

Optimum Salinity for Growth of Tropical Eel *Anguilla marmorata* Quoy & Gaimard, 1824 in Nursery Culture

ROWENA CADIZ*, REX FERDINAND TRAIFALGAR Institute of Aquaculture, College of Fisheries and Ocean Sciences, University of the Philippines Visayas, Miagao, Iloilo 5023, Philippines

*E-mail: recadiz@up.edu.ph | Received: 11/06/2020; Accepted: 27/11/2020

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Abstract

Due to the reduced supply of commercially important temperate eels, tropical species including *Anguilla marmorata* Quoy & Gaimard, 1824 are being targeted as an alternative to satisfy the high demand for eel products. However, optimum ecological conditions for its aquaculture remained understudied. The present study evaluated the salinity conditions optimum for the growth of *A. marmorata* in the nursery culture. Growth performances of elvers were assessed after 90 days of culture at 0, 10, 15, 20 and 25 ppt salinity. Results showed that survival and feed conversion ratio were not influenced by the culture salinity. Significant improvement in per cent weight gain, specific growth rate and daily length increase were observed at 15 and 20 ppt. Polynomial quadratic regression analysis indicates that salinity of 18.14 ppt is optimum for the growth of this eel in nursery culture. These findings can serve as a basis for developing brackish water nursery culture of the tropical eel *A. marmorata*.

Keywords: tropical eel, nursery culture, brackish water, growth rate

Introduction

Anguillid eels are among the high-valued fish species that are consumed globally. The East-Asian countries are considered as major consumers and producers of this commodity (Shiraishi and Crook, 2015). Despite the technological advances in aquaculture, of which more than 90 % of eel produced is derived, reduction in the global production of cultured *Anguilla* species has been reported (Monticini, 2014). While other aquaculture species have established hatchery and nursery technology, eel aquaculture remains reliant on wild caught *Anguilla* species juveniles.

The European eel Anguilla anguilla (Linnaeus, 1758) and the Japanese eel Anguilla japonica Temminck & Schlegel, 1846 are the dominant species used in industrial eel aquaculture (Crook, 2014). However, dramatic declines in the glass eel recruitment of these temperate species have raised concerns which led to the implementation of new catch and trade measures to limit their exploitation (Harrison et al., 2014). Due to the limited supply of these temperate anguillid eels, interests are currently focused on the utilisation of tropical species as an alternative (Muthmainnah et al., 2016). However, in contrast to the temperate eel species that information on their biology and culture are well-established, there is a dearth of information available on the biology of tropical eels despite comprising two-thirds of all the anguillid species (Arai, 2016).

The tropical eel Anguilla marmorata Quoy & Gaimard, 1824 has the widest geographic distribution among the freshwater anguillids (Miller et al., 2002; Arai, 2016) and has been reported to be the dominant anguillid species present in the Philippines (Aoyama et al., 2015; Shirotori et al., 2016). As resources are available, the Philippines is promoting the development of eel farming in the country. However, suitable ecological conditions for the culture of tropical eels such as *A. marmorata* are still unknown (Lou et al., 2013).

The shortage of fresh water in tropical archipelagic countries, as well as the resource use competition for domestic consumption and other agriculture activities, have increased the pressure to develop aquaculture in brackish water rather than in freshwater. Anguillid eels are viewed as freshwater fishes and eel farming is a freshwater or nearly freshwater-based activity. However, salinitydependent variations in the growth have been documented in several species. Tzeng et al. (2003) reported that Japanese eels grow better in estuarine than in freshwater environment due to the greater abundance of trophic resources and similar osmolality of the environment and internal medium of young fish which results in a lower cost of osmoregulation. Longfinned Anguilla dieffenbachii Gray, 1842 grows best in 0 ppt while the short finned Anguilla australis Richardson, 1841 has better growth in 17.5 ppt (Kearney et al., 2008). Lamson et al. (2009) reported that American eel Anguilla rostrata (Lesueur, 1817) grew 2.2 times longer and 5.3 times heavier in saltwater compared to freshwater.

