Effects of carbohydrate addition time on a biofloc system for white leg shrimp (*Litopenaeus vannamei*) at larvae and post larvae stage

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Abstract. Biofloc technology (BFT) was widely applied as a sustainable tool for use in aquaculture that simultaneously addresses environmental, social, and economic issues. This study aimed to evaluate the effects of carbon addition time on water quality, biofloc compositions, and *Litopenaeus vannamei* larvae performance. The experiment was conducted with four carbon addition times at Mysis-1 (Treatment 1), Mysis-3 (Treatment 2), Postlarvae-2 (PL-2) (Treatment 3) and Postlarvae-4 (PL-4) stage (Treatment 4). The larvae were stoked in 500L composite tanks at 30 ppt of salinity and 150 ind L^{-1} of density. According to the experimental designed treatments, refined sugar was daily applied as a carbon source to maintain the C/N ratio at 20:1. The results showed that the water quality, total bacteria and *Vibrio* spp. count, and biofloc performance in all treatments were in an appropriate range for larvae development. The treatment 2 (Mysis-3) was recorded with the highest growth, survival and productivity of PL-12 (10.47±0.12 mm, 66.5±5.5%, 99,709±8,231 ind m^{-3}, respectively), which were significantly different compared to others ($p < 0.05$). All the postlarvae-12 survived after formalin and salinity shock tests. These results suggested that sugar could be applied for white leg shrimp larvae rearing at Mysis-3 stage to promote the biofloc development, water quality management and larval performance.

Key Words: biofloc, carbon addition, growth performance, *Litopenaeus vannamei*.

Introduction. Biofloc technology (BFT) has been recently considered as an eco-friendly tool that simultaneously addresses environmental, social, and economic issues in aquaculture (Furtado et al 2015; Khoa et al 2020). The biofloc systems manipulate the carbon/nitrogen ratio and convert these metabolites to heterotrophic microorganisms and organic particles (Crab et al 2009; Bakhshi et al 2018). It could help to reduce the water use and risk of introduction and spread of pathogens (Furtado et al 2011). In addition, BFT can significantly improve feed conversion, shrimp growth performance, lowering the cost of production (Wasielesky et al 2006; Xu & Pan 2014; de Souza et al 2016), thus resulting in efficient and healthy culture of shrimp (Ray et al 2011; Khoa et al 2020). BFT was generated by using external carbon sources to stimulate the formation of microbial aggregates in the system (Avnimelech 1999) under negligible or zero-water exchange (Xu & Pan 2014). Previous studies demonstrated that carbon source and C/N ratio of culture water were considered to be critical factors affecting the performance of microbial communities which significantly impact water quality and biofloc production, hence, eventually affect feed utilization and shrimp performance (Avnimelech 1999; Ebeling et al 2006; Xu et al 2016; Gomes Vilani et al 2016). Importantly, these parameters are specific-species and developmental stage applications (Mugwanya et al 2021).

White leg shrimp (*Litopenaeus vannamei*) is a commercially important species in aquaculture and being widely cultured in the world. This species shows high stocking density tolerance and is the most compatible shrimp for biofloc systems (Panigrahi et al 2019; Khoa et al 2020). Numerous studies have investigated and optimized the appropriate biofloc conditions for nursery and grow-out of *L. vannamei* (Furtado et al 2015; Prangnell et al 2016; Xu et al 2016; Khoa et al 2020). Recently, BFT was applied in larval rearing of *L. vannamei* (Tao et al 2021), in which, sugar was used as a carbon source and C/N ratio at 20:1 was applied. Relative to conventional larval rearing methods, the larval survival rate and productivity were significantly improved when reared with bioflocs (Tao et al 2020; Tao et al 2021). In order to improve further the techniques of *L. vannamei* larviculture in BFT systems, this study was conducted to evaluate the effects of carbon addition time on water quality and *L. vannamei* larval and postlarval performance in BFT system.

