Salinity effect evaluation on the survival rate and hematology of snakeskin gourami juvenile *Trichopodus pectoralis*

Evaluasi pengaruh salinitas terhadap sintasan dan gambaran darah benih ikan sepat siam *Trichopodus pectoralis*

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ABSTRACT

Snakeskin gourami *Trichopodus pectoralis* an endemic fish that was highly demanded by Indonesian society. The declining in population stock was the main problem cause its availability still relies upon the nature catch. The increase of its production of culture was still constrained by the slow rate of growth and survival rate in the seed phase. This study aimed to determine the optimal salinity and its effect on growth performance, survival rate, osmotic activity, and blood description of the snakeskin gourami *Trichopodus pectoralis*. A complete randomized design (CRD) was used in this study with four different treatments and each treatment consisted of three replications. The salinity test consists of 0 g/L, 1 g/L, 3 g/L, and 5 g/L. The experimental fish sized at 0.49 ± 0.13 g and length 2.96 ± 0.42 cm with a stock density of 25 fish/aquarium (1 fish/L). During 28-day of rearing, the experimental fish was fed using *Tubifex* sp. as restricted based on the FR (5%) of biomass per weekly sampling. The results showed that the optimal survival rate and growth showed the best at salinity 3 g/L because it showed the most stable blood level so that the fish could avoid stress response.

Keywords: salinity, survival rate, growth, hematological, *Trichopodus pectoralis*

ABSTRAK

Ikan sepat siam *Trichopodus pectoralis* merupakan ikan endemik yang sangat diminati masyarakat Indonesia. Pemenuhan kebutuhan ikan sepat siam masih mengandalkan hasil tangkapan dari alam sehingga populasi di alam mengalami penurunan. Peningkatan produksi ikan sepat siam dari budidaya masih terkendala dengan sintasan dan pertumbuhan yang lambat pada fase benih. Tujuan penelitian ini adalah menentukan salinitas optimum dan pengaruhnya terhadap performa sintasan, pertumbuhan, kerja osmotik, dan gambaran darah benih ikan sepat siam. Penelitian dilakukan dengan metode rancangan acak lengkap (RAL), empat perlakuan dan masing-masing 3 kali ulangan. Perlakuan yang diujikan adalah salinitas 0 g/L, salinitas 1 g/L, salinitas 3 g/L, dan 5 g/L.Ikan yang digunakan berukuran 0,49 ± 0,13 g dan panjang 2,96 ± 0,42 cm, padat tebar masing-masing 25 ekor/akuarium (1 ekor/L). Selama 28 hari masa pemeliharaan, ikan sepat siam diberi pakan berupa cacing sutra (*Tubifex* sp.) secara *restricted* berdasarkan FR 5% dari biomassa per sampling mingguan. Hasil penelitian menunjukkan sintasan dan pertumbuhan terbaik dihasilkan pada perlakuan salinitas 3 g/L. Hal itu diduga karena gambaran darah pada perlakuan 3 g/L stabil sehingga terhindar dari respon stres.

Kata kunci: salinitas, sintasan, pertumbuhan, gambaran darah, sepat siam

INTRODUCTION

The snakeskin gourami *Trichopodus pectoralis* is an endemic species which highly popular in Indonesia. The economic importance of this species can be recognized through the market price which ranges from IDR 25,000–35,000/kg (Diniya *et al.*, 2013). The fulfillment of the market demand still depends on the natural catch, thus it leads to the decrease of population. The downturn of the population is indicated through the overall size of this species which is getting smaller (Ath-thar *et al.,* 2014) so that several attempts are profoundly required to maintain the sustainability of snakeskin gourami.

The advancement of local species in Indonesia is continuously managed. The culture of Indonesian local species is starting to manage because the high capture in their natural habitat causes a huge decline in the population. Sukadi *et al.* (2009) mentioned that the development of Indonesian local species is desperately required to keep biodiversity through on-farm conservation and food resilience. The production of snakeskin gourami is still inhibited by the low survival and the slow growth of this species in the seed phase. The problem solving is pursued through numerous studies about biological reproduction (Oktafia *et al.*, 2014; Ath-Thor *et al.,* 2014), feed management (Murjani, 2011; Siswanto *et al.*, 2016), and growth performance of the snakeskin gourami (Morioka *et al.*, 2010).

