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Influence of light wavelength and intensity on the survival and somatic growth of the early larval stage of sutchi catfish *Pangasianodon hypophthalmus*

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Abstract: The present study examined the influence of various light wavelengths and intensities on the survival and somatic growth of sutchi catfish *Pangasianodon hypophthalmus* larvae during their most critical period. Two-day-old larvae (initial total length, TL_i = 3 mm; initial body weight, BW_i = 0.002 g) at a stocking density of 10 larvae per litre were reared in triplicate under five light wavelengths (white as a control, blue, green, yellow and red wavelengths) of four light intensities (1.40×10^{-3}, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 μmol/m²/s). The larvae showed a tendency towards higher survival rates (*SR*) and production index (*PI*) under longer light wavelengths (yellow and red wavelengths) and at a light intensity of 1.40×10^{-3} μmol/m²/s. Furthermore, the larvae reared under red wavelength showed a tendency towards higher specific growth rates for both length and body weight (SGR_L and SGR_{BW}) than those reared under other light wavelengths. These findings provide fundamental information regarding the optimum light conditions for larval rearing of sutchi catfish, as well as providing insight about the adaptation of the larvae towards the environmental light in the wild.

Keywords: Growth, light intensity, light wavelength, *Pangasianodon hypophthalmus*

Introduction

The sutchi catfish *Pangasianodon hypophthalmus* is a species of pangasiid catfish (family Pangasiidae) native to the lower Mekong Basin, which includes parts of Thailand, Cambodia, Vietnam and the Lao People's Democratic Republic, and the Chao Phraya River in Thailand (Roberts and Vidthayanon, 1991; Rainboth, 1996; Poulsen *et al*., 2004; Rohul Amin *et al*., 2005). In addition to its importance to fisheries and aquaculture sectors in these areas, especially the Mekong Delta of Vietnam (Baird *et al*., 2004; Phan *et al*., 2009; Bui *et al*., 2010; De Silva and Phuong, 2011; Nguyen *et al*., 2015), sutchi catfish has been introduced for aquaculture in Malaysia, Indonesia, the Philippines, Taiwan, Bangladesh and India (Roberts and Vidthayanon, 1991; Van Zalinge *et al*., 2002; Poulsen *et al*., 2004; Ali *et al*., 2005; Rohul Amin *et al*., 2005). The preference for sutchi catfish over other pangasiids for aquaculture is due to its relatively high growth rates, market demand, market price and adaptability to local conditions, especially in captivity (Subagja *et al*., 1999; Hassan *et al*., 2011). Additionally, sutchi catfish able to tolerate oxygen deficiency (hypoxia) (Lefevre *et al*., 2011a, 2011b) and is highly fecund, but with small egg size in comparison to other pangasiids (e.g. *Pangasius bocourti* and *Pangasius djambal*) producing larger egg in size (Legendre *et al*., 1998; Islam, 2005; Morioka *et al*., 2010; Vidthayanon and Hogan, 2013).

The artificial propagation of sutchi catfish in hatcheries has been successful, but larval rearing of the species still remains difficult because of low survival rates associated with cannibalism (Subagja *et al*., 1999; Ali *et al*., 2005; Slembrouck *et al*., 2009). The first seven days of life are the most critical larval period, with low survival rates attributed to cannibalism observed through day seven (Subagja *et al*., 1999). During this early larval stage, strong cannibalistic behaviour was shown, with the larvae reported to entwine with each other, forming a long string or star-shaped figures and eventually causing high individual mortality and in part responsible for deadly bacterial outbreaks within the rearing system (Subagja *et al*., 1999; Baras *et al*., 2010; Mukai *et al*., 2013a). According to Baras *et al*. (2010), this cannibalistic behaviour is governed intimately by the presence of the oral spines, which prevent the larvae

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from closing fully their mouth and thus cause them to grasp each other.

