Kasetsart J. (Nat. Sci.) 36 : 278 - 284 (2002)

Salinity Levels on Survival Rate and Development of Mud Crab *(Scylla olivacea)* from Zoea to Megalopa and from Megalopa to Crab Stage

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ABSTRACT

Effect of salinity on survival rate and development of mud crab (*Scylla olivacea*) were conducted into two phases: phase I from zoea 1 to megalopa and phase II from megalopa to the crab stage. In phase I, four different levels of salinity, 28, 30, 32 and 34 parts per thousand (ppt) were used. The results showed that zoea reared at 28, 30, 32 and 34 ppt developed to megalopa stage with survival rates of 13.16, 22.19, 8.25 and 7.08 percent respectively. At 30 ppt, zoea developed to megalopa stage with better survival rate than at the other salinity levels ($P<0.05$). In all salinity levels, high mortality rate of zoea occured during the development of zoea 1 to zoea 3 and was higher than that of zoea developed from stage 3 to megalopa. Duration for development of zoea 1 to megalopa at all salinity levels were 23.64-24.22 days which were not significantly different ($P>0.05$) among treatments.

In phase II, the effect of salinity on survival rate and developmental period of megalopa to the fifth crab stage (C5) at salinity levels of 12, 16, 20, 32, 36 and 40 ppt were studied. The results showed that survival rates of the crab developed from megalopa at 16, 20, 32 ppt were 80.95, 76.19, and 80.95 percent respectively. These figures were significantly higher ($P<0.05$) than those at 12, 36, and 40 ppt. Time used for megalopa to develop into the fifth crab stage (C5) was influenced by salinity. The developmental period to the fifth crab stage from megalopa were 29.14, 28.25, 30.10, 31.75, 32.55, and 43.50 days, respectively. Aging also had influenced on time for crab development. The more advanced crab stage, the longer time used for the development of crab to the next stage.

Key words: *Scylla olivacea*, salinity, zoea, megalopa, crab

INTRODUCTION

The portunid crab *scylla* known as mangrove or mud crab, is the largest species of edible crab in the Indo-Pacific region. Its habitat is in brackish waters between the lower intertidal zone and offshore such as mangrove estuaries and creeks. (Hyland *et al*. 1984; Macintosh, 1988). Mud crab is important to socio-economic of Australia, Japan, Taiwan, Indonesia, Philippines and Thailand (Keenan, 1999). The export value for mud crab of Thailand is approximately 180 million baths with the total volume of 3,867 tons. Mud crabs produced in Thailand are mostly wild catches. However, due to the degradation of natural habitats, increasing of water pollution, and overexploitation of crab, the wild population of this shellfish decreased significantly.

Development of mud crab aquaculture has started for some times but not much success has been acheived due to mass mortality of seed crab especially during the zoea and megalopa stage.

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Therefore, it is necessary to identify the certain factors affecting survival and development of mud crab seed. Salinity is one of the factors related to the rearing conditions needed to be determined. This study was conducted to define optimal salinity levels for survival and development of mud crab *scylla olivaceae* in two developmental periods. Experiment 1: study from zoea 1 to the megalopa stage and experiment 2: study from megalopa to the crab stage.

MATERIALS AND METHODS

Experiment 1

An ovigerous female crab was raised in 50-L plastic tanks at the salinity 30 parts per thousand (ppt) and fed daily with chopped fishes and squids until spawning. After the eggs hatched into zoea1, the zoea were randomly stocked in 12, 31.0×43.5×29.5 cm styrofoarm boxes at density of 400 zoea/box. Each box was allocated to each salinity level, 28, 30, 32 and 34 ppt at three replicates/ treatment. Each box contained 15 liter of water at its respective salinity. At the stocking, the zoea were gradually acclimated to the increasing salinity until reaching the required salinity. Zoea were fed with rotifer 60 ind/mL and artemia 10 ind/mL. Dead zoea were removed and saline water was totally changed daily prior to feeding the zoea. Alive zoea were picking up in a wide-bore pipette and recorded daily. The survival rate and the development of zoea were examined by counting the number and identifying the stage of the larvae (zoea 2, 3, 4, 5 and megalopa). Time (days) for the development of zoea 1 to megalopa stage in each salinity level was recorded and used as indicator for zoea development.

