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## Effects of photoperiods on growth and quality of white leg shrimp (*Litopenaeus vannamei*) in biofloc system

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### ABSTRACT

This study is aimed to determine appropriate photoperiod regime for growth of white leg shrimp (*Litopenaeus vannamei*) in super-intensive tank culture system applying biofloc technology. The experiment was set up indoor and randomly designed with different photoperiod treatments including (i) 24-hour in dark condition (0L:24D); (ii) 6-hour light: 18-hour dark (6L:18D); (iii) 12-hour light: 12-hour dark (12L:12D); (iv) 18-hour light: 6-hour dark (18L:6D) and (v) 24-hour light (24L:0D). In the photoperiod treatments, a 55w compact fluorescent (light intensity ~ 6000 lux) was installed above each culture tank. Shrimp with initial weight and body length at 0.84 g and 4.45 cm, respectively were stocked at the density of 150 inds/m<sup>3</sup> in biofloc system (C:N=15:1) in 300 L tanks at salinity of 15‰. The results showed that after 90 days of culture, water quality was in suitable range for shrimp growth. Mean body weight of shrimp was from 18.28 to 22.96 g each in different treatments. In which, mean body weight of shrimp in total darkness condition (i) was the lowest and significantly lower than the others ( $p < 0.05$ ). The highest survival rate of shrimp was recorded in continuous photoperiod treatment (v) (i.e. 71.9%), and there was no significant difference compared to the treatment 12L:12D (66.7%) and 18L:6D (68.9%). Similarly, shrimp biomass reached the highest in the continuous photoperiod (v) treatment (2.47 kg/m<sup>3</sup>), there was no significant difference from the treatment 18L:6D (2.23kg/m<sup>3</sup>) but it was significantly higher than the others. Besides, shrimp in the photoperiod treatments (v) showed darker color compared to shrimp in total darkness condition (i).

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## 1 INTRODUCTION

*Litopenaeus vannamei* (the Pacific White shrimp) has been known as the most common reared species in intensive and super-intensive systems in Vietnam and the world. For Vietnam, the Mekong Delta is the main farming region which covers over 90% of the culture area and 60% of annual produc-

tion of the whole country. Vietnam had 230,000 hectares of shrimp ponds with 56,000 tons of total production in 1991, increasing to 600,479 hectares and 304,257 tons in 2005. In 2013, Vietnam reached 475,854 tons in shrimp production and 652,613 hectares in total area, of which white leg shrimp covered for 9.8% of the culture area and up to 51.7% of production (Directorate of Fisheries,

2013). White leg shrimp culture has been practiced mainly with intensive systems in the region. In the recent years, the new and advanced biofloc technology has been developed and preliminarily applied in a few companies with intensive or super-intensive systems (Avnimelech, 2005a, 2005b).

Several studies on the effect of light condition on several species have been carried out. Tidwell *et al.* (2001) indicated that continual light conditions showed a positive impact on survival of fresh water prawn juveniles during nursery phase. The studies on photoperiod and light intensity on survival, development and cannibalism of larvae of crab were also done by Gardner and Maguire (1998), and Xiaowu *et al.* (2011). Sanudin *et al.* (2014) suggested that the feeding activity of the shrimp (*Litopenaeus vannamei*) at small stage (0.5 cm in length) was affected by light condition whereas feeding activity, growth and survival of bigger sized shrimp (> 1.0 cm in length) were not affected by light and photoperiod regimes. Hoang *et al.* (2003) reported that the effect of light intensity on shrimp *Penaeus merguensis* growth was stronger and more immediate than that of photoperiod. Neal *et al.* (2010) reported that low or natural light condition did not affect the growth and survival of *Litopenaeus vannamei* in zero-exchange mixed biofloc systems.

Although different fields have been documented on the effects of light condition on shrimp growth, the studies on the photoperiod effecting on the development of bioflocs and water quality as well as shrimp growth performance are rare and required further studies. This study focusing on the effect of photoperiod on development of bioflocs and growth performance of white leg shrimps, *Litopenaeus vannamei*, is to find out an appropriate photoperiod that can help improve the yield of shrimp and reduce the production cost during shrimp culture.

## 2 MATERIALS AND METHODS

### 2.1 Experimental design

Brine water (80-100 ppt) sourced from Bac Lieu salt fields was used to dilute with tap water to obtain the salinity of 15 ppt, then, was treated with chlorine at the concentration of 30 ppm at least 4 days before use. The experiment was set up indoor with fluorescent compact light at the intensity of 6,000 lux. The triplicate experiment was designed with 5 treatments of photoperiod as follows: treatment (i) 24-hour in dark condition (0L:24D); (ii) 6-hour light: 18-hour dark (6L:18D); (iii) 12-hour light: 12-hour dark (12L:12D); (iv) 18-hour light: 6-

hour dark (18L:6D) and (v) 24-hour light (24L:0D).

Fifteen composite tanks of 0.5 m<sup>3</sup> each were used; each tank was filled with 250 L of 15 ppt brackish water. Shrimp postlarvae (PL15) were stocked at the density of 150 PL/m<sup>3</sup> (50 PL/tank). Aeration was supplied continuously during culture.

