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Survival and growth of early phyllosoma stages of *Panulirus homarus* under different salinity regimes

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Abstract

The survival and growth of phyllosoma larvae of the spiny lobster, *Panulirus homarus* were studied by exposing them to salinity regimes of 25, 27, 29, 30 (ambient), 31, 33 and 35 psu. Survival was low below 30 psu. Larvae in salinity range of 25 to 29 psu survived for 15 to 25 days whereas maximum survival of 55 to 60 days was obtained in salinities 31 and 33 psu. Growth represented as attainment of larval stages was proportional to longevity in the salinity range of 27 to 35 psu whereas it was retarded at 25 psu. The phyllosoma did not grow beyond stage II in 25 psu, even though it survived up to 15 days. The larvae reared in 31 and 33 psu salinity could reach Vb stage in 55 to 60 days.

Keywords: *Panulirus homarus*, phyllosoma, salinity, survival, developmental stage

Introduction

The early larval development of the spiny lobster *Panulirus homarus* till VI stage has been documented by Radhakrishanan and Vijayakumaran (1995). Scientists at the National Institute of Ocean Technology (NIOT), Chennai have further successfully reared the phyllosoma larvae of *P. homarus* and *P. ornatus* up to the VIII stage (DOD Annual Report, 2003-2004) on mixed diet of freshly hatched *Artemia nauplii*, adult enriched *Artemia*, chopped clam and mussel (Jha *et al*., 2006). However, high mortalities experienced during larval development highlights the need for further research on culture parameters to enhance larval survival in the culture system. The early life history of *P.homarus* is characterized by a pelagic larval phase in oceanic waters (Prasad *et al.*, 1975). Keys to the success of the mass production of pueruli may lie either in our ability to mimic these oceanic conditions or in the capacity of phyllosoma to adapt to the hatchery environment. Many Indian marine hatcheries experience seasonal variation in the salinity of intake water which affects seed production of fish and shellfish. The changes in salinity may be detrimental to the survival and development of larvae. In the early stage of larval development, it is essential to determine the tolerance of phyllosoma to shifts in seawater quality variables such as the salinity to design suitable culture system that maximizes larval survival. The effects of salinity changes on marine invertebrates range from sub-lethal to lethal levels depending on the magnitude of the change in the salinity and the tolerance of the species concerned (Torres *et al.*, 2008). Many coastal crustaceans are euryhaline and can withstand large shifts in environmental salinity unlike their stenohaline oceanic counterparts that live in or actively select isohaline waters (Willmer *et al.,* 2005). In euryhaline species such as *Carcinus maenas*, larval development is not affected by salinities ranging from 25 to 32 psu (Anger *et al.,* 1998). In contrast, the stenohaline *Pandalus borealis*, known to have an optimal salinity of 31 psu, does not complete larval development at 25 psu (Weinberg, 1982). In culture, sub-lethal salinities may result in delayed development and reduced growth (Anger *et al.,* 1998; Hereu and Calazans 2000; Pechenik *et al.,* 2000; Kumlu *et al.*

2001). The effect of salinity on the variability in the number of larval instars is reported in the sand lobster, *Scyllarus americanus* (Robertson, 1968). This experiment examines the effect of salinity on the survival and growth of early stage of cultured Indian *P. homarus* phyllosoma larvae with the aim of optimising culture conditions.

Material and Methods

Phyllosoma of *P. homarus* were obtained from captive breeders maintained at the Seafront laboratory of NIOT, Chennai, India. Initially spiny lobsters were collected from the wild (off Kovalam, Chennai, southeast coast of India) and maintained in 0.7 and 2.5 m^3 fiberglass tanks and in 5.0 m^3 rectangular cement tanks. A captive broodstock of *P. homarus* was developed and maintained at the seafront laboratory. The ambient seawater, directly pumped from subsurface in the intertidal area, had a salinity of 30.04 ± 0.04 psu during the course of the trials. To study the effect of salinity on the survival and growth of phyllosoma, the newly-hatched active larvae were cultured under different salinity regimes of 25, 27, 29, 30 (ambient), 31, 33 and 35 psu. The salinities were made up by adding non-chlorinated freshwater or raw crystal salt to the ambient seawater. The positively phototactic larvae were collected on the first day of hatching from a captive ovigerous female *P. homarus,* measuring 57.9 mm in carapace length.