Light wavelength, light intensity and photoperiod are factors that affect the fish survival, somatic growth and behaviour (Hecht and Pienaar, 1993; Appelbaum and McGeer, 1998; Boeuf and Le Bail, 1999; Tamazouzt *et al*., 2000; Biswas *et al*., 2005; Karakatsouli *et al*., 2007, 2008, 2010; Luchiari and Freire, 2009). For example, previous studies have reported that sutchi catfish larvae showed lower cannibalism and higher growth rates under dark or dim light conditions compared to bright light conditions (Mukai, 2011a, 2011b; Mukai *et al*., 2013a). However, these studies only considered the rearing of larvae under white fluorescent light. In the fish's natural water habitats, light conditions differ from those of terrestrial in terms of both light wavelength and intensity (Bone *et al*., 1995; Evans, 2004). Therefore, the present study examined the influence of various light wavelengths and intensities on the survival and somatic growth of sutchi catfish larvae during their most critical period, which is the first seven days after hatching. The findings may provide fundamental information regarding the optimum light conditions for larval rearing of sutchi catfish, as well as providing insight about the adaptation of the larvae towards environmental light in the wild.

Materials and methods

Larval fish and keeping conditions

Fertilized sutchi catfish eggs (from the same spawn of a single female broodfish and the sperms of a single male fish) were obtained from a private fish farm in Kuala Berang, Terengganu, Malaysia. The eggs were incubated in 320 L (120 cm length × 60 cm width × 45 cm depth) glass aquaria and hatched about 27 h after artificial fertilization at 27°C. The larvae were reared in the glass aquaria, with a water temperature between 26°C and 27.5°C. Freshly hatched brine shrimp *Artemia* sp. nauplii were given to the larvae starting from the second day after hatching.

Larval rearing experiment

Larvae were reared with continuous aeration under various light wavelengths and intensities in a dark room at a laboratory of Kulliyyah of Science, International Islamic University Malaysia. Two-day-old larvae (initial total length, TL_i = 3 mm; initial body weight, BW_i = 0.002 g) were randomly transferred into 60 acrylic aquaria (5 light wavelengths × 4 light intensities × 3 replications for each combination of light condition) containing 10 L of de-chlorinated freshwater at a stocking density of 10 larvae per litre (L). The experimental acrylic aquaria were illuminated with various light wavelengths from light emitting diodes (LEDs; 10 W high-power LED, Wayjun Technology Co., Ltd., Shenzhen). The colours of light (wavelengths) used for the rearing experiment were white (broad spectrum, with double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (630 nm). The light intensities were adjusted to 1.40×10^{-3}, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 μmol/m²/s respectively using a dimmer (Songyuan Electronics Technology Co., Guangdong) and neutral-density filters (ND×8, Hoya Corporation, Tokyo). Light wavelengths and intensities were both measured at the water surface using a modular USB spectrophotometer (USB4000-UV-VIS, Ocean Optics, Inc., Florida). The light conditions were switched on at 0600 h and off at 1800 h every day during the experimental period.

Throughout the experiment, the larvae in each experimental treatment were fed in excess with 5,000 individuals of freshly hatched *Artemia* sp. nauplii twice daily, with some *Artemia* sp. nauplii remaining at the next feeding time (Mukai, 2011a, 2011b). In order to maintain water quality, about 10% of the water in each aquarium was changed once per day to remove fish waste. All aquaria were exposed to fluorescent light from the laboratory for 10 min during these water changes. Dead larvae were removed daily at the time of water renewal and counted. Water temperature throughout the experiment was ranged between 26°C and 27.5°C.

At the end of the experiment (when the larvae were seven-day-old), all the remaining larvae were slightly anesthetized (25 ppm) with TRANSMORE® (Nika Trading Co., Selangor Darul Ehsan) and individually counted, measured and weighed. A micrometer (MBM11100, Nikon Corporation, Tokyo, Japan) was used to measure the total length of the larvae under a stereo microscope. Survival rates (*SR*), specific growth rates for length (SGR_L) (Lopez and Castello-Orvay, 1995), specific growth rates for body weight (SGR_{BW}) and production index (*PI*) (Rosas *et al*., 1999; Zacharia and Kakati, 2002) were calculated as follows:

$$SR = (n_f/n_i) \times 100\%$$

where *SR* is the survival rates of the larvae (%), n_i is the initial number of the larvae and n_f is the final number of the larvae.

$SGR_L = [(\ln TL_f - \ln TL_i)/d] \times 100\%$

where SGR_L is the specific growth rates for length (%/d), TL_i is the initial total length of the larvae (mm), TL_f is the final total length of the larvae (mm) and d is the number of days the experiment lasted.