The study about snakeskin gourami with the physiological approach is one of the efforts to boost growth performance, especially in the nursery phase. Rachmawati *et al.* (2012) stated that salinity could affect feed utilization and growth. Besides, Darwis *et al.* (2009) also described that salinity is one of the environmental factors which closely related to feed digestibility and fish growth. The salinity studies towards the growth performance of freshwater species were widely conducted, for example on marble goby *Oxyeleotris marmorata* (Ardi *et al.*, 2016), synodontis (Wijaya & Gustino, 2011), and tinfoil barb *Barbonymus schwanenfeldii* (Nurjanah, 2014). A study by Kucuk (2013) on *Carassius auratus* and *Carassius carassius* resulted in the best growth performance in 12 g/L of salinity. However, the optimum salinity to support the growth performance of snakeskin gourami seed in the nursery phase is limited. Therefore, this study aimed to determine the optimum salinity and the effect on growth performance, survival, osmotic mechanism, and blood profile of snakeskin gourami.

MATERIALS AND METHODS

Experimental design

The experimental design in this study was a complete randomized design with four treatments and three replications. The determination of the initial salinity concentration was tested through the preliminary study using five different salinity levels (3 g/L, 6 g/L, 9 g/L, 12 g/L, and 15 g/L). The determination of the salinity level was done based on the minimal and maximal tolerance of freshwater species. The result of the preliminary study for six days presented that 3 g/L of salinity level resulted in a 100% survival rate, while the 6 g/L of salinity levels resulted in 60% of survival. The result was used as a guide to set the treatment in the main study (0 g/L, 1 g/L, 3 g/L, and 5 g/L).

Rearing container

The rearing container was an aquarium sized in 40 cm×25 cm×25 cm and it was filled 25 L of water. The salinity water was obtained from the mixture of marine water and freshwater. The snakeskin gourami seed was collected from Jambi with the average weight and length was 0.49±0.13 g and 2.96 ± 0.42 cm, respectively. The fish seed was reared with 25 ind/aquarium of stocking density (I ind/L). During the 28-day of rearing, the fish were fed using *Tubifex* sp. The feeding method was restricted to 5% of FR each weekly sampling. The feeding frequency was three times a day.

Parameters

The observed parameters were bodyweight & length, survival rate, and blood profiles (osmotic performance, glucose, hematocrit, erythrocyte, and hemoglobin. The environmental parameters consisted of temperature, dissolved oxygen, pH, and ammonia. The sampling of body weight & length and environmental parameters were managed weekly, whereas the survival rate and blood profiles were calculated at the end of the study. The measurement of body weight & length, specific growth rate, survival rate, survival rate, osmotic performance, hematocrit, and erythrocyte referred to Ardi *et al.* (2016).

The absolute weight & length is the margin amongst the final weight/length and initial weight/

length. The later parameters were calculated using this following formula:

$$W = Wt - Wo \ (g)$$
$$P = Pt - Po \ (cm)$$

Note:
W   =   Weight gain (g)
Wt =   Final weight (g)
Wo =   Initial weight (g)
P   =   Length gain (cm)
Pt   =   Final length (cm)
Po   =   Initial length (cm)

The specific growth rate (SGR) is the length and weight gain each day. SGR was calculated using this following formula:

$$SGR = \frac{\ln Wt - \ln Wo}{t} \times 100$$

Note :
SGR = Specific growth rate (%/day)
W_t = Final average weight (g)
W_0 = Initial average weight(g)
 t   = Rearing period (day)

The survival rate is the percentage of the final population and the initial population. This parameter was calculated using this formula below:

$$SR \ (\%) = \frac{Nt}{No} \times 100$$

Note :
SR   =   Survival rate
Nt   =   Final population (individuals)
N_0 =   Initial population (individuals)

Data analysis

Data were tabulated and analyzed using analysis of variance (ANOVA) at 95% confidence level. The programs used for data analysis were Microsoft Excel 2010 and SPSS 16. A significant difference would be analyzed further using Duncan posthoc test. The environmental parameters were analyzed descriptively.