Shrimp was fed 4 times a day with Grobest feed (40-42% crude protein). Daily feeding amount was in range of 3-19% of shrimp body weight. Alkalinity was maintained at 140 mg CaCO<sub>3</sub> per m<sup>3</sup> by supplying regularly NaHCO<sub>3</sub>. Besides, there was adjustment of feed quantity according to survival rate, uneaten feed and observation of shrimp gut at sampling time. No siphon and no antibiotic were used.

Biofloc was created and maintained in tanks by supplying rice flour as carbon source to ensure C:N ratio at 15:1 (Avnimelech, 1999). For preparation, rice flour solution was heated up at 60°C in closed vessel, then, incubated in room temperature for 48 hours before adding into the tanks every 4 days. Rice flour was added based on the feeding rate (Avnimelech, 2012).

### 2.2 Sampling for shrimp growth evaluation

Shrimps were collected every 2 weeks to evaluate survival rate, growth rate, specific growth rate and production (g/m<sup>3</sup>), based on the following equations:

$$\text{Daily weight gain (g/day)} = (W_2 - W_1)/T$$

$$\text{Daily length gain (cm/day)} = (L_2 - L_1)/T$$

$$\text{Survival rate (\%)} = 100 \times (\text{No. of shrimp harvested} / \text{No. of shrimps stocked})$$

$$\text{Specific Growth Rate (SGR) (\%/day)} = 100 \times (\ln(W_2) - \ln(W_1))/T$$

$$\text{Specific Growth Rate (SGR}_L\text{) (\%/day)} = 100 \times (\ln(L_2) - \ln(L_1))/T$$

Where:  $W_1$  is initial weight (g),  $W_2$  is harvest weight (g),  $L_1$  is initial length (cm),  $L_2$  is harvest length (cm),  $T$  is the culture period (days).

### 2.3 Sampling and water quality parameters measurement

Water parameters such as temperature and pH were measured every 15 days by pH meter at 7:00 AM and 14:00 PM. Concentrations of nitrite, TAN and alkalinity were also measured every 15 days by SERA test kit at 7:00 AM.

Biofloc parameters including floc size and floc volume (FVI) were measured at 15-day intervals.

Length and width of flocs were measured under microscope, FVI was determined by filling 1 L of cultured tank water into Imhoff cone and waited for settlement within 20 minutes and checked for the flocs volume. Total bacteria count and *Vibrio* density in experimental water were determined every 30 days. NA medium was used for total bacteria count and TCBS was used for *Vibrio* count (Phạm Thị Tuyết Ngân and Trần Suong Ngọc, 2014). Water samples were analyzed every 15 days to determine the concentration of Chlorophyll-*a* following the method of Nusch (1980).

The shrimp sensory properties were evaluated following the method described in the study of Meilgaard *et al.* (1999). Shrimp flesh quality was determined through analysis of biochemical composition

**Table 1: Light intensity in the treatments**

Treatments	Light intensity (lux)				
	6:00	9:00	12:00	15:00	18:00
(0L:24D)	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>	0 <sup>a</sup>
(6L:18D)	6,145±33 <sup>b</sup>	6,160±25 <sup>b</sup>	6,199±10 <sup>b</sup>	0 <sup>a</sup>	0 <sup>a</sup>
(12L:12D)	6,132±22 <sup>b</sup>	6,181±17 <sup>b</sup>	6,223±15 <sup>b</sup>	6,160±45 <sup>b</sup>	6,114±10 <sup>b</sup>
(18L:6D)	6,149±16 <sup>b</sup>	6,171±31 <sup>b</sup>	6,216±13 <sup>b</sup>	6,150±20 <sup>b</sup>	6,122±23 <sup>b</sup>
(24L:0D)	6,159±15 <sup>b</sup>	6,159±7 <sup>b</sup>	6,227±8 <sup>b</sup>	6,152±17 <sup>b</sup>	6,126±19 <sup>b</sup>

Values of the same column with the same letters are not significant difference ( $p > 0.05$ )

### 3.1.2 Temperature and pH

Table 2 displayed that average water temperature fluctuated from 28.3 to 29.3°C, with the mean value of 28.3±0.61°C in the morning and 29.3±0.78°C in the afternoon. Temperature in the morning and the afternoon are not highly varied due to the experiment was set up indoor conditions. According

to Trần Việt Mỹ (2009), temperature range of 26-30°C does not affect the growth of white leg shrimp. Average pH in the treatments fluctuated from 7.86 to 8.07. This author recommended that the suitable pH for white leg shrimp culture should be within the range of 7.5-8.5. In general, the variations of temperature and pH in present study were in suitable range for growth of white leg shrimp.

## 3 RESULTS AND DISCUSSIONS

### 3.1 Environmental parameters

#### 3.1.1 Light intensity

Average light intensity fluctuated from 6,114-6,227 lux (Table 1). Light intensity was not significantly different ( $p > 0.05$ ) among treatments under controlled condition. According to Lavens and Sorgeloos (1996), light intensity strongly affects growth of algae and biofloc.

to Trần Việt Mỹ (2009), temperature range of 26-30°C does not affect the growth of white leg shrimp. Average pH in the treatments fluctuated from 7.86 to 8.07. This author recommended that the suitable pH for white leg shrimp culture should be within the range of 7.5-8.5. In general, the variations of temperature and pH in present study were in suitable range for growth of white leg shrimp.