Optimal salinity for growth and metabolic rates of fishes may be influenced by factors such as species and developmental stage (Morgan and Iwama, 1991). Identification of the optimum salinity for the growth and survival of a particular eel species and stage will result in efficient utilisation of resource requirements, particularly the seed stocks from the wild and freshwater resource. Little is known about the effect of salinity on the growth of the elvers of *A. marmorata*. Hence, the present study was aimed to evaluate the effect of salinity on the growth performance of *A. marmorata* elvers and determine the optimum salinity for the nursery culture of this tropical anguillid.

Materials and Methods

Experimental animals

Glass eels were purchased from a collector in Buayan River in General Santos, Philippines in November 2018 and transported in freshwater to the facilities of the Institute of Aquaculture-College of Fisheries and Ocean Sciences University of the Philippines Visayas. The glass eels were stocked in a 10-ton capacity concrete tank and acclimatised to the facility conditions. The eels were fed to satiation two times a day with artemia and minced fish (Decapterus macrosoma Bleeker, 1851) for a month before gradually weaned to a dough-type diet (47 % crude protein) composed of minced fish (70 %); alpha potato starch (20 %); yeast (3 %); vitamin/mineral mix (2 %) and attractant (5 %) (FAO, 1987). Following the acclimatisation and weaning, the glass eels has transformed into elvers and A. marmorata were sorted out from the stock according to the classification described by Leander et al. (2012). All applicable international, national, and/or institutional guidelines for the care and use of animals were strictly followed by the authors during the conduct of the experiment.

Effects of salinity on the growth parameters of Anguilla marmorata

Groups of 20 A. marmorata elvers, were randomly distributed into 50 \times 40 \times 30 cm (length \times width \times height) 20 experimental tanks containing 20 L aerated static freshwater. Five salinity treatments were tested (0 ppt, 10 ppt, 15 ppt, 20 ppt and 25 ppt) with each treatment replicated four times in a complete randomised design (CRD). After stocking, salinity was gradually increased by 5 ppt.day⁻¹ until experimental conditions were achieved. When all treatments reached the desired salinity condition, eels were immobilised through hypothermic anaesthesia for the measurement of initial weight $(0.60 \pm 0.06 \text{ g})$ and total length $(6.56 \pm 0.23 \text{ cm})$. This method was described by Iwama and Ackerman (1994). During sampling, all animals in every replicate of each treatment were collected for the measurement of weight and total length. There were 20 animals per tank. Initial feeding rate was at 15 % of biomass per day given early in the morning and late in the afternoon. Excess feeds were collected an hour after provided for the correction of feed intake. Static water was replaced every 2 days while the faecal matter was siphoned out daily. Total ammonia nitrogen (0.05-0.07 mg.L⁻¹), nitrite (<0.01 mg.L⁻¹), dissolved oxygen (6-7 mg.L⁻¹), pH (7.1-8.4), and temperature (29-30 °C) were monitored regularly. Experimental tanks were installed in a shaded room subjected to natural photoperiod. Bulk weight of the animals was obtained every 15 days for the adjustment of feeding rate. The experiment was terminated at 90 days and the following growth parameters were calculated:

Survival rate (%) =
$$\frac{Number of fish survived}{Number of fish stocked} \times 100$$

Weight gain (%) = $\frac{Final weight - Initial weight}{Initial weight} \times 100$

Specific growth rate (%)

 $=\frac{\ln Final \ weight - \ln Initial \ weight}{Days} \times 100$

$$Daily \ length \ increase \ = \frac{Final \ length \ - \ Initial \ length}{Days}$$

$$Feed \ conversion \ ratio = \frac{Total \ feed \ intake}{Weight \ gain}$$

The data was processed as follows; all animals in every replicate tank of each treatment were evaluated individually for the measurement of weight and length. Then the data obtained for each replicate were processed to generate the treatment means, n = 4. A similar analysis was employed in all treatments.

Statistical analysis

All means generated for the treatments were compared and analysed using one-way analysis of variance (ANOVA) and Tukey's post hoc test using Statistical Package for the Social Sciences (SPSS) software (P < 0.05). Results are presented as mean \pm SEM (standard error of the mean) with n = 4 experimental tanks. Data on weight gain and salinity were subjected to polynomial quadratic regression analysis to determine the optimum salinity for maximum growth.

