#### Research Article

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# Effects of domestication and temperature on the growth and survival of the giant freshwater prawn (*Macrobrachium rosenbergii*) postlarvae

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**Abstract:** To understand the growth and survival during the postlarval stage of *Macrobrachium rosenbergii*, commonly known as the giant freshwater prawn, two experiments were performed which explored the differences in growth and survival rates between wild and captive postlarvae and the effect of temperature on survival and growth. The postlarvae reared at 27 and 30°C grew significantly throughout the experimental periods. The survival rates in the wild and captive postlarvae were similar at approximately 60%. The highest survival rate of 58% was found at 27°C.

**Keywords:** Decapoda, growth, survival, temperature, tropical waters

### 1 Introduction

The giant freshwater prawn *Macrobrachium rosenbergii* population is distributed in tropical and subtropical areas throughout the Indo-Pacific region [1,2]. Among approximately 200 species of the genus *Macrobrachium*, the most important culturable species is *M. rosenbergii*. *Macrobrachium rosenbergii* is an amphidromous species living in various water environments, such as fresh and estuarine waters connected to the sea [3]. Matured females migrate from freshwater environments to estuarine environments for spawning. Larval development occurs in estuaries

and moves to freshwater, and the adults spend their lives in freshwater [1].

In Brunei Darussalam, giant freshwater prawns can be found widely in rivers, streams, estuaries, and mangrove areas. It is locally known as "Udang Galah" and is a popular delicacy of the country. The market price of the prawn is approximately 10-15 USD per kilogram, depending on its body size. Hatchery production of M. rosenbergii in Brunei Darussalam began in 1982 [4], though the current hatchery and production rate for the species are not well developed. Currently, the species is captured in the wild and sold in local markets due to the unstable culturing production of the species in Brunei Darussalam. Therefore, it was found that the prawn landing has declined drastically from 600 to 70 kg per day [5]. This decline is detrimental because the country continues to obtain seeds from the wild, especially berried females, which could ultimately lead to a decline in the wild population [6,7]. Therefore, it is important to examine baseline information such as growth and survival in aquaculture and the enhancement of M. rosenbergii in Brunei Darussalam.

Environmental factors such as temperature and salinity play important roles in the development and growth of decapod crustaceans [8-14]. Macrobrachium rosenbergii can tolerate a wide range of temperatures (14–35°C) and salinities (0–25 ppt) [13–15]. The geographical distribution, acclimatization of ectothermic organisms, and range of thermal tolerance are influenced by temperature [11–14,16,17]. Ectotherms have a limited range of thermal tolerance in tropical regions because of limited seasonal fluctuation, causing them to be vulnerable to global warming circumstances [17]. High temperatures can impact egg development in M. americanum [18] and growth and survival in M. amazonicum [11]. In M. rosenbergii, the survival, growth, and molting are influenced by salinity, temperature, and pH [9,13]. The higher temperatures result in an increased larval heartbeat, while larval activity decreased at higher temperatures and salinities in M. rosenbergii [14]. However, there is still limited information available on the

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effect of temperature on the survival and growth of M. rosenbergii during the postlarval stage [19,20]. Furthermore, little information is also available on differences in growth and survival between wild and captive M. rosenbergii.

In the present study, two independent experiments were performed to understand the growth and survival of the giant freshwater prawn M. rosenbergii; the first experiment examined differences in growth and survival rates between wild and captive postlarvae, and the second experiment determined the effect of temperature on survival and growth under laboratory conditions. These experiments provide a baseline data for the aquaculture of the giant freshwater prawn M. rosenbergii and other related species.