**Table 2: The average of temperature and pH in treatments**

Treatments	Temperature (°C)		pH	
	Morning	Afternoon	Morning	Afternoon
(0L:24D)	28.3±0.1	28.9±0.2	8.03±0.15	8.07±0.13
(6L:18D)	28.6±0.1	29.3±0.1	7.86±0.08	7.99±0.01
(12L:12D)	28.6±0.1	29.1±0.1	7.90±0.03	7.93±0.02
(18L:6D)	28.6±0.1	29.2±0.1	7.94±0.01	7.59±0.03
(24L:0D)	28.4±0.0	29.0±0.1	7.94±0.01	7.55±0.02

### 3.1.3 Concentrations of TAN, nitrite and alkalinity

The results showed that nitrite concentration was 1.87-2.33 mg/L and increased gradually in the first 15 days, then decreased maybe due to the growth of *Nitrobacteria* in tank water medium. Nitrite was the lowest in the treatment 24L:0D and the highest in the treatment 18L:6D. The nitrite concentration was insignificant difference among treatments

( $p > 0.05$ ). Alcaraz *et al.* (1999) reported that lethal concentration (LC-50) of  $\text{NO}_2^-$  for white shrimp *Penaeus setiferus* in 48 hours was 240 mg/L.

Average concentration of TAN fluctuated from 0.2-0.3 mg/L and there was no significant difference of TAN among treatments ( $p > 0.05$ ). TAN had the tendency of declining throughout the culture within 30 days. Whetston (2002) recommended that shrimp grow well in TAN concentration lower than 2 mg/L. It is recommended that the safe dose

of LC-50 of NO<sub>2</sub><sup>-</sup>, would be 12 mg/L. Variation of TAN and nitrite in all treatments of the present study were in the appropriate range for shrimp growth.

**Table 3: Average concentration of TAN, nitrite and alkalinity in the treatments**

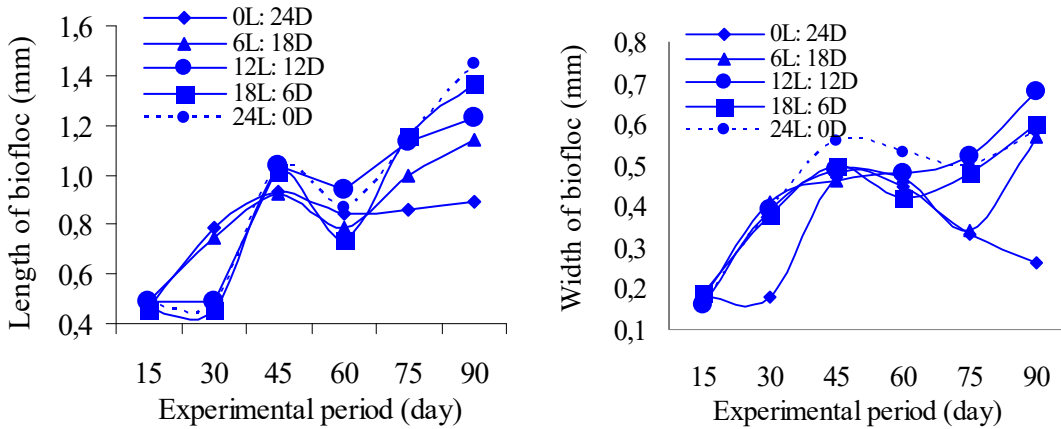
Treatments	Nitrite (mg/L)	TAN (mg/L)	Alkalinity (mg CaCO <sub>3</sub> /L)
(0L:24D)	2.17±0.4	0.2±0.1	104±6.0
(6L:18D)	2.17±0.5	0.2±0.0	109±1.7
(12L:12D)	1.93±0.2	0.2±0.1	113±8.6
(18L:6D)	2.33±0.5	0.3±0.1	109±3.5
(24L:0D)	1.87±0.1	0.3±0.0	111±3.5

Alkalinity varied 109-113 mg CaCO<sub>3</sub>/L, and there was no significant difference among the treatments, thanks to frequently adding of NaHCO<sub>3</sub>. According to Chanratchakool *et al.* (2003), the appropriate alkalinity for shrimp was 80-120 mg/L. Plinio (2013) reported that high alkalinity made the optimal condition for biofloc formation and bacteria growth. Alkalinity below 40 mg/L would cause un-

adjustable pH level. pH was significantly changed on day and night time and caused negative effect on shrimp health.

3.1.4 Biofloc parameters in the treatments

Figure 1 showed that the variation in biofloc dimension tends to increase to the end of the experiment. In the first 15 days of culture, biofloc size was 0.48-0.49 mm in length and 0.16-0.19 mm in width. The size continuously increased to 0.74-0.94 mm in length and 0.46-0.56 mm in width at day 60, finally reached 0.86-1.45 mm and 0.57-0.68 mm in length and width, respectively after 90 days. Treatment 0L:24D had the smallest biofloc size (0.89 × 0.26 mm) and it was significantly smaller than those in the other photoperiod treatments (p<0.05). Treatments 0L:24D and 6L:18D had larger biofloc size in the first 30 days than the others. However, they were decreased gradually and became smaller size than those in the others at the end of experiment. The results implied that light intensity and photoperiod affected the flocs formation and size.