Results

Mortality was not recorded during the acclimation of elvers to the experimental salinities. Following the 90-day growth trial, survival was not affected by variations in culture salinity (Fig. 1). Mean survival rates percent were at 96.67 ± 3.33 , 95.00 ± 3.35 , 93.75 ± 3.10 , 76.67 ± 9.27 and 88.75 ± 2.39 for 0, 10, 15, 20 and 25 ppt respectively.



Fig. 1. Survival of Anguilla marmorata elvers reared at different salinities for 90 days. Data are mean \pm SEM (n = 4 tanks). Values are not significantly different (P > 0.05).

Significantly higher per cent weight gain was recorded in elvers reared at higher salinities than in freshwater (Fig. 2). Weight of elvers at 0, 10, 15, 20 and 25 ppt increased by $36.84 \pm 1.72 \%$, $48.31 \pm 0.69 \%$, $58.14 \pm 2.10 \%$, $57.52 \pm 1.47 \%$ and $51.83 \pm 4.99 \%$ respectively. Specific growth rate (SGR) follows similar trend with that of weight gain. Calculated SGR (% body weight.day⁻¹) for 0 ppt was 0.35 ± 0.010 , 0.44 ± 0.005 for 10 ppt, 0.51 ± 0.014 for 15 ppt, 0.50 ± 0.012 for 20 ppt and 0.46 ± 0.020 for 25 ppt (Fig. 3). Fish held at 15 ppt (0.023 ± 0.0004 cm.day⁻¹) and 20 ppt (0.023 ± 0.0010 cm.day⁻¹) showed significantly higher length increase when compared with that at 0 ppt (0.018 ± 0.0020 cm.day⁻¹) only (Fig. 4).

Feed conversion ratio (FCR) did not vary significantly among treatments (Fig. 5). FCR for 0 ppt is 5.37 ± 0.23 , 5.54 ± 0.57 for 10 ppt, 4.42 ± 0.23 for 15 ppt, 4.37 ± 0.20 for 20 ppt and 4.35 ± 0.56 for 25 ppt.



Salinity (ppt)

Fig. 2. Weight gain of Anguilla marmorata elvers reared at different salinities for 90 days. Data are mean \pm SEM (n = 4 tanks). Values with different labels are significantly different (P < 0.05).



Fig. 3. Specific growth rate (SGR) of Anguilla marmorata elvers reared at different salinities for 90 days. Data are mean \pm SEM (n = 4 tanks). Values with different labels are significantly different (P < 0.05).



Fig. 4. Length increase of Anguilla marmorata elvers reared at different salinities for 90 days. Data are mean \pm SEM (n = 4 tanks). Values with different labels are significantly different (P < 0.05).



Fig. 5. Feed conversion ratio (FCR) of Anguilla marmorata elvers reared at different salinities for 90 days. Data are mean \pm SEM (n = 4 tanks). Values are not significantly different (P > 0.05).

Optimum salinity for maximum growth was estimated using polynomial quadratic regression analysis and was determined at 18.14 ppt (Fig. 6).



Fig. 6. Polynomial quadratic regression analysis of weight gain of *Anguilla marmorata* elvers at different salinities. Estimated optimum salinity for weight gain was determined at 18.14 ppt.

Discussion

The results of the present study prove that A. marmorata elvers can be reared in a wide range of salinity conditions without significantly affecting survival. Similar findings were also reported for glass eels of shortfin, A. australis and longfin A. dieffenbachii (Kearney et al., 2008), A. anguilla (Edeline et al., 2005) and Anguilla bicolor bicolor McClelland, 1844 (Lukas et al., 2017; Taqwa et al., 2018). The glass eel stage is known to reside in estuaries where variable and dynamic chemical conditions, especially salinity is very common. This ability to inhabit environment with varying salinity is a common characteristic of both tropical and temperate eel species. Further, it has been suggested that the migration of A. marmorata to freshwater environment is not obligatory but rather a manifestation of opportunistic behaviour to maximise resource utilisation for survival, growth and reproduction (Arai and Chino, 2018).

