Fish Aqua Sci 13(4), 308-314, 2010

KOSFAS FISHERIES AND AQUATIC SCIENCES
The Korean Society of Fisheries and Aquatic Science

Effects of Rearing Temperature on Larval Survival and Growth and on Reproductive Traits of *Palaemon serrifer* (Decapoda: Caridea: Palaemonidae)

Sunghan Kim
Department of Aquaculture and Aquatic Resources, Kunsan National University, Kunsan 573-701, Korea

Larvae of *Palaemon serrifer* were reared in the laboratory under three different temperature regimes (15°C, 20°C, 25°C) to study the effects of rearing temperature on larval survival and growth, as well as other traits such as embryo volume, number of embryos (fecundity), incubation period, development. Mode and development period. Growth pattern was analyzed by measuring the molt increment and intermolt period. The intermolt period consistently increased with size and instar number and was shortest at 25°C. However, molt increments generally decreased with instar number. Number of embryos varied from 552 to 1355. The relationship between the number of embryos and carapace length was expressed by the equation (fecundity) y=2.7744x+0.208 (R^2=0.7961). Egg volume was a primary factor affecting other life-history traits. Egg volume was 0.078 m^3, which is relatively small thus embryos exhibited a relatively short incubation period and a comparatively short development period, and the nutritional mode was planktotrophic. Brood production was followed by a fast parturitional pattern. Most ovigerous females had mature ovaries when the parturial molt occurred soon after eclosion.

Key word: Egg volume

Introduction

Crustacean larvae grow discontinuously in a saltatory pattern resulting from the molting process (Hartnoll, 1982). Little is known about the early life history of most crustaceans due to the difficulty of measuring larval growth *in situ* (Anger 1991; Anger and Moreira 1998). Thus, estimations of larval growth are generally based on rearing experiments in the laboratory (Costlow et al., 1960; Hartnoll and Mohamedeen, 1987). Crustacean growth includes the intermolt period and molt increments which are influenced by rearing temperature and instar number (Rothlisberg, 1979; Hartnoll).

Rearing experiments with decapod larvae have primarily been conducted under laboratory conditions to observe the relationship between molt increment, intermolt period, instar number, and water temperature (Rothlisberg, 1979; Hartnoll and Mohamedeen, 1987). Lebour (1927) was one of the first researchers to describe the early development stages of larvae. Later, individual larvae were cultured and environmental variables were controlled (Costlow and Bookout, 1959; Costlow et al., 1960).

In situ temperature is known to greatly influence the survival and growth of decapod larvae, and many studies on its influences have been conducted (Hartnoll and Mohamedeen, 1987; Mohamedeen and Hartnoll, 1989). In most studies the larval molt increment increased with increasing temperature, whereas the intermolt period decreased, thus increasing the growth rate (Hartnoll and Mohamedeen, 1987; Mohamedeen and Hartnoll, 1989). However, the effects of temperature on molt increment vary among species (Hartnoll and Mohamedeen, 1987; Mohamedeen and Hartnoll, 1989).

Early life-history traits in marine organisms are crucial and can influence other aspects of life history (Bauer, 2004). Generally, growth and reproduction require a trade-off in energy allocation by the individual organism (Kim and Hong, 2004). Embryo size is a key determinant that influences other traits such as fecundity, incubation period, larval develop-

*Corresponding author: w0827@kunsan.ac.kr

ment mode and the larval period (Bauer, 2004). The nutritional modes of larvae have been correlated with embryo size (Vance 1973). Thorson (1950) noted a trend toward increased egg size and non-feeding, demersal development along gradients of increasing latitude and water depth.

Research on Antarctic and deep-sea invertebrates has revealed several exceptions to Thorson's idea (Pearse et al., 1991). The evolution of direct development involves dramatic increases in egg size (Emlet et al., 1987; Wray and Raff, 1991). Smaller eggs usually undergo planktotrophic development, whereas larger, more yolky eggs are likely to exhibit lecithotrophic development (Bauer, 2004).