Material and Method

Experimental design. This study was conducted in College of Aquaculture, Can Tho University, Vietnam during May to July 2020. *L. vannamei* nauplii were provided by Chau Phi Company, Ninh Thuan province of Vietnam (were obtained by SIS broodstocks, Hawaii, USA). After 3 hours of acclimation in 500 L tank, the healthy and active nauplii were collected and treated with 200 ppm of formalin for 30 seconds then used for the experiment. The 30 ppt salinity water used for the larval rearing was prepared from brine (80 ppt of salinity) and tap water. Before using, water was treated with 50 ppm of chlorine followed by strong aeration for 48h. Sodium bicarbonate ($NaHCO_3$) was added to maintain the alkalinity level at 160 mg $CaCO_3$ L^{-1} (Tao et al 2015) throughout the culture. The water was filtered through a 1 µm mesh size filter prior to fill to larval tanks.

Refined sugar (Bien Hoa Company, Viet Nam; containing 55.54% of C and 0.19% of N) was daily applied as a carbon source to promote the development of biofloc. Carbon source provided was recommended by Avnimelech (2014) in order to maintain C:N = 20:1 in BFT system. Before introducing to rearing tanks, sugar was mixed with water and commercial probiotic (1 g m^{-3}) then incubated for 24 h with strong aeration (Tao et al 2021).

Shrimp larvae were initially stocked in 500-L composite tanks at 150 ind L^{-1} of density. Four treatments were randomly set up in triplicate, in which, carbon was added at different stages of larvae of Mysis-1 (Treatment 1), Mysis-3 (Treatment 2), Postlarvae 2 (Treatment 3), and Postlarvae 4 (Treatment 4).

Rearing management. At Zoea 1 and Zoea 2 stage, *Chaetoceros* sp. was added every 3 h to maintain a density of 60,000-120,000 cell mL^{-1}. From Zoea 3, shrimp larvae were fed 0.4 g m^{-3} of artificial feed (INVE, Thailand; with a mixture of 50% Lansy ZM + 50% Frippak-1) every 3 h. At Mysis stage, shrimp larvae were alternatively fed every 3 h with artificial feed (50% Lansy ZM + 50% Frippak-2, 1-1.5 gm^{-3}), and umbrella *Artemia* (2 g m^{-3}). From PL1 to PL6, alternatively fed every 3 h with Frippak-150 (2-3 g m^{-3}) and newly hatched *Artemia* (2-3 g m^{-3}/feeding; PL7 to PL12 alternatively fed every 3 h with Lansy PL (2-4 g m^{-3}) and newly hatched *Artemia* (3-4 g m^{-3}) (Tao et al 2021). During the rearing, siphoning was only conducted at the late Zoea 3 stage, however, there were no siphoning required during Mysis stage towards the end of the experiment. Rearing water was not exchanged during larval culture but only supplied with treated brackish water to compensate for the evaporation loss. During culture, morderate aeration was continuously applied to larval rearing tanks.

Sampling

Water quality parameters. Measurements of temperature and pH were performed twice a day at 8:00 AM and 2:00 PM using HI-98127 Multi-Parameter Waterproof Meter (HANNA Instruments, Ltd.). Total ammonia nitrogen (TAN), and nitrite (NO_2^-) were also monitored every 3 days by the Indophenol Blue colorimetric method and Griess-Ilosvay method, respectively (APHA 2005).

Bacterial count. Total bacterial density and *Vibrio* spp. density in rearing water were weekly sampled, and in shrimp gut (PL-12) were sampled at the end of the experiment. Total bacterial density was checked using Nutrient Agar containing 1.5% NaCl (NA), while *Vibrio* spp. density was analyzed using TCBS (Thiosulfat Citrate Bile Salt Surcose) agar (Huys 2002).

