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The Effect of Salinity on the Growth and Albumin Content of Striped Snakehead (*Channa striata* Bloch, 1793)

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ABSTRACT The striped snakehead (*Channa striata* Bloch, 1793) is a freshwater fish which has long been popular as a food fish. Commonly found in freshwater bodies such as rivers, swamps and lakes, striped snakeheads have a high albumin protein content which can help to accelerate wound healing, facilitate the circulation of body fluids and maintain osmotic pressure in the body. Salinity is one of the limiting factors that can affect fish growth and survival. Changes in salinity could directly affect striped snakehead growth and metabolism. The purpose of this study was to determine the effect of different salinity treatments on the growth and albumin content of juvenile striped snakeheads. The research was conducted in November-December 2021 at the Water Quality and Aquatic Biology Laboratory, Faculty of Animal Husbandry and Fisheries, Tadulako University, Indonesia. The study applied 5 salinity treatments (2 ppt, 4 ppt, 6 ppt, 8 ppt and 10 ppt) with 8 replicates for 30 days. Net weight gain, specific growth rate (weight and length) and survival rate did not differ significantly between treatments (ANOVA, P>0.05). The Tukey post-hoc test indicated significantly higher growth under the 8 ppt salinity treatment (1.04±0,19 cm) with 100% survival rate. Albumin content increased linearly with salinity from 2 ppt (3.6-4.0%) to 10 ppt (4.9-5.3 %).

Keywords: Albumin content; *Channa striata*; growth; salinity

INTRODUCTION

The striped snakehead *Channa striata* is a freshwater fish found in much of Indonesia, especially in Sumatra, Java, Kalimantan and Sulawesi (MMAF, 2013). The global distribution of this fish covers many other Asian countries, including India, Myanmar, Bangladesh, Laos, Vietnam, Thailand, Cambodia and Malaysia (Courtenay & Williams 2004; Muslim & Syaifudin, 2012). In addition, striped snakeheads have been introduced to several regions including Madagascar, the Philippines, eastern Indonesia, New Caledonia, Fiji (Froese & Pauly, 2023) and Taiwan (Li *et al.*, 2016).

In addition to a high overall crude protein content, striped snakeheads are rich in albumin the percentage composition varies but can reach 6.22% (Jamal, 2022). Albumin is a protein which is effective in accelerating wound healing, facilitating the circulation of body fluids and maintaining the body's osmotic pressure (Fuadi *et al.*, 2017), while the mucus is also though to have would healing properties (Kwan & Ismail, 2018). Striped snakeheads are cultured full-cycle and reared from wild fry to adult stages to meet community protein needs and to supply the fisheries processing industry (Muslim, 2007[a,b]; Sakuro & Yulisman, 2016).

The high demand for striped snakeheads as a food fish and for pharmacuetical and medical purposes, especially as a source of albumin, not only means that this fish has a high economic value but also drives unsustainable levels of exploitation that can threaten striped snakehead populations. To meet this demand it is necessary to domesticate this fish and develop striped snakehead aquaculture, including in Central Sulawesi (Ndobe *et al.*, 2013; 2014). Several limiting factors are thought to influence the growth and survival of fish, one of which is salinity; therefore, changes in salinity levels will have a direct effect on the growth and metabolism of fish in general (Nordlie, 2009), and striped snakeheads in particular.

Research on the biometric and physiological responses of snakehead fry has shown that salinity has a significant effect (Purnamawati *et al.*, 2019). In their study, 3 ppt salinity gave the best results (survival rate 77%, specific growth rate 5.62% and albumin content 4.52 g/100 ml), followed by 6 ppt, 9 ppt and 0 ppt salinity. Other studies have also indicated that salinity can affect snakehead growth, survival (Prakoso *et al.*, 2018) and albumin levels (Fuadi *et al.*, 2017). It is likely that salinity treatments could further improve albumen levels and/or growth at specific stages of striped snakehead development. Furthermore, *Channa striata* exhibits considerable variation in genetic characters between populations (Rahim *et al.*, 2012; Song *et al.*, 2013; Tan *et al.*, 2015), which likely also affects phenotype, including salinitiy tolerance and albumin production. Therefore, there is a need for further research on the effect of differences in salinity on albumin levels and growth of striped snakeheads at different life stages, from different populations, and under different conditions.