The brood-production pattern is related to ovarian development, embryonic development, and the molting cycle (Bauer, 2004). Patterns of brood production have been classified into three types: slow successive parturials, fast successive parturials, and alternate parturials (Bauer, 2000). Caridean shrimps generally undergo the larval development period as a complete plankton stage in the water from zoea to post-larvae before settlement (Bauer, 2004).

Palaemon serrifer is distributed in intertidal regions, particularly in the tide pools of the Yellow Sea and the Southern Sea of Korea (Holthuis, 1950; Kim, 1977). *Palaemon serrifer* exhibits the plank-totrophic nutritional mode which is the general pattern of caridean shrimps and it is hypothesized to have small eggs (Bauer, 2004).

The goal of this study was to examine the survival rate and the effects of temperature and instar number on growth (both molt increment and intermolt period), to describe larval life-history traits and to analyze the relationship of these variables to other life-history traits in the larvae of *Palaemon serrifer*.

Materials and Methods

Ovigerous females of *P. serrifer* were collected in the intertidal regions of Dongback Island. Water temperature was measured at the sampling site at the time of collection.

Ovigerous females were carried to the laboratory and reared at 20°C with 32 ppt salinity under a 12:12 L:D (hours of light:dark) photoregime. Specimens were then divided into three groups to receive three different temperature regimes (15°C, 20°C, 25°C) at a salinity of 31 to 32 ppt under a 12:12 (L:D) photoregime. Fish were fed chopped clams and squid daily. Seawater of the rearing tank was exchanged every other day, and detritus and defecated materials were removed daily. Fifty larvae were randomly selected and transferred individually to separate glass vials for individual culture. Seawater in the vials was exchanged with filtered sea water daily. Larvae were fed newly hatched *Artemia* nauplii. The carapace length of exuviae at molt was measured with a microscope, and molt increment was calcu-lated from measurements of carapace length of exuviae.

Additionally, intermolt period was measured during larval development from zoea 1 stage to post-larvae. Intermolt period was reported as days between molts, and molt increment was described by the equation :

$$\text{molt increment (\%)} = (\text{CL at postmolt} - \text{CL at premolt}) / \text{CL at premolt} \times 100$$

A regression equation was used to describe the relationships between instar number and growth components such as intermolt period and molt increment. All regression lines exhibited significant goodness of fit. Intermolt periods and molt increments were log-transformed due to non-homogeneity of variance. After performing regression analysis among intermolt periods, molt increments, and instar linear regression equations were calculated for the three different temperature regimes ANCOVA was used to detect differences in the slopes of the regression equation relating the intermolt period and molt increment (Zar, 1996). Intercepts of the re-gression equations were compared if no significant difference between slopes was found.

Ovarian development was divided into three stages based on the following morphological criteria: stage 1, immature ovary, thin and translucent; stage 2, intermediate, green ovary filling almost all of the cepholothorax volume; stage 3, mature ovary filling almost all of the cephalothorax, prespawning stage.

Carapace lengths of the female adult and larvae were measured from the posterior rim of the eye socket to the posterior lateral edge of the carapace. Fecundity (number of eggs) was determined by the regression equation relating the log-transformed number of eggs and carapace length for eyeless embryos of ovigerous females. Eggs containing embryos were subsampled to calculate egg volume. Eggs were measured with a stereomicroscope along the major and minor axes, and the volume was calculated using the formula $4/3\pi r_1 r_2^2$, where r_1 is half the major axis and r_2 is half the minor axis.

Results

Water temperature variation and survival rate

Water temperature ranged between 7 and 28°C and increased steadily from April to August then declined in autumn and winter (Fig. 1A). The highest temperature was 28°C in August, and the lowest temperature was 7°C in January.

Survival rate of larvae was highest at 25°C and lowest at 15°C (Fig. 1B). Survival rate of larvae reared at 15°C declined rapidly from the start of instar 2 and all larvae died by instar 3 the last instar. The survival rate gradually decreased with time at 20°C and 25°C and was 55% and 68%, respectively, until the post-larva stage. These results suggest that the highest production of larvae should occur from June to September.