Biofloc performance. Floc sizes (mm) of 30 samples of each tank were randomly collected and measured using Nikon ECLIPSE Ti2 microscope with a DS-Qi2 camera (Nikon

Corporation, Tokyo, Japan). Floc volume (ml L^{-1}) was monitored through collecting a 1 L water sample in an Imhoff cone, which was left undisturbed for 30 min; settled floc was read and noted as mL L^{-1} (Tao et al 2020). Biofloc performance was evaluated at PL4, PL8 and PL12 stages. At the end of the experiment, biofloc proximal compositions (including protein, lipid, and ash) were determined following the standard methods described by AOAC (2016), and plankton community compositions were determined following Shirota (1966).

Zootechnical parameters. The body length of shrimp larvae and postlarvae were determined at Zoea 3, Mysis 3, PL4, PL8, and PL12, by a random sampling of 30 shrimp of each tank to measure under the Nikon ECLIPSE Ti2 microscope with a DS-Qi2 camera (Nikon, Japan).

Survival rate and productivity of shrimp were determined after harvesting (PL12) as:

Survival rate (%) = (harvested shrimps/initial stocked shrimps) x 100

Productivity (ind L^{-1}) = number of shrimp collected/tank volume

The quality of postlarae was evaluated by application of stress tests for PL12 followed a standard of TCVN 10257: 2014 (MOST 2014), consisting of formalin shock test (100 ppm) and salinity – freshwater shock test (Tao et al 2021).

Data analysis. Microsoft Excel 2010 (Microsoft Corporation, USA) and SPSS 22.0 (IBM corporation, USA) were used to sort and analyze the data. One-way ANOVA and Duncan's test were applied to compare the mean. All differences were considered at $\alpha = 0.05$.

Results

Water quality parameters. During the experiment time, the environmental parameters such as temperature, pH and alkalinity were not significantly different among treatments ($p > 0.05$) (Table 1). In which, water temperature ranged from 29.1 to 29.7°C and pH was at 7.73-8.17. The alkalinity was stable at 155.6-156.2 mg $CaCO_3$ L^{-1} due to frequent additions of $NaHCO_3$. These parameters were in acceptable range for shrimp larviculture (Ho & Lu 2003; Hai et al 2017; Tao et al 2021). However, there are a significant difference in the levels of TAN and nitrite among treatments, whereby the lowest levels of TAN and nitrite were recorded in the treatments I (Mysis-1) while the treatment 4 (Postlarvae 4) showed the highest levels of TAN and nitrite ($p < 0.05$) (Table 1). According to Ebeling et al (2006), in the biofloc system, the nitrification process by chemoautotrophic bacteria resulted in the reduction of NO_2-N while the removal of TAN was obtained by heterotrophic bacteria. The addition of carbon could increase the total heterotrophic bacterial density in the system, having the ability to consume NH_3-N and NO_3-N for growth and multiplication (Avnimelech 1999). When the C/N ratio was well balanced, TAN and NO_2-N concentration in the biofloc system gets effectively reduced after the formation of biofloc by both heterotrophic assimilation and autotrophic nitrification (Santhana et al 2018; Panigrahi et al 2019).

Table 1

Water quality parameters

Parameters		*Treatments*			
		1	*2*	*3*	*4*
Temperature (°C)	8 am	29.1±0.4	29.2±0.4	29.0±0.5	29.3±0.3
	2 pm	29.7±0.5	29.5±0.5	29.6±0.4	29.6±0.5
pH	8 am	7.86±0.25	7.78±0.19	7.73±0.19	7.76±0.2
	2 pm	8.12±0.17	8.13±0.14	8.15±0.13	8.17±0.14
Alkalinity (mg $CaCO_3$ L^{-1})		156.2 ± 0.2^a	155.9 ± 0.3^a	156.2 ± 0.9^a	155.6 ± 0.7^a
TAN (mg L^{-1})		0.30 ± 0.02^a	0.37 ± 0.11^{ab}	0.47 ± 0.07^{bc}	0.51 ± 0.1^c
NO_2^- (mg L^{-1})		0.29 ± 0.02^a	0.36 ± 0.03^{ab}	0.42 ± 0.06^{bc}	0.49 ± 0.1^c

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b < c, $p < 0.05$).

