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Instars Number and Instars Duration during Larval Life Cycle of *Macrobrachium gangeticum* (Bate)

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Abstract

This experiment was conducted in 250 ml beakers and 300 L plastic pools to find out the instars duration (duration between two moults), total duration and instars number (larval moult) during the larval cycle of *Macrobrachium gangeticum.* The instars duration from stages I-V was 8-14 days (10.4 ± 2.88 days) recorded in 250 ml beakers which was comparatively longer than that of the plastic pools at 5-7 days (5.75± 0.957 days). Similar trend with a slight increase of 12-16 days (14.2 ±1.788 days) in instars duration in beaker was observed between stages V-VI and found much shorter duration of 5-6 days (5.25±0.5 days) in plastic pools. Further, a comparatively longer instars duration of 19-27 days (22.6 ±1.788 days) was observed between stages VI-XI in beaker than in plastic pools of 8-10 days (9.0±1.154 days). Same trend existed at later stages XI-PL, which was slightly longer in both treatments i.e. 9-10 days (9.6 ± 0.547 days), in beakers and 2-3 days (2.5±0.577 days) in plastic pools. The total instars duration of 49-62 days (57.4± 4.929 days) from stage I-PL was recorded in 250 ml beakers which was comparatively much longer than that of 300 l plastic pools of 20-25 days (22.5 ± 2.380 days).

The instars number (number of moults) varied significantly during the larval cycle of *M. gangeticum* carried out in 250 ml beakers. Zoea I passed through 11 distinct larval stages to attain post larval stage. A minimum of 4 moults from stages I-V, and a maximum of 5-7 moults (5.8±1.095) between stages V-VI, thereafter 5-9 moults (7.4±2.28) from stages VI-XI and 4-5 moults (4.25±0.44) between stages XI-PL were recorded in this species.

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Introduction

The moulting in prawns initiate immediately after the larvae come out of the eggshell. The nauplii of brackish water prawn (*Penaeus monodon)* passed through four distinct larval stages i.e. nauplii, protozoea, mysis and post larvae, whereas in freshwater, prawn zoea hatched out directly from the eggshell, passed through distinct zoeal stages before metamorphosing into post larvae (Kanaujia 2006; Kanaujia et al. 2001; 2007). Growth and development of the Decapods crustaceans depend upon the process of moulting. They eat food to develop their body tissue, since their body is covered with hard chitinous, it is difficult for them to expand their body. Therefore, frequent moulting occurs to stretch their body to attain growth. The moulting process starts immediately after hatching of zoea stage I from the eggs, which is frequent in early stages and gradually slowed down at later stages. In the end there is no moult, animal became sluggish and inactive, and died due to ciliates and fungal infection (New and Singholka 1985; New and Valenti 2000). Further larger freshwater prawns *Macrobrachium* species are mostly estuary bound and they need some amount of salinity for the completion of their larval cycle (Kanaujia 1998; 1999; Kanaujia and Mohanty 1992; 2001; Kanaujia et al. 2001; 2002; 2005; 2007). The prawns of this group grew, reached maturity, spawned and incubated the fertilized eggs and larvae hatched out in freshwater under natural riverine and freshwater pond conditions. The water current drifts out these larvae to the estuary for the completion of the different zoeal stages (Ibrahim 1962; Kanaujia and Mohanty 1992; Kanaujia et al. 2005). As soon as zoea attained post larval stage they left the saline zone and ascended to the upper riverine stretches against water current and migrated for long distance in the riverine system. This group of prawns passed through distinct larval stages through several moults in different durations depending upon the food, water temperatures and water quality (Kanaujia and Mohanty 1992; Kanaujia 2006; Kanaujia et al. 2005; 2007; Prasad and Kanaujia 2006). The present study was conducted under laboratory and hatchery conditions to find out the number of moults (instars number) and instars duration between subsequent moults in gangetic prawn *Macrobrachium gangeticum*.