**Fig. 1: Average length and width of biofloc in different treatments**

Figure 2 showed the biofloc volume gradually increased till the end of the experiment and fluctuated from 1.0-25.3 mL/L among treatments. In the first 15 days, FVI was as low as 1.0-1.8 mL/L. However, after 60 days, FVI increased to 6.7-11.3 mL/L, then 13.3-25.3 mL/L at the end of experiment. The treatment 6L:18D had the highest FVI

(25.3 mL/L) while treatment 0L:24D with the lowest FVI (13.3 mL/L). There was insignificant difference in FVI among treatments (p>0.05). According to Avnimelech (2012), the suitable FVI in shrimp pond was 15-50 mL/L. Thus, the FVI in this experiment was suitable for growth of white leg shrimp.

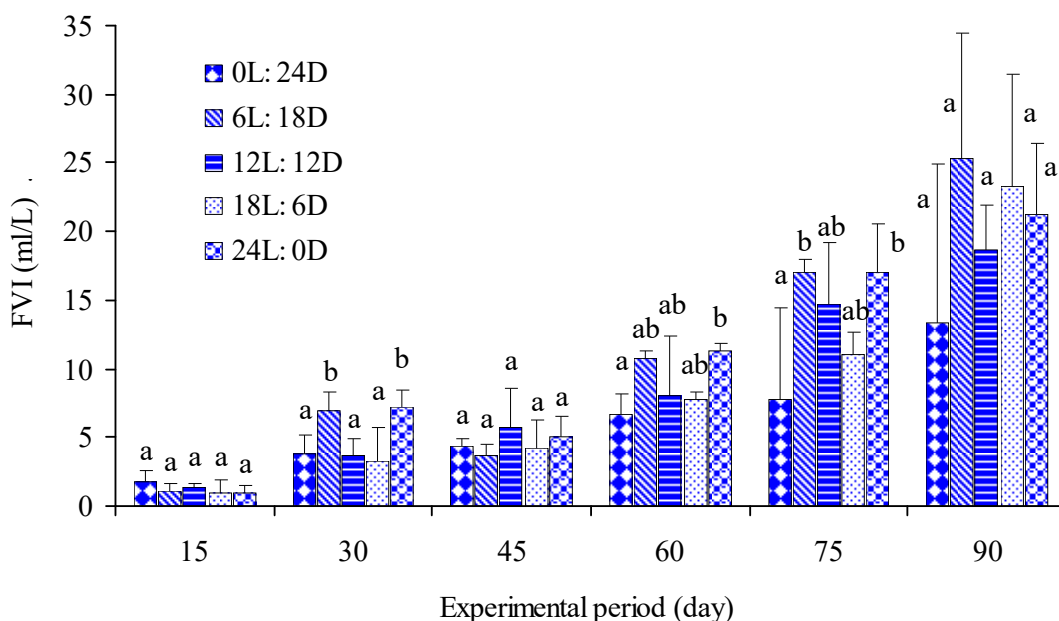


Fig. 2: The floc volume in different treatments after 90 days of culture

3.1.5 Chlorophyll-a concentration

Table 4 showed the variation of chlorophyll-a concentration in different photoperiod treatments. Besides, treatment 24L:0D obtained the highest chlorophyll-a level during cultured period (from 18-187 µg/L), and it was significantly higher than those in other treatments (p<0.05). In contrast, the treatment 24D:0L with total darkness showed the

lowest chlorophyll-a concentration. This result indicated that increase of photoperiod period increased chlorophyll-a concentrations in the treatments 6L:18D, 12L:12D, 18L:6D and 24L:0D. Perhaps, longer photoperiod period is to enhance algae development, therefore, to rise the chlorophyll-a concentration; according to Baloi *et al.* (2013), chlorophyll-a level was influenced by light photoperiod.

Table 4: Chlorophyll-a concentration (µg/L) in different treatments

Treatments	Culture duration (day)					
	15	30	45	60	75	90
(0L:24D)	5.5±2 <sup>a</sup>	3.7±0.8 <sup>a</sup>	9.5±4.2 <sup>a</sup>	11.6±1.1 <sup>a</sup>	7.7±7.7 <sup>a</sup>	9.3±8.3 <sup>a</sup>
(6L:18D)	9.6±4.3 <sup>ab</sup>	4.7±0.5 <sup>a</sup>	9.4±3.8 <sup>a</sup>	12.5±0.5 <sup>a</sup>	6.4±2.5 <sup>a</sup>	13.7±3.7 <sup>a</sup>
(12L:12D)	11.1±3.5 <sup>ab</sup>	6.1±1.2 <sup>a</sup>	13.0±1.4 <sup>a</sup>	17.5±2.7 <sup>a</sup>	12.8±2.9 <sup>a</sup>	16.8±3.0 <sup>a</sup>
(18L:6D)	14.8±2.3 <sup>bc</sup>	16.6±8.2 <sup>a</sup>	17.7±4.1 <sup>a</sup>	20.5±67.9 <sup>a</sup>	19.4±5.2 <sup>a</sup>	48.3±2.1 <sup>b</sup>
(24L:0D)	18.0±3.6 <sup>c</sup>	63.9±24.8 <sup>b</sup>	116±88.5 <sup>b</sup>	187±53.2 <sup>b</sup>	116±32.9 <sup>b</sup>	82.5±31.6 <sup>c</sup>

Values in the same column with the same letters are not significant difference (p>0.05)

3.1.6 Total bacteria and Vibrio in the treatments during 90 days of culture

The results in Table 5 showed that average bacteria count at day 30 was from 438 to 805×10<sup>3</sup> CFU/mL, and it was not significantly different among treatments (p>0.05). The total bacteria count reached 815-8,900×10<sup>3</sup> CFU/mL and 796-17,100×10<sup>3</sup> CFU/mL at day 60 and day 90, respectively. It is noted that the total bacteria in treatment 6L:18D after 60 days and 90 days of rearing was significantly higher than other treatments. This photoper-

iod is probably suitable for development of total bacteria though no reference was found to convince this. Rajkumar *et al.* (2016) reported that white leg shrimp farming in biofloc system with total bacteria of 148×10<sup>6</sup> CFU/mL had no effect on shrimp survival rate.