Total bacterial count and Vibrio spp. count

Total bacterial count. The total bacteria density among treatments gradually increased during the experimental period (Table 2). The results showed that there was no significant difference in total bacteria density in tanks until day 7. However, from day 14 through day 21, total bacteria density was significantly increased in Mysis-1 and Mysis-3 treatments ($p < 0.05$*)* compared to PL-2 and PL-4 treatments. Besides, the highest total bacteria densities in shrimp and biofloc were observed in Mysis-1 treatment (1.50±0.10 x 10^4 CFU g^{-1} and 13.30±1.30 x 10^4 CFU g^{-1}, respectively), while these two parameters were the lowest in PL-4 (1.16±0.15 x 10^4 CFU g^{-1} and 6.10±0.53 x 10^4 CFU g^{-1}, respectively) ($p < 0.05$).

Table 2
Total bacterial density among treatments during the experimental period (10^4 CFU mL^{-1} in water, 10^4 CFU g^{-1} in shrimp and biofloc)

Sampling day	*Treatments*			
	1	*2*	*3*	*4*
Day 7 - Water	0.05 ± 0.02^a	0.04 ± 0.01^a	0.03 ± 0.01^a	0.02 ± 0.02^a
Day 14 - Water	0.87 ± 0.04^b	0.88 ± 0.08^b	0.72 ± 0.25^{ab}	0.56 ± 0.11^a
Day 21 - Water	1.46 ± 0.10^b	1.37 ± 0.09^b	1.10 ± 0.07^a	0.98 ± 0.11^a
Shrimp	1.50 ± 0.10^b	1.47 ± 0.25^{ab}	1.17 ± 0.06^a	1.16 ± 0.15^a
Biofloc	13.30 ± 1.30^c	11.53 ± 1.12^c	8.83 ± 0.59^b	6.10 ± 0.53^a

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b < c, $p < 0.05$).

Vibrio spp. count. No statistical difference in *Vibrio* density in water during 21 days, in shrimp and biofloc was recorded between Mysis-1 and Mysis-3 treatments ($p > 0.05$), but was significantly lower than PL-2 and PL-4 treatments ($p < 0.05$) (Table 3). It indicated that the carbon addition time affected the growth of microbial population in the biofloc systems. Early addition of carbon will promote the growth of total bacteria and inhibit the growth of *Vibrio* bacteria. The results were in a suitable range and did not harm to the shrimp larvae (Tao et al 2021). Moreover, the competitive approach of bacteria, including heterotrophic bacteria for space and nutrients with the *Vibrio* population and also due to the disruption of cell to cell communication of *Vibrio* population by the biofloc reduced the *Vibrio* count in the biofloc systems (Bossier & Ekasari 2017; Santhana et al 2018).

Table 3
Vibrio spp. density among treatments during the experimental period (10^2 CFU mL^{-1} in water, 10^2 CFU g^{-1} in shrimp and biofloc)

Parameters	*Treatments*			
	Mysis-1	*Mysis-3*	*PL-2*	*PL-4*
Day 7 - Water	0.03 ± 0.01^a	0.03 ± 0.02^a	0.05 ± 0.02^a	0.06 ± 0.02^a
Day 14 - Water	0.53 ± 0.15^a	0.77 ± 0.15^a	1.13 ± 0.25^b	1.33 ± 0.15^b
Day 21 - Water	2.63 ± 0.06^a	2.60 ± 0.26^a	3.23 ± 0.42^b	3.47 ± 0.21^b
Shrimp	0.67 ± 0.21^a	0.87 ± 0.15^a	1.40 ± 0.20^b	1.67 ± 0.15^b
Biofloc	1.33 ± 0.15^a	1.53 ± 0.15^a	1.80 ± 0.10^b	1.93 ± 0.15^b

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

The size and composition of the bioflocs

Biofloc volume. the volume of biofloc monitored in treatments was gradually increased during culture and no significant change among treatments until PL-4 stages (0.1-0.13 mL L^{-1}). However, from stage PL-8 through PL-12, the highest biofloc volumes were observed in Mysis-1 treatment and Mysis-3 treatment (1.9-2.07 mL L^{-1}), that were statistically higher than PL-2 treatment and PL-4 treatment (1.23-1.37 mL L^{-1}) ($p < 0.05$).

