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The combined effects of temperature and salinity on survival of postlarvae tiger prawn *Penaeus monodon* under laboratory conditions

Nilnaj Chaitanawisuti[1*], Wannanee Santhaweesuk[1], Gullaya Wattayakorn[2]

[1]Aquatic Resources Research Institute, Chulalongkorn University, Bangkok, Thailand; [*]Corresponding Author: cnilnaj@chula.ac.th
[2]Department of Marine Science, Faculty of Science, Chulalongkorn University, Bangkok, Thailand

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ABSTRACT

A 3 x 4 two-factor factorial experimental design with three replications per treatment was conducted to determine the effects three temperatures viz., room temperature (29˚C ± 1˚C) and elevated temperatures (33˚C and 35˚C ± 0.5˚C) and four salinities (25, 30, 33 and 35 ppt) on survival of postlarvae tiger prawn *Penaeus monodon* under laboratory conditions. Results showed that temperature had a greater influence on survival of postlarvae *P. monodon* than salinity and low water temperature (29˚C) gave better larval survival than higher water temperatures (33˚C and 35˚C). In addition, the PL1 had better tolerance to high temperatures than PL15. At the termination of the experiment, the best survival of PL1 was found at 29˚C for all salinities tested with an average of 95.8%, while the best survival of PL15 was also found at 29˚C for all salinities tested with an average of 70.6%. Based on survival at temperatures and salinities tested, the best salinity and temperature combination for the culture of *P. monodon* PL1 and PL15 were 29˚C at 25, 30, 33 and 35 ppt, and 29˚C at 25 and 30 ppt, respectively.

Keywords: *Penaeus monodon*; Salinity; Temperature; Postlarvae; Survival

1. INTRODUCTION

The tiger prawn *Penaeus monodon* is the most important penaeid species for mass hatchery production and land-based aquaculture system in Thialand and various countries in Asia. Shrimp farming is potentially affected by adverse physical, chemical, and biological conditions [1,2]. Early life stages are the most sensitive phase in the complex life cycle of marine invertebrates and to maximise their survival larvae should be reared close to optimal conditions. Salinity and temperature are two of the most important abiotic factors affecting the growth and survival of aquatic organisms. Larval stages of most penaeid shrimp species occur in full strength seawater and at stable water temperatures. Hence, it is generally accepted that penaeid shrimps are not equipped with the capabilities of withstanding major environmental changes during their larval development. In addition, it is also well known that the response to these environmental parameters is species-specific and that salinity and temperature may also interact to influence growth and survival. It is generally agreed that temperature has a more pronounced effect on growth and survival of penaeids [3]. Many studies have reported on the effects of different environmental factors, in particular, salinity and temperature on development, survival and growth of various economical penaeid species [1,3-8]. Laboratory and field studies of responses of eggs and larvae of marine organisms to the combined effects of temperature and salinity would lead to a greater understanding of the significance of these factors on survival during early larval development. Thus, defining these optimal conditions for culture of euryhaline marine species may be the fundamental for developing rearing protocol for penaeid species. The purpose of the present study was to determine the survival of postlarvae *Penaeus monodon* at different combination of three temperatures (29˚C, 33˚C and 35˚C) and four salinities (25, 30, 33 and 35 ppt) during the early life stage development.

2. MATERIALS AND METHODS

The combined effect of salinity and temperature on survival of *P. monodon* postlarvae (PL) was determined in the laboratory using a 3 × 4 two-factor factorial experimental design was conducted with three temperatures viz., room temperature (29˚C ± 1˚C) and elevated temperature (33˚C and 35˚C ± 0.5˚C) and four salinities (25, 30, 33 and 35 ppt). The experiment followed a completely randomized design, using the postlarvae stage (PL1 and PL15) from different spawners from private hatchery. Four batched of larvae were stocked into 10^{-1} cylindrical glass tanks at a density of 100 L^{-1} and they were acclimated to four salinity levels (25, 30, 33 and 35 ppt) by lowering the salinity at a rate of 5 $ppt \cdot h^{-1}$. The required salinity was obtained either by diluting seawater with freshwater or by mixing filtered seawater with sea salt to keep variation within ±1 ppt. They were then acclimated to different temperatures at a rate of 5˚$C \cdot h^{-1}$. A static-water system consisting of 36 aquaria (1 L glass beakers) with gently aeration was used and each aquarium held 20 individuals of postlarvae per treatment. Temperature was maintained within to ±0.5˚C using thermostatically controlled water baths. Each combination of temperature-salinity was conducted with three replications per treatment. Salinity and temperature were measured each morning using a portable refractometer and a mercury thermometer, respectively. All cultures were covered with aluminium foil to prevent evaporation. Gently aeration was provided in the container during the experiment. There was no feeding and water exchange during experiment for all treatments. Each experimental unit was initially examined the dead juveniles after 6 h, and every 12 h thereafter. The experiments lasted 96 h. The number of larvae sank down to bottom of the aquaria was considered as dead as well as the juveniles did not react to the touch of a needle. The mean percentage of survival was calculated by combining the data from three replicates at the end of the experiment. Data from each treatment were subject to a two-way ANOVA. When overall differences were significant at less than 5% level, Tukey test was used to compare the mean values between individual treatments. The square-root transformation of the sine-arc before analyzing the values given in percentages was used. Statistical analysis was performed using SPSS (statistic package for social science) 10.0.

