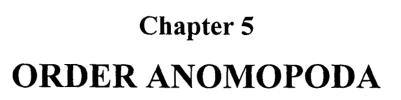
# ORDER ANOMOPODA

Britto Joseph. K "Studies on the biology of freshwater Cladocera: Crustacea" Thesis. Department of Zoology, St. Thomas College Thrissur, University of Calicut, 2007



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### Family Daphniidae Straus, 1820

### 5.1 Ceriodaphnia cornuta Sars, 1885

*Ceriodaphnia cornuta*, a common littoral species found in the freshwater ponds of India (Gurney, 1906, 1907; Arora, 1931; Brehm, 1936) is considered a synonym of *C. rigauadi* Richard by Rzoska (1956) and Nayar (1971). This species was also reported from Meghalaya (Patil, 1976); Bihar (Nasar, 1977); West Bengal (Sharma, 1978; Karnataka (Patil and Gouder, 1988), Chennai, Tamil Nadu (Raghunathan, 1990); Kerala (Michael and Sharma, 1988); Lake Kolleru, Andhra Pradesh (Durga Prasad and Padmavathi 2003a) and Thekkady, Kerala (Babu and Nayar, 2004).

Studies on the biology of *C. cornuta* were initiated in India by Michael (1962) from West Bengal; Murugan (1975b) from Tamil Nadu; Kanaujia (1988a) from Orissa and Babu and Nayar (1993) from Kerala. Although, the above papers give good accounts of the life history of parthenogenetic females; information about the ephippia bearing females and males of this species is scanty. Hence a detailed laboratory study on the life cycle of this species has been made.

#### 5. 1. 1 External Morphology

#### Parthenogenetic female (Plate 10. Fig. C)

Body oval-shaped; head small, distinctly separated from rest of the body by a conspicuous cervical sinus, ventral margin of the head produced into a short rostrum (Plate 12. Fig. A). Eye large, ocellus small. Antennules short, fusiform and not extending beyond the tip of rostrum, with a central sensory seta somewhat distal to the middle and a group of sensory setae on the apex. The antennae well developed, with 4-segmented dorsal ramus and 3-segmented ventral ramus; setation of antenna: (0-0-1-3)/(1-1-3). Valves with distinct polygonal marking (Plate 12. Fig. A), margins smooth, posterodorsal corner produced into a blunt process. Postabdomen short, with 5-6 sharply pointed anal spines; claws smooth, without basal spine (Plate 12. Fig. B). Mean size:  $0.533 \times 0.395$  mm.

Male (Plate 10. Fig. F)

Smaller than female; body quadrangular in outline with somewhat straight dorsal margin; antennule longer than that of female, and with two sensory hairs (Plate 12. Fig. D). The first thoracic leg with a prehensile hook and a long jointed filament emerging through the ventral margin of valves (Plate 12. Fig. C). Mean size:  $0.362 \times 0.216$  mm.

#### Ephippial female (Plate 10. Fig. D)

The general features of ephippial female similar to that of parthenogenetic female except in their smaller size and more rounded body. Ephippium dorsally placed, oval, single and white in colour. Ephippium forms a part of the carapace, made up of honey-comb pattern of ornamentation (Plate 10. Fig. E). The egg is single within the ephippium with a mean size of  $0.204 \times 0.140$  mm. Mean size of ephippial female:  $0.468 \times 0.280$  mm. Size of ephippium:  $0.341 \times 0.264$  mm.

#### 5.1.2 Reproduction

The population developed in the laboratory culture comprised asexually reproducing females, ephippia bearing females and males. Among this the parthenogenetic females dominated the culture throughout the period of study.

The ephippial females appeared in the stock culture during January 2005 when the mean water temperature was 22-24°C and the culture became crowded. They were formed from the parthenogenetic females in the stock culture. These ephippial females again resumed parthenogenetic reproduction after 3-4 days. The carapace of the ephippial females get modified as ephippium and are released into the medium along with moulting. The ephippium floated on the surface for a few minutes and afterwards get adhered to the side walls of the container or sank to the bottom. The ephippium enclosed a single egg with a mean size of  $0.204 \times 0.140$  mm. The development of ephippial egg as well as the ephippium took place simultaneously. In the absence of synchronization of these two processes, some of the ephippia were released without eggs within it.

The sudden appearance and subsequent disappearance of males and ephippial females were observed in stock culture. Although, males are rarely

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obtained from the field, they appeared in stock culture when the stock culture became crowded.

#### Life cycle of parthenogenetic female

The features characteristic of the reproduction and life cycle of C. cornuta are given as follows.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a mean birth size (SaB) of  $0.232 \times 0.120$  mm. The first moulting occurred at an interval 23.10 hrs, while the second moulting took place after an interval of 23.0 hrs. The total pre-adult duration was 46.1 hrs and the mean pre-adult duration (PID) was 23.05 hrs.

#### Attainment of maturity

Although, the ovary was clearly visible at  $32 \pm 1.2$  hrs of life towards the end of  $2^{nd}$  pre-adult instar (Plate 10. Fig. B), they attained maturity and started to bear eggs after completion of  $2^{nd}$  moult at 46.1 hrs (Table 11). Hence the age at first reproduction (AFR) was 1.91 days. The SFR was  $0.426 \times 0.288$  mm.

#### Egg production

The eggs were deposited into the brood pouch after the completion of two moults. There was a gradual increase in the size of eggs and attained a mean size of 0.152 mm, due to the accumulation of yolk. They became oval shaped with green colour. During this primiparous instar (3<sup>rd</sup> instar) the eggs

were produced with an initial number of 2.0 eggs/ brood. The primiparous instar was completed in 24.2 hrs and the first generation time (FGT) is calculated as 70.3 hrs.

The females continued reproduction during the succeeding instars; and there was a steady increase in egg production to 9.3 eggs/ brood. This maximum clutch size (E max) is attained in the 8<sup>th</sup> instar (Table 11). The egg production in *C. cornuta* showed a single peak with maximum production in 8<sup>th</sup> instar (Fig. 6 b).

Subsequently there was a gradual decline in the rate of egg production upto  $11^{\text{th}}$  instar. There was a sharp fall in egg production immediately after this instar and showed variation till the penultimate instar ( $16^{\text{th}}$ ). However, the last instar ( $17^{\text{th}}$ ) was without egg production; and they continued moulting until their death (Table 11).

Each clutch produced in the adult instar consisted of 1 to 2 rows of eggs on either side of the brood pouch; with more number of eggs occupying in the upper row. The adult instar duration varied from 24.2 to 34.6 hrs (Table 11) with a mean AID of 30.0 hrs.

#### Fecundity

The relationship of egg production with instar number is represented in Fig. 6 b. The range of egg production of a single female was from 2 to 12 with an egg production of 3.36 eggs/day of adult life. The cumulative number of eggs produced ( $\Sigma$ mx) during the entire life span was 63.0 (Table 11). Fourteen broods were produced during the entire life with an average of

4.5 eggs /brood. The  $\Sigma mx$  of *C. cornuta* is linearly correlated with instar number (Fig. 6 c). The rate of egg production (REP) of *C. cornuta* is calculated as 4.8039.

#### 5.1.3. Growth

The first pre-adult neonates had a mean TL of 0.232 mm and the  $2^{nd}$  instar had TL of 0.352 mm. Primiparous stage was attained during  $3^{rd}$  instar when the mean TL was 0.426 mm. The maximum mean TL of 0.696 mm was attained at  $17^{th}$  instar (Table 12).

The mean CH during the first pre-adult instar was 0.120 mm and attained CH of 0.248 mm at 2<sup>nd</sup> instar. They attained a mean CH of 0.288 mm during the primiparous condition. Maximum CH was attained in 17<sup>th</sup> instar with CH of 0.498 mm (Table 12). During the life span each individual has undergone two pre-adult and fifteen adult moults.

The increment of TL and CH during each instar is given in Table 12. Maximum growth increment recorded during the life cycle was in 2<sup>nd</sup> instar with TL of 51.72 % and CH of 106.67 % respectively. Although, growth increment decreased subsequently, the most significant decrease in growth increment occurred only after 7<sup>th</sup> instar.

The relationship between TL, CH and instar number of *C. cornuta* has been represented in Fig. 6 a. The correlation coefficients of life history characters are given in Table 13. The TL and CH shows positive correlation (r = 0.990).

#### 5. 1. 4 Embryonic Development

The stages of embryonic development of *C. cornuta* are represented in Plate 11. The most conspicuous features of the developmental stages are given below.

Stage I: This stage is recognized by the presence of oval-shaped egg which is yellow-green in colour with a central fat droplet. Mean duration: 1.05 hrs. Mean size: 0.158 mm.

**Stage II:** The embryo is more elongated. The outer area of the embryo remains transparent; while the inner granular area is opaque due to the centrally placed yolk. Mean duration: 1.25 hrs. Mean size: 0.168 mm.

**Stage III:** The embryo shows cellular divisions. The anterior region is elongated. Mean duration: 4.7 hrs. Mean size: 0.172 mm.

**Stage IV:** During this stage head become more distinct with the appearance of rudiment of antennae. The outer area of embryo remains transparent, while the inner area is opaque due to the presence of yolk. Mean duration: 2.6 hrs. Mean size: 0.182 mm.

Stage V: During this stage head became more distinct with appearance of rudiments of antenna and thoracic legs. Mean duration: 2.0 hrs. Mean size: 0.187 mm

**Stage VI:** This stage can be recognized by the presence of a pink eye. The yolk granules start to disappear and the rudiments of antennae, thoracic legs and postabdomen are clearly visible. Mean duration: 1.5 hrs. Mean size: 0.206 mm

**Stage VII:** This stage is recognized by the presence of a black eye. The yolk granules were completely absent and the antennae, thoracic legs and postabdomen are more distinct. Mean duration: 7.9 hrs. Mean size: 0.216 mm

**Stage VIII:** During this stage eye gets enlarged, antennae gets segmented, postabdomen and shell become more distinct. Mean duration: 2.4 hrs. Mean size: 0.243 mm.

#### **Release of neonates**

The neonates are released from the brood pouch after completion of the embryonic development by the intermittent movements of its postabdomen. After this the female underwent moulting within duration of 40.0 minutes. The embryonic development of primiparous instar of *C. cornuta* was completed in 23.4 hrs.

#### 5. 1. 5 Life span and Survivorship

The survivorship curve (Fig. 6 d) indicates the relationship of age (days) and percentage survival in *C. cornuta*. This shows that the survival was higher during the pre-reproductive phase and declined during the reproductive period.

The mean life span ( $\Sigma$ lx) of female is calculated as 8.23 days, while the maximum life span (L max) of females observed during the present study was 20.66 days.

### 5. 2 Scapholeberis kingi Sars, 1903

Scapholeberis kingi is a hyponeustonic cladoceran often found associated with the surface films of shallow water bodies. *S. kingi* is a darkly pigmented cladoceran which is considered a distasteful species (Dodson and Frey, 1991). This species was described first by O. F. Müller (1776) from Denmark. However, the name *Scapholeberis* was coined by Schoedler (1858).

The first report of this species from India is that of Gurney (1907) from Culcutta. Subsequently reported from Kashmir and Nilagiri Hills, (Brehm, 1936); Dharwad, (Patil and Gouder, 1989); and Andhra Pradesh, (Durga Prasad and Padmavathy, 2003a).

The only available report on the biology of this species is that of Murugan and Sivaramakrishnan (1976) from Tamil Nadu.

#### 5. 2. 1. External Morphology

#### **Parthenogenetic female** (Plate 13. Fig. C)

Body quadrangular, broadly rounded dorsally, maximum height behind the middle. Head relatively small and slightly depressed, rostrum rounded and slightly projecting ventrally (Plate 12. Fig. F). Eye rather large; small ocellus, situated closer to the rostrum than to the eye. Antennules short, attached to the posterior margin of the rostrum, with long sensory setae on the anterior surface and a group of 5-7 sensory setae on the apex. Antennae with 3segmented dorsal ramus and 3-segmented ventral ramus; setation of antenna: (1-1-3)/(0, 1, 3). Valves with faint reticulations, posterior margin straight vertically with distinct posterodorsal corner; ventral margin straight horizontally with a series of sub-marginal branched setae; the posteroventral corner of each valve produced into a short spine (Plate 13. Fig. A). Postabdomen short and broad, dorsal distal margin rounded with 5 marginal denticles, decreasing in size proximally (Plate 12. Fig. G); lateral surface has rows of spinules; claw slightly curved dorsally, with a series of spinules on the concave surface. Mean size:  $0.591 \times 0.383$  mm

#### Ephippial female (Plate 13. Fig. D)

The body length and carapace height of ephippial female of almost same dimensions. Ephippium dorsally placed, with straight posterodorsal margin and semi-circular ventral margin, having several rows of honeycomb ornamentation; darkly pigmented; enclosing one resting egg with elliptical shape (Plate 13. Fig. E; Plate 33. Fig. C). Ephippial females were also seen with ephippia having no eggs within it. Mean size: 0.648  $\times$  0.609 mm. Mean size of ephippium: 0.478 $\times$ 0.303 mm.