The average density of *Vibrio* was 0.37-0.92×10<sup>3</sup> CFU/mL at day 30, then increased up to 2.2-9.0×10<sup>3</sup> CFU/mL and 2.7-13.5×10<sup>3</sup> CFU/mL at day 60 and day 90, respectively. In this period, *Vibrio* in treatment 18L:6D reached the highest number at 13.5×10<sup>3</sup> CFU/mL and it was signifi-



cantly ( $p < 0.05$ ) higher than the others. In general, the density of total bacteria in treatment 6L:18D and *Vibrio* in 0L:24L, 18L:6D exceeded threshold for shrimp farming (Rajkumar *et al.*, 2016). However, *Vibrio* per total bacteria ratio was at low level

from 0.05 to 0.68% (Table 7). It was stated that shrimp still grow well when *Vibrio* density was higher than  $10^4$  CFU/mL (Phạm Thị Tuyết Ngân and Trần Suong Ngọc, 2014).

**Table 5: Total bacteria count ( $10^3$ CFU/mL)**

Treatments	Culture duration (day)			
	0	30	60	90
(0L:24D)	2.9±1.4 <sup>a</sup>	790±28.2 <sup>a</sup>	1.325±35.4 <sup>a</sup>	3.560±56.6 <sup>a</sup>
(6L:18D)	2.9±1.4 <sup>a</sup>	800±28.2 <sup>a</sup>	5.100±848 <sup>b</sup>	7.100±1.131 <sup>b</sup>
(12L:12D)	2.9±1.4 <sup>a</sup>	775±148 <sup>a</sup>	815±49.5 <sup>a</sup>	3.325±176 <sup>a</sup>
(18L:6D)	2.9±1.4 <sup>a</sup>	795±7.0 <sup>a</sup>	1.165±262 <sup>a</sup>	3.275±106 <sup>a</sup>
(24L:0D)	2.9±1.4 <sup>a</sup>	805±77.8 <sup>a</sup>	1.395±77.8 <sup>a</sup>	3.615±120 <sup>a</sup>

Values in the same column with the same letters are not significant difference ( $p > 0.05$ )

**Table 6: *Vibrio* density ( $10^3$  CFU/mL) during 90 days of culture period**

Treatments	Culture duration (day)			
	0	30	60	90
(0L:24D)	0.06±0.0	0.69±0.08 <sup>b</sup>	8.95±0.07 <sup>d</sup>	11.1±1.48 <sup>b</sup>
(6L:18D)	0.06±0.0	0.37±0.06 <sup>a</sup>	2.15±0.21 <sup>a</sup>	2.70±0.28 <sup>a</sup>
(12L:12D)	0.06±0.0	0.79±0.13 <sup>bc</sup>	5.90±0.28 <sup>c</sup>	8.60±0.57 <sup>b</sup>
(18L:6D)	0.06±0.0	0.82±0.05 <sup>bc</sup>	6.60±0.42 <sup>c</sup>	13.5±2.12 <sup>c</sup>
(24L:0D)	0.06±0.0	0.92±0.06 <sup>c</sup>	3.15±0.49 <sup>b</sup>	3.40±0.57 <sup>a</sup>

Values in same column with the same letter were not significant difference ( $p > 0.05$ )

**Table 7: *Vibrio* and total bacteria ratio (%) in different treatments**

Treatments	Time (day)			
	0	30	60	90
(0L:24D)	0.19±0.03 <sup>a</sup>	0.09±0.01 <sup>a</sup>	0.68±0.02 <sup>cd</sup>	0.31±0.04 <sup>b</sup>
(6L:18D)	0.19±0.03 <sup>a</sup>	0.05±0.01 <sup>a</sup>	0.05±0.01 <sup>a</sup>	0.04±0.01 <sup>a</sup>
(12L:12D)	0.19±0.03 <sup>a</sup>	0.10±0.00 <sup>a</sup>	0.73±0.01 <sup>d</sup>	0.26±0.03 <sup>b</sup>
(18L:6D)	0.19±0.03 <sup>a</sup>	0.11±0.01 <sup>a</sup>	0.58±0.09 <sup>c</sup>	0.42±0.05 <sup>c</sup>
(24L:0D)	0.19±0.03 <sup>a</sup>	0.12±0.02 <sup>a</sup>	0.23±0.02 <sup>b</sup>	0.09±0.01 <sup>a</sup>

Values in the same column with the same letter are not significant difference ( $p > 0.05$ )