Table 4

Biofloc volume from each treatment (mL L^{-1})

Parameter		*Treatments*			
		1	*2*	*3*	*4*
Biofloc volume (mL L^{-1})	PL-4 stage	0.13 ± 0.06^{a}	0.10 ± 0.01^{a}	0.13 ± 0.06^{a}	0.13 ± 0.05^{a}
	PL-8 stage	1.17 ± 0.25^{b}	1.00 ± 0.10^{b}	0.53 ± 0.15^{a}	0.57 ± 0.15^{a}
	PL-12 stage	2.07 ± 0.15^{b}	1.90 ± 0.36^{b}	1.37 ± 0.12^{a}	1.23 ± 0.15^{a}

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

Besides, the biofloc dimension (width x length) in the treatments intended to increase by size gradually during culture and was not significantly different until PL-8 stage (0.15-0.19 mm in width and 0.26-0.33 mm in length) (Table 5). However, the biofloc dimension in Mysis-1 treatment at stage PL-12 reached 0.28 mm in width and 0.49 mm in length, which was significant greater than other treatments, excepting Mysis-3 treatment ($p < 0.05$*).*

Table 5

Biofloc dimension in each treatment during the experimental periods (mm)

Biofloc dimension		*Treatments*			
		1	*2*	*3*	*4*
Width (mm)	PL-4 stage	0.15 ± 0.01^{a}	0.15 ± 0.03^{a}	0.16 ± 0.03^{a}	0.16 ± 0.03^{a}
	PL-8 stage	0.15 ± 0.02^{a}	0.15 ± 0.01^{a}	0.19 ± 0.02^{a}	0.18 ± 0.03^{a}
	PL-12 stage	0.28 ± 0.07^{b}	0.21 ± 0.05^{ab}	0.19 ± 0.02^{a}	0.21 ± 0.01^{ab}
Length (mm)	PL-4 stage	0.30 ± 0.01^{a}	0.26 ± 0.05^{a}	0.24 ± 0.05^{a}	0.27 ± 0.03^{a}
	PL-8 stage	0.26 ± 0.01^{a}	0.25 ± 0.03^{a}	0.33 ± 0.05^{a}	0.27 ± 0.06^{a}
	PL-12 stage	0.49 ± 0.07^{b}	0.35 ± 0.06^{a}	0.28 ± 0.06^{a}	0.29 ± 0.03^{a}

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

In biofloc systems, the size volume and size of biofloc are related to the biofloc uptake potential by the aquatic animals, the digestibility of the flocs as well as the nutritional value of the floc (De Schryver et al 2008). Moreover, floc size can play an important role in the quality of biofloc in terms of nutritional composition and nitrogen retention by the animals (Ekasari et al 2014). Additionally, regarding particle size or floc abundance, the rupture of the floc structure affects the distribution and interaction of nitrifying bacteria with reflections in the nitrification process (Souza et al 2019); in this study, smaller size biofloc showed remarkably lower nitrification compared to larger size biofloc (de Sousa Carvalho et al 2006; Souza et al 2019). The results for biofloc volume and size in this study correlated with TAN and nitrite levels, as higher biofloc size and volume reflected in the lower levels of TAN and nitrite (Khoa et al 2020).