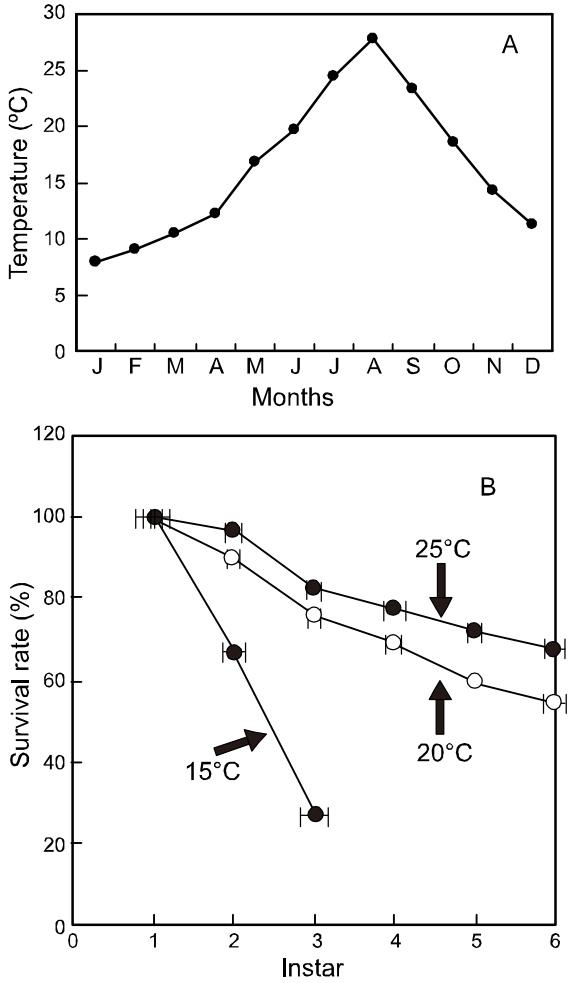

Fig. 1. *Palaemon serrifer*. Monthly variation of water temperature (A) Survival rate (%) per instar of the larvae of reared at 15, 20, 25°C (B).

Growth components (Intermolt period, IP and molt increment, MI)

Growth of larvae was highly influenced by temperature. Larvae exhibited faster growth at 25°C than at 20°C and 15°C.

Intermolt period lasted 19, 19, and 13 days for instars 1, 2, and 3, respectively, at 15°C, and 3, 7, 9, 12, 14,and 19 and 2, 4, 6, 9, 13 for instars 1 to 6 at 20°C and 25°C, respectively (Fig. 2A). The longest intermolt period occurred at 15°C, and the shortest was at 25°C. As instar number increased, molt increment decreased. Of the three different temperature regimes, the maximum molt increment occurred at 25°C (Fig 2B). The regression relationship between intermolt period and instar was shown using logarithmically transformed linear equations (Fig. 3A) as follows:

15°C, $\log IP = 0.2168IT + 0.6563$,
($r^2 = 0.95$, $n = 3$, $P<0.05$)
20°C, $\log IP = 0.1439IT + 0.4598$
($r^2 = 0.91$, $n = 6$, $P<0.05$)
25°C, $\log IP = 0.1588IT + 0.2426$,
($r^2 = 0.94$, $n = 6$, $P<0.05$)