Experiment 2

The effect of salinity on the survival and development of megalopa to crab stage was investigated. Each of eighteen styrofoarm boxes divided into seven compartments using seven punched plastic cups as the cell to prevent cannibalism. The cup had a diameter of 8 cm. Each of three replicates of divided box was filled with 15 liters of water at salinity levels of 12, 16, 20, 32, 36 and 40 ppt. A megalopa was randomized into each cup and daily fed with newly hatch artemia 10 ind/ mL and chopped fishes soon after daily change of new water . Survival rate (%) and time (days) of the larvae molted to the next stage were determined.

Analysis of variance

Survival rate (%) and time (days) used for the development of mud crab larvae at various salinity levels in both experiments were subjected to analysis of variance. If the differences were significant ($P<0.05$), then Duncan 'new multiple range test (DMRT) was conducted to find out which treatments were different.

RESULTS AND DISCUSSION

Experiment 1

The survival of zoea 1 that successfully developed to megalopa stage at salinity level of 28, 30, 32 and 34 ppt were 13.16, 22.91, 8.25 and 7.08 percent, respectively (Table 1, and Figure1). The zoea reared at 30 ppt developed to megalopa stage with significantly higher survival rate ($P<0.05$) than those at the other levels. At this salinity level, zoea 1 survived into zoea 2, 3, 4, 5 and megalopa at percentages of 51.75, 35.58, 31.75, 30.08 and 22.91 respectively. There were no significant differences ($P>0.05$) in survival rates of zoea at various stages of development when reared at 28, 32 and 34 ppt. High mortality of zoea was observed at 34 ppt. The result agreed with Heasman and Fielder (1983) and Hoang (1999) who found that optimum salinity level for survival of *Scylla serrata* could be varied from 30-34 ppt. They also reported that survival and megalopa production depended on the ability of the zoea to maintain the body fluid in the isotonic environment. Zoea raised in the hypo or hypertonic solution would lose some energy for adjusting body fluid, the energy left for molting might not be

enough and cause mortality in zoea. This study showed that zoea raised at 30 ppt had the highest survival rate, this probably due to the tested zoea was in isotonic of 30 ppt water which was right for the zoea to hatch.

At all salinity levels, the accumulate mortality occurring during zoea 1 developing to zoea 3 were significantly higher ($P<0.057$) than those of the other developmental stages. Worner (1997) reported that in the wild, mass mortality of the early stage larvae always occured in marine animals that produced abundance of larvae. Furthermore, the early stage larvae, zoea 1 in this study had not yet developed the eyestalk which was the organ producing water balance regulating hormone. When crab larvae developed to zoea 3, 4, 5, the rate of mortality was found to decrease and survival rates were not significantly differences ($P>0.05$). The reason was that zoea at these stages were acclimated and adjusted to the salinity so that the efficiency in controlling fluid-balancing was better as appeared in *Macrobthalmus setosus* (Nongnut, 1999).

Time for the developing of zoea 1 to

Table 1 Mean percent survival of zoea 1 (Z1) to megalopa (M) reared under four different salinity ; 28, 30, 32 and 34 parts per thousand (ppt).

Treatment	Z1	% survival				
		Z1-Z2	Z1-Z3	Z1-Z4	Z1-Z5	Z1-M
1. (28 ppt)	$100\pm0.00^{a,A}$	$37.58\pm7.60^{b,B}$	$18.50\pm5.71^{b,C}$	$15.75\pm6.66^{b,C}$	$14.50\pm6.08^{b,C}$	$13.16\pm6.14^{b,C}$
2. (30 ppt)	$100\pm0.00^{a,A}$	$51.75\pm1.08^{a,B}$	$31.75\pm3.07^{a,C}$	$35.58\pm3.68^{a,CD}$	$30.08\pm3.39^{a,D}$	$22.91\pm2.40^{a,E}$
3. (32 ppt)	$100\pm0.00^{a,A}$	$30.08\pm5.39^{b,B}$	$14.16\pm5.13^{b,C}$	$11.00\pm3.76^{b,C}$	$9.75\pm4.54^{b,C}$	$8.25\pm3.47^{b,C}$
4. (34 ppt)	$100\pm0.00^{a,A}$	$31.66\pm2.02^{b,B}$	$13.33\pm3.14^{b,C}$	$9.58\pm1.87^{b,D}$	$8.91\pm1.50^{b,D}$	$7.08\pm0.62^{b,D}$