RESULTS AND DISCUSSIONS

Results
Survival rate
The highest survival rate of snakeskin gourami was 97.33% and the lowest was 62.00%. The statistic analysis presented significant results amongst treatment (P<0.05). The overall result of the survival rate is presented in Figure 1 below.

Growth performance
The growth performance of snakeskin gourami indicated a significant difference among treatments. The 3 g/L of salinity level showed a significant difference amongst treatments (P<0.05). The SGR, body weight, and body
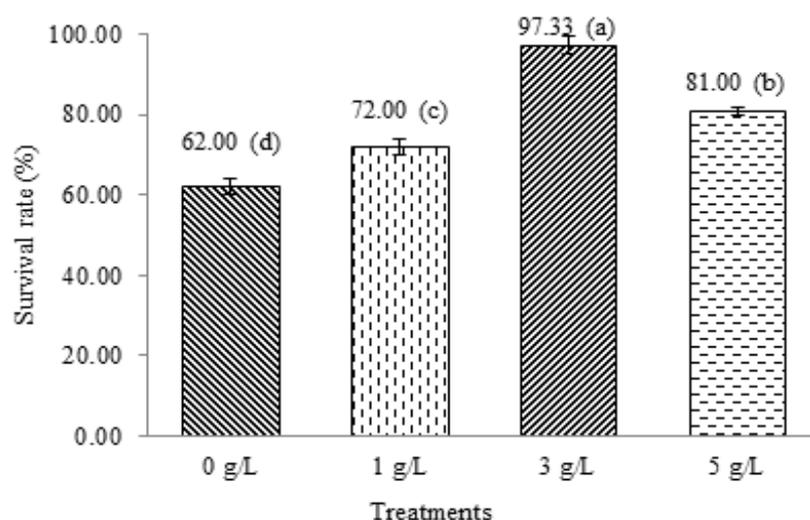