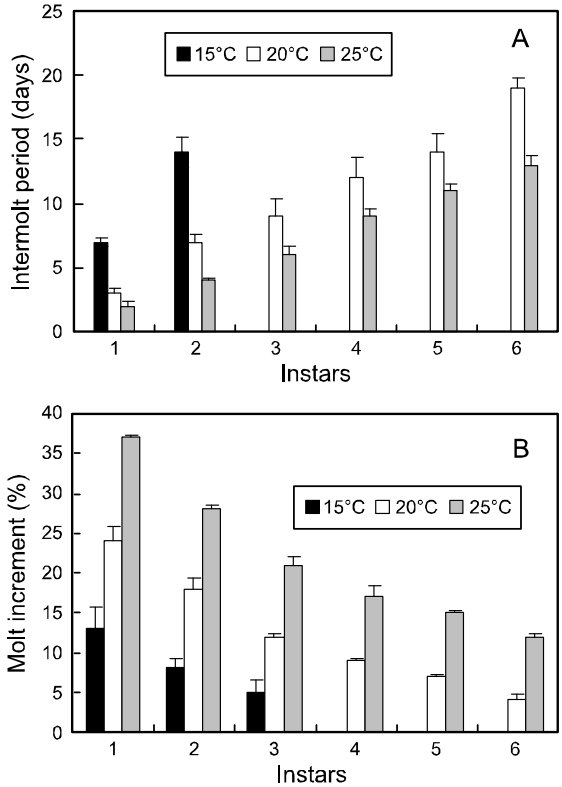

Fig. 2. *Palaemon serrifer.* Intermolt period plotted against instar number in (A) and molt increment plotted against instar numbers (B) for larvae reared at 15, 20 and 25°C.

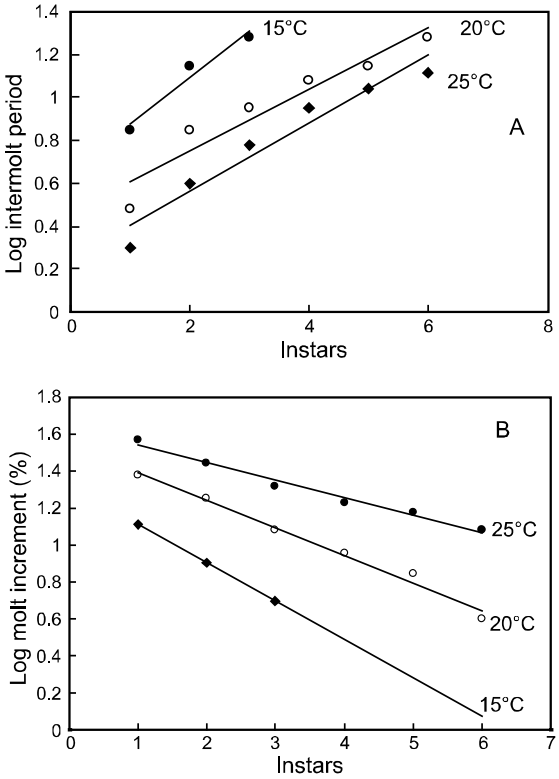

Fig. 3. *Palaemon serrifer*. Regression relationship between log intermolt period and instars (A) and regression relationship between log molt increment and instars (B).

These regression equations exhibited a high deterministic coefficient (r^2>0.9) and a high linear regression relationship. At all temperatures, the slopes of the linear regression equations showed a significant positive relationship, suggesting that the intermolt period gradually increased with instar number. The ANCOVA conducted on the difference in the slopes of the regression lines between instar number and the intermolt period revealed no significant difference. This implies that the intermolt period decreases as instar number increases with temperature.

The logarithmically transformed regression equations (Fig. 3B) below showed a linear relationship between instar and molt increment:

15°C, logMI = -0.2075IT + 1.3203
(r^2 = 0.99, n = 6, P<0.05)
20°C, logMI = -0.1499IT + 1.544
(r^2 = 0.98, n = 6, P<0.05)
25°C, logMI = -0.09571IT + 1.6389
(r^2 = 0.98, n = 3, P<0.05)

The slopes of the linear regression equations under all three temperature regimes were significantly negative, which implies that molt increment tends to decrease as instar number increases. The results of the ANCOVA for differences in the slopes of the linear regressions of instar and molt increments among the three different temperatures were significant (ANCOVA, P<0.001), suggesting that molt increment decreases as instar number increases with temperature.