Note : Data are expressed as mean ± SD
Mean in the same rows (capital letter) or column (lower case) with the same letter are not significantly different ($P>0.05$)

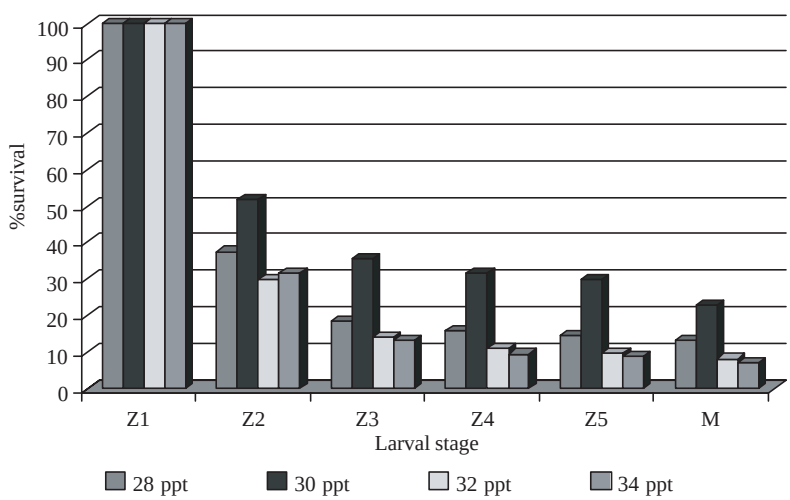

Figure 1 Percentage of zoea surviving reared under four different levels of salinity; 28, 30, 32 and 34 parts per thousand (ppt).

megalopa at the salinity levels of 28, 30, 32 and 34 ppt were 23.64, 24.22, 23.76 and 23.95 days, respectively (Table 2). There were no significant differences (P>0.05) of the development period among treatments. This was probably because zoea was the stage of crab whose life cycle developed in marine environment, therefore, salinity levels might not have much effect on its development (Nongnut, 1999).

Experiment 2

The survival rates of the fifth crab stage developed from megalopa at the salinity levels of 12, 16, 20, 32, 36 and 40 ppt were 42.85, 80.95, 76.91, 80.95, 52.38 and 9.52 percent, respectively (Table 3 and Figure 2). Megalopa in different treatments were found to tolerate salinity and developed to crab stage differently. Survival rate of crab at all stages developed from megalopa were not significantly different among salinity levels from16-32 ppt (P>0.05) and were significantly higher (P<0.05) than those at 12 and 40 ppt. High mortality of megalopa when developed to crab 1 were recorded at 40 ppt. It showed that crab stage tended to have greater tolerance to wide ranges of salinity particularly between 16-32 ppt. This result agreed with the study in *Scylla serrata* by Trans *et al*. (1998). However, too low or too high salinity as evidented in 12 and 40 ppt, will affect survival rate of crab. This was because crab had to spend more energy to regulate body fluid at extreme salinity. Furthermore, crab might have mineral deficiency if reared at too low salinity levels (Holliday, 1969).

At salinity levels of 12, 36 and 40 ppt , the mortalities of crab 1 developing from megalopa were high similar to that occured in Zoea 2

Table 2 The average developmental period (days) from zoea 1 to megalopa reared under four different salinity.

Treatment	Developmental period (days)
1 (28 ppt)	23.64 ± 0.78^{a}
2 (30 ppt)	24.22 ± 0.71^{a}
3 (32 ppt)	23.76 ± 0.30^{a}
4 (34 ppt)	23.95 ± 0.48^{a}

Note : Mean in the same column with the same letter are not significantly different (P>0.05)

Table 3 Mean percent survival of megalopa (M) to crab 1, 2, 3, 4 and 5 (C1-C5) reared under six different salinity ; 12, 16, 20, 32, 36 and 40 parts per thousand (ppt).