3. RESULTS

Table 1 showed percentage survival of *P. monodon* postlarvae through 96 h under different temperature and salinity combinations. Results showed that temperature had a greater influence on survival of *P. monodon* postlarvae than salinity and low water temperature (29˚C) gave better larval survival than higher water temperatures (33˚C and 35˚C). In addition, the PL1 had better tolerance to high temperatures than PL15. Statistical analyses of the survival data by means of two-way ANOVA (**Table 2**) showed that temperature exerted a significantly higher influence on the survival at different stages of *P. monodon* from PL1-PL15 stages ($P < 0.05$). The effect of temperature and temperature-salinity interaction was significant at 5% level. At the termination of the experiment, the best survival of PL1 was found at

Table 1. Percentage survival of *P. monodon* postlarvae through 96 h under different temperature and salinity combinations.

Temperature (˚C)	Salinity (ppt)	Survival (%)	
		PL1	PL15
29	25	95.7 ± 0.42^a	75.9 ± 1.28^a
	30	96.6 ± 0.84^a	69.9 ± 1.27^b
	33	94.1 ± 0.49^a	65.2 ± 0.99^c
	35	97.1 ± 0.57^a	71.4 ± 0.85^a
33	25	87.9 ± 1.06^b	69.0 ± 1.34^b
	30	86.2 ± 0.98^b	65.1 ± 0.78^c
	33	87.3 ± 0.71^b	66.5 ± 0.78^c
	35	86.9 ± 1.49^b	68.2 ± 0.42^b
35	25	64.6 ± 0.78^c	37.1 ± 1.56^e
	30	69.2 ± 0.98^c	43.7 ± 1.49^d
	33	68.9 ± 1.34^c	47.1 ± 1.27^d
	35	76.3 ± 0.70^c	44.6 ± 0.85^d

Mean in the same column with different superscript letters are significantly different ($P < 0.05$).

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Table 2. Results of two-way ANOVA for survival of *P. monodon* postlarvae through 96 h under different temperature and salinity combinations at 95% confidence interval.

Parameters	Sum of square	*df*	Mean square	F-value	P-value
Postlarvae stage (PL1)					
Corrected model	4246.765[a]	11	386.070	293.589	0.000
Intercept	89377.215	1	89377.215	67967.464	0.000
Temperature	3907.922	2	1953.961	1485.902	0.000
Salinity	41.515	3	13.838	10.523	0.001
Temperature × salinity	297.328	6	49.555	37.684	0.000
Error	15.780	12	1.315		
Total	93639.760	24			
Corrected Total	4262.545	23			
Postlarvae stage (PL15)					
Corrected model	2992.991[a]	11	272.090	289.329	0.000
Intercept	170302.954	1	170302.954	181093.083	0.000
Temperature	2843.192	2	1421.596	1511.666	0.000
Salinity	55.355	3	18.452	19.621	0.000
Temperature × salinity	94.444	6	15.741	16.738	0.000
Error	11.285	12	0.940		
Total	173307.230	24			
Corrected total	3004.276	23			

R Squared = 0.996 (Adjusted R Squared = 0.993).

29˚C for all salinities with an average of 95.8% (94.1% - 97.1%), while the lowest survival of PL1 was found at 35˚C for all salinities with an average of 69.8% (64.6% - 76.3%). In contrast, the best survival of PL15 was found at 29˚C for all salinities with an average of 70.6% (65.2% - 75.9%), while the lowest survival was found at 35˚C for all salinities with an average of 43.1% (37.1% - 47.1%).

