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EGG HATCHING RATES OF BROWN-MARBLED GROUPER, *EPINEPHELUS FUSCOGUTTATUS* UNDER DIFFERENT LIGHT WAVELENGTHS AND INTENSITIES

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ABSTRACT This study was conducted to examine the egg hatching rates of brown-marbled grouper, *Epinephelus fuscoguttatus* under different light conditions in terms of light wavelengths and intensities. Results of two-way ANOVA showed significant difference for light wavelengths (p<0.05), while light intensities showed no significant difference. There was no interaction between light wavelengths and intensities. Post-hoc Tukey test showed that hatching rates under green light wavelength were significantly higher than white and yellow light wavelengths (p<0.05). When white light 2.7 µmoles/m²/s was set as the control, the hatching ratio of the brown-marbled grouper under green light 2.7 µmoles/m²/s was almost three times higher than the control. The lowest ratio of hatching rates was observed under the white light 13.3 µmoles/m²/s with 70 percent lower ratio of hatching rates than the control light. Eggs hatching under other light conditions mostly showed higher ratios than the control light.

ABSTRAK Kajian ini dijalankan untuk mengkaji kadar penetasan telur kerapu harimau, *Epinephelus fuscoguttatus* di bawah keadaan cahaya yang berbeza dari segi jarak gelombang dan keamatan. Keputusan ANOVA dua hala menunjukkan panjang gelombang cahaya menunjukkan perbezaan yang signifikan (p<0.05), manakala tiada perbezaan yang signifikan bagi keamatan cahaya. Panjang gelombang cahaya dan keamatan cahaya tidak berkaitan antara satu sama lain dalam mempengaruhi penetasan telur ikan kerapu harimau. Keputusan ujian *post-hoc* Tukey telah menunjukkan kadar penetasan di bawah panjang gelombang cahaya hijau adalah jauh lebih signifikan daripada cahaya putih dan cahaya panjang gelombang kuning (p<0.05). Apabila cahaya putih 2.7 µmol/m²/s ditetapkan sebagai kawalan, nisbah penetasan ikan kerapu harimau di bawah cahaya hijau 2.7 µmol/m²/s adalah hampir tiga kali ganda lebih tinggi daripada kawalan. Nisbah kadar penetasan paling rendah diperhatikan di bawah cahaya putih 13.3 µmol/m²/s, dengan nisbah kadar penetasan 70 peratus lebih rendah daripada kawalan. Telur menetas di bawah keadaan cahaya yang lain kebanyakannya menunjukkan nisbah yang lebih tinggi daripada kawalan.

(**Keywords:** Brown-marbled grouper, *Epinephelus fuscoguttatus*, light wavelengths, light intensities, egg hatching rates)

INTRODUCTION

Egg hatching rates vary depending on several factors such as temperature [1], salinity [2], dissolved oxygen [3], and light environment [4]. Light conditions affect the growth and development of aquatic living things [5]. Some studies have been done on various light conditions for incubation of teleost eggs. For example, studies of light intensities on haddock (*Melanogrammus aeglefinus*) eggs show that the eggs are not affected by light intensity [4]. Haddock eggs stay in the upper 50 m in the sea and the eggs are exposed to highly variable light environment. Therefore, haddock eggs are adaptive to wider range of light intensity [4]. Incubations of rainbow trout (*Salmo irideus* Gibbons) and Atlantic halibut (*Hippoglossus hippoglossus*) eggs are optimum under dim light conditions [6 and 7]. Incubation of Walleye Pollock (*Theragra chalcogamma*) eggs is optimum under dark condition [8]. Studies of egg incubation under different light wavelengths show that hatching rates of Senegal sole (*Solea senegalensis*) and zebrafish (*Danio rerio*) are the highest when the eggs are incubated under blue light wavelength [9 and 10]. Studies of egg incubation show that light conditions are species-specific and related to their natural habitats [4, 6, 7, 8, 9 and 10].

Brown-marbled grouper, *Epinephelus fuscoguttatus* is widely distributed in the Indo-Pacific Ocean including the Red Sea [11]. It is an important species for aquaculture in Southeast Asia [12 and 13]. However, only few studies have been done on its early developmental stage [14]. The optimum light condition for its egg incubation is still unknown. Therefore, this study was conducted to examine the optimum light wavelength and intensity for brown-marbled grouper egg incubation.