$SGR_{BW} = [(\ln BW_f - \ln BW_i)/d] \times 100\%$

where SGR_{BW} is the specific growth rates for body weight (%/d), BW_i is the initial body weight of the larvae (g), BW_f is the final weight of the larvae (g) and d is the number of days the experiment lasted.

$PI = [(BW_f - BW_i)/d] \times SR$

where PI is the production index, BW_i is the initial body weight (g) of the larvae, BW_f is the final body weight (g) of the larvae, d is the number of days the experiment lasted and SR is the survival rates of the larvae (%).

Statistical analysis

Statistical analysis was done on arcsine transformed data, analysed by two-way analysis of variance (ANOVA) using IBM SPSS Statistics ver. 20 (IBM Corp., New York) with light wavelength and light intensity as the factors. Tukey's multiple comparison test was performed when the P values were significant ($P < 0.05$).

Results

Survival rates

The survival rates of sutchi catfish larvae reared under various light wavelengths and intensities are shown in Figure 1. Although there were no statistically significant differences among larvae reared under various light conditions, the larvae showed a tendency towards higher SR under longer light wavelengths (yellow and red wavelengths) and at a light intensity of 1.40×10^{-3} µmol/m²/s (Tab. 1).

Specific growth rates

The specific growth rates (SGR) for length and body weight of sutchi catfish larvae are shown in Figures 2 and 3, respectively. Although there were no significant differences among larvae reared under various light conditions, the results of two-way ANOVA suggested a significant interaction between light wavelength and light intensity on SGR_L. This indicated that the effect of light wavelength on SGR_L was depended on the light intensity. Additionally, a tendency of higher SGR_L was observed in larvae reared under red wavelength than those of the other light wavelengths.

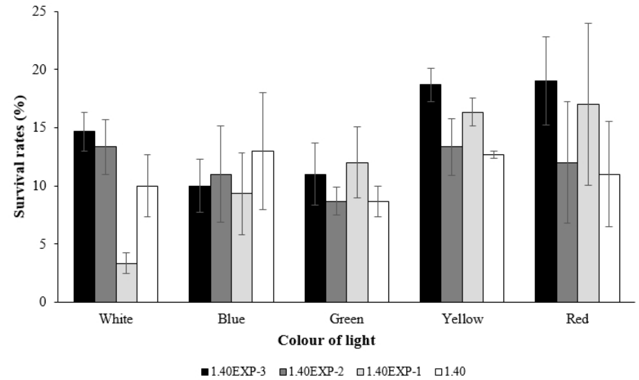

Fig. 1: Survival rates (%) of sutchi catfish larvae under various light wavelengths and intensities. Wavelengths were white (double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (632 nm), with intensities of 1.40×10^{-3}, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 µmol/m²/s. Data are presented as mean ± SE.

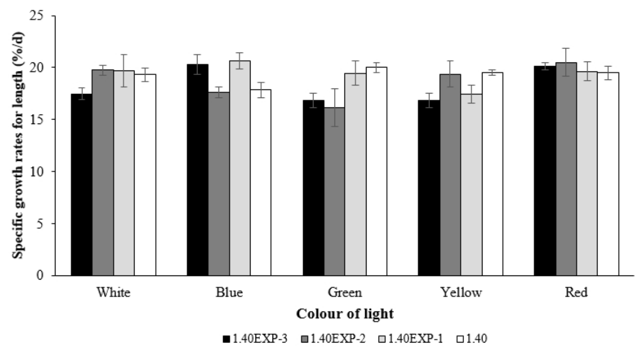

Fig. 2: Specific growth rates for length (SGR_L) of sutchi catfish larvae under various light wavelengths and intensities. Wavelengths were white (double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (632 nm), with intensities of 1.40×10^{-3}, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 µmol/m²/s. Data are presented as mean ± SE.