4. DISCUSSION

The present study showed that temperature had a greater influence to postlarvae tiger prawn *P. monodon* than salinity and low water temperature (29˚C) gave better larval survival than higher water temperatures (33˚C and 35˚C). In addition, the PL1 had better tolerance to high temperatures than PL15. The best survival of PL1 was found at 29˚C for all salinities tested with an average of 95.8% as compared to 70.6% for those of PL15. The elevated temperature might be suitable for growth of aquatic animals, but it had an inverse effect on survival [3]. Consistent with our study, temperature, salinity, shrimp size and the interaction of these parameters significantly influence the specific oxygen consumption of shrimp *Litopenaeus vannamei*. At 25˚C and 30˚C oxygen consumption was more stable at salinities 13‰ and 25‰ for all groups. At 20˚C and salinity below 25‰ oxygen consumption was higher, possibly due to the reduced hyperosmoregulatory ability in lower temperatures [9]. Maximum survival of the larval stages of *P. merguiensis* during the protozoeal stages also had been reported at 35 ppt followed by 48% at 33˚C and 45% at 29˚C and salinity exerted a greater influence than temperature on the survival and development of larvae. Based on the results, the best temperature-salinity combination for larval survival and metamorphosis is 33˚C and 35 ppt. A salinity range of 30 - 35 ppt is ideal for larval development [2]. A reduction in temperature level was also found resulting in a decreased osmoregulatory capacity of farmed shrimp *P. stylirostris* at low salinity and at high salinity, respectively, below and above the isoosmotic point (26.2 ppt). Furthermore, the sensitiveness of osmoregulation to temperature changes was dependent on the developmental stage of the shrimp. Subadults were more sensitive than juvenile animals [10]. Similar effects temperature and salinity on various marine and freshwater penaeid shrimp species are found. Juveniles of *P. vannamei* have

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their best survival between temperatures of 20˚C and 30˚C and salinities above 20%. Best growth was obtained between temperatures of 25˚C and 35˚C, with little difference being noted among salinities. Survival and growth coincide best at around 28˚C to 30˚C and 33% to 40%. In addition, many studies also indicated the effect of high temperature on development and growth of penaeid species but not for survival [8]. Development rate from the naupliar stage to the protozoal stage of *Metapenaeus monoceros* was best at a salinity level of 35 $g \cdot L^{-1}$ combined with temperatures of 28˚C and 32˚C and larval activity was found to be best at 28˚C and 32˚C at 35 $g \cdot L^{-1}$ and 40 $g \cdot L^{-1}$, as compared to that at 24˚C in all salinity levels [4]. Growth of the prawn *Macrobrachium rosenbergii* was also increased as temperature increased from 26˚C to 30˚C then the growth declined at the highest temperature (34˚C). Also as salinity increased from 0 to 16 ppt, growth of females decreased at all temperatures tested. It was clearly found that optimum level of both temperature and salinity for growth, reproduction and hatching success of *M. rosenbergii* was 30˚C temperature and 6 ppt salinity [5]. In addition, larvae *Pandalus jordani* were shown to have a wide tolerance to salinity, especially in the early stages, but a relatively narrow tolerance to temperature. The optimal temperatures for survival, 8˚C to 11˚C, were also optimal for growth as reflected by maximal growth increments and body size. It is therefore felt that fluctuations in temperature as seen within and between successive larval seasons would have profound effects on larval survival, growth rates and size at metamorphosis to the benthic juvenile phase [1]. Temperature also exerted a greater influence than the salinity on the growth and survival of *P. semisulcatus* during the larval development. The range of temperature in which the larvae showed high survival and growth is relatively narrow as compared to that of salinity. At all salinity levels, survival to PL1 (69% - 77%) was higher at 26˚C as compared to 30˚C (44% - 73%) and 34˚C (14% - 21%). However, daily growth rate at 30˚C and 34˚C was about 60% higher than at 26˚C. Larval development was also 3 - 4 days faster at 30˚C and 34˚C. Based on the survival and growth results, the best salinity and temperature combination for the culture of *P. semisulcatus* were 30 ppt and 30˚C [3]. Jackson and Burford (2003) found that salinity did not have a significant effect on growth or survival of *P. semisulcatus* larvae above 28‰. At 28‰, both growth rate and survival decreased, and there was significantly lower survival at the higher temperatures (32˚C and 29˚C).

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