Materials and Methods

The experiment was conducted under laboratory conditions at room temperature (28-31ºC) with the provision of sufficient natural sunlight for a few hours in the evening (4-6 PM) and artificial flourescent tube light during day time for 12 hours (6AM-18 PM). The newly hatched individual zoea stage I was inoculated in 20 individual glass beakers (250 ml) cleaned properly and filled with 12 ppt brackish water. Each beaker was half dipped into 20 L plastic tub half filled with freshwater to maintain water temperature. Initial size of the larvae was recorded using an occular micrometer through a compound microscope placed on cavity slide. The larvae were fed twice daily with freshly hatched *Artemia* nauplii at 15 nauplii per prawn larva in the morning at 6AM and in the evening at 6PM. Once the larvae attained stage VI, the *Artemia* nauplii was fed 4 times at 6 hours interval with the same quantity. To maintain suitable water quality, the water medium was changed daily in the morning between 6-7 AM and individual larva was removed from individual beaker using a wide mouthed dropper and released into another beaker filled in fresh medium of same salinity. While shaking the beaker, before water exchange, moulting shell and larva were observed daily with the help of hand lens. Once larva attained stage VI, it was transferred to 500 ml beaker to provide more space to the developing larva. Once the moulting shell was observed, the size and distinguishing features of individual's larval stages were observed through a compound microscope. In this process size, characters, number of instars and instars duration of individual larva from each beaker were recorded till the larva metamorphosed into post larvae.

Further observation was made on instars duration between two subsequent larval stages from four larval rearing tanks operated for seed production in 300l plastic pools. To observe distinguishing stages, individual larva from the plastic pools were collected daily in the morning hours between 6-7AM using a wide mouthed dropper and kept on cavity slide placed under a compound microscope. During observation of the larval stages, a bigger size larva was segregated and collected. Once the larva attained subsequent stage the total duration between two stages was recorded. In this process the duration of each larval stage was recorded from zoea stage I - post larvae and compared it with instars duration observed in beakers.

Results

The data on instars duration collected from 20 individual larvae carried out in 250 ml beakers are presented in figure 1. Variations in instars duration from stage I -V were observed in the larva of individual beaker that ranged from 2-4 days. Maximum instars duration of 12-16 days was recorded between the zoea stages V-VI. Instars duration of 3-4 days from stages VI-VII was comparatively shorter than the earlier stages V-VI. More or less similar instars duration of 2-9 days was recorded from stages VII – X and a maximum of 9-10 days (9.6 ± 0.547days) between stages XI- PL.

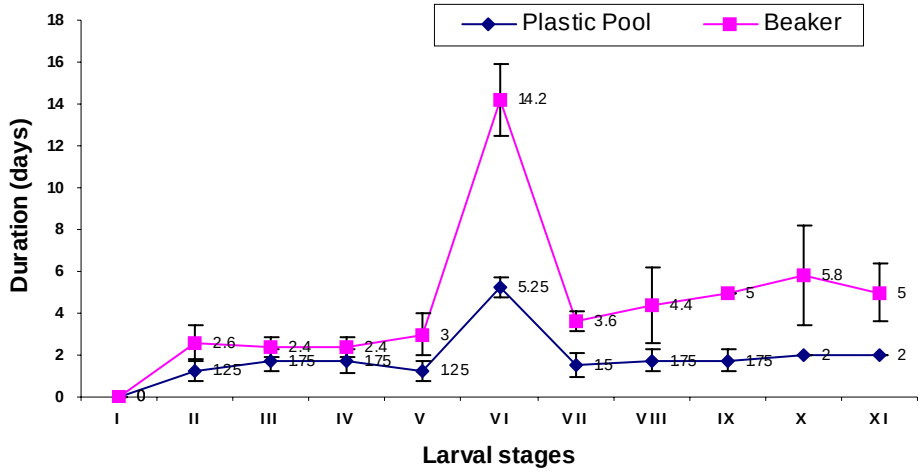

Figure 1. Mean instars duration between two consecutive larval stages from stages I-Pl of *M. gangeticum* in 300 L plastic pools and 250 ml beakers

The data on average instars duration recorded from 300 L plastic pools are presented in figure 1. Zoea attained stages I - II within 1.25±0.50 days and similar trend was observed from stages II – III (1.75 ±0.50 days), III - IV (1.75 ±0.57 days) and IV - V (1.25± 0.50 days). Maximum 5.25 ±0.50 days was recorded between zoea stages V -VI. Thereafter instars duration was comparatively shorter than that of stages V-VI and found more or less similar with gradual increase from stage VI–PL which ranged from 1.5 ±0.57–2.5± 0.57days (Fig. 1).