Fecundity and embryo volume

Total number of eggs counted varied from 552 to 1,355.The relationship between the number of eggs and carapace length was expressed by the equation (fecundity) y=2.7744x+0.208 (R^2=0.7961). The relationship between carapace length and the number of embryos was positive, and the slope was 2.7 (Fig. 4A). Embryo volume was 0.078 mm^3. Egg volume increased as embryo development proceeded from stage 1 to stage 5 (Fig. 4B). Egg volume gradually increased from embryo stage 1 to stage 5. A significant difference [0.78-0.15 m^3] occurred among embryonic development stages (P<0.05, ANOVA).

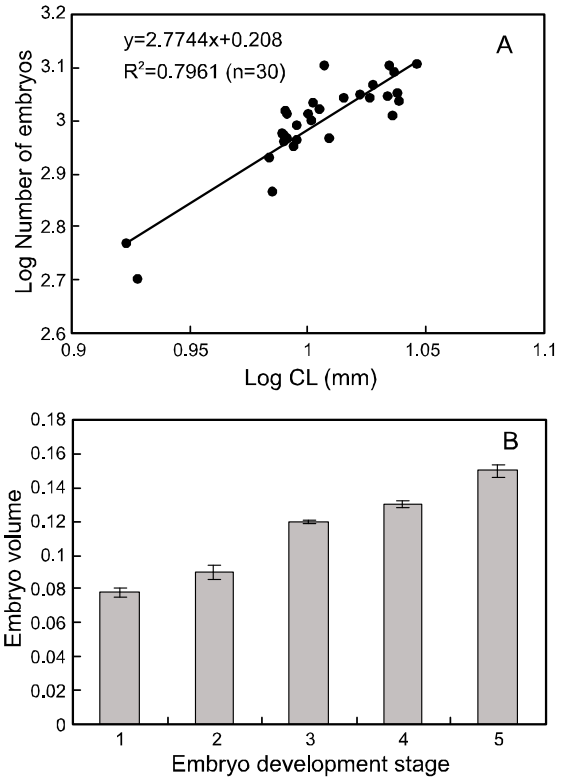

Fig. 4. *Palaemon serrifer*. Relationship between log carapace length (mm) and log number of embryos (A) and embryo volume increment with embryo developmental stages (vertical bar, standard deviation) (B).

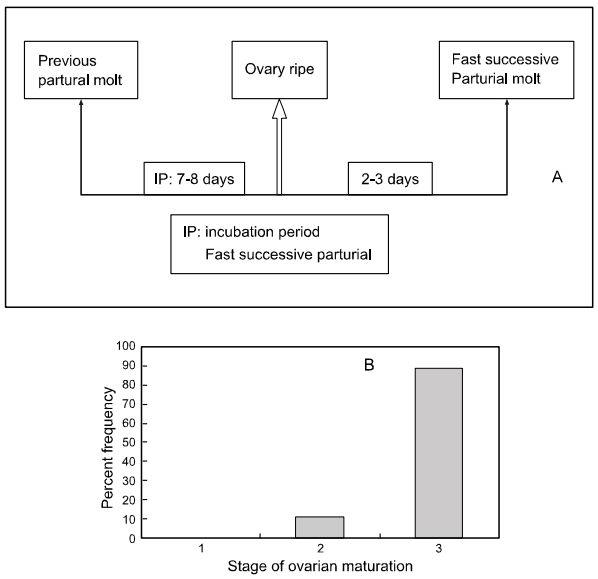

Fig. 5. *Palaemon serrrifer*. Pattern of brood production, schedule of molting, spawning, embryo incubation, hatching, ovarian maturation (A). Stage of ovarian maturation of laboratory females upon hatching of brooded embryos (B).

Pattern of brood production

The incubation period was between 7 and 8 days at 20°C. The pattern of brood production was fast, and successive parturial molts occurred (Fig. 5A). Most ovigerous females had mature ovaries when they released larvae at eclosion (Fig. 5B).