#### 2 Materials and methods

#### 2.1 Experimental specimens and acclimation

To examine differences in growth and survival rates between captive and wild postlarvae, freshwater giant prawns were collected from the Tutong River (4°48'16"N, 114°39'40"E) and from a local hatchery in Brunei Darussalam (4°22′05"N, 114°27′40"E). Two types of postlarvae were collected: (1) second-generation captive postlarvae and (2) wild postlarvae. A total of 200 second-generation captive postlarvae were collected on December 5, 2018. A total of 30 individuals of wild postlarvae were collected on December 4, 2018. All captive and wild postlarvae were transported using polyethylene plastic bags with aerated water and were placed in styrofoam boxes to prevent excess shaking during transport to the Universiti Brunei Darussalam (Gadong, Brunei Darussalam) for experiments and then transferred to temporary tanks  $(130 \text{ cm} \times 80 \text{ cm} \times 46 \text{ cm}, 270 \text{ L})$  for 11-12days at the ambient temperature of 27°C for acclimation. Postlarvae were initially fed with Artemia and gradually shifted to artificial pellets (Star Feed 5004-S, Charoen Pokphand Malaysia, containing 40% protein) twice daily (9:00 and 16:00 h).

Furthermore, to determine the effect of temperature on the survival and growth, a total of 200 captive postlarvae were compared with the first experiment with acclimation under the same conditions. Experiments were performed by subdividing postlarvae into three tanks during the experimental period.

## 2.2 Differences in growth and survival rates between wild and captive postlarvae

A total of 12 captive postlarvae and 14 wild postlarvae were transferred to the experimental tanks and reared at 27°C (±1°C) with 0 psu in salinity (freshwater) and were observed for 80 days (December 2018 to March 2019). The optimal feeding rate of all crustaceans, including postlarvae, is 10-20% of their biomass [1]. The feeding rate of the postlarvae was calculated as follows:

Food (g) = Number of individuals 
$$\times$$
 Average body weight (g)  $\times$  20%. (1)

The feeding rate was further split into 40 and 60% for the morning (9:00) and afternoon (16:00) routines, respectively. During the observation period, the dissolved oxygen (DO) (YSI EcoSense DO200, Yellow Springs, USA), water temperature (YSI EcoSense DO200), and pH (Campbell Scientific ISFET pH Probe, Logan, USA) were measured twice daily.

The growth rates of all the individuals were examined by measuring their weight and length every 20 days. The body length (BL) of each specimen was measured from the tip of the rostrum to the telson to the nearest 0.01 mm. The body weight (BW) of each specimen was examined to the nearest 0.01 g. The weight gain (WG) and survival rate of the prawns were calculated by using the following formulae:

$$WG(g) = Wf(g) - Wi(g), \tag{2}$$

Survival rate (%) =

Number of specimens/Survived initial number of specimens × 100,

where Wf is the final wet weight and Wi is the initial wet weight.

## 2.3 Effect of temperature on survival and growth

Three temperature regimes, 27, 30, and 33°C ( $\pm$ 1°C), with 0 psu in salinity (freshwater) were chosen for the thermal tolerance experiment. The DO and pH values were  $6.41 \pm 0.73$  (mean  $\pm$  SD) mg L<sup>-1</sup> and  $6.66 \pm 0.95$ , respectively. The temperature in each tank was controlled using a thermostat. Each tank was aerated in every corner of the tank with a total of 6 aeration tubes. A color stabilizer (blue) was added to each tank to mimic the natural environment for postlarvae and to prevent stress and cannibalism. In each tank, 3 nylon nets ( $51 \, \text{cm} \times 44 \, \text{cm} \times 35 \, \text{cm}$ ) were placed to prevent predation and cannibalism among postlarvae. Nylon substrates were added to each net to increase the surface area and provide hiding spots during the molting period. Thirty-six postlarvae were assigned to each net, and 12 post-larvae were further separated into 3 nets ( $0.04 \, \text{individual cm}^{-3}$ ).

The BW and BL of each specimen were examined every 20 days for 80 days (December 2018 to March 2019). The average daily growth (ADG) (g  $d^{-1}$ ) and specific growth rate (SGR) (g  $d^{-1}$ ) were calculated by using the following formulae:

ADG = (Wf – Wi) Wi<sup>-1</sup> × 100, (4)  
SGR = [LnWf (g) – Ln Wi (g)]
$$t^{-1}$$
 × 100.

#### 2.4 Statistical analysis

Differences in BL and BW between wild and captive postlarvae for every 20 days were examined by means of the Student's *t*-test. Differences in BW between each measurement every 20 days were also examined by means of the Student's *t*-test. Analysis of covariance (ANCOVA) was used to compare two regression lines by testing the effect of a categorical factor on a dependent variable in the growth patterns of BL and BW between wild and captive post larvae and BW between 27 and 30°C.