As shown in figure 2 the instars duration between stages I -V in 250 ml beakers ranged from 8-14 days with a mean of 10.4 ± 2.880 days. Slight increase in instars duration of 12-16 days with an average of 14.2 ±

1.788 days was recorded between stages V –VI. Instars duration of 19-27 days with an average of 22.6± 2.880 days was found in stages VI –XI which was comparatively longer than those of the earlier stages. Instars duration of 9-10 days (9.6.± 0.547 days) in 20 larvae was recorded during later stages XI -PL. A total duration of 49- 62 days with an average of 57.4 ± 4.929 days was recorded from stages I -PL the trend of instars duration indicated short duration during early stages and a gradual increase at later stages (Fig. 2).

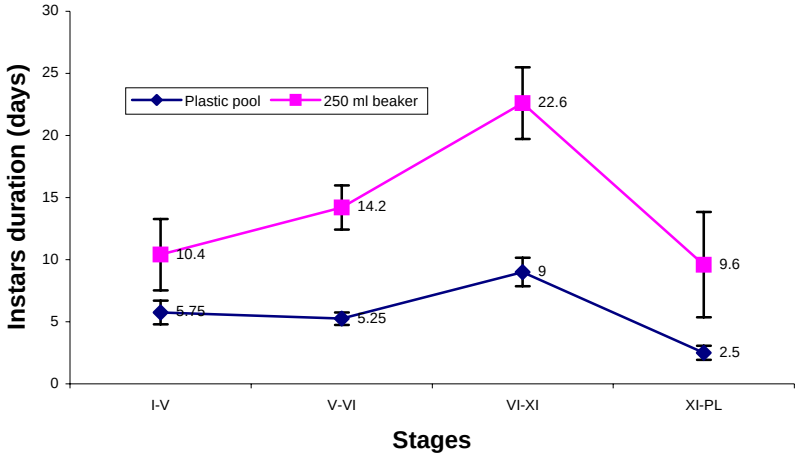

Figure 2. Mean instars duration between stages I-V, V-VI, VI-XI and XI-PL of *M. gangeticum* in 300 L plastic pools and 250 ml beakers

While considering early stages I-V, middle stage V-VI and later stages VI-PL in 300 l plastic pools, the total duration between stages I -V ranged from 5-7 days with a mean of 5.75 ± 0.957days (Fig. 2). A slightly short duration of 5-6 days with an average of 5.25 ± 0.50 days was recorded between stages V –VI in the middle and comparatively longer than those of early stages (I-V). The duration from stages VI – XI recorded 8-10 days with an average of 9.0 ±1.154 days and 2-3 days with an average of 2.5 ±0.577 days from XI-PL (Fig. 2). Total instars duration 20-25 days (22.5±2.380 days) in 300 L plastic pool was found much shorter than that of 250 ml beakers.