### 3.2 Growth rate of white leg shrimp

#### 3.2.1 Growth rate in length

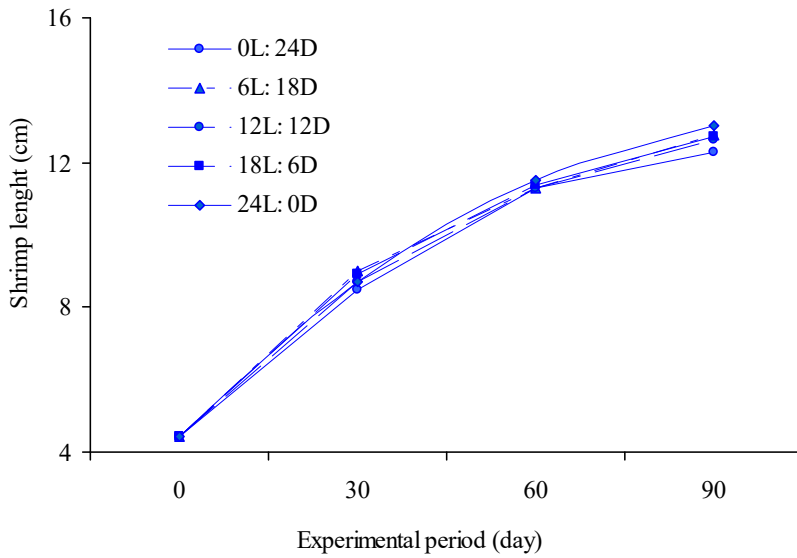
The body length of shrimps after 90 cultured days in all treatments varied in the range of 12.35-13.0 cm (Table 8) and daily length gain reached 0.09-

0.1 cm/day (or 1.13-1.19 %/day). Shrimp in treatment 24L:0D showed the highest length (13.0 cm), this was similar to those in total darkness condition (i) ( $p > 0.05$ ) but significantly different to the other treatments ( $p < 0.05$ ).

**Table 8: Average growth rate in body length of shrimps after 90 days of culture period**

Treatments	L <sub>1</sub> (cm/ind)	L <sub>2</sub> (cm/ind)	DLG (cm/day)	SGR <sub>L</sub> (%/day)
(0L:24D)	4.45±0.96 <sup>a</sup>	12.35±0.21 <sup>a</sup>	0.09±0.01 <sup>a</sup>	1.13±0.02 <sup>a</sup>
(6L:18D)	4.45±0.96 <sup>a</sup>	12.77±0.21 <sup>ab</sup>	0.09±0.02 <sup>a</sup>	1.19±0.02 <sup>a</sup>
(12L:12D)	4.45±0.96 <sup>a</sup>	12.60±0.17 <sup>ab</sup>	0.09±0.01 <sup>a</sup>	1.16±0.01 <sup>a</sup>
(18L:6D)	4.45±0.96 <sup>a</sup>	12.73±0.39 <sup>ab</sup>	0.09±0.02 <sup>a</sup>	1.17±0.02 <sup>a</sup>
(24L:0D)	4.45±0.96 <sup>a</sup>	13.00±0.17 <sup>b</sup>	0.10±0.01 <sup>a</sup>	1.22±0.02 <sup>a</sup>

Values in same column with the same letters were not significantly different ( $p > 0.05$ )

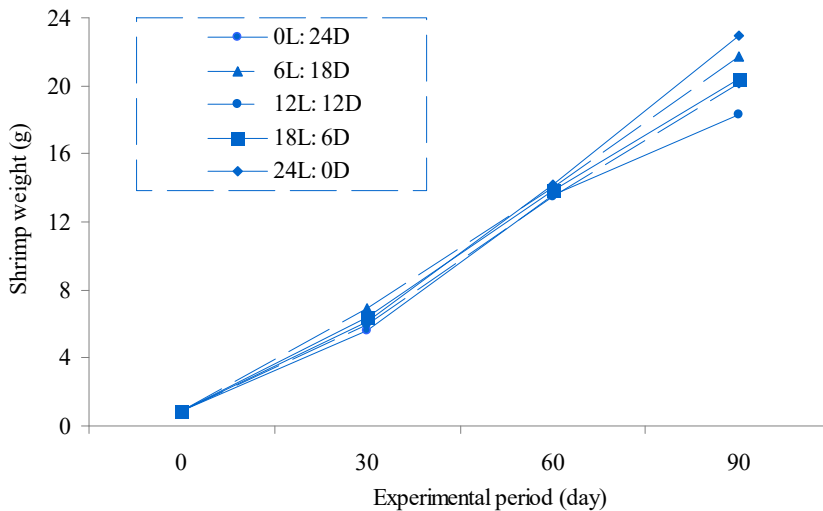


**Fig. 3: Body length of shrimp during culture period**

**3.2.2 Weight gain of shrimps in the treatments**

After 90 days of culture, body weight of shrimp was 18.28-22.96 g, and daily weight gain was in the range of 0.20-0.25 g/day (or 3.42-3.68 %/day). Shrimps in the treatment 24L:0D reached the highest weight (22.96 g/ind) and daily weight gain (0.25 g/day or 3.68%/day). It was significantly different compared to those in total darkness treatment with the corresponding numbers of 18.28 g/ind, 0.2 g/day or 3.42%/day. However, there was no significant difference ( $p > 0.05$ ) from other treatments. In general, shrimp in treatment 24L:0D showed the highest body weight and growth rate

compared to the others. Neal *et al.* (2010) reported that natural light at 710 lux or 12-hour light at 50 lux did not affect growth rate of *Litopenaeus vannamei* under biofloc condition, only high density of 364 shrimp per  $m^2$  affect to growth. Biao *et al.* (2013) reported that white leg shrimp farming with different light regimes had effects to shrimp growth. Wang *et al.* (2004) revealed that light intensity affected the growth of the shrimp mainly by influencing feed conversion efficiency, or energy allocation to growth. Therefore, the shrimp may grow better in organically rich earthen ponds that have lower light intensity 500-1300 lux.