The aim of this research was to determine the effect of different salinity treatments on the growth and albumin protein levels of snakehead fingerlings from Poso in Central Sulawesi cultivated in a controlled environment. The results will contribute to the body of knowledge on striped snakeheads and inform domestication of this species, thereby benefitting stakeholders involved in striped snakehead research, fishfarming, and utilization, in particular as a source of albumin.

MATERIALS AND METHODS

The experimental stage of the research (rearing snakehead fingerlings) was conducted from November to December 2021 in the Water Quality and Aquatic Biology Laboratory, Faculty of Animal Husbandry and Fisheries. Snakehead albumin levels were measured at the Chemistry Laboratory, Faculty of Mathematics and Natural Sciences. Both laboratories are in Tadulako University, Palu, Central Sulawesi.

Materials

Experimental fish

The experimental fish used in this study were striped snakehead (*Channa striata*) fingerlings (n=40) with an initial length of 7-8 cm. The fingerlings were obtained from fishfarmers in Poso regency, Central Sulawesi, Indonesia. The fingerlings were allocated at random between the treatments and the between-treatment differences in initial weight and length were not statistically significant at the 95% confidence level ($p > 0.05$).

Controlled rearing environment

This study used aquaria (n=40) measuring 30x30x30 cm^3 and equipped with aerations systems for rearing the striped snakehead fingerlings. Before use each aquarium was thoroughly cleaned with detergent and rinsed with freshwater and then dried. The aquaria were then filled with a mixture of seawater and freshwater to obtain the desired salinity for each treatment. The water was filtered before use to avoid contamination with dirt or pathogens. The salinity was monitored daily to ensure each experimental unit remained at the correct salinity level.

Methods

Acclimatization and stocking

The striped snakehead fingerlings were initially acclimatized in styrofoam tanks for 2 days. The fingerlings allocated to each treatment were acclimated through raising the salinity by 2 ppt per day until the correct salinity level was reached. One fish was placed in each aquarium, equivalent to a stocking density of 1 fish/5 L. Live feed (blue panchax *Aplocheilus panchax*, locally called *kepala timah*) was provided to satiation twice a day, in the morning and evening, throughout the 30-day experimental period.

Variables measured

The experimental fish were weighed (digital scales, precision 0.01g) and measured (total length, calipers, precision 0.1 mm) at the start of the experiment (day 0) and on days 7,14,21 and 28 of the experimental period. The date of any fingerling mortality was recorded. In addition to the monitoring to maintain salinity, water quality parameters monitored daily were temperature (°C), pH, and dissolved oxygen (DO, mg/L). Ammonia levels (mg/L) were measured at the start of the experimental period, on day 14 and on day 28.

Parameters calculated

Net and specific (daily) growth parameters were calculated to evaluate weight gain and increase in length. Survival rate was also calculated.

The net growth parameters were calculated using the following equations:

$$W = Wt - W0$$

where :
W = Net weight gain (g)
$WtWt$ = fingerling weight at the end of the experiment (g)
$W0W0$ = fingerling weight at the start of the experiment (g)

and

$$L = Lt - L0$$

where :
L = Net increase in length (mm)
$LtLt$ = length at the end of the experiment (mm)
$L0L0$ = length at the start of the experiment (mm)