Figure 1. The survival rate of snakeskin gourami *Trichopodus pectoralis* at different salinity levels. The different superscript indicates a significant difference among treatment (P<0.05).
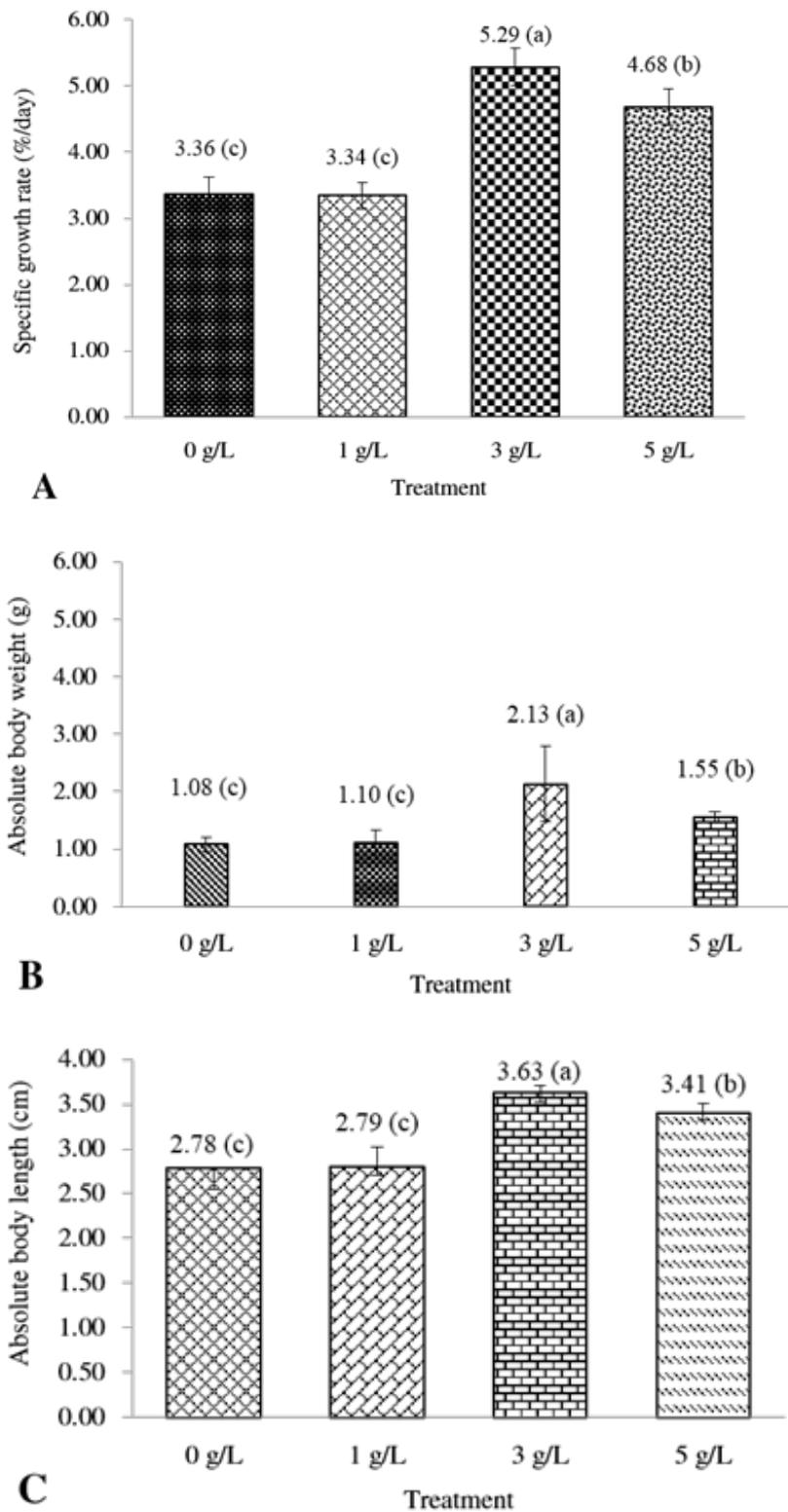
Figure 2. Specific growth rate (A), weight growth (B), and length growth (C) of snakeskin gourami reared in various levels of salinity. The different superscript indicates a significant difference among treatment (P<0.05).
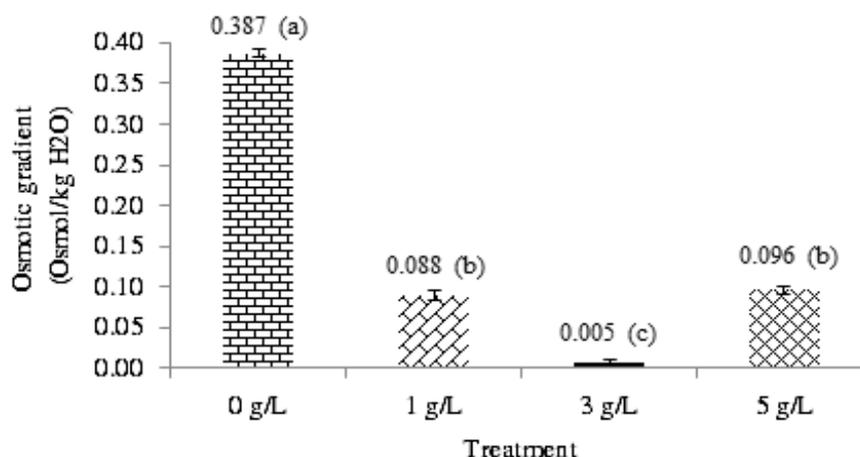
Figure 3. The osmotic gradient of snakeskin gourami in various levels of salinity. The different superscript indicates a significant difference among treatment (P<0.05).

length of the 3 g/L treatment were 5.29%, 2.13 g,and 3.63 cm. The SGR, body weight, and body length are shown in Figure 2.

Osmotic gradient

The osmotic gradient of the 0 g/L treatment showed the highest result (0,387 Osmol/kg H₂O), whereas the lowest osmotic gradient as shown in the 3 g/L treatment (0,005 Osmol/kg H₂O). Statistically, the results were different significantly among treatment.