#### 5.2.2 Reproduction

S. kingi formed an aggregation in the surface film of water when collected from Site 4, as well as when cultured in the laboratory indicating that they are adapted for hyponeustonic life. The presence of specialized setae on the ventral surface of the valves assists their movements in the surface water film (Plate 12. Fig. F).

The population developed during the laboratory culture comprised asexually reproducing females and ephippia bearing females, while the males were not observed. Among this the parthenogenetic females dominated the culture. Ephippial females appeared in December 2003 when the water temperature was 23°C and the population became crowded. The presence of ephippial females could be identified due to the dark pigmentation of ephippium. The ephippium is placed dorsally with a straight posterodorsal margin and a semicircular ventral margin (Plate 13.Fig. D). Ephippial females disappeared suddenly from the stock culture after a brief period of appearance.

#### Life cycle of parthenogenetic female

The parthenogenetic reproduction was the dominant method of reproduction in *S. kingi* throughout the period of study. The features characteristic of the reproduction and life cycle are given as follows.

#### Moulting

S. kingi underwent moulting towards the termination of each instar with casting off old carapace as exuvium. The detachment of old carapace is initiated from the posteroventral region of the body. During this event the animal remained in the water surface with its ventral side facing upwards to enable movement through the surface film. The casting off the exuvium is aided by the jerky movements of the body. These movements resulted in the detachment of old carapace (exuvium) first from the postabdomen and head; and subsequently from the remaining portion attached to the ventral side of the body and finally from the posterodorsal part. The exuvium is shed as a whole and the moulting process is completed in a mean duration of 2.0 minutes. There was an increase in size followed by each moult indicating that growth in *S. kingi* is associated with moulting.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a mean birth size (SaB) of  $0.290 \times 0.187$  mm. The first and second moulting occurred in an interval of 32.0 hrs each. The total pre-adult instar duration was 64.0 hrs and the mean PID was 32.0 hrs.

#### Attainment of maturity

The ovary of the parthenogenetic females were conspicuous as a pair of orange coloured elongated sacs on either side of the alimentary canal at 48  $\pm$  2.0 hrs of life during the 2<sup>nd</sup> pre-adult instar (Plate 13. Fig.B). The ovary appeared orange coloured due to the accumulation of lipid prior to egg production. However, they started to bear eggs after the second moult, and hence the AFR was 2.7 days. The SFR was 0.483× 0.283 mm.

#### **Egg production**

The mature females bear eggs after the  $2^{nd}$  moulting. In the primiparous instar ( $3^{rd}$  instar), there is a lower egg production. The eggs were subsequently deposited into the brood pouch which measured a mean length of 0.160 mm. Soon after the deposition into the brood pouch there is an increase in egg size due to the initiation of embryonic development. The primiparous instar was completed in 34.5 hrs. The time required for the release of the first batch of neonates (FGT) is 98.5 hrs.

There was a steady increase in egg production during the subsequent instars and attained a maximum clutch size of 14 eggs/ brood. This maximum clutch size (Emax) was attained in the  $10^{\text{th}}$  instar (Table 14). The egg production of *S. kingi* showed a single peak with maximum egg production in  $10^{\text{th}}$  instar followed by a gradual decline (Fig. 7 b). The egg production continued throughout the life span (Table 14).

Each clutch in the early adult instar consisted of 1 to 2 rows of eggs which were deposited on either side of the brood pouch. The female continued moulting till the end of life span. The adult instar duration varied from 34.5 to 39.5, with a mean adult duration (AID) of 36.68 hrs.

#### Fecundity

The relationship between egg production and instar number is represented in Fig. 7 b. The range of egg production of a single female varied from 1 to 16 with an egg production of 4.85 eggs/day of adult life. The cumulative number of eggs ( $\Sigma$ mx) produced during the entire life was 81.5 (Table 14). Eleven broods were produced during the entire life with a mean of 7.40 eggs/ brood. The cumulative egg production ( $\Sigma$ mx) is linearly correlated with instar number (Fig.7 c). The rate of egg production (REP) of *S. kingi* is calculated as 7.3434.

#### 5. 2. 3 Growth

The first pre-adult neonates had a mean TL of 0.290 mm and  $2^{nd}$  instar had TL of 0.376 mm. Primiparous stage was attained during the  $3^{rd}$  instar

when the mean TL was 0.483 mm. The maximum mean TL of 0.731 mm was attained at the end of 13<sup>th</sup> instar (Table 15).

The mean CH during the first pre-adult instar was 0.187 mm and attained CH of 0.226 mm at 2<sup>nd</sup> instar. They attained a mean CH of 0.283 mm during the primiparous instar. Maximum CH was attained in 13<sup>th</sup> instar with CH of 0.496 mm (Table 15). During the life span each individual has undergone two pre-adult and eleven adult moults.

The increment of TL and CH during each instar is given in Table 15. Maximum growth increment recorded during the life cycle was in 2<sup>nd</sup> instar with TL of 29.66% and in 3<sup>rd</sup> instar with CH of 25.22% respectively, and the growth increment decreased in the succeeding instars.

The relationship between TL, CH and instar number of *S. kingi* is represented in Fig. 7 a. A higher growth rate is noticed during the early instars than in the later phase of life cycle. The correlation coefficients for some life history characters are given in Table 16. The TL and CH are positively correlated (r = 0.990).

#### 5. 2. 4 Embryonic Development

The transfer of egg from the ovary into the brood pouch could be regarded as the first event of embryonic development. In *S. kingi* the egg transfer took place after  $2^{nd}$  moult within a short duration of 48-50 minutes. A similar pattern was also observed in the succeeding instars.

The stages of embryonic development in *S. kingi* are represented in Plate 14. The most conspicuous features of the developmental stages are given below.

Stage I: This stage is recognized by the presence of oval-shaped eggs with yellow-green colour. A fat globule was centrally placed. Mean duration: 1.4 hrs. Mean size: 0.116 mm

**Stage II:** This stage is recognized by the elongated embryo with yellow-green colour. The outer area of the embryo is transparent while the inner area is granular with a centrally located fat globule. Mean duration: 1. 2 hrs. Mean size: 0.146 mm

**Stage III:** The anterior region of embryo is elongated and cellular divisions are initiated. The inner granular zone of the embryo contained abundant fat droplets.

Mean duration: 6.2 hrs. Mean size: 0.198 mm.

**Stage IV:** The embryo has elongated anterio-posteriorly and shows cellular divisions. The head lobe and antennary rudiments appear. The yolk is concentrated in the centre during this stage.

Mean duration: 10.6 hrs. Mean size: 0.210 mm

Stage V: This stage can be recognized by the presence of pink eye. The head lobe, antennae and rudiments of thoracic legs become more clearly visible.

Mean duration: 3.20 hrs. Mean size: 0.214 mm

Stage VI: The eye, head, antennae, postabdomen and valves become more distinct.

Mean duration: 7.0 hrs. Mean size: 0.216 mm

Stage VII: This stage is characterized by the presence of distinct eye, antennae and postabdomen. The antennae are segmented and bear small setae.

Mean duration: 3.0 hrs. Mean length: 0.232 mm

Stage VIII: This stage is characterized by the presence of distinct eye, antennules, antennae, thoracic legs and postabdomen.

Mean duration: 1.10 hrs. Mean length: 0.242 mm

#### **Release of neonates:**

The embryonic development of primiparous instar of *S. kingi* was completed in 33.7 hrs. The neonates were released from the brood pouch by the jerking movements of postabdomen and they resembled the adult with a mean length of 0.290 mm.

#### 5. 2. 5 Life span and Survivorship

The survivorship curve (Fig.7 d) indicates the relationship of age (days) and percentage survival of *S. kingi*. The mean life span ( $\Sigma$ lx) of female is calculated as 8.14 days; while the maximum life span (Lmax) observed during the present study was 19.47 days.

### 5. 3 Simocephalus serrulatus (Koch, 1841)

*Simocephalus serrulatus* is a large cladoceran most often found among the littoral weeds and sediments of ponds. The first report of this species from India is by Michael and Sharma (1988) from Meghalaya and Tamil Nadu; and later recorded from West Bengal and Southern Tamil Nadu by Venkataraman (1995, 1999).

Our information on the biology and life cycle of *Simocephalus* is based on the studies by Murugan and Sivaramakrishnan (1973) and Murugan (1977) on *S. acutirostratus* from Tamil Nadu; further by Kanaujia (1987, 1988b) on *S. vetulus* from Orissa and Sharma and Sharma (1989) on *S. exspinosus* from Shillong, Meghalaya. Subsequently Babu and Nayar (1997) made a study on the life cycle of male and parthenogenetic females of *S. serrulatus* collected from Kerala. The present study is a detailed investigation on the life cycle of *S. serrulatus*.

#### 5. 3. 1 External Morphology

#### Parthenogenetic female (Plate 16. Fig. B)

Body rhomboidal and slightly widened behind (Plate 15. Fig. A.); dorsal margin of carapace evenly arched while ventral margin bulging in middle, with a blunt posterior protuberance slightly above median axis of body. Head projecting and acute anteriorly with serrations on the apex; rostrum short (Plate 15. Fig. B). Eye comparatively large, ocellus small and having an oval or triangular shape, situated closer to the apex of rostrum than to the eye. Antennules short, serrated on the anterior margin and with a group of terminal sensory setae (Plate 15.Fig. B). Antennal formula: (0-0-1-3)/ (1-1-3). Valves with faint reticulations. Postabdomen broad, post-anal margin armed with 8 anal denticles, decreasing in size proximally up to the pre-anal corner, lateral surface covered with scattered groups of short spinules; claw long and slender with two groups of spinules on its concave surface (Plate 15. Fig. C). Mean size: 1.730 × 1.150 mm.

#### Male

The body smaller than female, quadrangular shaped, without distinct posterior protuberance (Plate 15. Fig. D). The antennules bear two sensory setae on the middle (Plate 15. Fig. E). Postabdomen with reduced pre-anal margin, post-anal margin armed with 3 anal denticles, decreasing in size proximally up to the pre-anal corner (Plate 15, Fig. F). Mean size: 0.680 mm.

#### Ephippial female (Plate 16. Fig. C)

The ephippial females smaller than parthenogenetic females; and without the blunt posterior spine of carapace. The ephippium triangular, orange-yellow coloured, with honeycomb ornamentation, darkly pigmented, enclosing a single egg (Plate 16. Fig. D).

The egg distinctly orange coloured, measured a size of  $0.390 \times 0.200$ mm, and encased by an inner transparent and an outer thick leathery membrane (Plate16. Fig. E). Size of ephippial female:  $1.55 \times 1.21$  mm. Size NB 5624 5953 BRIS of ephippium:  $0.975 \times 0.650$  mm.



#### 5.3.2 Reproduction

The population developed during the laboratory culture comprised asexually reproducing females, males and ephippia bearing females. Among this the parthenogenetic females dominated the culture. The number of ephippial females increased in the stock culture during December 2004 when the water temperature was 22-24°C.

#### **Production of ephippial female**

The ephippial females appeared in the stock culture from December 2004 to February 2005 when the culture became crowded. Generally the ephippial production is initiated immediately after the pre-adult moults. The development of egg and the ephippium most often took place simultaneously and the eggs were deposited into the ephippium. However, due to absence of synchronization of egg release and ephippial development some of the ephippia were found to be produced without eggs within it (Plate 16. Fig. F).

The ephippium appeared as triangular structure with melanin pigmentation (Plate 16. Fig. D). Two membranes were found to enclose the ephippial egg (Plate 16. Fig. E). The ephippial development was completed in duration of 65-70 hrs. When the ephippial females were transferred to fresh culture medium, many of them cast off their ephippia along with moult. The newly released ephippia floated on the surface of the medium for some time and then sank to the bottom or get adhered to the side walls of the container. After the release of the first ephippium the ephippial females continued production of ephippia for 2-3 generations and then resumed parthenogenetic reproduction.