Instars number in beakers

The number of moults (instars number) recorded between two subsequent stages from stages I-PL is indicated in figure 3. One moult was

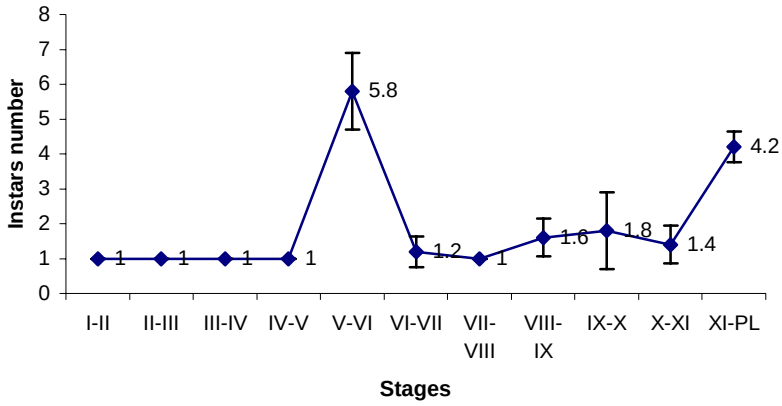

Figure 3: Mean number of instars between two consecutive larval stages of *Macrobrachium gangeticum* reared in 250 ml beakers

observed in between subsequent stages from stages I-V and a maximum of 5-7 moults between stages V-VI. Thereafter 1-2 moults between stages VI -VII and 1 moult between stages VI -IX and 1-3 moults between stages IX -X, it was reduced further to 1-2 moults between stages X-XI and recorded a maximum of 4-5 moults between stages XI -PL. As indicated in figure 4, while considering the total moults during early, middle and later stages from stages I –PL. The total instars number in 20 trials during stages I -V recorded 4 days that increased to 5-7 moults with an average of 5.8 ± 1.09 moults between stages V-VI and a slight increase in the number of 5-9 moults with a mean of 7.4± 2.28 moults recorded during stages VI-XI. Further 4-5 moults with a mean of 4.25 ± 0.44 moults were recorded between stages XI -PL. The total number of moults from stages I -PL ranged from 18- 25 moults with a mean of 21.0 ± 3.31 moults during the larval cycle of *M. gangeticum*. As shown in figures 1 and 4, the number of moults as well as the duration at each larval stage recorded a minimum at early stages, which increased gradually at later stages.

Discussion

Instars duration

The instars duration during each larval stage and the total duration of 49-62 days (58 ± 5.029 days) recorded during the larval cycle of *M. gangeticum* in 250 ml beakers were comparatively much longer than those

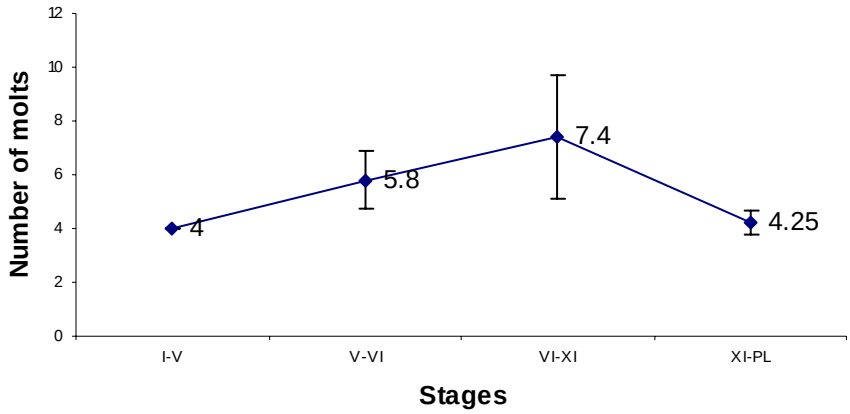

Figure 4. Mean number of moults between stages I-V, V-VI, VI-XI and XI-PL in 250 ml beakers