Discussion

Water temperature variation and survival rate

Water temperature followed the typical pattern of warm temperate waters and varied greatly with season. An estimation of variability in survival rate over time with a specific instar is required to assess survival rate. Knowledge of how many larvae successfully reach the post-larval stage is crucial for understanding recruitment success. Temperature greatly influences larval survival rate (Hartnoll, 1982; Sastry, 1983). Most larvae hatch at times when they will encounter foods such as phytoplankton in optimum quantities (Thorson, 1950; Bauer, 1992). In warm temperate waters, the phytoplankton bloom occurs in spring, when water temperatures have warmed sufficiently and larval hatching occurs.

We observed that more than 50% of the larvae survived to the post-larvae stage between 20ºC and 25°C. The optimum temperature range for maximum larval survival of *P. serrifer* occurred in spring and summer, largely overlapping with that for larvae of other species.

Larvae of *Crangon crangon, Palaemon elegans* and *Processa nouveli* have been reported to exhibit higher survival rates at 20°C compared with 15°C (Rochanaburanon and Williamson, 1976). Hartnoll and Mohamedeen (1987) reported similar results for larvae of the British crabs *Inachus dorsettensis* and *Pilumnus hirtellus*. For *Palaemon serratus* the optimum survival occurred between 22°C and 26°C (Reeve, 1969).

Intermolt period and molt increment

Intermolt period increased with instar number, and it was longest at 15°C and shortest at 25°C. This is a typical growth pattern in crustaceans, in which the intermolt period steadily increases with premolt size at all temperatures (Hartnoll, 1982). The longer intermolt period reflects the necessity of larvae to spend additional time accumulating sufficient nutriation (Mauchline, 1976, 1977; Hartnoll, 1982). These results suggest that the intermolt period of larvae was influenced by temperature and size.

Intermolt period continuously increased at three different rearing temperatures, whereas the molt increment decreased as the instar number increased.

Temperature influences the molt increment as well as the intermolt period in crustaceans (Hartnoll, 1982). We found that the molt increment increased per instar with temperature. For *Orconectes limnosis, Carcinus, maenas*, and *Phronima sedentaria*, temperature did not influence molt increment (Kracht, 1974; Larval, 1975), whereas for *Eriocheir sinensis* (Leersnyder, 1972) and *Rhithropanopeus harrisii* (Hartnoll, 1978), molt increment increased with increasing temperature from 15-20°C to 20-30°C, which is consistent with the molt increment pattern of *P. serrifer* larvae. Research on *P. elegans* and *P. serratus* found similar results (Salama and Hartnoll, 1992; Reeve, 1969; Forster, 1973).

Embryo volume, fecundity and pattern of brood production

In *P. serrifer*, the egg volume was rather small in comparison with other palaemonid species such as *Palaemon northropi* (Rankin 1898), *Palaemonetes intermedius* (Holthuis, 1949) *Leander tenuicornis* (Say, 1818), *and Macrobrachium ohione* (Smith, 1874), which showed volumes of 0.200 mm^3, 0.294 mm^3, 0.163 mm^3, and 0.080 mm^3, respectively (Corey and Reid, 1991).

The trade-off relationship between egg volume and fecundity is explained by energy allocation in females

(Bauer, 2004; Kim and Hong, 2004). Egg volume is correlated with life-history traits and environmental variables in general, and it can be influenced and selected by those variables. For example, if the egg volume is relatively small, then high fecundity and a long larval development period with obligate planktotrophy ensue (Bauer, 2004).

However, in the present study, although embryo volume was rather low (0.078 m^3 in the first larval stage), and the fecundity (number of eggs) was not high, the larval nutritional mode of *P. serrifer* was planktotrophic and there were six larval stages, implying a small number of stages compared with the 12 stages observed in *Palaemon. gravieri* (Kim, 2005). This indicates that the larval developmental period was relatively short.