**Ethical approval:** Our protocols followed the ethical guidelines for the use of animals of Universiti Brunei Darussalam (UBD) and were approved by the animal ethics committee at UBD.

#### 3 Results

# 3.1 Differences in survival rates and growth between wild and captive postlarvae

During the initial experimental period between 0 and 20 days, the survival rates of wild and captive postlarvae decreased from 100% (n = 14) to 64.3% (n = 9) and 100% (n = 12) to 58.3% (n = 7), respectively (Figure 1). Thereafter, the survival rates remained constant at 64.3% in wild and 58.3% in the captive postlarvae throughout the experiment, dropping slightly to 57.1% in the wild postlarvae at 80 days (Figure 1). There was no significant difference in survival rates between wild and captive prawns throughout the experiment (p > 0.05).

The initial and final BLs in wild prawns were 1.70  $\pm$  0.68 cm (mean  $\pm$  SD) and 3.35  $\pm$  0.68 cm, respectively, and the initial and final BLs in captive prawns were 1.72  $\pm$  0.83 and 3.81  $\pm$  0.83 cm, respectively. The initial and final

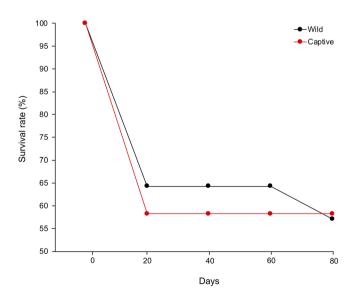


Figure 1: Survival rates in wild (red line) and captive (black line) postlarvae in the giant freshwater prawn Macrobrachium rosenbergii.

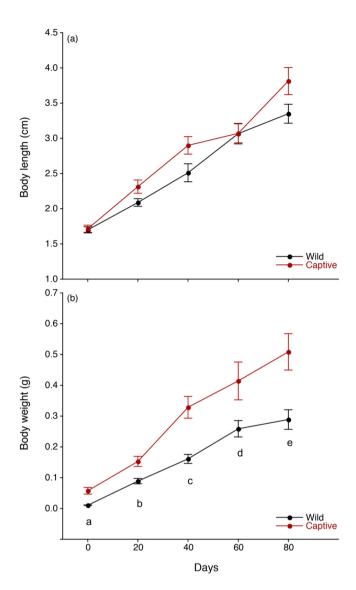


Figure 2: Growth patterns of body length (a) and body weight (b) in wild (black line) and captive (red line) postlarvae in the giant freshwater prawn *Macrobrachium rosenbergii*. The results are expressed as the mean  $\pm$  standard error (SE). The letters (a–e) indicate a statistically significant difference at p < 0.05 between wild and captive postlarvae.

BWs in wild and captive prawns were  $0.01 \pm 0.003$  g (mean  $\pm$  SD) and  $0.0575 \pm 0.192$  g and  $0.289 \pm 0.09$  g and  $0.509 \pm 0.157$  g, respectively. There were significant differences in BLs and BWs between the initial and final stages of the experiment (p < 0.0001). The BLs and BWs tend to increase throughout the experimental period (p < 0.0001) (Figure 2). There were no significant differences found in BLs between wild and captive postlarvae every 20 days (0, 20, 40, 60, 80 days) (p > 0.05) (Figure 2a). Significant differences were found in BW between wild and captive postlarvae every 20 days (p < 0.05-0.0001) (Figure 2b). No significant differences were found in regression slopes in BW and BL between wild and captive postlarvae (p > 0.05).