#### Life cycle of parthenogenetic female

In the laboratory culture the females were generally found clinging on to the side walls of the container with the help of their antennal hooks. The detritus getting attached to their body is continuously removed by the movements of well developed postabdomen. They continued water filtration and feeding even when they remained stationary or while clinging to the substratum. Defecation also took place frequently.

The population of *S. serrulatus* developed during laboratory culture comprised an abundant number of parthenogenetic females. The features characteristic of the reproduction and life cycle are given as follows.

#### Moulting

The stages of moulting are represented in Plate 16. Figs. G-I. *S. serrulatus* underwent moulting towards the termination of each instar with casting off old carapace as exuvium. The detachment of old carapace is initiated from the posteroventral region of the body. The casting off the exuvium is aided by the intermittent jerky movements of the body, which is initiated from the extension of postabdomen. These movements resulted in the detachment of exuvium first from the postabdomen and head; and subsequently from the remaining portion attached to the ventral part of the body and finally from the posterodorsal part. The exuvium is shed as a whole and the moulting was completed in a mean duration of 2.5 minutes. There was an increase in size followed by each moult (Table 18), which indicates that in *S. serrulatus* growth is associated with moulting.

#### **Pre-adult** instar

The neonates produced from the parthenogenetic females had a mean birth size (SaB) of  $0.576 \times 0.296$  mm. The first, second and third moulting occurred in a uniform duration. The total pre-adult instar duration was 130.0 hrs and the mean pre-adult instar duration (PID) was 43.33 hrs. After the third moulting they attained sexual maturity and started reproduction.

#### Attainment of maturity

The ovaries of parthenogenetic females were clearly visible at  $110 \pm 3$  hrs of life. In mature females ovaries are seen as a pair of elongated sacs on each side of alimentary canal (Plate 16. Fig. B). After completion of pre-adult instar the contents of ovaries are discharged into the brood pouch through the opening present in the posterior end. This discharged mass soon attains a spherical shape to form the eggs, after completion of the  $3^{rd}$  moult in 130.0 hrs (Plate 16. Fig. B). Hence the AFR was 5.42 days. The size at first reproduction (SFR) was 1.144 × 0.880 mm.

#### **Egg production**

The females attain the primiparous instar (4<sup>th</sup> instar), and starts the egg production with an initial clutch size of 5.2 eggs/brood (Table 17). The eggs were deposited into the brood pouch after completion of three moults which contained yolk. The eggs were yellow-green coloured and measured a mean size of  $0.225 \times 0.172$  mm. The duration of primiparous instar was 44.0 hrs and the first generation time (FGT) is calculated as 174.0 hrs.

During the subsequent instars there was a steady increase in egg production to 26.2 eggs/ brood. This maximum clutch size (Emax) was attained in 11<sup>th</sup> instar (Table 17). The egg production of *S. serrulatus* showed a single peak with maximum egg production in the 11<sup>th</sup> instar followed by a sharp fall and continued throughout life (Fig. 8 b).

Each clutch in the early adult instars consisted of 1 to 3 rows of eggs placed on either side of brood pouch with higher number of eggs in each upper row. The female underwent moulting towards the end of each instar until death. The adult instar duration varied from 44.0 to 52.2 hrs with a mean AID of 48.04 hrs (Table 17).

#### Fecundity

The relationship of egg production with instar number is represented in Fig. 8 b. The range of egg production was from 4 to 30 with a mean 5.81 eggs per day of adult life. The cumulative number of eggs ( $\Sigma$ mx) produced during the entire life span was 151.3 (Table 17). Thirteen broods were produced during the entire life with a mean of 11.63 eggs\ brood.The  $\Sigma$ mx is linearly correlated with instar number (Fig. 8 c). The rate of egg production (REP) of *S. serrulatus* is calculated as 12.2752.

#### 5.3.3 Growth

The first pre-adult neonates had a mean TL of 0. 576 mm and the  $2^{nd}$  instar had TL of 0. 600 mm. During the  $3^{rd}$  instar it attained a TL of 1.108 mm. Primiparous stage was attained during the  $4^{th}$  instar when the mean TL

was 1.144 mm. The maximum mean TL of 2.280 mm was attained at the end of  $12^{\text{th}}$  instar and no further growth during succeeding instars (Table 18).

The mean CH during the first pre-adult instar was 0.296 mm and attained CH of 0.344 mm at 2<sup>nd</sup> instar. During the 3<sup>rd</sup> instar it attained a mean CH of 0.664mm. They attained a mean CH of 0.880 mm during the primiparous condition. Maximum CH was attained in 14<sup>th</sup> instar with CH of 1.586 mm and no further growth during succeeding instars (Table 17). During the life span each individual has undergone three pre-adult and thirteen adult moults.

The increment of TL and CH during each instar is given in Table 18. Maximum growth increment recorded during the life cycle was in 3<sup>rd</sup> instar with TL of 84.67% and CH of 93.02% respectively (Table 18).

The correlation coefficients of the life history characters studied in *S. serrulatus* are given in Table 19. The TL and CH are positively correlated (r = 0.988). The relationship between TL, CH and instar number of *S. serrulatus* has been represented in Fig. 8 a. A higher growth is noticed during the pre-reproductive instars than the reproductive phase of the life cycle.

#### 5. 3. 4 Embryonic Development

The stages of embryonic development of *S. serrulatus* are represented in Plate 17. The most conspicuous features of the developmental stages are given below. **Stage I:** This stage is recognized by the presence of spherical egg which is yellow-green in colour, containing yolk granules. Mean duration: 1.2 hrs. Mean size: 0.216 mm.

**Stage II:** This stage is recognized by the appearance of two distinct zones in the embryo. The outer zone becomes transparent while the inner region becomes opaque due to the accumulation of yolk towards the centre. Mean duration: 3.3 hrs. Mean size: 0.234 mm.

**Stage III:** The embryo is slightly elongated during this stage and shows cellular divisions. The divisions were clearly seen in the inner area due to the presence of yellow coloured yolk which contains abundant fat globules. Mean duration: 9.5 hrs. Mean size: 0.280 mm

**Stage IV:** The cellular divisions become more distinct and the embryo is more elongated in the anterio-posterior axis during this stage. Mean duration: 6.5 hrs. Mean size: 0.292 mm.

Stage V: During this stage the rudiments of head and antennae starts to differentiate. The embryo is covered by a membrane. Mean duration: 3.5 hrs. Mean size: 0.302 mm.

**Stage VI:** The embryonic membrane is cast off during this stage. This is followed by the appearance of pink eyes. The rudiments of head, antennae and legs become more distinct. Mean duration: 4.5 hrs. Mean size: 0.307 mm.

**Stage VII:** This stage could be recognized by the presence of black eye. The thoracic legs and postabdomen are visible. The segmentation of antennae is more distinct. Mean duration: 8.0 hrs. Mean size: 0.420 mm.

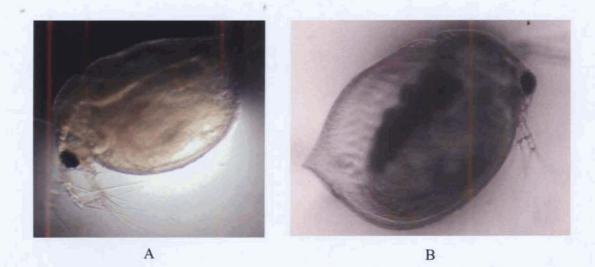
**Stage VIII:** This stage could be recognized by the disappearance of yolk and appearance of terminal setae in antennules, antennae and thoracic legs. Mean duration: 2.3 hrs. Mean size: 0.456 mm.

**Stage IX:** This stage could be recognized by the complete disappearance of yolk and appearance of distinct antennae, antennules, thoracic legs and postabdomen. Movements are also observed in the embryo during this stage. Mean duration: 1.7 hrs. Mean size: 0.496 mm.

**Release of neonates**: After completing the embryonic development the neonates were released from the brood pouch of the female by the jerking movements of its postabdomen. The neonates are also capable of movement and come out through the posteroventral region of the carapace. They swim in water by the movements of their antennae immediately after release. The embryonic development of primiparous instar of *S. serrulatus* was completed in duration of 40.5 hrs followed by subsequent moulting. Moulting was completed within duration of 3.5 minutes and it cast off the exuvia by the intermittent jerky movements of the body (Plate 16. Fig. I).

#### 5. 3. 5 Life span and Survivorship

The survivorship curve (Fig. 8 d) indicates the relationship of age (days) and percentage survival of *S. serrulatus*. As evident from the data survival was higher near the age of maturity and declined slowly further after maturity. The mean life span ( $\Sigma$ lx) of female is calculated as 13.37 days; while the maximum life span (Lmax) observed during the present study was 31.44 days.







*Ceriodaphnia cornuta* Fig. A. Pre-adult (0.367 mm), B. Female with ovary (0.387 mm), C. Parthenogenetic female (0.432 mm), D. Ephippial female (0.465 mm), E. Ephippium with egg (0.343 mm), F. Male (0.358 mm).



Stage-I (0.156 mm)

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Stage-II (0.162 mm)



Stage-III (0.176 mm)



Stage-IV (0.180 mm)



Stage-V (0.188 mm)



Stage-VI (0.202 mm)



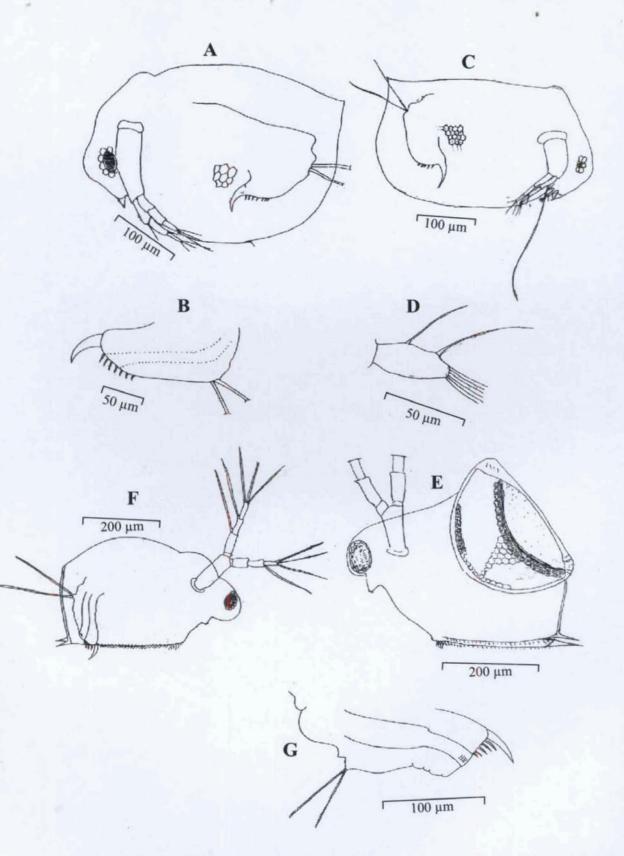
Stage-VII (0.212 mm)



Stage-VIII (0.230 mm)

Plate 12

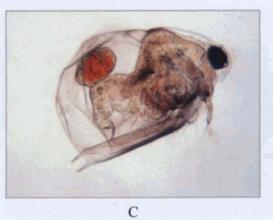
A



Ceriodaphnia cornuta Sars Fig. A. Female, B. Postabdomen of female, C. Male, D. Antennule of male. Scapholeberis kingi Sars E. Ephippial female, F. Female, G. Postabdomen of female.