recorded in 300 L plastic pools in 20-25 days (23.25 ± 2.75 days). Twenty to twenty six days total instars duration recorded during the larval cycle of *M. gangaticum* in 300 L plastic pool was much earlier than that of the observations made in 250 ml beakers of 49-62 days. It may be concluded that as compared to plastic pools, the animal takes longer duration as well as more number of instars in small containers for the completion of the larval life cycle. This may be perhaps due to small containers (250 ml beakers) used for larval rearing. The instars duration at different larval stages recorded in 300l plastic pool varied significantly. The total duration during larval cycle from stages I-PL recorded as the period of occurrence of first post larvae during which, most of the larvae in the tanks were found in advanced stages. More or less similar observation during 19-26 days and 22 days were made by Kanaujia et al. (2005; 2007) and Prasad (2005) in the same species. However variation in total instars duration/occurrence of first few post larvae were similar with that of *M. rosenbergii* as recorded by Uno and Sao (1969) and differed from those of other *Macrobrachium* species. Kanaujia and Mohanty (1992; 1999) recorded longer duration with wide variations in occurrence of the first few post larvae in *M. malcolmsonii* (40 and 53 days) in natural seawater and 41 days in artificial seawater. Further, Mohapatra (2001) obtained within 39-42 days, Sankolli et al. (1984) in 22-35 days, Rao (1991) in 52 days and Yadav (1993) within 46-72 days in *M. malcolmsonii*. Such variations in instars duration during the larval cycle of three *Macrobrachium* species may be due to variations in genetic constitution.

The variations in total instar duration at different larval stages for attaining post-larval stage in *M. gangeticum* were recorded much less than those of *M. malcolmsonii* and *M. rosenbergii* (Ling 1964; 1969); Kanaujia and Mohanty 1992; Kanaujia 1998; 1999). The instars duration between the stages is monitored through moulting, which occurs in all larvae uniformly depending upon the feed and water quality and delay in moulting at later stages either due to genetical variations or unhealthy conditions of the mother prawns (Sankaran and Nair 1992).

Instars number (moults)

In the present study 11 larval stages with 16 moults were recorded in *M. gangeticum.* The number of moults undertaken by the larvae at each stage varied significantly. Ling (1969) recorded 8 larval stages in *M. rosenbergii*, while Uno and Sao (1969) and Gomez and Kasahara (1987) recorded 11 and 17, respectively. Kewalramani et al. (1971) recorded 15 moults and 16 zoeal stages in *M. malcolmsonii* to reach post larval stage. Sankolli et al. (1984) obtained post larvae after passing through 18-20 moults. Later on many workers recorded 11 distinct zoeal stages before attaining post larval stage in *M. malcolmsonii* (Rao 1991; Kanaujia and Mohanty 1992).

Growth of freshwater prawn as well as most of the crustaceans depends on the frequency of their moulting. Visible growth in prawns is registered only at the time of moulting, which occurred immediately after all the stages of life cycle (Rao and Tripathi 1993; Kanaujia and Mohanty 1992; Kanaujia et al. 2005; 2007). The frequency of moulting is dependent on the age, quality and quantity of food consumed and environmental parameters. Moulting process is also evident in larval stages. The number of larval stages and the frequency of moults by the larvae to metamorphose into post-larval stage in different species varied significantly. The growth rate in *M. gangeticum* was observed to be more or less uniform during early zoeal stages (I–V), which is similar to that of *M. malcolmsonii* recorded by Kanaujia and Mohanty (1992), Yadav (1993), Choudhuri (1995) and Kanaujia (1998; 1999). Malecha (1985) observed the occurrence of II-V larval stages on any single day in *M. rosenbergii*. Feed density, particle size, water exchange and husbandry found responsible factors. Similar observations are made in *M. gangeticum*. Sandifer and Smith (1979) reported that the number of larval stages in *Macrobrachium* species is non-synchronous in nature and depends on various factors. Eighteen to twenty five moults with eleven distinct larval stages were recorded in *M. gangeticum* to attain post larval stage, which is similar to that of *M. malcolmsonii*

reported by Rao (1991), Kanaujia and Mohanty (1992), Kanaujia (1998; 1999; 2003) and Mohapatra (2001). But 16 larval stages and 15 moults in *M. malcolmsonii* were reported by Kewalramani et al. (1971) and 11 stages by several other workers. Stocking density, food and water quality were reported to be important factors influencing larval growth, moulting and post larval production under hatchery conditions (Rao 1991; Kanaujia 1998; 1999; Yadav 1993; Sharma 1994; Kanaujia and Mohanty 1992; Mitra 2001; Mohapatra 2001).

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