Treatment	M	% survival				
		M-C1	M-C2	M-C3	M-C4	M-C5
1. (12 ppt)	$100\pm0.00^{a,A}$	$57.14\pm50.70^{b,B}$	$47.61\pm51.17^{b,BC}$	$42.85\pm50.70^{b,BC}$	$42.85\pm50.70^{b,BC}$	$42.85\pm50.70^{b,BC}$
2. (16 ppt)	$100\pm0.00^{a,A}$	$95.23\pm21.82^{a,A}$	$90.47\pm30.07^{a,A}$	$85.71\pm35.85^{a,A}$	$85.71\pm35.85^{a,A}$	$80.95\pm40.23^{a,A}$
3. (20 ppt)	$100\pm0.00^{a,A}$	$95.23\pm21.82^{a,AB}$	$95.23\pm21.82^{a,AB}$	$95.23\pm21.82^{a,AB}$	$90.47\pm30.07^{a,AB}$	$79.19\pm43.64^{a,B}$
4. (32 ppt)	$100\pm0.00^{a,A}$	$95.23\pm21.82^{a,A}$	$95.23\pm21.82^{a,A}$	$95.23\pm21.82^{a,A}$	$95.23\pm21.82^{a,A}$	$80.95\pm40.23^{a,A}$
5. (36 ppt)	$100\pm0.00^{a,A}$	$52.38\pm51.17^{b,B}$	$52.38\pm51.17^{b,B}$	$52.38\pm51.17^{b,B}$	$52.38\pm51.17^{b,B}$	$52.38\pm51.17^{ab,B}$
6. (40 ppt)	$100\pm0.00^{a,A}$	$9.52\pm30.07^{c,B}$	$9.52\pm30.07^{c,B}$	$9.52\pm30.07^{c,B}$	$9.52\pm30.07^{c,B}$	$9.52\pm30.07^{c,B}$

Note : Data are expressed as mean ± SD

Mean in the same rows (capital letter) or column (lower case) with the same letter are not significantly different (P>0.05)

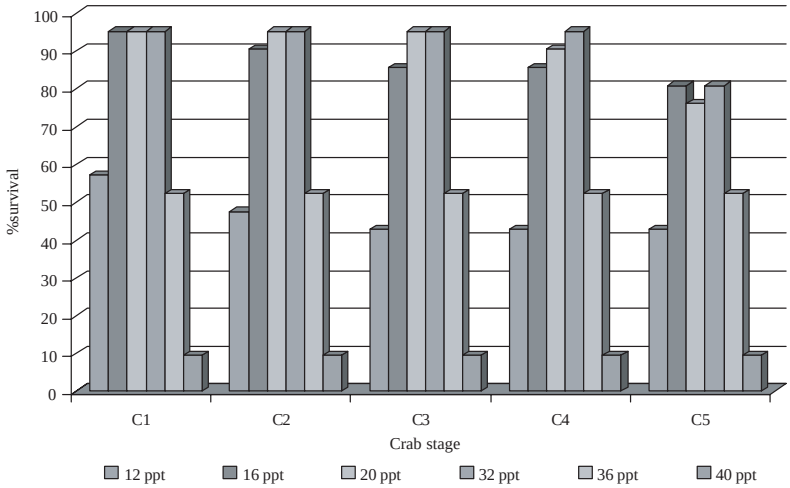

Figure 2 Percentage of megalopa surviving reared under six different levels of salinity; 12, 16, 20, 32, 36 and 40 parts per thousand (ppt).

developing from Zoea l. After developing to crab 1 stage, mortality of further crab stages were low. Therefore, there were no significant differenes ($P>0.05$) in the survival rates of crab at each development stage. The study also showed that salinity levels had effect on time for the development of megalopa to crab 5. The developmental period to crab 5 from megalopa raised at salinity level of 16, 20, 32, 36 and 40 ppt were 28.25, 30.10, 31.75, 32.55 and 43.50 days, respectively (Table 4 and Figure 3). At higher salinity, the trend for longer time was needed for the development of megalopa to crab stage. Crab age also influenced time for development to the next stages. The more advanced crab stage, the longer times spent for the development. This was because the setting period during the molting process in the older crab was longer than that of the younger one (Marichamy and Rajapackiam,1991).

Table 4 The average developmental period (days) from megalopa (M) to crab 1, 2, 3, 4 and 5 (C1-C5) reared under six different salinity ; 12, 16, 20, 32, 36 and 40 parts per thousand (ppt).