A



D

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Scapholeberis kingi Fig. A.Neonate - Ventral view (0.330 mm), B. Pre-adult (0.518 mm), C. Parthenogenetic female with egg (0.768 mm), D. Ephippial female (0.642 mm), E. Ephippium (0.482 mm)



Stage-I (0.114 mm)



Stage-II (0.138 mm)



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Stage-III (0.204 mm)



Stage-IV (0.212 mm)



Stage-V (0.212 mm)



Stage-VI (0.216 mm)



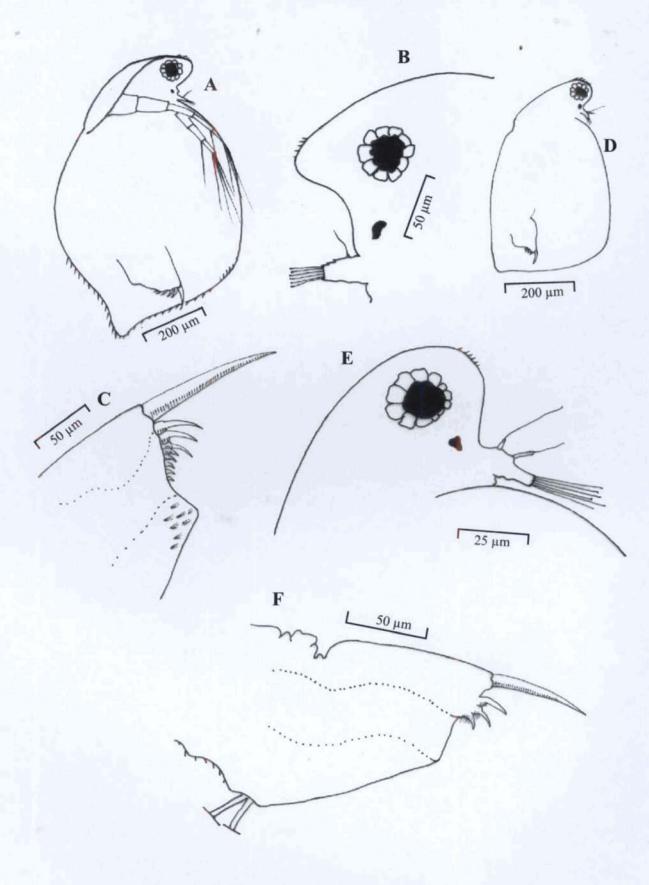
Stage-VII (0.228 mm)



Stage-VIII (0.252 mm)

Scapholeberis kingi Embryonic development

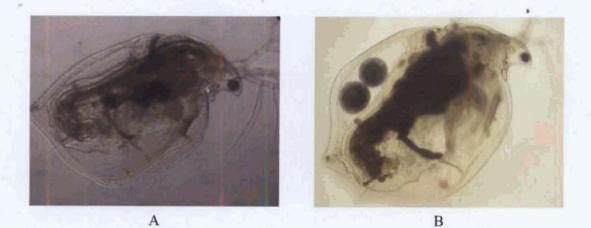
3

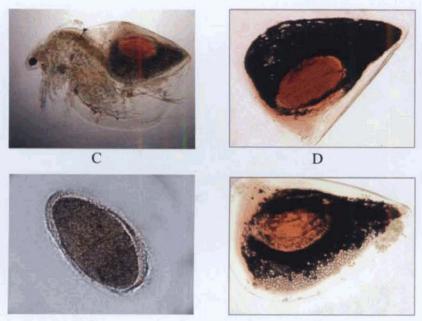


Simocephalus serrulatus (Koch)

Fig. A. Female B. Head of female C. Postabdomen of female D. Male E. Head of male F. Postabdomen of male

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E

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F



#### Simocephalus serrulatus

Fig. A. Female with ovary (1.102 mm) B. Parthenogenetic female (1.680 mm)
C. Ephippial female (1.558 mm) D. Ephippium with egg (0.988 mm)
E. Ephippial egg enlarged (0.390 mm) F. Ephippium without egg (0.920 mm)
G. Initiation of moulting H. Detachment of carapace
I. Casting off exuvium



Stage I 0.220 mm)



Stage II (0.226mm)



Stage III ( 0.286 mm)



Stage IV (0.296 mm)



Stage V (0.298 mm)



Stage VI (0.304 mm)





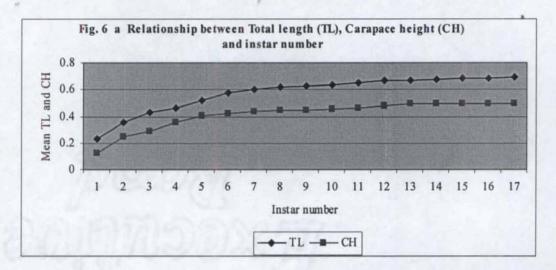
Stage IX (0.487 mm).

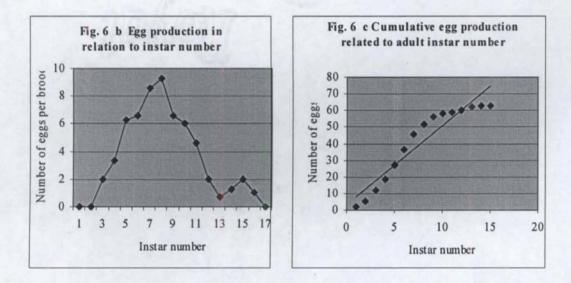


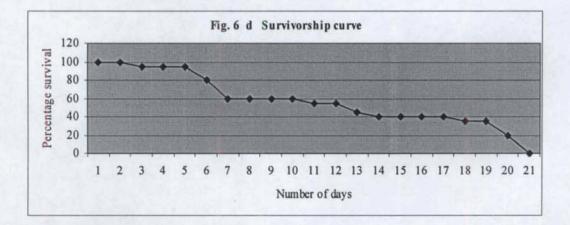
Stage VIII (0.457 mm)

Simocephalus serrulatus Embryonic development

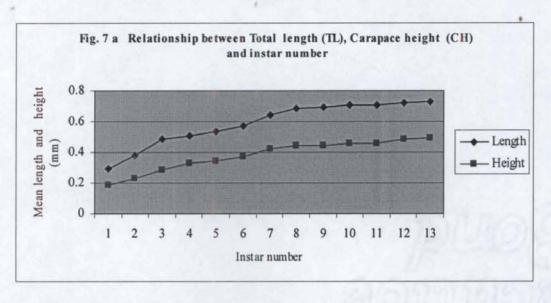
### Ceriodaphnia cornuta

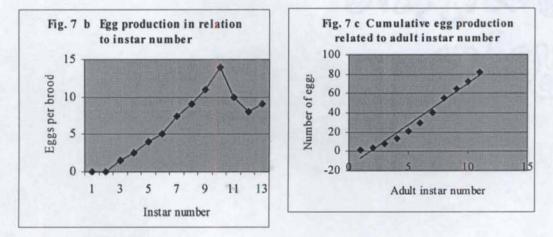


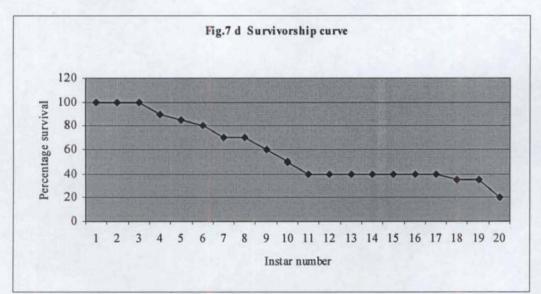




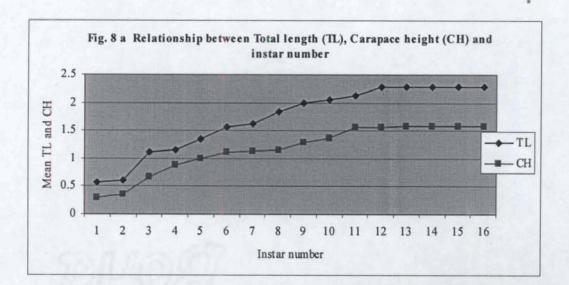
## Scapholeberis kingi

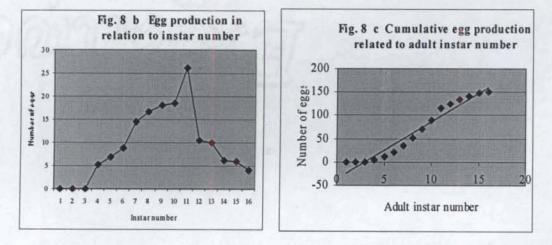


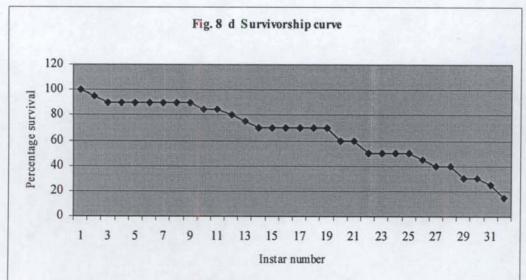


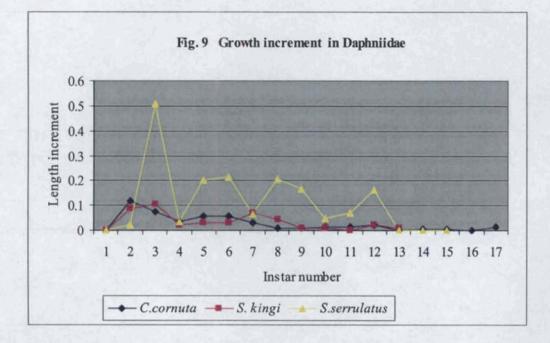


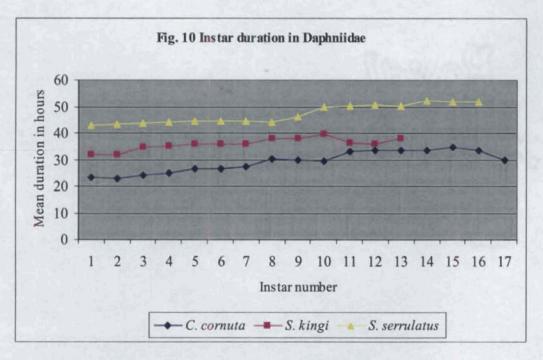
# Simocephalus serrulatus











# Family Moinidae Goulden, 1968

## 5. 4 Moina brachiata (Jurine, 1820)

*Moina brachiata* is primarily an inhabitant of temporary ponds and ditches. This species was described first by Jurine (1820) from Switzerland as *Monoculus brachiatus*. The first report of this species in India is by Brehm (1936) from Kashmir. Subsequent reports are that of Biswas (1971) and Nayar (1971) from Rajasthan, Patil (1976) from Meghalaya and Chandrasekhar and Kodarkar (1994) from Andhra Pradesh.

Our information on the biology and life cycle of *Moina* is based on the studies made in *Moina micrura* by Murugan (1975a) from Tamil Nadu and Thresiamma *et al.* (1991) from Kerala. Information on the biology of *M. brachiata* is scanty; hence made an investigation on its life cycle.

#### 5. 4. 1. External Morphology

### Parthenogenetic female (Plate 18. Fig. A)

Head rather broad; supraocular depression shallow. Eye situated near the dorsal margin of head; ocellus absent (Plate 20. Fig. A). Antennules long and thin (Plate 18. Fig. B), with several setae arranged in rings; long sensory seta at 1/3<sup>rd</sup> distance from head, and with 8 terminal setae (Plate 20. Fig. B). Valves nearly spherical, with granulated surface, ventral margin with

38-40 setae. Antennae well developed, with 4-segmented dorsal ramus and 3-segmented ventral ramus; antennary setation: (0-0-1-3)/(1-1-3). Postabdomen with 11 feathered teeth, and one long bident tooth; claw with pecten and few hairs on the convex surface (Plate 20. Fig. C). Mean size:  $1.158 \times 0.710$  mm.

#### Male (Plate 18. Fig. C)

Oblong and rectangular body, antennules long, bent about  $1/3^{rd}$  distance from the base with two sensory setae originating near the bend (Plate 18. Fig. D). The distal end of antennules with 6 long curved hooks that are grouped together. The 1<sup>st</sup> thoracic leg with a long hook. Size : 0.816 mm.

## **Ephippial female**

Ephippial female similar to parthenogenetic female. Ephippium bright yellow, with distinct reticulations along the posterior and ventral margins (Plate 18. Fig. E). Egg single, situated in a spherical depression. Mean size of ephippium: 0.512 mm.

#### 5.4.2 Reproduction

A laboratory culture of *M. brachiata* was prepared by hatching the resting eggs present in the dried soil sample collected from Site-7 as mentioned in section 3.2.2 (7). The neonates appeared on  $6^{th}$  day, indicating hatching of the resting eggs present in the soil sample. Several parthenogenetic females appeared in the culture from which a stock culture was developed for further studies.

#### Life cycle of parthenogenetic female

The population developed in the laboratory comprised only parthenogenetic females. Males and ephippial females appeared after one month. The sporadic appearance and disappearance of population was one of the interesting aspect observed in this species during the study. The features characteristic of the reproduction and life cycle of *M. brachiata* is given as follows.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a mean birth size (SaB) of  $0.496 \times 0.256$ mm. Both the first and second moulting occurred in same duration of 11.0 hrs each. The total pre-adult instar duration was 22.0 hrs. The mean duration of pre-adult instar (PID) was 11.0 hrs.