Biofloc compositions. The plankton compositions and their relative abundance (%) were consisting of Bacillariophyta, Cyanophyta, Euglenophyta, Pyrophyta, Chlorophyta, Protozoa (Table 6). In which, each treatment was recorded with 16-24 species; whereby Protozoa (37.5-47.06%), Bacillariophyta (29.17-31.82%), and Chlorophyta (5.88-12.5%) were recorded as predominant species in the biofloc system.

Table 6
The biofloc compositions and relative abundance (%) of phytoplankton and zooplankton community structure in all treatments at during the trial period

Variables	*Treatments*							
	1		*2*		*3*		*4*	
	Number of species	*%*	*Number of species*	*%*	*Number of species*	*%*	*Number of species*	*%*
Bacillariophyta	7	29.17	7	31.82	5	29.41	5	31.25
Cyanophyta	3	12.50	0	0.00	1	5.88	1	6.25
Euglenophyta	1	4.17	1	4.55	1	5.88	2	12.50
Pyrophyta	1	4.17	2	9.09	1	5.88	1	6.25
Chlorophyta	3	12.50	2	9.09	1	5.88	1	6.25
Protozoa	9	37.50	10	45.45	8	47.06	6	37.50
Total	24	100	22	100	17	100	16	100

Biofloc contained from 29.1 to 30.8% of protein, 2-2.33% of lipids, and 28.1-28.8% of ash (Table 7). Proteins in the treatments of Mysis-1 (30.8%) and Mysis-3 (30.4%) were significantly higher than PL-2 and PL-4 treatments (29.1-29.2%) ($p < 0.05$). The highest lipid value was recorded in Mysis-1 treatment, which was statistically higher than others ($p < 0.05$). Similarly, the ash content in Mysis-1 treatment (28.8%) was also higher than other treatments (28.1-28.4%) ($p < 0.05$).

Table 7
Proximate nutritional compositions of biofloc (dry weight)

Parameter	*Treatments*			
	1	*2*	*3*	*4*
Protein (%)	30.8 ± 0.46^{b}	30.4 ± 0.26^{b}	29.2 ± 0.21^{a}	29.1 ± 0.20^{a}
Lipid (%)	2.33 ± 0.15^{b}	2.13 ± 0.15^{ab}	2.10 ± 0.10^{ab}	2.00 ± 0.10^{a}
Ash (%)	28.8 ± 0.31^{b}	28.5 ± 0.20^{ab}	28.4 ± 0.15^{ab}	28.1 ± 0.15^{a}

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

BFT provided a supplemental natural food source containing protein, lipid, mineral, and vitamin, which beneficially affected shrimp production (Wasielesky et al 2006; Xu & Pan 2012). The microbial abundance (algae, bacteria, plankton, etc.) in biofloc system could help to enhance the nutrient removal process as well as the proximate nutritional compositions of biofloc (Khoa et al 2012). The values in protein (29.1-30.8%) and ash (28.1-28.8%) of this study were in range with studies described by Tao et al (2021) with 29% of protein, or 27.3-31.6% (Xu & Pan 2012). However, the lipid content (2-2.33%) was significantly lower compared to 3.37-3.34% (Khoa et al 2020), 3.7-4.2% (Xu & Pan 2012) and 5.4-5.7% (Tao et al 2021). It could be explained by the use of different carbon sources and C/N ratios, differences in microbial population structures in biofloc systems (Xu & Pan 2012; Deng et al 2018; Khoa et al 2020).

Growth performance of shrimp larvae and postlarvae

Growth of shrimps. No significant difference in length of shrimp larvae among the treatments was recorded from Zoea 3 through Mysis 3 stage (3.48-3.57 mm ($p > 0.05$). At postlarave 12 stage, the Mysis-3 treatment was observed with a significant higher value in length (10.47 mm) compared to others (9.72-9.95 mm) ($p < 0.05$) (Table 8).