The specific growth rate (SGR) parameters were calculated using the following formulas:

$$SGR_W = \frac{\mathrm{Ln(Wt)} - \mathrm{Ln(W0)}}{\Delta t} \times 100\ \%$$

where :
SGR_W = specific growth rate in terms of weight (%)
Ln(Wt) = natural logarithm of fingerling weight at the end of the experiment (g)
$Ln(W_{0)}$ = natural logarithm of fingerling weight at the start of the experiment (g)
$\Delta\Delta t$ = length of time (days) since the start of the experiment

and

$$SGR_L = SGR\ Panjang\ = \frac{Ln(Lend) - Ln(Lstart)}{\Delta t} \times 100$$

where :
SGR_L = specific growth rate in length (%)
Ln(*Lend*) = natural logarithm of fingerling length at the end of the experiment (cm)
Ln(*Lstart*) = natural logarithm of fingerling length at the start of the experiment (cm)
$\Delta t\Delta t$ = length of time (days) since the start of the experiment

Survival rate for each treatment was calculated using the following equation:

$$SR = \frac{Nt}{N0} \times 100\ \%$$

where :
SR = fingerling survival rate (%)
Nt = number of live fingerlings at the end of the experiment
N0 = number of fish at the start of the experiment

Albumin content

Albumin was extracted from two fingerlings in each treatment at random. The albumin content in the flesh of each fingerling was calculated using the following formula:

$$\% \text{ Albumin} = [(\text{mL extract} \times N \times P)/50] \times 6.25$$

where :
mL = extract obtained (mL);
N = quantity of nitrogen based on the standard curve (mg/ml);
P = dilution level of the albumin extract

Statistical data analysis

Statistical data analyses were conducted and graphics produced in Microsoft Excel 2010 and Minitab 16. The between treatment differences in the four growth parameters and the albumin content were evaluated using analysis of variance (ANOVA) at the 95% confidence level (α=0.05). Statistically significant differences were further analyzed using the Tukey post-hoc test. Linear regression of albumin content against salinity was implemented in Excel 2010. Survival rate and water quality data were analyzed descriptively.

RESULTS AND DISCUSSION

Striped snakehead fingerling weight gain

The mean net weight gain varied between treatments (Figure 1), however these differences were not statistically significant (p = 0.739 > 0.05). The high level of variation between individual fingerlings within each treatment is reflected in the high standard deviation.

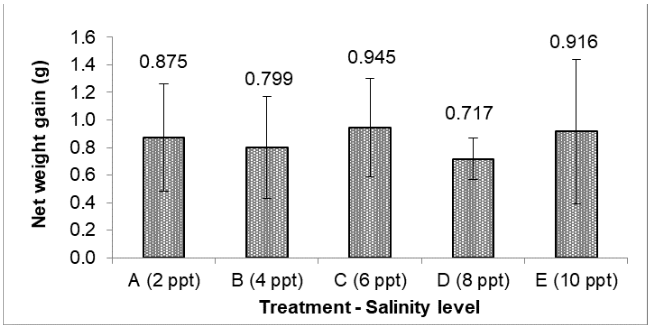

Figure 1. Net weight gain of striped snakehead fingerlings reared for 28 days under different salinity treatments (error bars indicate standard deviation).

The specific growth rate (SGR_W) in terms of weight showed a similar pattern to the net weight gain (Figure 2), in that growth rates varied more between individuals within treatments than between treatments. There was no statistically significant difference between the treatments (p = 0.731 > 0.05).

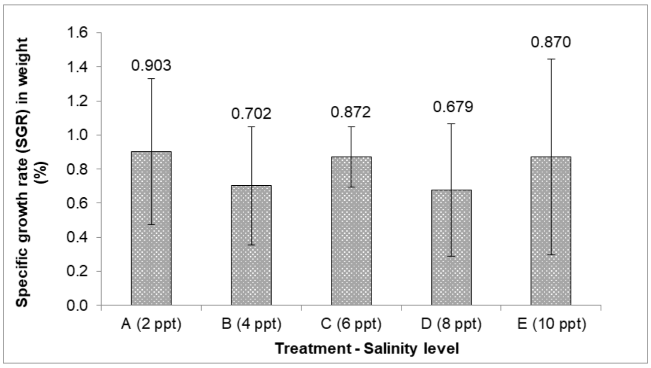

Figure 2. Specific growth rate (SGR) in weight of striped snakehead fingerlings reared for 28 days under different salinity treatments (error bars indicate standard deviation).