Growth rate is an important factor in aquaculture as it determines the period required by the fish to reach market size. In the present study, growth of elvers was found to be influenced by the salinity of the culture environment. Anguilla marmorata elvers were observed to grow faster at higher salinities than in pure freshwater with optimum salinity determined at 18.14 ppt. Similar findings were also documented in other laboratory and field studies on different anguillid species. New Zealand short finned A. australis has better growth in 17.5 ppt than in 0 and 35 ppt as reported by Kearney et al. (2008). According to Côté et al. (2009) A. rostrata glass eels collected from two different sources grew significantly faster in brackish water than in freshwater. Anguilla bicolor bicolor glass eels starved for 14 days at different salinities exhibited lower decline in biomass at 10 ppt with optimum salinity range estimated to be 5.00 -13.40 ppt (Lukas et al., 2017). The present result is the first time to provide evidence that the brackish water environment favours the optimum growth of juvenile A. marmorata. The present results conform to the findings of Arai et al. (2013), showing that some populations of A. marmorata are purely estuarine residents and do not migrate upstream in freshwater habitats. Further, it has been hypothesised that the efficient growth in brackish water salinities is a conserved trait that reflects the marine evolutionary origin of these freshwater eel species (Tesch, 1977). Similarly, better growth in intermediate salinity conditions was also exhibited by other euryhaline fishes such as pompano, Trachinotus blochii (Lacepède, 1801) (Hamed et al., 2016); fat snook, Centropomus parallelus Poey, 1860 (Tsuzuki et al., 2007); turbot, Scophthalmus maximus (Linnaeus, 1758) (Imsland et al., 2001) and most tilapia species except for red tilapia (Boeuf and Payan, 2001). The SGR of A. marmorata elvers obtained in the present study were similar to those documented for other long fin anguillid species reared in laboratory conditions (Rodríguez et al., 2005; Kearney et al., 2008).

The effects of salinity in the growth of fishes, including eels are often attributed to several physiological factors such as metabolic cost of osmoregulation, feed intake and conversion and growth hormone (GH) stimulation. Osmoregulation or the process of maintaining salt and water homeostasis in the fish body involve energy demanding synthesis and operation of various enzymes and transport related proteins (Tseng and Hwang, 2008). According to Boeuf and Payan (2001), osmoregulation appears to utilise 20-50 % of animal's total energetic expenditure. Previous studies the utilisation proteins, demonstrated of carbohydrates and lipids as energy suppliers during fish acclimation to different salinities (Rocha et al., 2005; Gracia-López et al., 2006; Rocha et al., 2007). However; in a condition wherein gradients between blood and water are minimal (isosmotic), energetic cost of osmoregulation will be lower resulting in more energy spent for growth (Boeuf and Payan, 2001). According to Tzeng et al. (2003), the isosmotic point of eel's body fluid corresponds to water salinity of 10.5 to 14 ppt which is close to that of brackish water. This suggests that the cost of osmoregulation will be lower if eels are held in brackish water than in fresh or seawater. In the wild, better growth in brackish water environment has also been reported for anguillid eels. from the lower energetic Aside cost of osmoregulation, the higher habitat productivity in brackish water is also a factor associated with enhanced eel growth in this environment (Tzeng et al., 2003; Jessop et al., 2004).

Growth rate of fish species at different environmental condition has also been reported to be controlled by feed intake. The higher growth of *A. anguilla* glass eels in brackishwater conditions compared to freshwater

conditions regardless of previous salinity preference have been attributed to the greater appetite and feeding activity (Edeline and Elie, 2004; Edeline et al., 2005). This higher feeding activity in the higher salinity treatment has been linked with the increased production of growth hormone (GH). Osmoregulation of fishes in seawater is known to be regulated by GH (Boeuf and Payan, 2001; Sakamoto and McCormick, 2006). Also, cannibalism due to enhanced feeding behaviour contributed to the suppression of slow growers and increased apparent growth rates in seawater reared glass eels (Edeline and Elie, 2004). In the present study, survival was similar among the treatments and the observed enhancement of growth could be attributed to better feed utilisation as influenced by the culture salinity.

Conclusion

The present study proves that *A. marmorata* elvers can be reared in environmental salinity of 0 to 25 ppt without significantly affecting survival however; optimum salinity for growth is 18.14 ppt. These findings could serve as a basis for establishing brackish water nursery culture of this tropical anguillid which will be beneficial in speeding up production cycle and promoting efficient utilisation of limited resources.

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