**Fig. 4: Weight gain of shrimp during culture duration**

The present study is different to Sanudin *et al.* (2014) who suggested that the feeding activity of

the small shrimp (0.5 cm in length) was affected by light condition whereas feeding activity, growth

and survival of bigger sized shrimp (> 1.0 cm in length) were not affected by light and photoperiod regimes. The difference could be due to biofloc application. It is noted that the biofloc size in total

darkness treatment was significantly smaller compared to illumination treatment. And smaller floc particles seem to be favorable to smaller shrimp.

**Table 9: Average weight gain of shrimps after 90 days**

Treatments	W <sub>1</sub> (g/ind)	W <sub>2</sub> (g/ind)	DWG (g/day)	SGR (%/day)
(0L:24D)	0.84±0.17	18.28±0.67 <sup>a</sup>	0.20±0.01 <sup>a</sup>	3.42±0.04 <sup>a</sup>
(6L:18D)	0.84±0.17	21.32±0.94 <sup>b</sup>	0.23±0.01 <sup>b</sup>	3.59±0.05 <sup>b</sup>
(12L:12D)	0.84±0.17	20.15±1.12 <sup>b</sup>	0.23±0.01 <sup>b</sup>	3.58±0.06 <sup>b</sup>
(18L:6D)	0.84±0.17	21.67±1.08 <sup>b</sup>	0.23±0.02 <sup>b</sup>	3.61±0.05 <sup>b</sup>
(24L:0D)	0.84±0.17	22.96±0.73 <sup>b</sup>	0.25±0.01 <sup>b</sup>	3.68±0.04 <sup>b</sup>

Values in same column with the same letter were not significant difference ( $p > 0.05$ )

**3.3 Survival rate, biomass and feed conversion rate (FCR)**

Survival rate of shrimp after 90 cultured days was from 55.6 to 71.9 % (Table 10). The highest survival rate (71.9%) was under treatment 24L:0D, and it was not significantly different ( $p > 0.05$ ) compared to treatment 12L:12D (66.7%) and 18L:6D (68.9%). However, it was significantly different ( $p < 0.05$ ) to the total darkness and treatment 6L:18D. Similarly, shrimp biomass fluctuated from 1.51 to 2.47 kg/m<sup>3</sup>. Treatment 24L:0D presented the highest shrimp biomass (2.47 kg/m<sup>3</sup>)

and it was not significantly different compared to treatments 18L:6D and 12L:12D, but it was significantly greater than other treatments ( $p < 0.05$ ). Neal *et al.* (2010) reported that natural light at 710 lux or 12-hour light at 50 lux did not affect the survival rate and FCR of *Litopenaeus vannamei* under biofloc condition.

FCR values reached 1.74-2.84 in which treatment 24L:0D has the lowest FCR values compared to the treatment 6L:18D and total darkness treatment but it was not significantly different to the others ( $p > 0.05$ ).

**Table 10: Average survival rate, biomass and FCR during 90 days of culture**

Treatments	Survival rate (%)	FCR	Biomass (kg/m <sup>3</sup> )
(0L:24D)	55.6±6.3 <sup>a</sup>	2.84±0.18 <sup>c</sup>	1.51±0.13 <sup>a</sup>
(6L:18D)	57.8±8.0 <sup>ab</sup>	2.33±0.37 <sup>b</sup>	1.82±0.27 <sup>ab</sup>
(12L:12D)	66.7±2.3 <sup>abc</sup>	1.88±0.21 <sup>a</sup>	2.12±0.17 <sup>bc</sup>
(18L:6D)	68.9±7.7 <sup>bc</sup>	1.89±0.1 <sup>a</sup>	2.23±0.15 <sup>c</sup>
(24L:0D)	71.9±3.4 <sup>c</sup>	1.74±0.13 <sup>a</sup>	2.47±0.16 <sup>c</sup>

Values in the same column with the same letters are not significant difference ( $p > 0.05$ )

**3.4 Quality and approximate composition of white leg shrimp**

*3.4.1 Quality of white leg shrimp*

Table 11 showed the score of sensory property evaluation on color of fresh and after boiling shrimp, fluctuated from 6.71-7.57. The shrimp color of fresh and boiled shrimp of total darkness treatment (i) had the lowest point (6.71), and it was significantly lower than other treatments ( $p < 0.05$ ). In other photoperiod treatments, there was no significant difference in shrimp color (either fresh or boiled shrimp). In general, the longer the photoperiod was, the higher the color scores were. However, the fresh shrimps under the treatments 18L:6D and 24L:0D had lower score than those under the treatment 12L:12D.

was not significantly different among the treatments ( $p > 0.05$ ). The pattern of boiled shrimp fluctuated from 7.19-8.0; under total darkness condition, shrimps had the lowest point in smelling. Again, the longer the photoperiod was, the higher the scores of smelling were. However, shrimps under continuous photoperiod treatment 24L:0D presented lower smelling point than those under other treatments, significant difference was not found.