#### Attainment of maturity

The ovary became conspicuous during the middle of  $2^{nd}$  pre-adult instar at 14.0±1 hrs. However, they started to bear eggs after completion of  $2^{nd}$  moult at 21.0 hrs; and hence age at first reproduction (AFR) was 0.92 days. The size at first reproduction (SFR) was 0.758 ×0.508 mm.

### **Egg production**

The eggs were deposited into the brood pouch within 10 minutes after completion of  $2^{nd}$  moult. The size of the egg gets increased immediately after its deposition. During this primiparous instar ( $3^{rd}$  instar) egg production started with a mean number of 6.4 eggs/ brood. The primiparous instar was completed in 21.0 hrs and the FGT is calculated as 43.0 hrs.

During the subsequent instars egg production gets almost doubled to attain 12.4 eggs/ brood in the 4<sup>th</sup> instar. The egg production increased further and attained a peak of 15.6 eggs/ brood (Table 20). This maximum clutch size (E max) was attained in 5<sup>th</sup> instar (Fig. 11 b). The egg production in *M. brachiata* showed two peaks with maximum egg production during the 5<sup>th</sup> and 10<sup>th</sup> instar. The egg production decreased further and continued till the end of life span. The duration of adult instars steadily increased throughout the life cycle from 21.0 to 35.0 hrs. The mean duration (AID) was 25.85 hrs.

#### Fecundity

The relationship between egg production and instar number is shown in Fig. 11 b). The range of egg production of a single female varied from 6 to 20 with 10.78 eggs/day of adult life. The cumulative number of eggs ( $\Sigma$ mx) produced during the entire life span was 116.3 (Table 20). Ten broods were produced with a mean of 11.63 eggs /brood. The cumulative egg production ( $\Sigma$ mx) of *M. brachiata* is linearly correlated with instar number (Fig.11 c). The rate of egg production (REP) of *M. brachiata* is calculated as 11.771.

#### 5.4.3 Growth

The first pre-adult neonates had a mean TL of 0.528 mm and the  $2^{nd}$  instar had TL of 0.636mm. Primiparous stage was attained during the  $3^{rd}$  instar when the mean TL was 0.758 mm, with an increment of 19.18%. The maximum mean TL of 1.302 mm was attained at the end of  $12^{th}$  instar (Table 21). The mean CH during the first pre-adult instar was 0.272 mm and

attained CH of 0.332 mm at second instar. They attained a mean CH of 0.508 mm during the primiparous condition, with an increment of 53.01%. Maximum CH was attained in 12<sup>th</sup> instar with CH of 0.752 mm (Table 21). During the life span each individual has undergone two pre-adult and ten adult moults.

The size increment during each instar has been represented in Table-21. Maximum growth increment recorded during the life cycle was with CH of 53.01% in 3<sup>rd</sup> however the growth increment decreased further after 5<sup>th</sup> instar. The relationship between TL, CH and instar number of *M. brachiata* is represented in Fig. 11 a. The correlation coefficients of life history parameters given in Table 22, shows a positive correlation between TL and CH (r = 0.952).

#### 5. 4. 4 Embryonic Development

The stages of embryonic development in *M. brachiata* are represented in Plate 19. The most conspicuous features of the developmental stages are given below.

**Stage I:** This stage is recognized by the presence of spherical egg which appears granular. Mean duration: 3.0 hrs. Mean size: 0.133 mm

Stage II: The embryo is elongated in anterio-posterior axis and shows cellular divisions. The embryo is covered by membrane. Mean duration: 5.0 hrs. Mean size: 0.191 mm

**Stage III:** The head lobe and antennary buds made their first appearance during this stage. Mean duration: 3.70 hrs. Mean size: 0.206 mm.

Stage IV: The head lobe and antennary bud become more distinct during this stage. The cellular divisions are more clearly seen. Mean duration: 2.30 hrs. Mean size: 0.208 mm.

**Stage V:** During this stage the head lobe and the rudiment of antennae become more distinct. The eye, thoracic legs and postabdomen started to differentiate. Mean duration: 1.50 hrs. Mean size: 0.310 mm.

**Stage VI:** The embryonic membrane is cast off just before to this stage. The embryo can be recognized by the presence of eye and segmented antennae and thoracic legs. Mean duration: 2.10 hrs. Mean size: 0.426 mm

Stage VII: Setae appear on the antennae, antennules and thoracic legs. The valves become distinct. Mean duration: 1.50 hrs. Mean size: 0.446 mm

**Stage VIII:** During this stage the development is completed. The neonates resembled the adult in external appearance and were released within a short duration. Mean duration: 1.20 hrs Mean size: 0.496 mm.

**Release of neonates:** The embryonic development of primiparous instar of *M. brachiata* was completed in a mean duration of 20.3 hrs, followed by moulting.

#### 5. 4. 5 Life span and Survivorship

The survivorship curve (Fig. 11 d) indicates the relationship of age (days) and percentage survival of *M. brachiata*. The mean life span ( $\Sigma$ lx) of female is calculated as 5.63 days; while the maximum life span (L max) observed during the present study was 11.69 days.

## 5. 5 Moinodaphnia macleayi (King, 1853)

Moinodaphnia macleayi was described first by King (1853); while the name Moinodaphnia was coined by Herrick (1887). M. macleayi is considered a "missing link" by Herrick (1887) between Moina and the other daphniids; because it possessed an ocellus and a supposed abdominal process.

Although, *M. macleayi* is widely distributed in tropics; it was reported first from Kerala by Brehm (1953) in a pond at Jagady, Thiruvananthapuram. Subsequent reports are from West Bengal (Sharma, 1978) and Lake Kolleru, Andhra Pradesh (Durga Prasad and Padmavathy, 2003a). There is no report of the studies on biology of this species from the country and hence made the present study on reproduction, life cycle, growth and embryonic development of both males and females reared in the laboratory.

#### 5. 5. 1 External Morphology

#### Parthenogenetic female (Plate 21. Fig. C)

Body broadly rounded in outline. Head small, triangular with a flat ventral margin. Eye large and fills the tip of the head, presence of a slight supra-ocular depression above the eye; ocellus distinct (Plate 20. Fig. D). Antennules long and thin, arise from the ventral margin just behind the eye; with a long lateral seta and a group of nine terminal setae (Plate 20. Fig. E). Antennae thin, dorsal ramus 4-segmented and ventral ramus 3-segmented; setation of antenna: (0-0-1-4)/(1-1-3). Shell broadly rounded and slightly reticulated; valves with series of short marginal spines. Postabdomen well developed, distal end elongated, having 11 feathered setae and one bident tooth; claw straight, with fine setae on concave margin (Plate 20. Fig. F). Mean size:  $0.825 \times 0.592$  mm

#### Male (Plate 21. Fig. D)

Smaller than female. Head elongated, with large eye; ocellus small (Plate 20. Fig. G). Antennules long and curved inward; sensory seta originate about  $1/3^{rd}$  distance away from head, terminal setae comprise one long seta and 4-6 short setae. (Plate 20. Fig. H). First leg with large curved hook. Postabdomen similar to female with 6 feathered teeth (Plate 20. Fig. I). Mean size:  $0.600 \times 0.416$  mm

#### Ephippial female (Plate 21. Fig. E)

Ephippial female similar to that of parthenogenetic female, except for the presence of reticulations in the valves. Ephippium white, occupying a central position in between the valves (Plate 21. Fig. F). Egg single (Plate 21. Fig. H), surrounded by polygonal ornamentation (Plate 21. Fig. G). Mean size:  $0.840 \times 0.496$  mm. Mean size of ephippium:  $0.565 \times 0.541$  mm

#### 5. 5. 2 Reproduction

The population developed during the laboratory culture comprised asexual females, ephippia bearing females and males. The parthenogenetic and sexual reproduction was observed throughout the period of study. The neonates produced from a clutch were all females, all males or both males and females. However, parthenogenetic females dominated the stock culture. The appearance of males was followed by the production of ephippial females in the culture. The carapace of ephippial female gets modified as an ephippium which was subsequently released into the medium during moulting. The ephippium floated on the surface for a few minutes and then sank to the bottom upon getting wet. During the course of development the egg changed from an oval to spherical shape. The egg had mean initial size of  $0.260 \times 0.162$  mm. The ephippia were produced by the females during their early part of life cycle. After completing 1-2 ephippial generations they resumed the parthenogenetic reproduction.

#### Life cycle of Male

*M. macleayi* male was reported for the first time from India by Venkataraman (1995). Males were not obtained from the field collection during the present study. However, a good number of males were produced in the laboratory culture throughout the period of study. Males could be distinguished shortly after birth, due to the presence of a prominent curved antennule and their relative smaller size. Twenty neonates of less than 12 hrs of age were sorted out and reared individually for life cycle studies following the methodology cited in section 3. 2. 3.

The neonates had a mean birth size (SaB) of  $0.380 \times 0.218$  mm. *M. macleyai* male during the entire lifespan, underwent only 2 moults. The 1<sup>st</sup> and 2<sup>nd</sup> moulting occurred in the same duration of 42 hrs each. The neonates attained TL of 0.568 and CH of 0.272 mm after first moulting. The total pre-adult duration was 84 hrs (Table 25).

After the 2<sup>nd</sup> moulting they became sexually mature as indicated by the presence of yellow coloured testis located just below the alimentary canal. The size after second moult was  $0.668 \times 0.320$  mm. The maximum growth was observed during 2<sup>nd</sup>, 3<sup>rd</sup> and 4<sup>th</sup> day respectively and decreased further (Table 25). There was no moulting after attainment of maturity and growth was retarded further. The maximum size was  $0.762 \times 0.376$  mm recorded on  $12^{th}$  day (Table 25). The percentage increment of TL and CH during each day is represented in Table 25. The relationship between TL, CH and instar number is given in Fig.12 b. The mean life span ( $\Sigma$ lx) of male is calculated as 5.60 days, while the maximum life span observed during the present study (L max) was 12.19 days.

## Life cycle of parthenogenetic female

The features characteristic of the reproduction and life cycle of *M macleayi* is given as follows.

#### Moulting

The neonates underwent moulting towards the end of each instar followed by an increase in size, especially during the early instars. Moulting in adult instars was followed by the release of young ones. Soon after this the eggs released from the ovary are deposited into the brood pouch.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a birth size (SaB) of  $0.480 \times 0.296$  mm. The first moulting occurred at an interval of

23.26 hrs while the second moulting occurred at 23.56 hrs duration. The total pre-adult duration was 46.82 hrs and the mean PID was 23.41 hrs.

#### Attainment of maturity

The ovaries of parthenogenetic females were clearly visible at  $32\pm 0.5$  hrs of life as green coloured, elongated sacs on either side of the alimentary canal (Plate 21. Fig. B). However, the eggs were deposited into the brood pouch after the completion of  $2^{nd}$  moulting and the age at first reproduction (AFR) was 1.95 days. The size at first reproduction (SFR) was  $0.628 \times 0.432$  mm.

### Egg production

The parthenogenetic females started to bear eggs after completion of the  $2^{nd}$  moult at  $63.82\pm 1$  hour. The eggs attained a spherical shape soon after its deposition into the brood pouch and measured a size of  $0.148 \times 0.096$  mm. Egg production started during the primiparous instar ( $3^{rd}$  instar) with the production of 2.2 eggs/ brood. The primiparous instar had a mean duration of 29.3 hrs and the first generation time (FGT) is calculated as 76.0 hrs.

During the subsequent instars there was a sharp increase in egg production to 19.6 eggs/ brood. This maximum clutch size (Emax) was attained in the  $12^{\text{th}}$  instar (Table 23). The egg production of *M. macleayi* showed a single peak with maximum egg production in  $12^{\text{th}}$  instar followed by a sudden decrease (Fig. 12 c).

Each clutch in the early adult instar consisted of 1 to 4 rows of eggs on either side of brood pouch with 3 to 4 eggs in each row. The egg production continued throughout the life span.

#### Fecundity

Fig. 12 c indicates the relationship between egg production and instar number. The range of egg production of a single female varied from 2.0 to 26.0 with an egg production of 7.02 eggs per day of adult life. The cumulative number of eggs ( $\Sigma$ mx) produced during entire life was 150.9 (Table 23). Fifteen broods were produced during the entire life span with a mean of 10.06 eggs/ brood. The cumulative egg production ( $\Sigma$ mx) of *M. macleayi* is linearly correlated with adult instar number (Fig. 12 d). The rate of egg production (REP) of *M. macleayi* is calculated as 11.4968.