MATERIALS AND METHODS

Brown-marbled grouper eggs were obtained from the brood stock in the hatchery of Institute of Oceanography and Maritime Studies (INOCEM), International Islamic University Malaysia (IIUM). The eggs were incubated in 40 L aquaria [60 cm (L) x 29 cm (W) x 32 cm (H)] with triplicate in dark room, using light emitting diode (LED) (10W high power LED, Wayjun Technology Co., Ltd, Shenzhen, China) lamps with different light wavelengths: white (broad spectrum with double peaks 452, 557 nm) as control, blue (peak, 458 nm), green (528 nm), yellow (593 nm), and red (636 nm) (Figure 1). Light intensities for each wavelength were set at 0.53, 2.7, and 13.3 µmoles/m²/s. The spectral wavelengths and light intensities of LED lamps were measured at the water surface using a spectrometer (USB4000, Ocean Optics, Inc., Florida, USA). The water depth was 20 cm. The light conditions were set up as 0600 on and 1800 off. The water salinity and the temperature in the incubation aquaria were 31 ppt and 27.5-28.5 °C, respectively. *Nannochloropsis sp.* frozen paste (Nannochloropsis K2, New World Aqua, Busan, Korea) was put at the density of 1 million cells/ml in each aquarium. In general, brown-marbled grouper spawn at night [15]. The eggs hatched at 24 hours after spawning, and then the number of larvae was counted to examine the hatching rates. Statistical analysis was done using two-way ANOVA with two factors, light wavelengths and light intensities (SPSS version 15).