The experiment was conducted in 1.5 litre glass bowls with one litre of seawater. Ten active phyllosoma were stocked in each bowl and duplicates were run for all salinity regimes. Complete water exchange and removal of dead larvae were carried out daily in the morning. Mean (±SD) salinity levels during the course of the trial were 25.04 ± 0.02, 27.04 ± 0.01, 29.03 ± 0.01, 30.04 ± 0.04 (ambient), 31.03 ± 0.01, 33.03 ± 0.01, 35.03 ± 0.01 psu. Temperature and pH were measured daily while dissolved oxygen was measured once in a week and the values ranged from 27.5 to 31.0°C, 7.8 to 8.1 and 5.05- 5.55 mg/l respectively during the experiment.

Stage I-III phyllosoma were fed exclusively on freshly hatched *Artemia* nauplii (1–2 mm in length) at a rate of 6 nauplii/ml. Thereafter, a combination of freshly hatched *Artemia* nauplii (6 nauplii/ml) and sub adult, enriched (by feeding microalgae *Chlorella* sp. and *Isochrysis galbana*) and freshly killed *Artemia* were fed. Moultings were recorded by daily observation of exuvia. The size of phyllosoma was measured using its exuvia as well as dead larvae. The dead phyllosoma or exuvia were measured using a stereozoom microscope (Nikon SMZ 800 microscope). Dead larvae and exuvia were preserved in 4% buffered formalin for measuring its carapace length, carapace width, total length and to study its exopods in various phyllosoma stages. The experiment was continued for a maximum of 60 days, when the last surviving phyllosoma died. Statistical analysis (ANOVA, one way) was carried out for the survival percentage in different salinity regimes using MS-Excel data analysis tools. Mean survival percentage was also calculated for duplicates in each salinity regime.

Results

The mean survival percentage of phyllosoma larvae reared in different salinity regimes are given in Fig. 1. Under the lowest salinity of 25 psu, only 60% of phyllosoma survived up to the 5th day, whereas the highest mean survival of 95% was recorded in the 31 psu salinity. Similarly on the 10th day, the minimum mean survival of 45% and the maximum mean survival of 85% were observed in 25 and 31 psu respectively. Phyllosoma reared in lower salinity regimes of 25 and 27 psu did not survive beyond the 25th day, whereas all phyllosoma in 35 psu died on the 30th day. The minimum and the maximum mean survivals of 15 and 60% were

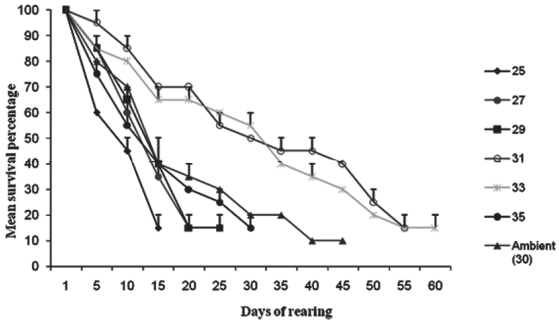

Fig. 1. Mean survival percentage (mean ±SE) of phyllosoma of *P. homarus* reared under different salinity regime

Table 1. Total length (mm) of different stages of phyllosoma larvae of *P. homarus* (Linnaeus, 1758) reared in laboratory in different salinity regimes (psu); the values are mean ± SD)

Stage	25	27	29	30(ambient)	31	33	35
I	1.45±0.02	1.45±0.02	1.45±0.02	1.45±0.02	1.45±0.02	1.45±0.02	.45±0.02
II	1.77±0.02	1.81±0.02	1.82±0.00	1.82±0.00	1.90±0.02	1.95±0.00	1.95±0.02
IIIa	-	2.10±0.01	2.12±0.05	2.01±0.02	2.10±0.00	2.22±0.00	2.25±0.00
IIIb	-	-	-	2.51±0.05	2.59±0.00	2.75±0.02	2.79±0.05
IIIc	-	-	-	2.65±0.00	2.70±0.02	2.89±0.00	-
IV a	-	-	-	2.91±0.00	3.05±0.02	3.11±0.00	-
IV b	-	-	-	-	3.41±0.00	3.49±0.02	-
V a	-	-	-	-	3.70±0.02	3.99±0.00	-
Vb	-	-	-	-	4.13±0.01	4.66±0.05	-

recorded in 29 and 33 psu respectively on the 25th day. On the 45th day, the minimum and the maximum mean survival of 10 and 25% were recorded in 30 and 31 psu respectively.