Blood profile

The blood profile analysis is required to observe the physiological responses or stress levels of the snakeskin gourami seed. The parameters consisted of blood glucose, hematocrit, and erythrocyte. The lowest result of blood glucose resulted in the 3 g/L treatment (74.79 mg/dL). The result was different significantly compared to the 0 g/L (97.85 mg/dL), 1 g/L (96.09 mg/dL), and 5 g/L (92.14 mg/dL) (P<0.05). The result is presented in Figure 4 below.

Water quality

The accomplishment of aquaculture is not separated from chemical and physical water quality management. The water temperature of the rearing media ranged from 27.4–29.4°C. The pH level ranged from 5.74–7.94. The pH level was considered normal and tolerable to support snakeskin growth, especially in the 3 g/L treatment.

Discussions

The survival rate is believed as the main aspect of aquaculture because the final population

directly affects the total production. After 28 days of rearing, the highest survival rate was the 3 g/L of salinity treatment (97.33%) and significantly different amongst treatments (P<0.05) (Figure 1). The decrease of snakeskin gourami survival in the 1 g/L of salinity level was presumably caused by the hyperosmotic condition so that it triggered the osmoregulation system to manage the osmotic balance. It was closely related to the stress condition. A high level of osmotic gradient between internal body fluids and the environment will disrupt the physiological process, increase stress, and cause death (Porchase *et al.*, 2009; Ostrowski *et al.*, 2011; O'Neill *et al.*, 2011). Optimization of salinity level was achieved in the 3 g/L of salinity treatment. The result was supported by Jomori *et al.* (2012) who reported the pacu *Piaractus mesopotamicus* which reared in a 2 g/L of salinity level and the survival rate reached 100%.

The specific growth rate of snakeskin gourami seed in the 3 g/L of salinity treatment was 5.48 ± 0.29%/day and the lowest was obtained in the 0 g/L (3.36 ± 0.26%/day) and 1 g/L(3.34 ± 0.20%/day) (P<0.05) (Figure 2). The 3 g/L of salinity treatment performed an isosmotic condition which supported an optimal growth. It presumably caused by the effective energy allocation which was majorly utilized for growth than basic metabolism. That might explain the low osmotic gradient in the 3 g/L of salinity treatment. The slow growth in the 0 g/L and 1 g/L of salinity treatment was reasonably triggered by the hypoosmotic of the rearing media. Thus, it lowered the metabolic function and growth of the snakeskin gourami seed. As for the 5 g/L of salinity treatment was apparently in hyperosmotic condition. Arjona *et*
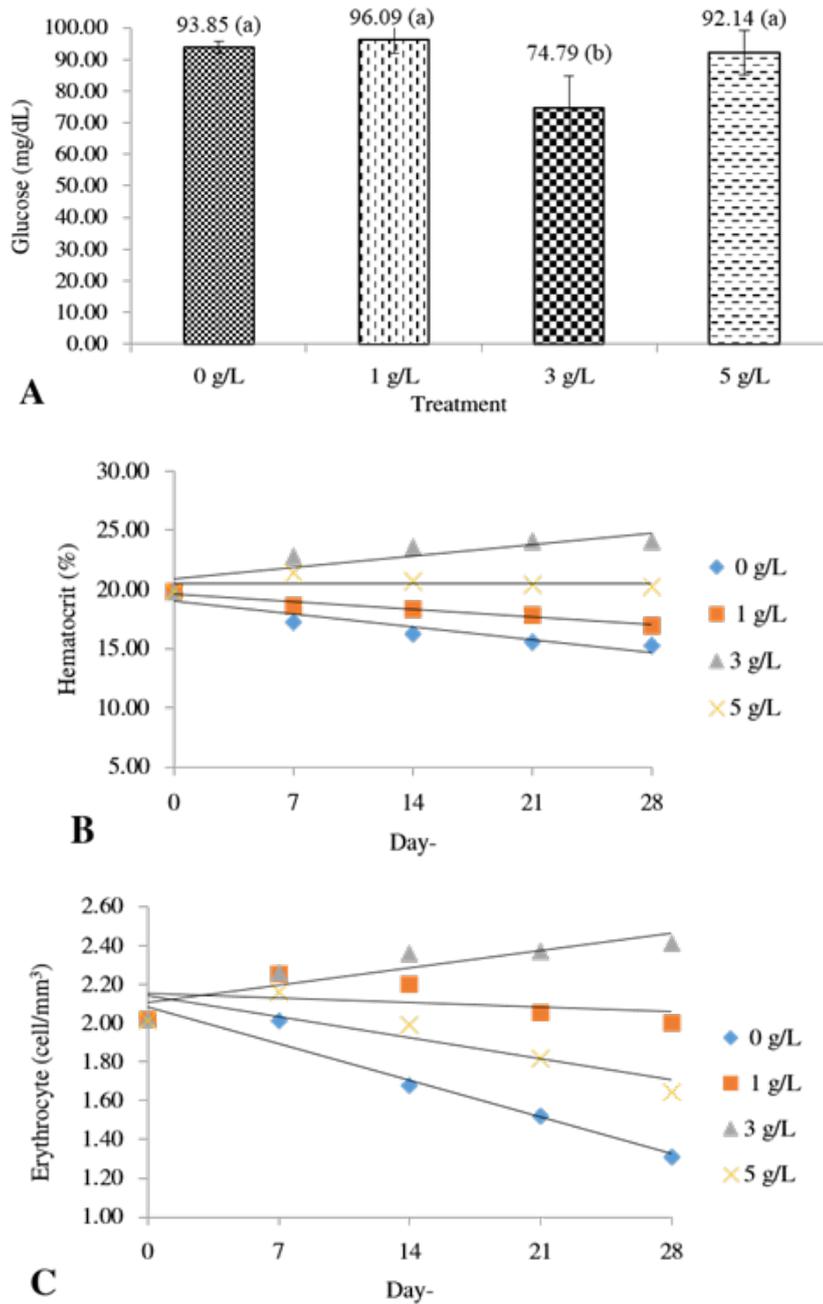
Figure 4. Blood profile of snakeskin gourami in various levels of salinity. The different superscript indicates a significant difference amongst treatment (P<0.05). A: Glocose, B: Hematocrit, C: Erythrocyte.