In wild prawns, significant increases in BW were found between 0 days (0.01  $\pm$  0.003 g; mean  $\pm$  SD) and 20 days (0.089  $\pm$  0.03 g), between 20 days and 40 days (0.161  $\pm$  0.04 g) and between 40 days and 60 days (0.259  $\pm$  0.08 g) (p < 0.01–0.0005), while there was no significant difference in BW between 60 days and 80 days (0.289  $\pm$  0.09 g) (p > 0.05). There were significant increases in BW between 0 days (0.0575  $\pm$  0.04 g; mean  $\pm$  SD) and 20 days (0.153  $\pm$  0.04 g) and between 20 days and 40 days (0.329  $\pm$  0.09 g) (p < 0.05–0.0005), while there were no significant differences in BW between 40 days and 60 days (0.414  $\pm$  0.16 g) or between 60 days and 80 days (0.509  $\pm$  0.16 g) (p > 0.05) in captive prawns. The WG based on the mean BW in wild and captive prawns

between 0 and 20 days, between 20 and 40 days, between 40 and 60 days, and between 60 and 80 days was 0.078 and 0.095 g, 0.072 and 0.176 g, 0.098 and 0.086 g, and 0.030 and 0.094 g, respectively. WG in captive prawns was higher than that in wild prawns for all periods except between 40 and 60 days.

# 3.2 Effect of temperature on survival and growth

The survival reduction was found during the initial experimental period between 0 and 20 days from 100% (n=36) to 63.9% (n=23) at 27°C and 100% (n=36) to 58.3% (n=21) at 30°C (Figure 3). All postlarvae died within 7 days during the initial 20 days at 33°C (Figure 3). There was no survival reduction found, and the survival rate was constant (58.3%) throughout the experiment after a slight reduction at 20 days (61.1%) at 27°C (Figure 3). In contrast, a survival reduction was found at 60 days (n=19, 52.8%) and the end of the experiment at 80 days (n=15, 41.7%) at 30°C (Figure 3).

The BWs at 27, 30, and 33°C at the beginning of the experiment were 0.16  $\pm$  0.13 (mean  $\pm$  SD) g, 0.13  $\pm$  0.10 and 0.13  $\pm$  0.11, respectively (Table 1). There were no significant differences in initial BW among the three temperature regimes (p > 0.05). Significant differences were found in BW between each experimental period (p < 0.05-0.0001),

**Table 1:** Production parameters of the giant freshwater prawn *Macrobrachium rosenbergii* reared in different temperature ranges for 80 days

-	Temperature				
	27°C	30°C	33°C		
Initial weight	0.16 ± 0.13	0.13±0.10	0.13 ± 0.11		
	(n = 36)	(n = 36)	(n = 36)		
Final weight	$2.14 \pm 1.67$	$2.37 \pm 1.68$	n.d		
	(n = 21)	(n = 15)	(n = 0)		
Weight gain	1.98	2.24	n.d		
Average daily growth	0.02	0.03	n.d		
Specific growth rate	3.24	3.63	n.d		

n – number of specimens; n.d – no data due to all specimens died within 7 days.

except between 40 days and 60 days (p>0.05) at 27°C (Table 2), suggesting successive growth throughout the experiment (Figure 4). There were significant differences in BW between each experimental period (p<0.01-0.0001), except between 60 days and 80 days (p>0.05) at 30°C (Table 3, Figure 4). The final BWs at 27 and 30°C were 2.14  $\pm$  1.67 and 2.37  $\pm$  1.68 g, respectively. There was no significant difference in the final BW between these two temperature regimes (p>0.05). WG, ADG, and SGR during 80 days at 27 and 30°C were 1.98 (g), 0.02 (g d<sup>-1</sup>), and 3.24 (g d<sup>-1</sup>) and 2.24 (g), 0.03 (g d<sup>-1</sup>), and 3.63 (g d<sup>-1</sup>), respectively (Table 1).

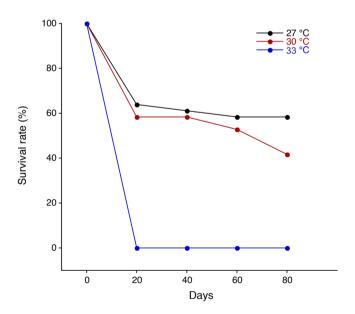


Figure 3: Survival rates of postlarvae in the giant freshwater prawn *Macrobrachium rosenbergii* reared in different temperature regimes of 27°C (black), 30°C (red), and 33°C (blue).