Treatment C gave the highest mean weight gain followed by treatment E which had the most variable weight gain. Treatment D gave the most consistent weight gain but with the lowest mean value. The SGR in weight was highest but very variable under treatment A and most consistent under treatment C.

Striped snakehead fingerling increase in length

The mean net growth in length differed significantly between treatments (p = 0.0015 < 0.05). Length increase was significantly higher under treatment D but did not differ significantly between the other four treatments (Figure 3).

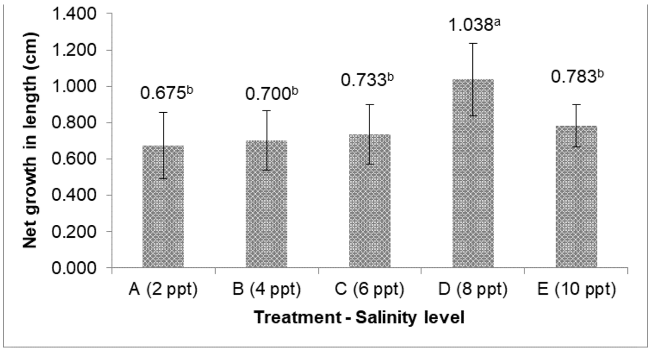

Figure 3. Net growth in length of striped snakehead fingerlings reared for 28 days under different salinity treatments (error bars indicate standard deviation; superscripts indicate significant differences at the 95% confidence level).

The mean specific growth rate in length (SGR_L) did not differ significantly between treatments (p = 0.286 > 0.05). The mean rate of increase in length was highest under treatment D, but the variation within treatments was greater than the variation between treatments (Figure 4).

Survival rate

The survival varied between treatments (Figure 5). However, the differences were not significant and did not show a trend with respect to salinity level (p = 0.519 > 0.05).

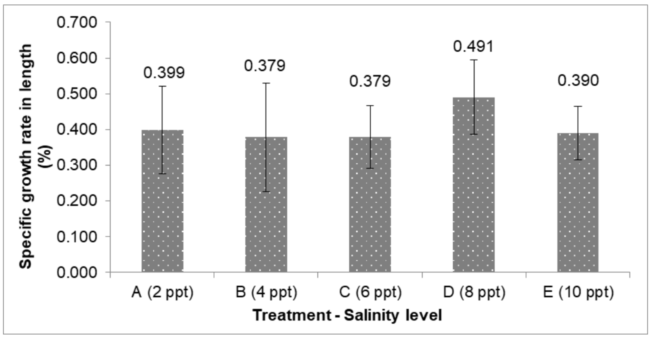

Figure 4. Specific growth rate (daily increase) in length of striped snakehead fingerlings reared for 28 days under different salinity treatments (error bars indicate standard deviation).

Albumin content

The albumin content varied significantly between treatments (p = 0.007 < 0.05). The linear regression of albumin content against salinity (Figure 6) shows the strong positive correlation ($R^2 \approx 0.86$; correlation factor = 92.7%) of albumin with salinity.

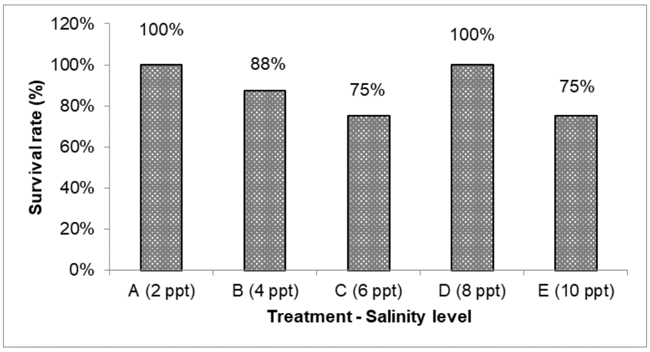

Figure 5. Survival rate of striped snakehead fingerlings reared for 28 days under different salinity treatments (n = 8 per treatment).