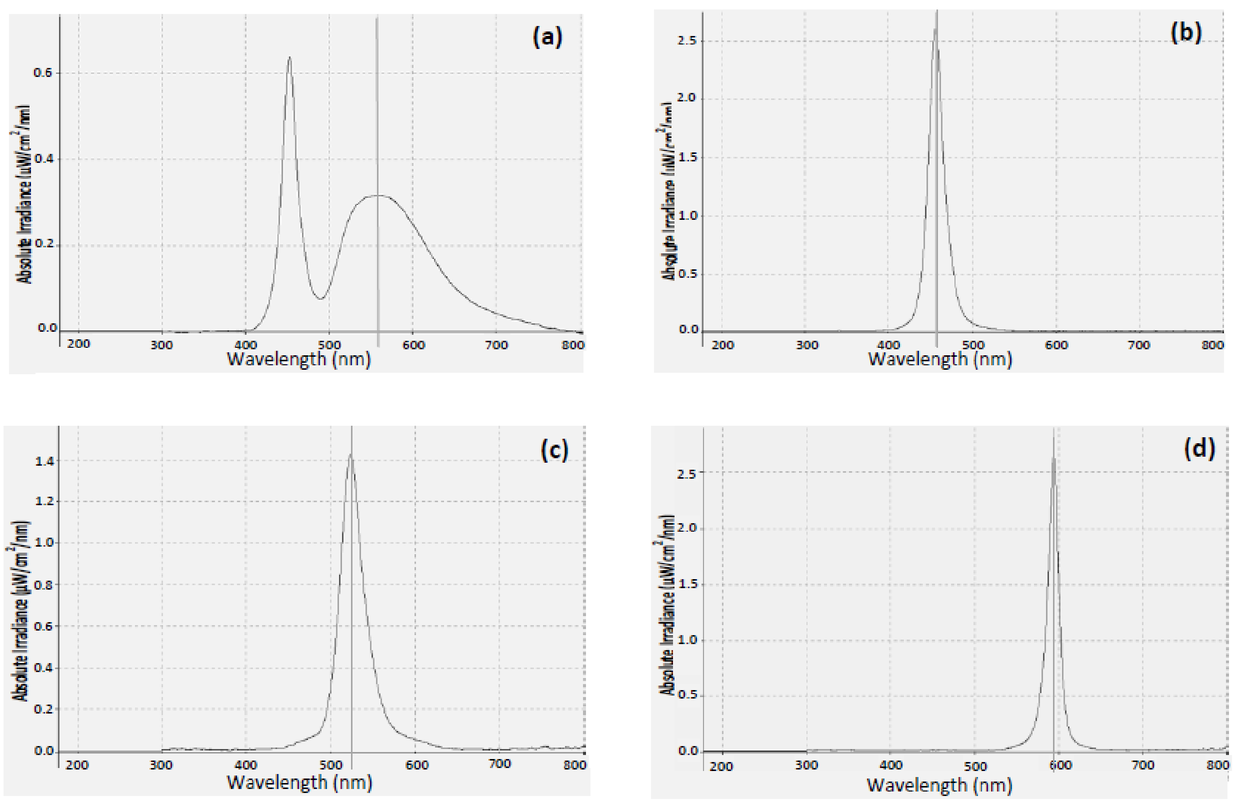

Figure 1. Graphs of light wavelengths (a) white, (b) blue, (c) green, (d) yellow and (e) red.

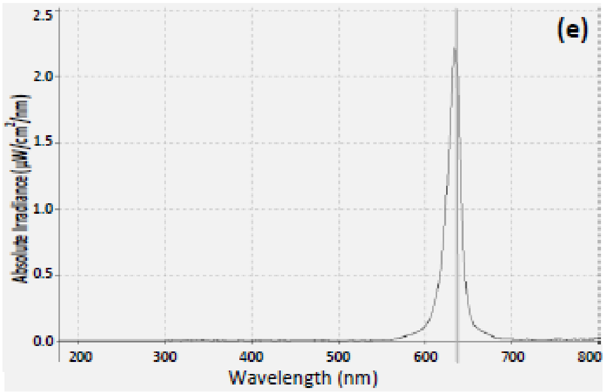

Figure 1 (Cont'd.). Graphs of light wavelengths (a) white, (b) blue, (c) green, (d) yellow and (e) red.

RESULTS

Results of two-way ANOVA showed significant difference for light wavelengths ($p<0.05$), while light intensities showed no significant difference (Table 1). There was no interaction between light wavelengths and intensities. Post-hoc Tukey test showed green light wavelength was significantly higher than white and yellow light wavelengths ($p<0.05$) (Table 2). Table 3 shows result of post-hoc Tukey test for light intensities. Although there was no significant difference, there was a tendency of higher hatching rates when the eggs were incubated under 2.7 µmoles/m²/s. Figure 2 shows the ratios of hatching rates under different light wavelengths and intensities relative to white light 2.7 µmoles/m²/s as control. The hatching ratios under green light 2.7 µmoles/m²/s showed almost three times higher than the control light. Blue light 13.3, green light 0.53 and 13.3, and red light 13.3 µmoles/m²/s showed double hatching ratios compared to the control.

Table 1. Two-way ANOVA of light intensities and wavelengths. Light intensities showed no significant difference while light wavelengths showed significant difference ($p<0.05$). There was no interaction between light intensities and wavelengths.

Source	***df***	**Mean Square**	***F***	***p***
Intensity	2	921	2.019	.150
Wavelength	4	2770	6.071	.001*
Intensity × Wavelength	8	473	1.036	.432

*Significant difference

Table 2. Post-hoc Tukey test for light wavelengths regarding hatching rates of brown-marbled grouper egg. Hatching rate under green light wavelength was significantly higher than hatching rates under white and yellow light wavelengths.

	White	Blue	Green	Yellow	Red
White	---	---	---	---	---
Blue	0.086	---	---	---	---
Green	0.001*	0.440	---	---	---
Yellow	0.961	0.299	0.007*	---	---
Red	0.097	1.000	0.405	0.329	---

*Significant difference ($p<0.05$)

Table 3. Post-hoc Tukey test for light intensities (µmoles/m²/s) regarding egg hatching rates of brown-marbled grouper. There was no significant difference between groups.