Many caridean females are continuous or successsive breeders (Bauer, 1976, 2004). *P. serrifer* was also observed to be a fast successive breeder. Continuous breeders are females that carry embryos near hatching. They have a mature pre-spawning ovary and undergo a parturial molt within one or two days of eclosion. Continuous (fast successive) breeding has been reported in females of many caridean species including *Heptacarpus sitchensis*, *H. palundicola* (Bauer, 1976, 2004), and *Macrobrachium rosenbergii* (Wickins and Beard, 1974).

In conclusion, egg volume appeared to be correlated with the number of embryos, larval type, and length of the development period. The egg volume of *P. serrifer* appeared to exhibit the greatest influence over egg production compared with other factors. However, the patterns of *P. serrifer* did not completely conform to the reported general patterns such as small embryo size, high fecundity, and long developmental period, thus demonstrating the need for further research on this species.

Acknowledgements

This research was supported by Tanhae Aquatic Resources Research Institute and the Kunsan National University.

References

Anger K. 1991. Developmental changes in the bioenergetics of decapod larvae. Mem Queensl Mus 31, 289-308.

Anger K and Moreira GS. 1998. Morphometric and reproductive traits of tropical carideans shrimps. J Crust Biol 18, 823-838.

Bauer RT. 1976. Mating behaviour and spermatophore transfer in the shrimp *Heptarcarpus pictus* (Stimpson) Decapoda: Caridea: Hippolytidae) J Nat Hist 10, 415-440.

Bauer R. 1992. Testing generalization about latitudinal variation in reproduction and recruitment patterns with sicyoniid and caridean shrimp species. Invert Reprod Develop 22, 193-202.

Bauer RT. 2000. Patterns of brood production in the grass shrimp *Palaemonetes pugio* (Decapoda: Caridea). Invert Reprod Develop 38, 107-113.

Bauer RT. 2004. Remarkable shrimps, adaptations and natural history of the carideans. Animal Natural History Series, Vol. 7, 282pp.

Corey S.and Reid DM. 1991. Comparative fecundity of decapod crustaceans I. The fecundity of thirty-three species of nine families of caridean shrimp. Crustaceana 60, 270-294.

Costlow JD Jr. and Bookout CG. 1959. The larval development of *Callinectes sapidus* Rathbun reared in the laboratory. Biol Bull (Woods Hole, Mass) 116, 373-396.

Costlow JD Jr, Bookout CG and Monroe R. 1960. The effect of salinity and temperature on larval development of *Sesarma cinereum* (Bosc) reared in the laboratory. Biol Bull (Woods Hole, Mass.) 118, 182-202.

Emlet RB, McEdward LR and Strathmann RR. 1987. Larval ecology of caridean shrimp. Crustaceana 60, 270-294.

Forster JRM. 1973. Growth experiments with the prawn *Palaemon serratus* Pennant fed with fresh and compound foods. Fish Invest Ser II 27, 1-16.

Hartnoll RG. 1978. The effect of salinity and temperature on the post larval growth of the crab *Rithropanopeus harrisii*. In: D. S McClusky and A. J. Berry editors-in-chief, Physiology and Behaviour of Marine Organisms, Pergamon Press Oxford, pp. 349-358.

Hartnoll RG. 1982. Growth. In: L. G. Abele ed, The biology of crustacea, 2. Embryology, morphology and genetics, Academic Press, New York, pp. 111-196.

Hartnoll RG and Mohamedeen H. 1987. Laboratory growth of the larvae of six British crabs. J Exp Mar Biol Ecol 107, 155-170.

Holthuis LB. 1950. The Palaemonidae collected by the Siboga and Snellius Expeditions with remarks on other species. 1. Subfamily Palaemonidae. The Decapoda of the Siboga Expedition. Part 10. Siboga Expedition $39a^9$, 1-268.

Kim HS. 1977. Illustrated Flora and Fauna of Korea. 19. Macrura. Samwha Publishing Company, Seoul, 694 pp.