Water quality

The ranges of the water quality variables measured (Table 1) show slight differences between treatments. However, none of these differences was statistically significant (p > 0.05) between units or treatments. In all units the values remained within suitable limits for the husbandry of tropical freshwater or brackish water fishes with temperatures of 27.0-30.5˚C (mean 29.4˚C), pH levels of 7.1-8.6 (mean 7.47), dissolved oxygen concentrations of 6.0-8.4 mg/L (mean 7.1 mg/L), and ammonia concentrations of 0.05-0.2 mg/l (mean 0.117 mg/L).

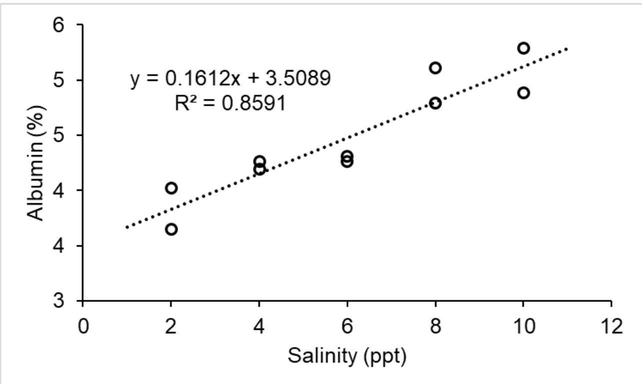

Figure 6. Linear regression of albumin content against salinity for striped snakehead fingerlings reared for 28 days under different salinity treatments.

DISCUSSION

Snakehead growth and survival

Combining the two measures of weight gain, no treatment can be considered superior to the others, as neither net weight gain nor specific growth rate (SGR_W) differed significantly between treatments. The increase in length was significantly higher under treatment D (8ppt), but the rate of increase in length (SGR_L) did not differ significantly between treatments. Survival rate also varied but the differences were also not statistically significant, with 100% survival at the lowest A, 2 ppt) and second highest (D, 8 ppt) salinity treatment levels. Raising salinity levels tends to increase the energy needed for osmoregulation in freshwater fishes, thereby reducing the proportion of energy obtained from feed that can be converted into growth (Prakoso *et al.*, 2018; Fauzi *et al.*, 2021). Looking at this from a different aspect, basic metabolism, in particular osmoregulation, should require more energy at salinity levels that differ from the salinity of the fluids in the fish body (Perry *et al.*, 2003; Evans *et al.*, 2005). Therefore, slower growth and increased mortality could be expected at salinity levels higher than the optimal range for a given species. Prakoso *et al.* (2018) report a decline in striped snakehead fry growth rates at 10 ppt salinity compared to 5 ppt salinity, This indicates that 10 ppt could represent an upper limit to the optimal range, although in this study the difference in fingerling growth rates from 0-10 ppt was not significant, with the exception of a higher increase in length (but not in weight) at 8 ppt.

There could be several reasons for the results, which demonstrate that juvenile freshwater striped snakehead fingerlings can tolerate a relatively wide range of salinity (0-10 ppt). Salinity tolerance to 10 ppt has been reported previously in striped snakehead fry (Prakoso *et al.*, 2018). The evolutionary advantages are clear, as such tolerance would facilitate migration between watersheds and colonization of new river basins, in addition to the well-known ability of striped snakeheads to travel overland in moist conditions (Li *et al.*, 2013). Furthermore, striped snakeheads belong to the Anabantidae and are capable of partial air breathing (Lefevre *et al.*, 2016). They have a high capacity to adapt to and survive in a wide range of environments, often with poor water quality such as calm or even stagnant waters with low oxygen levels (Li *et al.*, 2013). Other factors that can affect striped snakehead survival include high stocking densities, that can lead to competition for feed, space and oxygen, and can result in aggression including cannibalism (Ndobe *et al.*, 2013; 2014; Purnamawati, 2017).