#### **Instar duration**

Aithough, they had a uniform duration of pre-adult instars with an average (PID) of 23.41 hrs. The primiparous instar had a mean duration of 29.3 hrs. The duration of adult instars varied from 29.3 to 38.0 with a mean (AID) of 34.43 hrs (Table 23). *M. macleayi* passed through fifteen adult instars during the life span. Fig. 14 indicates the relationship between instar number and instar duration.

#### 5. 5. 3 Growth

The first pre-adult neonates had a mean TL of 0.480 mm and  $2^{nd}$  instar had TL of 0.584 mm. Primiparous stage was attained during the  $3^{rd}$  instar

when the mean TL was 0.628 mm; with an increment of 7.53%. The maximum mean TL of 1.104 mm was attained at the end of  $16^{th}$  instar and no growth observed during  $17^{th}$  instar (Table 24).

The mean CH during the first pre-adult instar was 0.296 mm and attained CH of 0.312 mm at second instar. They attained a mean CH of 0.432 mm during the primiparous condition; with an increment of 38.46%. Maximum CH was attained in 16<sup>th</sup> instar with CH of 0.786 mm (Table 24). During the life span each individual has undergone two pre-adult and fifteen adult moults.

The size increment during each instar is given in Table 24. Maximum growth increment recorded during the life cycle was with TL of 21.67% in  $2^{nd}$  instar and with CH of 38.46% in  $3^{rd}$  instar; and the growth increment decreased in the succeeding instars. The relationship between TL, CH and instar number of *M. macleayi* has been represented in Fig. 12 a. The correlation coefficients of the life history characters are given in Table 26, which shows a positive correlation between TL and CH (r = 0.992).

#### 5. 5. 4 Embryonic Development

The stages of embryonic development in *M. macleayi* are represented in Plate 22. The most conspicuous features of the developmental stages are given below.

**Stage I:** This stage is recognized by the presence of spherical egg, with less amount of yolk. Mean duration: 3.60 hrs. Mean size: 0.136 mm

**Stage II:** During this stage the embryo is elongated anterio-posteriorly. The elongation of the anterior region is clearly visible. Cellular divisions are initiated in the embryo. Mean duration: 5.10 hrs. Mean size: 0.153 mm.

Stage III: The head lobe appears and cellular divisions continued. Mean duration: 3.50 hrs. Mean size: 0.157 mm.

**Stage IV:** The head lobe is clearly distinct with the formation of rudimentary structures. Mean duration: 2.00 hrs. Mean size: 0.229 mm.

Stage V: The presence of pink eyes is noticed for the first time during this stage. The rudiments of antennae become distinct. The embryo contained less amount of yolk and was enclosed in a membrane. Mean duration: 2.4 hrs. Mean size: 0.241 mm.

Stage VI: The eyes undergo fusion and become black in colour. The rudiments of thoracic legs become distinct during this stage. Mean duration: 6.20 hrs. Mean size: 0.283 mm

**Stage VII:** The embryonic membrane is cast off just before this stage and the antennae get segmented followed by the development of terminal setae. The eye gets enlarged followed by the appearance of a small ocellus. The rudiments of thoracic legs and postabdomen become more distinct during this stage. Mean duration: 4.6 hrs. Mean size: 0.312 mm

**Stage VIII:** Yolk completely disappears and some movements are initiated in the embryo. The antennae, antennules, thoracic legs and postabdomen become distinct and the embryo resembles the adult in external morphology. Mean duration: 1.5 hrs. Mean size: 0.401 mm

**Release of neonates** 

The hatching could be regarded as the culmination of embryonic development. The neonates were released by the opening of the brood pouch by the extension of postabdominal processes. The total duration of embryonic development during the primiparous instar of *M. macleayi* was 28.90 hrs.

## 5. 5. 5 Life span and Survivorship

The survivorship curve (Fig. 12 e) indicates the relationship of age (days) and percentage survival of *M. macleayi*. The the mean life span ( $\Sigma$ lx) observed during the present study is 9.23 days while the maximum life span observed during the present study (Lmax) for the female was 23.45 days.



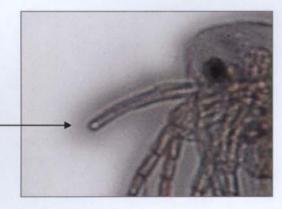


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Moina brachiata

Fig. A. Parthenogenetic female (1.128 mm), B. Female head enlarged, C. Male (0.726 mm), D. Male head enlarged ( $\rightarrow$  indicates the elongated antennule), E. Ephippium (0.504 mm).



Stage-I (0.112 mm)



Stage-II (0.195 mm)



Stage-III (0.204 mm)



Stage-IV (0.208 mm)



Stage-V (0. 306 mm)



Stage-VI (0.424 mm)



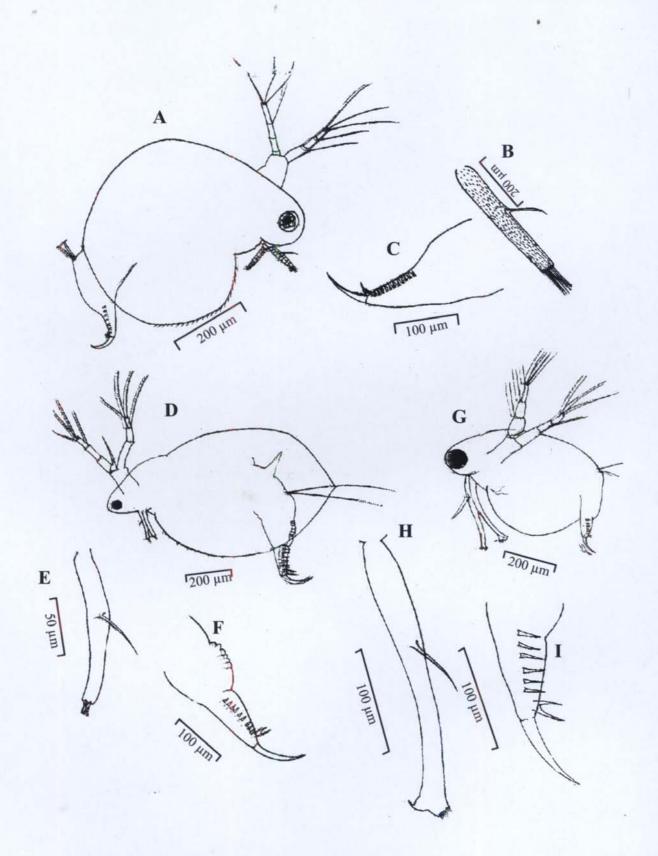
Stage-VII (0.450 mm)



Stage-VIII (0.492 mm)

Moina brachiata Embryonic development





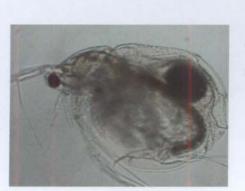
Moina brachiata (Jurine) Fig. A. Female, B. Antennule of female, C. Postabdomen of female Moinodaphnia macleayi (King) D. Female, E. Antennule of female, F. Postabdomen of female, G. Male, H. Antennule of male, I. Postabdomen of male.



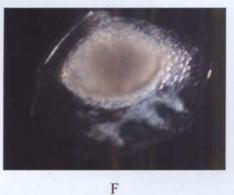
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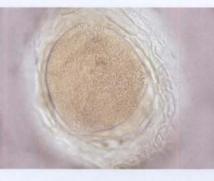


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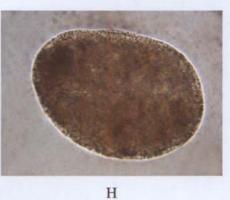








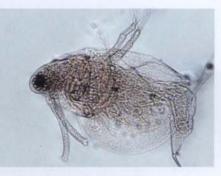
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Moinodaphnia macleayi

Fig. A. Pre-adult (0.526 mm), B. Female with ovary (0.580 mm),
C. Parthenogenetic female with egg (0.857 mm), D. Male (0.604 mm), E. Ephippial female (0.710 mm),
F. Ephippium (0.457 mm), G. Ephippium enlarged, H. Ephippial egg (0.265 mm).





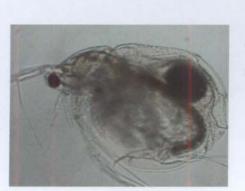
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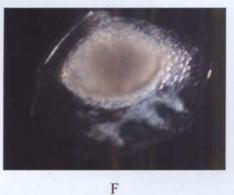
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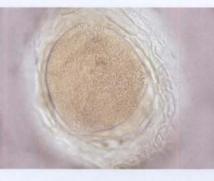


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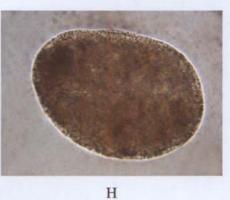








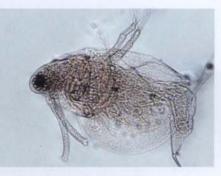
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Moinodaphnia macleayi

Fig. A. Pre-adult (0.526 mm), B. Female with ovary (0.580 mm),
C. Parthenogenetic female with egg (0.857 mm), D. Male (0.604 mm), E. Ephippial female (0.710 mm),
F. Ephippium (0.457 mm), G. Ephippium enlarged, H. Ephippial egg (0.265 mm).





D



Stage-I (0.138 mm)



Stage-II (0.153 mm)



Stage-III (0.158 mm)



Stage-IV (0.228 mm)



Stage-V (0.240 mm)



Stage-VI (0.284 mm)

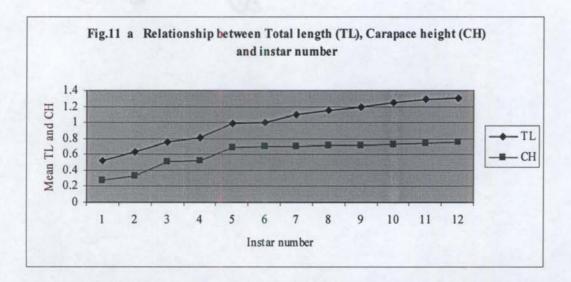


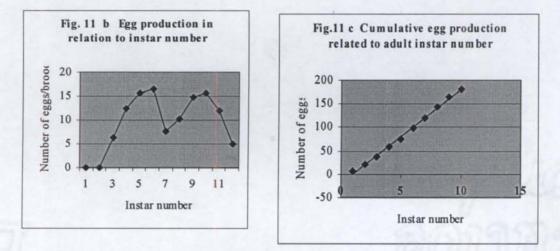
Stage-VII (0.312 mm)

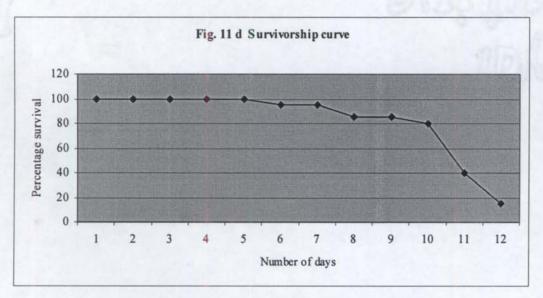


Stage-VIII (0.398 mm)