**Table 2:** Statistical results (*p*-value) of body weight in the freshwater giant prawn *Macrobrachium rosenbergii* between 20-day intervals reared at 27°C

	Day 0	Day 20	Day 40	Day 60	Day 80
Day 0	1	< 0.0001	<0.0001	< 0.0001	<0.0001
Day 20		1	< 0.0005	< 0.0005	< 0.0001
Day 40			1	0.388	< 0.05
Day 60				1	< 0.05
Day 80					1

**Table 3:** Statistical results (*p*-value) of body weight in the freshwater giant prawn *Macrobrachium rosenbergii* between 20-day intervals reared at 30°C

	Day 0	Day 20	Day 40	Day 60	Day 80
Day 0	1	<0.0001	<0.0001	<0.0001	<0.0001
Day 20		1	< 0.0005	< 0.0001	< 0.0001
Day 40			1	< 0.01	< 0.0005
Day 60				1	0.206
Day 80					1

#### 4 Discussion

# 4.1 Differences in growth and survival rates between wild and captive postlarvae

There was no difference between the survival rates of wild and captive postlarvae. The cultural environment can create significant selection pressures that can affect population growth and reproduction rates, collectively known as "fitness" [21]. The giant freshwater prawns are generally opportunistic omnivores that eat a wide range of food items efficiently [22]. Wild prawns feed on algae, molluscs, aquatic insects, worms, aquatic plants, and other crustaceans [23]. The cultivation of live feed to support aquaculture is costly and unpredictable, and hence, the usage of artificial feed has become popular,

particularly in shrimp culture [24]. *Macrobrachium rosen*bergii is a nonactive hunter and appears to capture food only by chance [25,26]. The captive postlarvae might be more acclimatized than the wild postlarvae. Environmental factors such as nutrition, social stress, or fear were found to play an important role in shaping the traits and phenotypes across several offspring generations; an effect is known as transgenerational epigenetic inheritance [27]. This suggests that the captive postlarvae would be more acclimatized than the wild postlarvae under experimental and cultural conditions. The captive postlarvae might also be acclimatized to artificial foods providing regular instead of predator-prey interactions in the wild. However, wild postlarvae might be undomesticated compared to captive larvae. Adult female prawns might experience less stressful conditions in the wild, and thus, transgenerational epigenetic information might

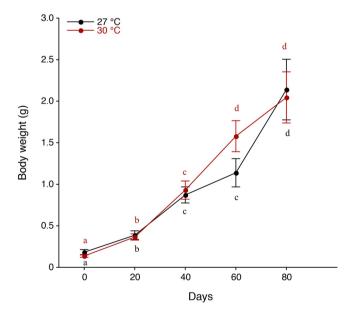


Figure 4: Growth patterns of body weight of postlarvae in the giant freshwater prawn *Macrobrachium rosenbergii* reared in different temperature regimes of 27°C (black) and 30°C (red). The results are expressed as the mean  $\pm$  standard error (SE). The letters a, b, c, and d indicate a statistically significant difference at p < 0.05 between each experimental period in different temperature regimes of 27°C (black) and 30°C (red).

not be transferred to their offspring [28]. Therefore, the wild postlarvae might need to adjust artificial pellets across generations. Moreover, the growth rate in captive prawns might be higher than that in wild postlarvae. The growth patterns were not significantly different between wild and captive postlarvae in BL and BW (Figure 2), and no significant differences were found in BLs between wild and captive postlarvae through the experimental periods (Figure 2a). However, the initial BW was different between wild and captive postlarvae, and significant differences were found in BW between wild and captive postlarvae through the experimental period (Figure 2b). These results suggest that wild postlarvae might be less nourished than captive postlarvae and that condition might be maintained throughout their growth period. Another explanation is that wild postlarvae might not be acclimatized to foodhunting behavior for artificial foods while maintaining opportunistic behavior in the wild. Furthermore, in relation to energy partitioning, the wild postlarvae adjusting to new captive environments might need to increase energy supply to acclimatize to new conditions from wild to experimental conditions. It is suggested that the wild prawn subsequently depletes the energy reserved for growth [29]. These results might suggest a reduction in growth rates, such as WG. Further studies would be required using wild and captive postlarvae from various environments in the wild and multiple hatcheries, respectively, to enable broader conclusions to be drawn.