In this case, the fingerlings were reared in separate aquaria, so intra-specific interactions were not a factor. One explanation could be related to feed, as the fingerlings in this study were fed *ad libitum* and feed consumption was not monitored. The fish that grew faster may

Table 1. Water quality parameter ranges in the striped snakehead fingerlings rearing units over 28 days of salinity treatments.

No.	Water quality parameter (unit)	Treatment (Salinity level)				
		A 2 ppt	B 4 ppt	C 6 ppt	D 8 ppt	E 10 ppt
1	Temperature (°C)	27.0-30.5	27.0-30.5	27.0-30.5	27.0-30.5	27.0-30.5
2	Alkalinity (pH)	7.2-7.7	7.1-8.5	7.2-7.9	7.2-8.5	7.1-8.6
3	Dissolved oxygen (mg/L)	6.1-8.4	6.0-7.6	6.0-7.6	6.0-8.4	6.0-7.7
4	Ammonia (mg/L)	0.05-0.2	0.05-0.2	0.05-0.1	0.05-0.2	0.05-0.2

have consumed more feed in order to compensate for any additional energy needs associated with metabolic challenges such as maintaining osmotic pressure under less than ideal conditions. Another explanation could be the life stage. Striped snakehead fingerlings are reported to grow quite slowly initially until they reach an inflection point where growth speeds up, as reported by Ndobe *et al*. (2013), where over 30 days growth (length and weight) was slow and highly variable within a small range, as observed in this study, but by 70 days of husbandry growth was much faster and also differed more between treatments. Once past the initial stage, they can quickly grow from around 8 to 12 cm (MMAF, 2014).

In this study growth was very variable between individuals. Such variation has been reported in other studies on striped snakehead culture (Ndobe *et al*., 2013; 2014; 2017; Serdiati *et al*., 2023), and could be simply due to genotypic and phenotypic variation between individuals. However, in this case the fingerlings had all been sourced from a freshwater facility (0 ppt salinity), and the adaptation period given to adjust to the various levels of salinity was quite short. It is possible that there may have been considerable individual variation in the rapidity and effectiveness of adaptation. In view of the relation between energy in (feed) and expended (activity, metabolism/maintenance and growth), activity levels may also have played a role, as this parameter was not measured. Fish expending less activity for any reason could have expended more energy on growth (Anggoro *et al*., 2013).

The ideal salinity for striped snakeheads in terms of minimum energy expenditure on osmotic regulation is not known. However, the results of this study indicate that salinity of 8ppt could be suitable or even within the optimal range. Additionally, metabolism and osmotic regulation are affected by fatty acids and other metabolites that can be influenced by albumin (Kovyrshina & Rudneva, 2012; Nurfaidah *et al*., 2021). Therefore, albumin levels could be causally related to growth through mechanisms related to energy balance and metabolic functioning.

Albumin content

Albumin functions to maintain blood osmotic pressure and helps transport metabolites while filtering body fluids (Nurfaidah *et al*., 2021), and prevents the accumulation of excessive liquid in the body (Suprayitno, 2014). Excreting excess salt can be energy intensive, and require metabolites, which could trigger the production of extra albumin (Purnamawati, 2017). Albumin is one of the proteins found in blood plasma that is synthesized in the liver (Infusino & Panteghini, 2013). Striped snakeheads from brackish water habitats have been found to contain higher levels of albumin compared to those in freshwater (Fuadi *et al*., 2017). Higher salinity means the fish are in a hypertonic state. When cells are in ideal conditions, all physiological processes will function normally. One way to maintain this condition under stress would be to reduce activity, so that more energy can be channeled to basic functions, while another is to increase the production of albumin because albumin can help regulate osmotic pressure and other bodily functions. The strongly correlated increase in albumin content with salinity in this study indicates a metabolic response by the fingerlings to their environment resulting in increased albumin production.

Water quality

The water quality monitoring verified that husbandry conditions were suitable for striped snakehead fingerlings throughout the experiment. Furthermore, conditions were very similar in all experimental units, despite some difference in the range or timing of fluctuations. Therefore, water quality should not have influenced the results in terms of between-treatment outcomes.