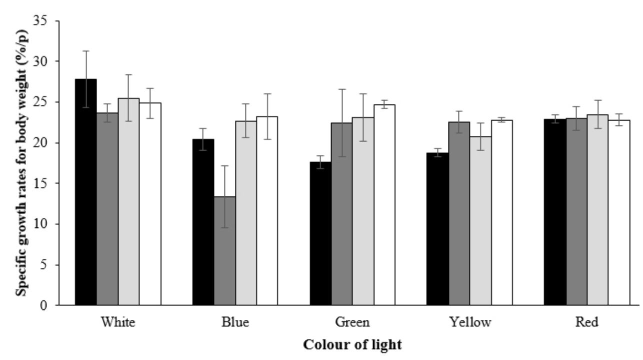

Fig. 3: Specific growth rates for body weight (SGR_{BW}) of sutchi catfish larvae under various light wavelengths and intensities. Wavelengths were white (double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (632 nm), with intensities of $.40\times10^{-3}$, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 µmol/m²/s. Data are presented as mean ± SE.

Tab. 1: Results of two-way ANOVA and Tukey's multiple comparison test regarding survival rates (*SR*), specific growth rates for length (SGR_L), specific growth rates for body weight (SGR_{BW}) and production index (*PI*) of sutchi catfish larvae under various light wavelengths; white (double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (632 nm), with intensities of 1.40×10^{-3}, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 µmol/m²/s.

Variable	Significance (*P* value)			Tukey's multiple comparison test								
				Light wavelength (represented as color)					Light intensity (µmol/m²/s)			
	LW	LI	LW×LI	White	Blue	Green	Yellow	Red	1.40×10^{-3}	1.40×10^{-2}	1.40×10^{-1}	1.40
Survival rates (%)	NS	NS	NS	10.33	10.83	10.08	15.25	14.75	14.67	11.67	11.60	11.07
SGR_L (%/d)	NS	NS	*	19.07	19.10	18.12	18.30	19.94	18.32	18.68	19.38	19.24
SGR_{BW} (%/d)	*	NS	NS	25.44^a	19.92^b	21.96^{ab}	21.20^{ab}	23.02^{ab}	21.51	20.97	23.09	23.66
Production index	NS	NS	*	0.0127	0.0087	0.0092	0.0128	0.0140	0.0135	0.0104	0.0107	0.0113

LW = Light wavelength; LI = Light intensity; LW×LI = interaction of light wavelength and light intensity. Mean values in the same row with no superscript in common differ significantly ($P < 0.05$). If the effects were significant, ANOVA was followed by Tukey's multiple comparison test. * $P < 0.05$; NS, not significant.

Two-way ANOVA showed significant differences among the groups of light wavelengths in terms of SGR_{BW}. The results of the Tukey's multiple comparison test showed that larvae reared under white wavelength attained significantly higher SGR_{BW} than those reared under blue wavelength (Tab. 1). In spite of this, the larvae showed a tendency towards better SGR_{BW} under red wavelength and all examined light intensities.

Production index

The production index of sutchi catfish larvae reared under various light wavelengths and intensities are shown in Figure 4. Light wavelengths and intensities did not have significant effects on the *PI* of the larvae; however, a tendency towards a higher *PI* was observed in larvae reared under red wavelengths with light intensity of 1.40×10^{-3} µmol/m²/s (Tab. 1). Additionally, two-way ANOVA showed a significant interaction between light wavelength and light intensity on *PI*, which implied that the effect of light wavelength on *PI* was depended on the light intensity.

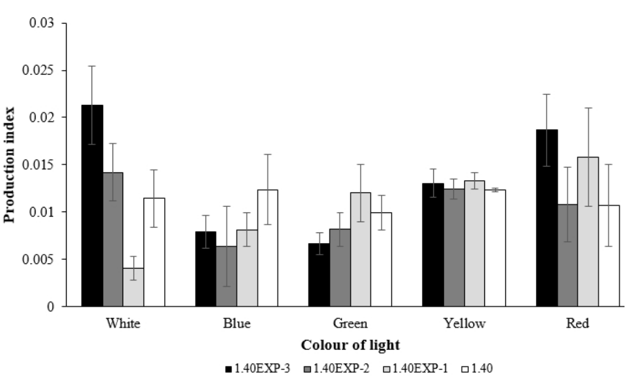

Fig. 4: Production index (*PI*) of sutchi catfish larvae under various light wavelengths and intensities. Wavelengths were white (double peaks at 446 and 566 nm), blue (454 nm), green (520 nm), yellow (590 nm) and red (632 nm), with intensities of $.40\times10^{-3}$, 1.40×10^{-2}, 1.40×10^{-1} and 1.40 µmol/m²/s. Data are presented as mean ± SE.