	0.53	2.7	13.3
0.53	---	---	---
2.7	0.147	---	---
13.3	0.878	0.331	---

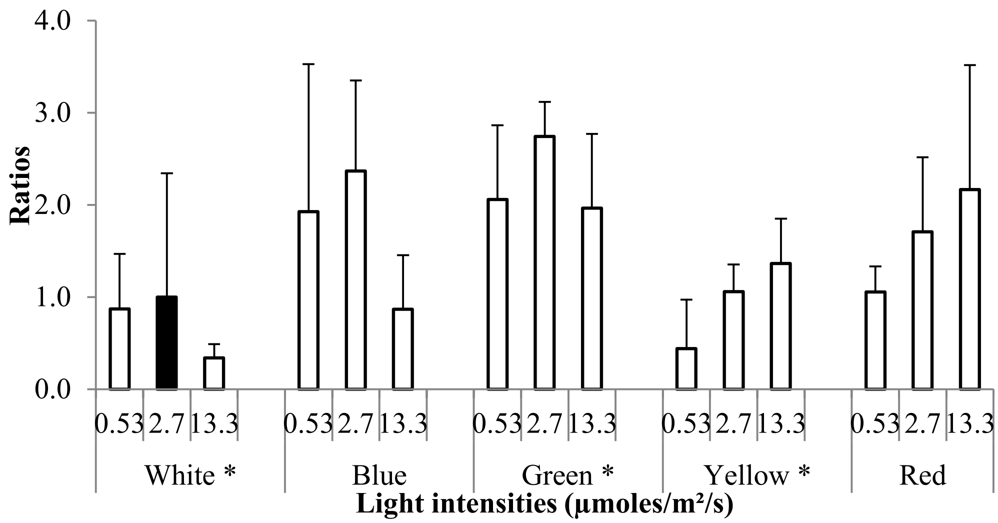

Figure 2. Results of egg hatching ratios of brown-marbled grouper, *Epinephelus fuscoguttatus*. Light conditions were blue, green, yellow, red and white with light intensities 0.53, 2.7, and 13.3 µmoles/m^2/s. Egg hatching ratios compared with white light as control (as white 2.7 µmoles/m^2/s is 1.0). *Post-hoc Tukey test showed the egg hatching ratios under green light were significantly higher than white and yellow light (p<0.05).

DISCUSSION

In this study, the egg hatching rates were influenced by light wavelengths and intensities. From our results (Figure 2), the highest ratio of hatching rates was shown when the eggs were put under the green light wavelength. White light wavelength, which represented the typical fluorescent lamp in most hatcheries, showed the lowest ratio of hatching rates. The lowest ratio of hatching rates was observed under the white light 13.3 µmoles/m²/s, with 70 percent lower ratio of hatching rates than the control light (white light 2.7 µmoles/m²/s). Yellow and red light 13.3 µmoles/m²/s showed slightly higher hatching ratios than the control light.

In terms of light wavelengths, the significantly higher hatching ratios under the green light wavelength might be due to the natural habitats of the brown-marbled grouper at their early life stage. Compared to white light wavelength, blue and green light wavelengths showed higher hatching ratios for brown-marbled grouper as these lights probably provided suitable lighting for the eggs to hatch.

Present results are similar to the incubation of zebrafish eggs which is optimum under shorter light wavelength (blue) [10]. The study suggests blue light wavelength relates to the development of pineal organ during embryo stage of zebrafish [10]. The pineal organ, which is one of the sensory organs located in the brain and associated with light sensitivity, might have developed at the embryo stage of brown-marbled grouper. Further investigation is needed to confirm the function of the pineal organ in the brown-marbled grouper embryo stage.

In terms of light intensities, there was a tendency of higher ratio of hatching rates under blue and green light 2.7 µmoles/m²/s, while under yellow and red light wavelengths the ratios of hatching rates tend to increase with light intensities. Although further studies are needed to investigate the effects of higher light intensities in yellow and red light wavelengths, results of post-hoc Tukey test showed that green light wavelength was significantly higher than white and yellow light wavelengths. Therefore, it can be suggested that green light 2.7 µmoles/m²/s is the optimum light condition for brown-marbled grouper egg incubation.

CONCLUSION

Egg hatching rates of brown-marbled grouper, *Epinephelus fuscoguttatus* were influenced by light conditions. In this study, green light 2.7 µmoles/m²/s was an optimum condition for the egg incubation of brown-marbled grouper. It can be estimated that the eggs stay in the water area where blue and green light are dominant.

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