al. (2009) explained that a physiological response generated by salinity fluctuation caused plenty of energy loss to maintain the homeostasis condition. The mechanism mentioned before generated the decline of metabolic function and hormone related to the osmoregulation process. A significant result of snakeskin gourami body weight appeared in the 3 g/L of salinity treatment (2.43 ± 0.65 g). The outcome was considered higher than the 0 g/L (1.08 ± 0.12 g), 1 g/L (1.10 ± 0.23 g), and 5 g/L (1.55 ± 0.09 g) in salinity treatment (Figure 2). Statistical analysis also presented a significant result in the 3 g/L treatment compared to the

other treatments (P<0.05). However, the result amongst 0 g/L and 1 g/L treatment did not differ significantly (P>0.05). This particular species also presented a higher weight growth compared to the Kalimantan snakeskin gourami (Ath-thar *et al.*, 2014).

Similarly, the significant result of snakeskin gourami body length resulted in the 3 g/L treatment (P<0.05) amongst treatment. Different salinity level presented different growth responses through specific growth rate, body weight, and body length. It's reasonable generated by physiological processes related to the salinity level, included the

metabolism and osmoregulation process. Salinity relates to the osmotic pressure, which in line with the ion solubility in water. The higher the salinity gets, the osmotic pressure will get higher as well (Patterson *et al.*, 2012). The gradient between the ion and total osmotic in a rearing medium determines the ion flow (diffusion) and water flow (osmosis) through the gill epithelium (Evans, 2011). A freshwater teleost species is hyperosmotic towards the rearing media. The internal ion concentration is higher than the media so that it experiences NaCl loss through the gill epithelium. However, when a species are exposed to a nonoptimal salinity level, it will release an abundance amount of energy to regulate the osmotic balance (Kucuk, 2013). One of the freshwater species which requires salinity is the common carp *Cyprinus carpio.* The growth and survival rate of common carp increased and feed efficiency was achieved in 12 g/L of salinity level (Jelkic *et al.*, 2014).