Discussion

The sutchi catfish is an important species for freshwater aquaculture; however, the rearing of its larvae is still constrained by low survival rates that are attributed to cannibalism. The results of the present study suggest that light wavelength and intensity influence the survival and somatic growth of sutchi catfish larvae. The larvae showed a tendency towards higher *SR* and *PI* under longer light wavelengths (yellow and red wavelengths) at a light intensity of 1.40×10^{-3} µmol/m²/s. Furthermore, the larvae reared under red wavelength showed a tendency towards higher specific growth rates for both length and body weight (SGR_L and SGR_{BW}) than those reared under other light wavelengths. These results agree with previous studies of freshwater fish species. For example, Giri *et al*. (2002) reported the highest survival rates when the larvae of freshwater catfish *Wallago attu* were reared under red light compared to those reared under other light conditions. Other fish species, such as rainbow trout *Oncorhynchus mykiss* (Karakatsouli *et al*., 2008) and the scaled and mirror common carp *Cyprinus carpio* (Karakatsouli *et al*., 2010), showed better SGR, weight gain and feed efficiency when reared under red light compared to white and blue light. Moreover, yellow perch *Perca flavescens* gained more in body length under red light compared to blue and white light (Head and Malison, 2000). In terms of light intensity, Mukai (2011a) reported that sutchi catfish larvae reared under 0.1 lux (equivalent to about 1.40×10^{-3} µmol/m²/s) of white fluorescent light showed higher survival rates and steadier growth compared to those reared under other light conditions (1, 10 and 100 lux).

Sutchi catfish larvae showed a tendency towards higher SGR_{BW} but lower *SR* under light intensity of 1.40 µmol/m²/s for all examined light wavelengths. These results imply that light intensity of 1.40 µmol/m²/s (the highest light intensity in the present study) induced higher cannibalistic behaviour in the larvae, eventually causing mortality among many individuals in the experimental aquaria. Mukai (2011a) reported observing a higher incidence of cannibalistic behaviour among *P. hypophthalmus* larvae reared under brighter light conditions (10 and 100 lux of white fluorescent light) than among those reared under dimmer light conditions (0.1 and 1 lux of white fluorescent light). The larvae reared under brighter light conditions showed frequent resting behaviour on the bottom of the aquaria and the other, more active larvae bit these resting individuals (Mukai, 2011a). Bodily fluids, such as blood, released from the skin surfaces of these injured larvae might have stimulated the cannibalism of the other larvae, thereby resulting in lower *SR* with higher SGR_{BW} of the larvae (Mukai, 2011a; Mukai *et al*., 2013a). In addition, African catfish *Clarias gariepinus* larvae were reported to show similar cannibalistic behaviour under such light conditions (Mukai and Lim, 2011; Mukai *et al*., 2013b).

The findings of the present study correlate with the light conditions in the natural environment of sutchi catfish larvae in the Mekong River. Wild sutchi catfish concentrate in areas such as rapids and sand banks interspersed with deep, rocky channels and pools during spawning season, depositing their eggs on the root systems of rheophilic tree species *Gimenila asiatica* (Van Zalinge *et al*., 2002). After hatching, the larvae drift downstream following the current to inundated floodplains for feeding (Poulsen *et al*., 2004). The feeding grounds consist of inundated forests and other vegetation (So *et al*., 2006) as well as high densities of phytoplankton and zooplankton (Van Zalinge *et al*., 2002). Since there is a high level of coloured or chromophoric dissolved organic matter (CDOM), suspended particles and chlorophyll (Lythgoe, 1984; Lythgoe and Partridge, 1989; Bowmaker, 1990) present in these waters, the freshwater environment in both the spawning and feeding grounds of the larvae are characterized by high turbidity and low light intensity. Moreover, the presence of CDOM, suspended particles and chlorophyll in these waters transmits light shifted towards longer light wavelengths such as those within the yellow and red colour spectrums.

Conclusion

The present study showed that higher survival and somatic growth performances of sutchi catfish larvae were observed when the larvae were reared under longer light wavelengths (yellow and red wavelengths) with a light intensity of 1.40×10^{-3} μmol/m²/s. In addition, the findings of the present study also correlate with the light conditions in the larvae's natural environment which are characterized as displacing towards longer light wavelengths and low in light intensity.

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