The survival rate on the 15th day was significantly ($p < 0.05$) different under the salinity regimes tested. Improved survival and growth was obtained in 31 and 33 psu salinities and the larvae reached Vb stage in both cases. The larvae reared in 33 psu survived till 60th days with mean survival of 15%.

The maximum stage of development attained in different salinity groups is given in Table 1. The maximum developmental stage was attained in the salinities 31 and 33 psu in which the larvae reached up to Vb stage whereas in 25 psu salinity, the larvae could reach up to stage II only. Larvae reared in 27 and 29 psu could reach up to IIIa stage whereas in 35 psu it could reach IIIb stage. Larvae reared in ambient salinity (30 psu) reached IVa stage.

The mean moulting of 95 and 85% was observed in 31-33 psu salinity whereas the lowest mean moulting of 15% was recorded in 25 psu during I-II stage (Fig. 2). There was statistically significant difference ($p < 0.05$) in the mean moulting percentage in different groups during I-II larval stage instars. It was observed that better moulting and survival was recorded in 31 and 33 psu and the larvae could reach to Vb stage in these salinity regimes.

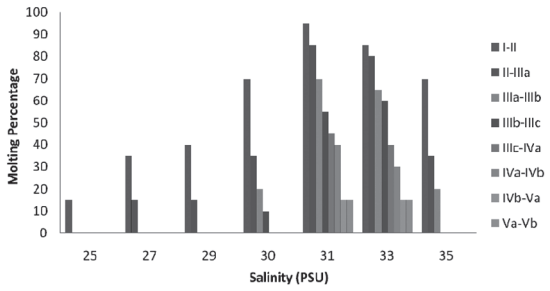

Fig. 2. Mean moulting percentage of phyllosoma of *P. homarus* (stages I-Vb) reared under different salinity regime

Discussion

The survival of any cultured marine organism is highly influenced by the quality of rearing water apart from other factors such as feeding and high stocking density. Denton and Jones (1982) reported that salinity was the sole factor affecting the growth of cultured shrimp *Penaeus merguiensis* under normal culture conditions. The present study reveals that salinity affects the survival and growth of phyllosoma of *P. homarus*. Maximum survival, longevity and growth were recorded in salinity range of 31 to 33 psu. The developmental stage attained under different salinity regimes were proportional to the survival of phyllosoma at 25 psu, the phyllosoma did not grow beyond stage II. As it is much below the optimal range for larval development in *P. homarus*. Parry and Potts (1965) reported that at low salinity, cultured organisms spend more energy to maintain the osmotic balance. The osmotic stress may lead to lower survival and reduction in the growth of phyllosoma larvae. The energy spent in maintaining osmotic balance may be the main cause for the decreased growth and lower survival of lobster larvae in lower salinities.

The phyllosoma larvae survived for a maximum of 60 days in the present study which is very a short period within a larval cycle that may extend up to 416 days in hatchery conditions for different species of spiny lobsters (Kittaka 1994; Booth 1996). Previous investigations attempted to relate the metabolic rate of aquatic organisms with their growth performances under different salinities (Anger *et al.* 1998; Pechenik *et al.,* 2000; Villareal *et al.,* 2003) but in the present study focus was on survival of phyllosoma larvae. There was statistically significant difference in the survival percentage of different groups on day 15 ($p < 0.05$). Further, there was statistically significant difference in the mean moulting percentage in different groups during moulting from I to II stage larval instars ($p < 0.05$). Phyllosoma reared in salinity regimes of 31 and 33 psu have shown better survival and growth. In nature phyllosoma are carried away offshore by water currents and wind action and dwell at oceanic salinities of around 35 psu most of their larval life. Therefore it should have survived better than the result obtained in this study in the highest salinity regime (35 psu) tested here and more trials at this salinity are required. The present study suggests that the optimum salinity range for rearing phyllosoma of *P. homarus* is 31-33 psu.

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