In this study, the environment factors did not directly affect the treatments, especially temperature, because the study was managed indoor in a well-controlled room. Dalvi *et al.* (2009), a nonoptimal water temperature potentially causes various pathophysiology diseases. A huge gradient between the water temperature and the internal body disrupts metabolism and growth. The particular condition causes a major energy allocation to regulate the adaptation process towards the unconducive environment and disturbs the metabolic system. Schulte (2011) mentioned that temperature has a major influence on the biological process of an organism from the cell form to the ecosystem because the affected aspects were various biological reactions and intermolecular interaction. Besides, the temperature is also involved in the physiological process and fish reproduction (Migaud *et al.*, 2013; Pankhurst & Munday, 2011).

The osmotic gradient in all treatments was varied. The initial osmotic gradient was 0.387 Osmol/kg H_2O. The final measurement of the osmotic gradient of 0 g/L, 1 g/L, 3 g/L, and 5 g/L were 0.387, 0.088, 0.005, and 0.09 Osmol/kg H_2O, respectively. A low level of an osmotic gradient indicates the low energy required to regulate the osmoregulation condition. On the contrary, a high level of osmotic gradient leads to lower survival. It is in line with Porchase *et al.* (2009) who mentioned that a high level of osmotic gradient potentially disrupts the basal physiological process, triggers stress, even causes death. The

osmotic gradient of all treatments presented a significant result (P<0.05) (Figure 3). Evans (2011) stated that the gap of ion and total osmotic concentration between the internal fish's body and the rearing media determines the diffusion and osmotic amount through the gill epithelium. On the other hand, the osmotic gradient of the 3 g/L of salinity level was low as a consequence of the approach to the isosmotic condition.

The physiological response can observe through the blood profile(Porchase *et al.,* 2009). The lowest blood glucose was resulted in the 3 g/L of salinity treatment (74.79 mg/dL) and differed significantly with the 0 g/L (97.85 mg/dL), 1 g/L (96.09 mg/dL), and 5 g/L (92.14 mg/dL) treatment (Figure 4). The other blood profile parameters of the 3 g/L of salinity treatment, hematocrit, and erythrocyte, were also the lowest. This particular condition requires an adequate amount of energy to form hemoglobin to attach oxygen. According to Royan *et al.* (2014), the blood glucose level increases at a certain level when exposed to a stress condition. A species live in a low oxygen concentration frequently experiences hematopoiesis, thus the erythrocyte level increased as an adaptation process to attach more oxygen (Ardi*et al.*, 2016).The low level of stress in the 3 g/L of salinity treatment was described by the blood glucose level and osmotic gradient. The highest body weight and the length was also explained that the 3 g/L of salinity level was optimum for snakeskin gourami seed rearing.

Dissolved oxygen (DO) is one of the crucial water quality parameters. The oxygen molecules are the electron acceptor in anaerobe metabolism, therefore an adequate concentration of oxygen supply is a must. The measured DO during the snakeskin gourami rearing was 2.65–4.35 mg/L. Moreover, the DO concentration will decline when the water temperature increases, vice versa. The DO concentration will decrease when the decomposition and respiration process of the aquatic species happened. It will be accompanied by the increase of free CO_2 and pH decrease. Froese & Pauly (2014) and Oliveira *et al.* (2012) reported several tolerable ranges for fish rearing, i.e. temperature26–30°C, pH 6–7, and dissolved oxygen 3.0–7.7 mg/L. Generally, the result of water quality measurement was the tolerable range of snakeskin gourami seed rearing.

Ammonia (NH_3) is inorganic water-soluble nitrogen. The ammonia sources are organic nitrogen decomposition (protein, urea, and feces) and inorganic nitrogen, which comes from the

organic decomposition by microbes or fungi. The increase of ammonia more than 0.3 mg/L will decrease the oxygen level and increase CO_2 in blood. The result of ammonia measurement was 0.0036–0.1987 mg/L. The ammonia concentration of more than 0.02 mg/L potentially causes long-term poisoning and damages the gill tissues (Ardi *et al.*, 2016).

CONCLUSION

The advance survival and growth of snakeskin gourami seed was obtained in the 3 g/L of salinity treatment. This particular treatment also showed the most stable blood profile, thus it was spared from the stress condition.

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