The flavour point of shrimp in treatments ranged from 7.14 to 7.86. Treatments 12L:12D and 0L:24D had the highest score (7.86), and it was significantly higher than 6L:18D (7.14). However, this value was not significantly different ( $p > 0.05$ ), compare to the treatments of 18L:6D and 24L:0D. The color of shrimp changed with photoperiod in different treatments, especially there was the difference of the shrimp color for both fresh and boil-

The average sensory point on smelling of fresh shrimp in the treatments was from 7.28-7.86, and it



ing pattern between total darkness condition and photoperiod treatments in the present study. Mar-

tinez *et al.* (2014) reported that the stronger the color was, the healthier the shrimp was.

**Table 11: Evaluation scores on color, smelling and flavor of shrimp**

Treatments	Fresh		Boiled		
	Color	Odor	Color	Odor	Flavor
(0L:24D)	6.71±0.49 <sup>a</sup>	7.28±0.49 <sup>a</sup>	6.71±0.49 <sup>a</sup>	7.14±0.69 <sup>a</sup>	7.14±0.69 <sup>a</sup>
(6L:18D)	7.43±0.53 <sup>b</sup>	7.71±0.49 <sup>a</sup>	7.14±0.38 <sup>ab</sup>	7.43±0.53 <sup>a</sup>	7.14±0.38 <sup>a</sup>
(12L:12D)	7.57±0.54 <sup>b</sup>	7.86±0.38 <sup>a</sup>	7.14±0.69 <sup>ab</sup>	7.86±0.38 <sup>b</sup>	7.86±0.38 <sup>b</sup>
(18L:6D)	7.42±0.53 <sup>b</sup>	7.57±0.53 <sup>a</sup>	7.43±0.54 <sup>b</sup>	8.00±0.00 <sup>b</sup>	7.57±0.53 <sup>ab</sup>
(24L:0D)	7.29±0.48 <sup>b</sup>	7.43±0.54 <sup>a</sup>	7.57±0.53 <sup>b</sup>	7.43±0.53 <sup>ab</sup>	7.29±0.49 <sup>ab</sup>

Values in the same column with the same letters are not significant difference ( $p > 0.05$ )

**3.4.2 Proximate compositions and toughness of white leg shrimp**

There was no significant difference in proximate compositions and toughness of shrimps in all treatments ( $p > 0.05$ ). Moisture fluctuated from 75.6-76% with the highest value in treatments 6L:18D and 18L:6D. Shrimp in the treatments

18L:6D and 24L:0D had the highest protein level (74.0-74.7%). The lowest lipid content was in the treatment 18L:6D. Ash varied from 6.2-6.7% and the lowest was found in treatment 24L:0D. Toughness varied from 477-535 g.cm. In general, compact light 55w (~ 6000 lux) with different photoperiods did not affect biochemical compositions and toughness of white leg shrimp.

**Table 12: Proximate compositions and toughness of shrimp in different treatments**

Treatments	Moisture (%)	Protein (%)	Lipid (%)	Ash (%)	Toughness (g.cm)
(0L:24D)	75.8±0.1 <sup>a</sup>	73.7±1.1 <sup>a</sup>	2.8±0.8 <sup>a</sup>	6.7±0.9 <sup>a</sup>	477±95 <sup>a</sup>
(6L:18D)	76.0±0.1 <sup>a</sup>	72.6±1.5 <sup>a</sup>	2.1±0.7 <sup>a</sup>	6.4±0.8 <sup>a</sup>	499±80 <sup>a</sup>
(12L:12D)	75.7±0.3 <sup>a</sup>	73.4±0.8 <sup>a</sup>	1.9±0.6 <sup>a</sup>	5.6±0.9 <sup>a</sup>	488±72 <sup>a</sup>
(18L:6D)	76.0±0.3 <sup>a</sup>	74.0±1.7 <sup>a</sup>	1.8±0.9 <sup>a</sup>	6.5±0.7 <sup>a</sup>	505±146 <sup>a</sup>
(24L:0D)	75.9±0.2 <sup>a</sup>	74.7±0.4 <sup>a</sup>	2.0±0.5 <sup>a</sup>	6.2±0.6 <sup>a</sup>	535±111 <sup>a</sup>

Values in the same column with the same letters are not significant difference ( $p > 0.05$ )

**4 CONCLUSIONS AND RECOMMENDATIONS**

All the water quality parameters in the treatments remained within the appropriated ranges for the culture of white leg shrimp. The photoperiod of treatment 24L:0D by compact light 55w (6.114-6.227 lux) showed the better growth rate (3.69%/day), FCR (1.68), survival rate, shrimp biomass and quality of shrimp (color and toughness) compared to the other treatments. However, those parameters in the treatments of 12L:12D and 18L:6D were not significantly different, compared to those of the 24L:0D. Thus, it is acceptable for the culture.

Applying biofloc technology in green house with 24L:0D of photoperiod to be considered as the effective condition for this experiment. This experiment proved that light intensity from different treatment was far below the light intensity in outdoor culture. Therefore, is it possible to propose a shelter system, through which light intensity per treatment could be controlled (e.g. 20%, 30% dark cover and thus less light penetration into water tanks). However, several factors such as energy,

operational expenses, and production goals must be considered. Prior to apply for large scale, production with different photoperiods should be tried and analyzed in term of financial aspects.

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