Kim SH and Hong SY. 2004. Reproductive biology of *Palaemon gravieri* (Decapoda: Caridea: Palaemonidae). J Crust Biol 24, 121-130.

Kim SH. 2005. Survival rate and growth of *Palaemon gravieri* larvae reared in the laboratory (Decapoda: Caridea: Palaemonidae) J Fish Sc Technol 8, 90-96.

Kracht D. 1974. Modification du rhythme des mues de la croissance totale et de l apparition des caractéres sexuels externs adltes chez l Ecrevisse *Orconectes limnosis* (Rafinesque) juvenile, élévée a 28 depuis l âge de un an. C. R. Hebd. Sean Acad Sci 178, 381-384.

Larval P. 1975. Une analyse multivariable du développement au laboratoire de *Phronima sedentaria* (Forsk.), Amphipode Hypéride. Etude de l'influence de la temperature et de la quantité de nourriture. Ann Inst Oceanogr 51, 5-41.

Lebour MV. 1927. Studies on the Plymouth Brachyura.1. The rearing of crabs in captivity, with a description of the larval stages of *Inachus dorsettensis, Macropodia logirostris* and *Maja squinado*. J Mar Biol Ass UK 14, 795-821.

Leersnyder M. 1972. Influence de la témperature sur la fréquence des mues et le taux de croissance d *Eriocheir sinensis* H. Milne Edwards (Crustacé Dévelopement au laboratorie de *Phronima sedentaria* (Forsk.) amphipode hypéride. Etude de ltempérature et de la quantité de nourriture. Ann Ins Océanogr 51, 5-41.

Mauchline J. 1976. The Hiatt growth diagram for crustacean. Mar Biol 35, 79-84.

Mauchline J. 1977. Growth in shrimps, crabs and lobsters--an assessment. J Cons Int Explor Mer 37, 162-169.

Mohamedeen H and Hartnoll. RG. 1989. Larval and postlarval growth of individually reared specimen of the common shore crab *Carcinus maenas* (L.). J Exp Mar Biol Ecol 134, 1-24.

Pearse JS, JB McClintock and Bosch I. 1991. Reproduction of Antarctic marine benthic invertebrates: Temposs, Modes and timing. Am Zool 31, 65-80.

Reeve MR. 1969. The laboratory culture of the prawn *Palaemon serratus* Fish Invest Ser. II, 26, 1-37.

Rochanaburanon T and Williamson DI. 1976. Laboratory survival of larvae of *Palaemon elegans* Rathke and other caridean shrimps in relation to their distribution and ecology. Estuar Coast Mar Sci 4, 83-91.

Rothlisberg PC. 1979. Combined effects of temperature and salinity on the survival and growth of the larvae of *Pandalus jordani* (Decapoda: Pandalidae). Mar Biol 54, 125-134.

Salama AJ and Hartnoll RG. 1992. Effects of food and feeding regime on the growth and survival of the prawn *Palaemon elegans* Rathke, 1837 (Decapoda, Caridea). Crustaceana 63, 11-22.

Sastry AN. 1983. Ecological aspects of reproduction. In: F. J. Vernberg & W. B. Verberg eds, The biology of crustacean, Vol. 8. Environmental adaptations. Academic Press, New York, pp. 179-270.

Thorson G. 1950. Reproductive and larval ecology of marine bottom invertebrates. Biol Rev 25, 1-45.

Vance RR. 1973a. On reproductive strategies in marine benthic invertebrates. Am Nat 107, 339-352.

Wickins JF and Beard TW. 1974. Observations on the breeding and growth of the giant freshwater prawn *Macrobrachium rosenbergii* (De Man) in the laboratory. Aquaculture 3, 159-174.

Wray GA. and Raff RA. 1991. The evolution of developmental strategy in marine invertebrates. Trends Ecol Evol 6, 45-50.

Zar JH. 1996. Biostatistical Analysis. Prentice-Hall, New Jersey, pp. 1-662.

(Received26 February 2010; Revised 17 November 2010; Accepted 22 November 2010)