Table 8

Body length of shrimp larvae and postlarvae (mm)

Larval stages	*Treatments*			
	1	*2*	*3*	*4*
Zoae 3	2.03 ± 0.04^{a}	2.02 ± 0.01^{a}	2.01 ± 0.01^{a}	2.02 ± 0.02^{a}
Mysis 3	3.51 ± 0.04^{a}	3.48 ± 0.11^{a}	3.57 ± 0.14^{a}	3.56 ± 0.09^{a}
Postlarvae 4	5.49 ± 0.15^{a}	5.72 ± 0.10^{ab}	5.75 ± 0.12^{b}	5.53 ± 0.15^{ab}
Postlarvae 8	8.26 ± 0.12^{a}	8.36 ± 0.02^{a}	8.12 ± 0.21^{a}	8.07 ± 0.24^{a}
Postlarvae 12	9.72 ± 0.25^{a}	10.47 ± 0.12^{b}	9.95 ± 0.04^{a}	9.79 ± 0.18^{a}

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

Survival rate and productivity (PL-12) of shrimp. After harvest, the shrimp survival in all treatments was above 50% (ranged from 52.2 to 66.5%) (Table 9). The highest survival was observed in Mysis-3 treatment (66.5%) which was not significantly different to Mysis-1 treatment (60.2%) ($p > 0.05$), but significantly higher than PL-2 treatment and PL-4 treatment (52.2-53%) ($p < 0.05$). Similarly, the productivity of PL-12 in the Mysis-3 treatment was the highest (99.709 ind m^{-3}), followed by Mysis-1 treatment (90,366 ind m^{-3}) ($p > 0.05$), PL-2 treatment and PL-4 treatment (78,366-79.489 ind m^{-3}) ($p < 0.05$).

Table 9

Final survival rate and productivity of shrimp from each treatment

Parameter	*Treatments*			
	1	*2*	*3*	*4*
Survival rate (%)	60.2 ± 2.9^{ab}	66.5 ± 5.5^{b}	52.2 ± 3.0^{a}	53.0 ± 9.0^{a}
Productivity (ind m^{-3})	$90{,}366 \pm 4{,}297^{ab}$	$99{,}709 \pm 8{,}231^{b}$	$78{,}366 \pm 4{,}567^{a}$	$79{,}489 \pm 13{,}565^{a}$

Mean values in the same row with different superscript differ significantly (ANOVA, Duncan's test, a < b, $p < 0.05$).

BFT could help to maintain good water quality conditions (Ebeling et al 2006; Ekasari et al 2014; Tao et al 2021), improve shrimp growth performance, digestive capacity, and immune responses (Xu & Pan 2012, 2014), hence enhance the survival, growth and productivity in shrimp larval rearing. The differences in growth, survival and productivity among treatments may due to the fact of water quality, biofloc compositions and nutrition (Hari et al 2004; Khoa et al 2020). Accordingly, biofloc was considered as an important source of food protein (Hari et al 2004), free amino acids, lipid, mineral, and vitamins (Xu & Pan 2012; Samocha et al 2019). Moreover, the biofloc could exert a positive effect on the immune system of cultured shrimp probably because of being rich in natural microbial bioactive compounds such as polysaccharides and carotenoids (Xu & Pan 2014). The results in shrimp survival, growth and productivity were similar to studies on white leg shrimp and tiger shrimp larvae rearing in biofloc systems (Tao et al 2019; Tao et al 2020; Tao et al 2021).

Postlarvae quality. After subjecting to shock test using formalin and salinity stress, all the treatments were observed with 100% of shrimp survival. This result was acceptable for postlarvae 12 quality (TCVN 10257: 2014) prior to transferring to the grow-out phase.

Conclusions and recommendations. In larval rearing of white leg shrimp in biofloc systems, it was suggested that refined sugar could be applied as a carbon source to stimulate the biofloc performance. In which, the application of sugar at Mysis-3 stage showed the best results in water quality, shrimp growth performance, survival, and productivity. Therefore, it could be applied to practical production of shrimp hatchery.

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