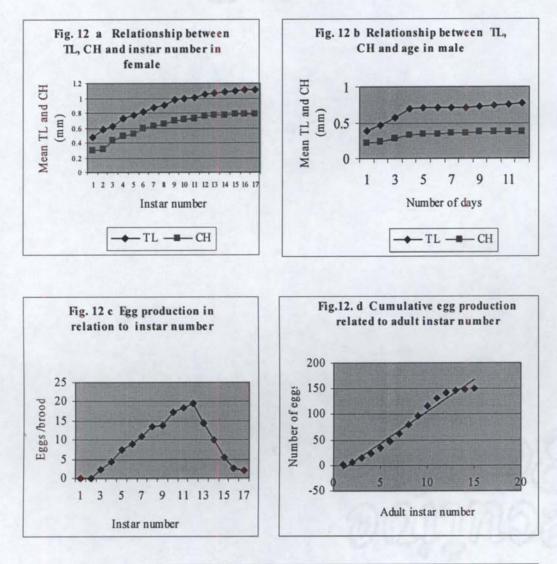
## Moina brachiata

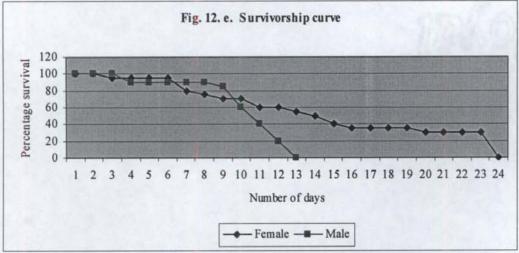


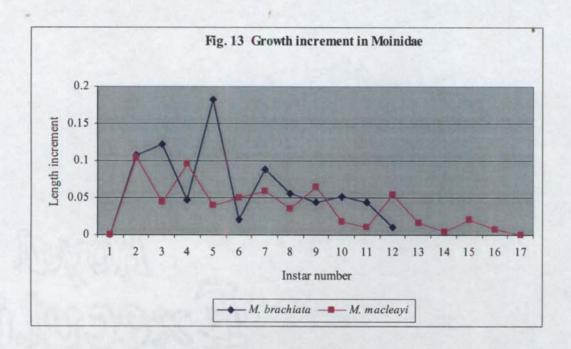


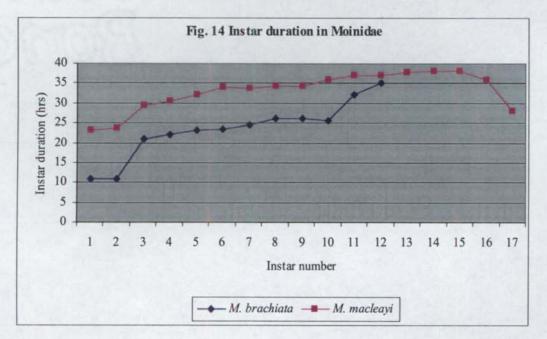


## Moinodaphnia macleayi









# Family Macrothricidae Norman and Brady, 1867

## 5. 6 Ilyocryptus spinifer Herrick, 1882

*Ilyocryptus spinifer* is an inhabitant of freshwater bodies which often prefers the muddy substratum. This species was first described by Herrick (1882) from USA. *I. spinifer* is also reported from Australia (Henry, 1922), Africa (Kořínek, 1984) and Asia (Rajapaksa and Fernando, 1982).

Many authors consider *I. spinifer* a pan-tropical species occurring throughout tropical and sub-tropical regions (Birge, 1918; Harding, 1957). Smirnov (1976) included it in a separate Family Ilyocryptidae.

*I. longiremis* Sars was considered a synonym of *I. spinifer* Herrick by Gurney (1907) who reported this species from Calcutta. Although, *I. spinifer* has been reported from different states in India, this species is reported first from Kerala by Michael and Sharma (1988). Subsequently, Patil and Gouder (1988) reported it from Karnataka, Durga Prasad and Padmavathy (2003a) from Andhra Pradesh and Babu and Nayar (2004) from Thekkady, Kerala.

The biology of *I. spinifer* has not been investigated so far and hence made a detailed description of its parthenogenetic reproduction.

#### 5. 6. 1 External Morphology

#### **Parthenogenetic female** (Plate 23. Fig. C)

Body oval shaped, with deeply arched ventral margin, length greater than width (Plate 25. Fig. A). Head small, slightly bluntly pointed anteroventrally, appears triangular in lateral view (Plate 23. Fig. D). Eye relatively large; ocellus small, situated about half way between eye and base of antennules. Antennules elongated and slender, 2-segmented, attached to posteroventral margin of the head, with a group of terminal sensory setae (Plate 25. Fig. C). Valves with straight dorsal margin, ventral and posterior margins evenly rounded, with a series of long plumose setae (Plate 25. Fig. B); hexagonal reticulations throughout shell surface. Antennae with 4segmented dorsal ramus and 3-segmented ventral ramus; antennary setation: (0-0-0-3)/(1-1-3). Postabdomen bilobed, with 25 marginal denticles and long anal spines; claw slender with two unequal basal spines (Plate 23. Fig. F; Plate 25. Fig. D); anal aperture opens in the middle of postabdomen. The carapace of the field animals were found profusely covered with detritus. Mean size:  $0.708 \times 0.586$  mm.

#### Ephippial female (Plate 23. Fig. G)

Ephippial female similar to parthenogenetic female in all respects except reticulation of the shell. Ephippium released with a major portion of carapace and has hexagonal reticulations. The ventral margin bears several setae. The ephippium encloses two eggs of almost same size (Plate 23. Fig. H). Mean size of ephippial female: 0.570 mm. Size of ephippium: 0.506 mm.

#### 5. 6. 2 Reproduction

The population developed during the laboratory culture comprised only asexually reproducing females. Males were not observed during the present study.

#### Life cycle of parthenogenetic female

Parthenogenetic reproduction was observed throughout the period of study. The neonates produced by parthenogenetic reproduction were completely asexual females. The features characteristic of the reproduction and life cycle of *I. spinifer* is given as follows.

#### Pre-adult instar

The neonates produced from parthenogenetic females had a birth size (SaB) of  $0.304 \times 0.208$  mm. The newly hatched young ones were found to avoid light and burrow into the bottom. Moulting was not observed during pre-adult instar and they retained their of old carapace. The period between the birth and first appearance of eggs inside the brood pouch is taken as the pre-adult instar duration of the species. The mean pre-adult instar duration (PID) was 68.0 hrs.

#### Attainment of maturity

Although, the development of ovary started very early in life, they were clearly visible at  $52.0 \pm 1.5$  hrs. The ovary appeared as elongated sacs on either side of the alimentary canal with dark-brown colour (Plate 23. Fig. B).

They started to bear eggs after 68.0 hrs. The AFR was 2.83 days and the SFR was  $0.584 \times 0.448$  mm

#### **Egg production**

One of the important features noted during the life cycle was the delayed deposition of each clutch of eggs into the brood pouch. The eggs were first deposited into brood pouch after 13.0 hrs during the primiparous instar. The same pattern of delayed egg production was observed in the successive instars.

The eggs were oval-shaped and measured a mean size of  $0.195 \times 0.126$  mm. During this primiparous instar (2<sup>nd</sup> instar) egg production started with an average of 2.0 eggs/ brood. The primiparous instar was completed in 52.0 hrs and the first generation time (FGT) is calculated as 120.0 hrs.

During the subsequent instars there was a sharp increase in egg production to 12.0 eggs/ brood (Table 27). The maximum clutch size (Emax) was attained in the  $6^{th}$  instar, with production of 12.0 eggs/ brood. However, the egg production of *I. spinifer* showed a bimodal pattern with two peaks. The egg production declined after  $6^{th}$  instar and again increased further to produce 7.5 eggs in  $10^{th}$  instar (Fig. 15 b). In *I. spinifer* egg production continued throughout the life span.

Each clutch during the adult instars consisted of 1 to 2 rows of eggs placed on either side of the brood pouch. Due to the absence of moulting the adult instar duration is taken as the time interval between the releases of successive batches of neonates. This includes the time for egg transfer, completion of embryonic development and subsequent release of young ones. The adult instar duration varied from 42.0-50.0 hrs. The mean adult duration (AID) is calculated as 45.96 hrs (Table 27).

## Fecundity

The relationship between egg production and instar number is represented in Fig. 15 b. The range of egg production of a single female was from 2 to 14 with an egg production of 2.69 eggs/ day of adult life. The cumulative number of eggs produced ( $\Sigma$ mx) during the entire life span was 61.5 (Table 27). Twelve broods were produced during the entire life with a mean of 5.12 eggs/ brood. The cumulative egg production ( $\Sigma$ mx) is linearly correlated instar number Fig. 15 c. The rate of egg production of *I. spinifer* is calculated as 5.7637.

#### 5. 6. 3 Growth

The first pre-adult neonates had a mean TL of 0.336 mm. The primiparous stage was attained during the  $2^{nd}$  instar when the mean TL was 0.584 mm. The maximum mean TL of 0.830 mm was attained at the end of  $13^{th}$  instar (Table 28).

The mean CH during the first pre-adult instar was 0.238 mm.. The mean CH of 0.448 mm was attained during the primiparous condition  $(2^{nd} \text{ instar})$ . Maximum mean CH of 0.662 mm was attained in  $13^{th}$  instar (Table 28). During the entire life span each individual has undergone one pre-adult and twelve adult instars.

The increment of TL and CH during each instar is given in Table 28. Maximum growth increment recorded during the life cycle was in 2<sup>nd</sup> instar with TL of 73.81% and CH of 88.24% respectively. This indicates maximum growth during the early (pre-reproductive) phase of life cycle than during the late (reproductive) phase.

The relationship between TL, CH and instar number of *I. spinifer* has been represented in Fig. 15 a. The correlation coefficients of life history characters are given in Table 29, which shows a positive correlation between TL and CH (r= 0.989).

#### 5. 6. 4 Embryonic Development

The stages of embryonic development of *I. spinifer* are represented in Plate 24. The most conspicuous features of the developmental stages are given below.

**Stage I:** This stage is recognized by the presence of an oval-shaped egg which is brown in colour.

Mean duration: 1.0 hrs. Mean size: 0.188 mm

**Stage II**: The embryo has two distinct areas. The outer area of the embryo appears transparent while the inner area is granular and opaque due to the concentration of yolk.

Mean duration: 3.5 hrs. Mean size 0. 190 mm

Stage III: The embryo started to elongate, the outer transparent area become more distinct and shows cellular divisions. Mean duration: 8 .0 hrs. Mean size 0. 202 mm

Stage IV: This stage is characterized by the appearance of rudiments of head and antennae.

Mean duration: 4.0 hrs. Mean size: 0. 218 mm

Stage V: This stage is characterized by the presence of rudiments of head and antennae. The embryo also shows rudiments of thoracic legs and postabdomen.

Mean duration: 7.5 hrs. Mean size: 0. 224 mm

**Stage VI:** This stage is recognized by the presence of pink eye. The antennae become segmented and thoracic legs become distinct.

Mean duration: 4.0 hrs. Mean size: 0. 258 mm.

Stage VII: This stage is characterized by the presence of black eye. The antennae, antennules, thoracic legs and postabdomen become more distinct.

Mean duration: 9.0 hrs. Mean size: 0. 288 mm

**Stage VIII:** During this stage the segmentation of antennae and development of setae is complete. The valves and the postabdomen become distinct. The embryo resembles the adult in external morphology.

Mean duration: 2.0 hrs. Mean size: 0.304 mm

### **Release of neonates**

The adult parthenogenetic females released neonates from the brood pouch by the opening of valves and movement of postabdomen. The total mean duration of primiparous instar was 52 hrs; out of which 13.0 hrs needed for the deposition of eggs and 39.0 hrs for the embryonic development. The delayed deposition of eggs (13.0-14.0 hrs) as observed in the present study may be due to the absence of moulting in this species.

#### **Retention of Carapace**

The carapace was found profusely covered with algae (*Chlorella*) in the laboratory culture. Absence of true moulting enables these animals to retain its old carapace without renewing encrustation. These retained carapaces appear as concentric rings over the shell, which appears as successive growth lines (Plate 23. Fig. E).

The retained carapaces along with encrustation provide an excellent camouflage. The slow movements exhibited by the species are due to this algal encrustation.

#### 5. 6. 5 Life Span and Survivorship

The survivorship curve (Fig. 15. d) indicated the relationship of age (days) and percentage survival of *I. spinifer*. The mean life span ( $\Sigma$ lx) is calculated as 11.43 days, while the maximum life span (L max) of female observed during the study (Lmax) was 25.81 days.

## 5. 7 Macrothrix triserialis (Brady, 1886)

Macrothrix triserialis is a littoral cladoceran most often found among submerged vegetation in water bodies especially ponds, paddy fields and marshes. This species was described first by Brady (1886) from Sri-Lanka. *M. triserialis* resembles *M. rosea* (Lievin, 1848) which is commonly recorded in Europe and North America.

Among Cladocera, Macrothricidae is one of the least studied families and hence many species have recently been re-described (Kotov, 1999; Silva-Briano *et al.* 1999). Recently Dumont *et al.* (2002) revised the group *Macrothrix rosea-triserialis*.

In India *M. triserialis* is reported from Bihar (Gurney, 1907). Later from Rajasthan (Biswas, 1971; Michael and Sharma, 1988), West Bengal (Michael and Sharma, 1988) and Karnataka (Patil and Gouder, 1988). The first report of this species from Kerala is that of Michael and Sharma (1988) based on the collections of Nayar, C.K.G from Irinjalakuda. Further report of this species is by Raghunathan (1989a) from Wynad and recently from Thekkady by Babu and Nayar (2004).

