotifer and *Artemia* in the ratio of 1:3. The 16th day feed was only *Artemia* with the density 6-8 per/ml up to end of experiment (40th day).

RESULTS AND DISCUSSION

The larvae growth among the batches of larvae from the same parents but exposed to different photoperiods showed low variation. Larvae reared under 16L: 8D had the highest growth followed by the 24L: 0D treatment, with the 12L: 12D larvae displaying the lowest growth (Fig. 1). The survival rate was approximately 80% for all three batches.

Continuous light reaction had effect on larval growth and survival, when compared to a different photoperiodic reaction. *A. percula* larvae fastest growth under 16L:8D photoperiodic reaction. The 24L:0D photoperiodic reaction showed slightly slow growth than the 16L: 8D photoperiodic reaction. However 12L:12D photoperiodic reaction showed slow growth than the 16L:8D and 24L:0D photoperiodic reactions. The increase in total length in 16L:8D and 24L:0D photoperiodic reactions due to longer period for feeding than 12L:12D photoperiodic reaction. The effect of photoperiod appears to be species specific and its influence may extend to other aspects of larval development beyond growth and survival. Many commercially important species have been shown to have improved growth under continuous light, e.g., sole, *Solea solea* [18], Atlantic salmon, *Salmo salar* [26], barramundi, *Lates calcarifer* [27], greenback flounder, *Rhombosolea tapirina* [28] and winter flounder, *Pleuronectes americanus* [21]. For other species, total darkness may improve larval growth, e.g., African catfish, *Clarias gariepinus* [9]. A study of the rockfish *Sebastes diploproa* was reported to have an optimum growth rate with a 16-hour light period [7].

Previous photoperiod history of fish may have an important influence on the growth response to light regimes [29, 30] and in some salmonids [31, 32]a decrease in photoperiod may have a growth depressing effect. Also in Atlantic halibut kept on continuous light for approximately 2 months [17]. As with photoperiod, the effect of light intensity is species specific.



Values are found significance at 5% level between total length and the treatment duration.
$R2= 0.991939$ (R2 - Regression Square)

Fig. 1: Total length comparison against age for three batches of *Amphiprion percula* larvae reared under three different light regimes

Some species benefit from higher light intensities, e.g., sand whiting, *Siliago ciliata* [33], whereas some perform better under low light intensity, e.g., African catfish [9]. The conclusion of this study is *A. percula* larvae growth enhancing and high growth was observed in 16L: 8D photoperiodic reaction. However, optimum lighting condition and feed is present for all the 16 hours, using this photoperiod for larval rearing should improve growth within a short period.

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