Specific competitive capacities between wild and captive-bred animals can occur by means of several factors. In captive-bred animals, there are fewer competitors than in wild animals [30]. The high rearing densities and the absence of predatory stimuli in captive environments may lead to aggressive foraging behavior [30]. Density-dependent survival in response to food limitation has been observed in *Penaeus esculentus* and *P. setiferus* [31,32], although no study has examined competition in *M. rosenbergii* between wild and captive breeds. We conducted the experiment using similar numbers of wild and captive postlarvae at the same densities with sufficient food. Therefore, the survival rate might not differ between wild and captive postlarvae in the same experimental environment.

# 4.2 Effect of temperature on survival and growth

The giant freshwater prawn is susceptible to various factors, such as ambient temperature, stocking density, physiochemical parameters, transport, and storage

in aquaculture [13,14,33]. Temperature is an important abiotic factor that can affect the survival and growth of aquatic organisms. The regulation of temperature by the growth rate is the main factor in determining the economic performance of prawn farming [34]. *Macrobrachium rosenbergii* could acclimate to a wide range of temperatures, with an optimal temperature range between 26 and 31°C [13,14,35]. Successive growth was found at 27 and 30°C, while their growth rates did not differ significantly in the present study. This study suggests that 27 and 30°C would be the optimal temperatures in *M. rosenbergii*.

There was a sudden decrease in the survival rates at 27 and 30°C over 20 days. The reduction in survival rates occurred after prawns were stocked even when the conditions were ideal [1]. The sudden change in temperature and pH during the stocking period can cause an increase in mortality in prawns [1]. It was also noted that the prawns reared at 27°C had the highest survival rate of 58%, while those reared at 30°C had the second-highest survival rate of 41% throughout the experiment. Paul et al. [36] found a survival rate of 60% after initial stocking days and a survival rate of 55% after 90 days in M. rosenbergii reared at 28°C. The current results and that of the previous study were similar in the survival rate in M. rosenbergii, so a survival rate of 60 % might be common in M. rosenbergii under culturing conditions at the optimal temperature.

The lethal thermal tolerance of *M. rosenbergii* is less than 14°C and more than 35°C [37]. However, the post-larvae reared at 33°C did not survive within a week after the experiment was initiated in the present study. This result suggests that the temperature range was the lethal thermal tolerance for the postlarvae to live during the rearing experiment. The postlarvae reared at 33°C may represent a predeath thermal point in which the locomotory movements became impaired because of presynaptic failure, and postlarvae might not tolerate such a temperature regime [38,39]. In addition, the higher respiratory rate at increased temperature may also contribute to mortality.

#### 5 Conclusion

This study showed that a temperature range between 27 and 30°C is optimal for postlarvae-to-juvenile *M. rosenbergii* in terms of survival and growth rate. The prawns reared at 33°C did not survive throughout the experiment. This would suggest the lethal temperature for survival for postlarval to juvenile stages. Precautions of temperature

fluctuations are essential to monitor in culturing the prawn, especially in the ponds, as they may affect the growth and survival rates.

The captive prawns exhibited significantly higher WG than the wild prawns. Although the wild prawns also showed WG throughout the entire experiment, it was significantly lower than that of the captive prawns. This weight reduction would be due to its parental lack of acclimation to captive conditions by means of a lack of transgenerational epigenetic inheritance [27]. The wild prawns exerted a lower growth rate due to their energy partitioning, whereby the energy used for growth might be replaced by acclimation to captive conditions.

Domesticated animals, such as farmed aquaculture crustaceans, have a higher survivability rate [30], while we found that there was no significant difference in the survival rates between wild and captive prawns. Our domesticated or farmed prawns might have similar survival rates because each prawn was reared at a density similar to that of sufficient food in this experiment.

This study might provide baseline information for further experiments to improve the aquaculture of the giant freshwater prawn *M. rosenbergii* and other prawns.

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**Author contributions:** JT: data curation, formal analysis, and writing – original draft; AS: data curation, formal analysis, and writing – original draft; MA: data curation, formal analysis, and writing–original draft; NY: conceptualization and data curation; TA: supervision, conceptualization, writing – review and editing, and funding acquisition.

**Conflict of interest:** The authors state no conflict of interest.

**Data availability statement:** All data generated or analyzed during this study are included in this published article.

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