Treatment	Developmental period (day)				
	M-C1	C1-C2	C2-C3	C3-C4	C4-C5
1. (12 ppt)	$9.83\pm1.46^{a,C}$	$4.10\pm0.31^{a,A}$	$4.22\pm0.66^{a,A}$	$4.55\pm0.88^{ab,A}$	$6.44\pm1.13^{ab,B}$
2. (16 ppt)	$9.70\pm1.41^{a,C}$	$3.73\pm0.87^{a,A}$	$4.27\pm0.82^{a,A}$	$4.44\pm0.78^{a,A}$	$7.11\pm1.36^{ab,B}$
3. (20 ppt)	$10.30\pm1.52^{a,C}$	$4.45\pm0.75^{a,A}$	$4.45\pm1.09^{a,A}$	$4.57\pm1.10^{ab,A}$	$6.37\pm1.14^{a,B}$
4. (32 ppt)	$11.30\pm1.45^{a,C}$	$4.35\pm0.81^{a,A}$	$3.90\pm0.91^{a,A}$	$4.85\pm0.67^{ab,A}$	$7.05\pm1.29^{ab,B}$
5. (36 ppt)	$13.54\pm1.50^{a,C}$	$4.00\pm0.63^{a,A}$	$4.27\pm0.64^{a,AB}$	$4.81\pm0.87^{ab,AB}$	$6.81\pm1.47^{ab,B}$
6. (40 ppt)	$20.00\pm2.82^{a,C}$	$4.50\pm0.70^{a,A}$	$5.00\pm0.70^{b,A}$	$5.50\pm0.70^{b,A}$	$8.00\pm1.41^{a,AB}$

Noet : Data are expressed as mean ± SD

Mean in the same rows (capital letter) or column (lower case) with the same letter are not significantly different ($P>0.05$)

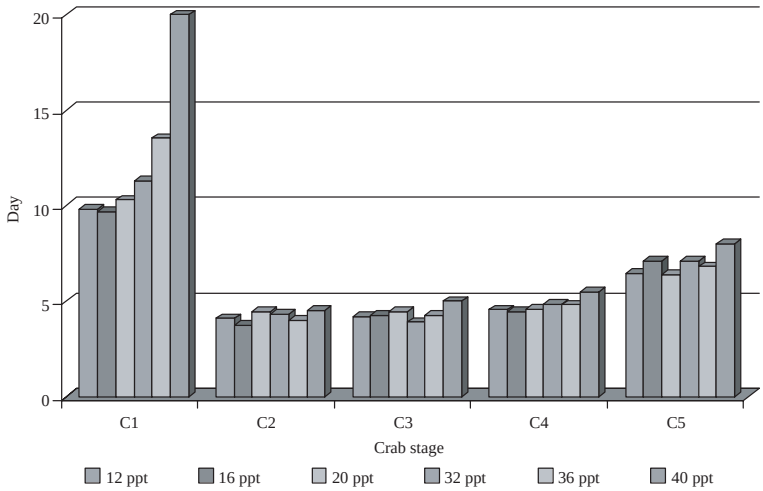

Figure 3 The average developmental period (days) from megalopa (M) to crab 1, 2, 3, 4 and 5 (C1-C5) reared under six different salinity.

CONCLUSION

Zoea reared at 30 ppt developed to megalopa stage with better survival rate (22.19 percent) than those at the other salinity levels of 28, 32 and 34 ppt. In all salinity levels, high mortality rate of zoea occured during the development of zoea 1 to zoea 3. Duration for development of zoea 1 to megalopa at all salinity levels were 23.64-24.22 days. For survival rates of crab developed from megalopa, the survival rates of crab were high among salinity levels from16-32 ppt (76.91-80.95 percent) and were significantly higher ($P<0.05$) than those at 12 and 40 ppt (42.85 and 9.52 percent, respectively). The developmental period to the fifth crab stage from megalopa was influenced by salinity. At higher salinity, the trend for longer time was needed.

ACKNOWLEDGEMENTS

This contribution is based on research funded by Danish Cooperation for Environment and Development (Danced) , TCE Project.

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Received date : 19/08/02
Accepted date : 30/09/02