Fish are poikilothermic or cold-blooded animals meaning that their body temperature is determined by the temperature of their environment. Thermal shock due to rapid temperature changes or temperatures outside their thermal niche can cause fish to become weak, lose weight, and act abnormally, and can even be fatal (Saifuddin, 2020). The temperature remained at 27.0-30.5°C throughout this study, well within the temperature range 26-32°C tolerated by striped snakeheads according to Courtenay & Williams (2004).

While many fish have a relatively narrow pH tolerance, the tolerance limits of most fish lie within pH 6.5 to 9 (Boyd & Lichkopper, 1991). Striped snakeheads can tolerate pH as low as 4.25 and as high as 9.4 (Saifuddin, 2020), although much lower or higher pH values tend to be fatal. During this research, the pH remained in the range 7.1-8.6 and therefore in the range suitable for striped snakeheads.

In general, dissolved oxygen levels less than 3 mg/L will have negative effects on aquatic biota, ranging from loss of appetite and poor feed digestion to dysfunction of physiological processes such as osmoregulation and metabolism, leading to reduced growth and even mortality (Alamsyah & Fujaya, 2014). Although striped snakeheads are anabantids that can obtain some oxygen from the air and are relatively resistant to low oxygen levels (Li *et al.*, 2013; Lefevre *et al.*, 2016), during this research the dissolved oxygen levels remained above 6 mg/L due to the aeration and low stocking level.

For most fish, ammonia levels should remain below 0.1 mg/L, as higher levels can affect kidney function, slow growth, and make fish more susceptible to disease (Boyd & Lichkopper, 1991). Ammonia levels during this research ranged from 0.05 to 0.2 mg/L, quite high for many fish. However, striped snakeheads have a high tolerance to many pollutants including nitrogenous compounds, and can tolerate levels of ammonia in excess of 0.2 mg/L (Meidiana *et al.*, 2022).

CONCLUSIONS AND RECOMMENDATIONS

Conclusion

This 30-day study on striped snakehead fingerlings from Poso in Central Sulawesi reared at salinities from 2 ppt to 10 ppt found no significant between treatment difference ($p>0.05$) in net weight gain, specific growth rates (weight and length) or survival rate (75-100%). Although net increase in length was significantly higher at 8ppt, with a survival rate of 100%, individual variation in growth was high in all treatments and the results indicate that striped snakehead fingerlings can be reared at all salinities within the tested range with minimal effects on growth and survival. However, the salinity treatment had a significant effect on albumin content, with a strong positive linear correlation, increasing from 3.6-4.0% at 2ppt to 4.9-5.3 % at 10ppt.

Recommendation

Rearing striped snakehead (*Channa striata*) in salinity levels up to 8-10 ppt for a month increased albumin content at the end of the period with apparently little if any ill effects on growth and survival. However, it is not known whether these fingerlings would retain this trait as they grow or lose it, or whether this will depend on conditions during the ongoing grow-out, or whether other populations would exhibit the same response. At larger (commercial) scales, for example in coastal ponds, salinity is likely to vary. Therefore, we recommend further experimental (laboratory) scale research on the benefits of rearing striped snakeheads from various populations through more advanced life stages to harvest size with different permanent or temporary salinity treatments. We also recommend trials with rigorous monitoring and control on striped snakehead grow-out in natural and controlled brackishwater water bodies.

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AUTHORS' CONTRIBUTIONS

SH: Conceived and designed the analysis; Collected the data; Contributed data or analysis tools; Performed the analysis; Wrote the paper. SN: Conceived and designed the analysis; Collected the data; Contributed data or analysis tools; Performed the analysis; Wrote the paper; corresponding author. SFM: Conceived and designed the analysis; Collected the data; Contributed data or analysis tools. AM: Collected the data; Performed the analysis, Contributed data or analysis tools. AHR: Conceived and designed the analysis; Contributed data or analysis tools. MM: Conceived and designed the analysis; Contributed data or analysis tools

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