The biology of *M. triserialis* has not been studied so far and hence made a detailed description of the life cycle of parthenogenetic female and male.

#### 5. 7. 1 External Morphology

#### Parthenogenetic female (Plate 26. Fig. A)

Body nearly oval; dorsal margin slightly arched, ventral margin deeply arched. Head large, separated from rest of body by a cervical depression (Plate 25. Fig. E); rostrum slightly pointed, labral plate serrated and almost straight posteriorly. Eye relatively large; ocellus small, situated nearer to the apex of the rostrum than to the eye. Antennules slender, cylindrical, with series of spinules, distal part armed with 3 spinules and a few terminal setae (Plate 25. Fig. F). Antennae relatively short, the longest seta of the antenna have a series of short setules and 3 spines in the middle. Shell produced into a sharp pointed angle posteriorly; ventral margin serrated and with a series of rather long setae. Postabdomen bilobed, with several anteriorly directed spines along its dorsal margin, lateral spines in rows; claw short, slightly curved dorsally, without basal spine; distal segment of natatorial setae short, with a group of long and fine setules attached to them (Plate 25. Fig. G). Mean size: 0.892 × 0.533 mm.

#### Male (Plate 26. Fig B)

Males smaller than female. Antennules elongated, with one strong bristle at the base anteriorly and 4 rows of rigid hairs in its proximal portion, distal part armed with 3 spines and a few terminal setae (Plate 25. Fig. H). The first thoracic leg bears a curved hook. Postabdomen relatively small, dorsal side forms a cylindrical tube, transversely truncated at the tip and with ejaculatory duct opening dorsally (Plate 25. Fig. I). Mean size:  $0.517 \times 0.308$ mm.

#### Ephippial female (Plate 26. Fig. C)

Ephippial female similar to the parthenogenetic female except reticulations in the shell. The ephippium appears transparent with striations, enclosing two eggs. Ephippium shed along with a major portion of the valve (Plate 26. Fig. D). Eggs large, spherical, brown coloured and granular containing abundant yolk (Plate 26. Fig. E). Mean size: 0.766 ×0.516 mm. Mean size of ephippium: 0.450×0.508 mm.

#### 5. 7. 2 Reproduction

The population developed during the laboratory culture comprised asexually reproducing females, ephippia bearing females and males. The life cycle is represented in Plate 27.

The neonates produced from a single clutch during the parthenogentic reproduction consist of males and females. They were found to be all females, all males or both males and females. The males could be distinguished during early stages due to their relative smaller size and elongated antennules. Production of *M. triserialis* males are often followed by the generation of ephippial females leading to sexual reproduction.

The ephippial females develop a transparent ephippium which encloses two eggs inside it. Detachment of ephippium always occurs during moulting, and the ephippium is released into the medium. The ephippia then sink to the bottom or most often get attached to the sides of the container. The ephippial females after completion of 2-3 generations resumed the parthenogenetic reproduction.

#### **Population growth**

The population density in the laboratory culture was estimated from 28-11-2004 to 24-12-2004. Numerical study was made by taking a subsample of 1 ml from the container. A counting chamber of 1 ml capacity was used for counting and calculated the number of individuals/ litre of water.

Table 41 shows that during the period of study, the population density of *M. triserialis* in the laboratory culture varied between 1124 and 16239 per litre of water. The growth of population is graphically represented in Fig. 17. It is evident from the figure that the population showed an increase in the initial period. After attaining its peak on 16<sup>th</sup> day, the population declined associated with the appearance of males and ephippial females. A sudden increase in the population was further noted on 26<sup>th</sup> day when the population comprised of only parthenogenetic females. The males and ephippial females however disappeared towards the end the period.

#### Life cycle of Male

Twenty male neonates were sorted out for the life history studies and cultured them individually in test tubes using the methodology mentioned in section 3. 2. 3.

The mean birth size (SaB) was  $0.266 \times 0.216$  mm. The first moulting occurred in duration of 36.0 hrs and there was no moulting further. The first pre-adult neonates had a mean TL of 0.408 mm and CH of 0.248 mm. They attained maturity on 2<sup>nd</sup> day after first moulting and attained mean TL

of 0.432 mm and CH of 0.256 mm (Table 32). The pre-adult duration was 36.0 hrs (1.5 days). The sexually mature individuals have yellow coloured testis.

The maximum percentage increment of TL was observed on  $2^{nd}$  day with 5.88%. The increment of TL showed variation subsequently (Table 32). Maximum size attained during the study was  $0.518 \times 0.318$  mm on  $9^{th}$  day. The mean life span of male ( $\Sigma$ lx) is calculated as 4.6 days while the maximum life span (Lmax) observed was 8.6 days.

#### Life cycle of parthenogenetic female

The majority of neonates produced were parthenogenetic females. The features characteristic of the reproduction and life cycle of parthenogenetic female is given as follows.

#### Moulting

*M. triserialis* underwent moulting towards the end of each instar. The release of whole part of the carapace as exuvium was observed in *M. triserialis* and the moulting was completed within duration of 50.0 minutes.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a mean birth size (SaB) of  $0.318 \times 0.207$  mm. Both the first and second moulting occurred in a uniform duration of 29.0 hrs. The total pre-adult duration was

58.0 hrs and the mean pre-adult duration (PID) was 29.0 hrs. They started to reproduce after second moulting.

#### Attainment of maturity

The ovary of parthenogenetic female was clearly visible at  $48\pm1$  hrs of life; and appeared as green coloured elongated structures on either side of alimentary canal. They started to bear eggs after completion of  $2^{nd}$  moult at 58.0 hrs; and hence the AFR was 2.42 days. The SFR was 0.608×0.376 mm.

#### Egg production

Eggs were deposited into the brood pouch in a mean duration of 50.0 minutes after completion of the second moult. The eggs attained spherical shape with yellow green colour and measured a mean size of  $0.175 \times 0.133$  mm. During this primiparous instar (3<sup>rd</sup> instar) egg production started with a mean of 5.5 eggs/brood. The primiparous instar was completed in mean duration of 30.5 hrs and the first generation time FGT is calculated as 88.5 hrs.

During the subsequent instars there was a steady increase in egg production to 20.4 eggs/ brood in  $6^{th}$  instar; followed by a slight decrease (Table 30). In the succeeding instars the egg production increased to attain a peak production of 24.2 eggs/ brood during  $10^{th}$  instar. This maximum clutch size (Emax) was attained in the  $10^{th}$  instar, followed by a slow decline till death (Fig. 16 c). The bimodal pattern of egg production is observed in *M. triserialis* with two peaks. They continued egg production throughout the life span.

Each clutch consisted of 1-2 rows of eggs deposited on either side of the brood pouch. The female underwent moulting towards the end of each adult instar. The adult instar durations varied from 30.5 and 36.5 hrs (Table 30); with a mean adult duration (AID) of 32.9 hrs. The relationship between instar number and instar duration is represented in Fig. 18 b.

#### Fecundity

The relationship of egg production with instar number is represented in Fig. 16 c. The range of egg production of a single female was from 4 to 26 with 13.24 eggs/day of adult life. The cumulative number of eggs produced ( $\Sigma$ mx) during entire life span was 181.5 (Table 30). Ten broods were produced during the entire life with a mean of 18.15 eggs/ brood. The  $\Sigma$ mx was linearly correlated with instar number Fig. 16 d. The rate of egg production REP of *M. triserialis* is calculated as 17.9329.

#### 5.7.3 Growth

The first pre-adult neonates had a mean TL of 0.410 mm and the second instar had TL of 0.464 mm. Primiparous stage was attained during the  $3^{rd}$  instar when the mean TL was 0.608 mm. The maximum mean TL of 0.966 mm was attained at the end of  $12^{th}$  instar (Table 31).

The mean CH during the first pre-adult instar was 0.248 mm and attained CH of 0.366 mm at second instar. A mean CH of 0.376 mm was attained during the primiparous condition. Maximum mean CH of 0.608 mm was attained in 12<sup>th</sup> instar (Table 31). During the life span each individual has undergone two pre-adult and ten adult moults. The relationship between TL,

CH and instar number of *M. triserialis* has been represented in Fig. 16 a. The correlation coefficients of life history characters are given in Table 33, which shows a positive correlation between TL and CH (r= 0.971).

The increment of TL and CH during each instar is given in Table 31. Maximum growth increment recorded during the life cycle was with TL of 31.03% in 3<sup>rd</sup> instar and with CH of 47.58% was observed in 2<sup>nd</sup> instar.

#### 5. 7. 4 Embryonic Development

The stages of embryonic development of *M. triserialis* are represented in Plate 28. The most conspicuous features of the developmental stages are given below.

**Stage I:** This stage is recognized by the presence of oval-shaped egg with dark granular yolk. Mean duration: 1.0 hrs. Mean size: 0.178 mm.

**Stage II**: The embryo has two distinct areas. The outer area of the embryo appears transparent while the yolk granules are concentrated at the inner granular zone. Mean duration: 1.5 hrs. Mean size: 0.180 mm.

**Stage III:** The embryo has elongated in anterio-posterior axis and shows cellular divisions. Mean duration: 4.0 hrs. Mean size: 0.182 mm.

**Stage IV:** The head lobe and antennary bud made their first appearance during this stage. Mean duration: 1.5 hrs. Mean size: 0.183 mm

Stage V: During this stage the head lobe and the rudiment of antennae become more distinct. The cellular divisions can be clearly seen. Mean duration: 3.0 hrs. Mean size: 0. 214 mm.

Stage VI: This stage can be recognized by the development of rudiments of head, antennae, antennules and postabdomen. Mean duration: 5.5 hrs. Mean size: 0.218 mm

**Stage VII:** This stage can be recognized by the presence of pink eye. Head become more distinct. Mean duration: 3.8 hrs. Mean size: 0.236 mm

**Stage VIII:** The eye become dark, ocellus appears. Setae appear on antennae, antennules and thoracic legs and postabdomen which become more distinct. Mean duration: 7.4 hrs. Mean size: 0.266 mm

**Stage IX:** The development of antennae, antennules, thoracic limbs and postabdomen is completed Mean duration: 2.0 hrs. Mean size: 0. 0.298 mm.

#### **Release of neonates**

Embryonic development of primiparous instar of *M. triserialis* was completed in a mean duration of 29.7 hrs and the neonates were released by the movement of postabdomen of the female.

#### 5. 7. 5 Life Span and Survivorship

The survivorship curve (Fig. 16 e) indicates the relationship of age (days) and percentage survival of *M. triserialis*. The mean life span ( $\Sigma$ lx) of female is calculated as 7.55 days, while the maximum life span (Lmax) of was 16.1 days. As evident from the data survival was higher near the age of maturity and declined further a few days after maturity.

# Plate 23

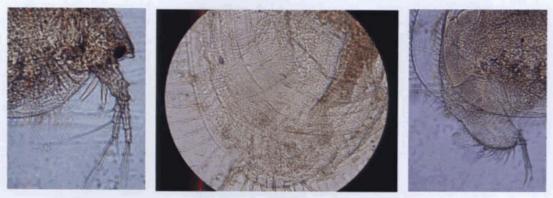


A

В

С

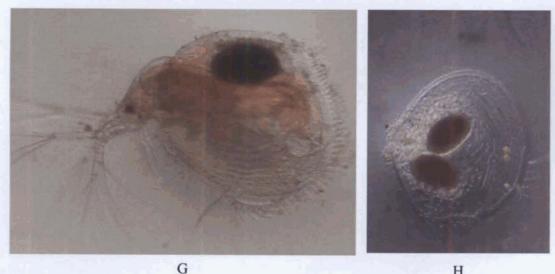
3



D

E

F



H

#### Ilyocryptus spinifer

Fig. A. Pre-adult (0.346 mm), **B**. Female with ovary (0.502 mm), **C**. Parthenogenetic female with egg (0.596 mm), **D**. Head enlarged, E. Part of shell enlarged to show growth lines, F. Postabdomen enlarged, G. Ephippial female (0.571 mm), H. Ephippium (0.506 mm).





Stage-I (0.187 mm)



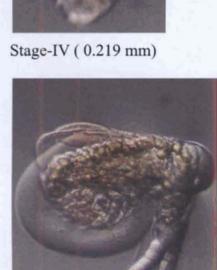
Stage-II (0.194 mm)



Stage-III (0.204 mm)



Stage-V ( 0.224 mm)



Stage-VI (0.259 mm)

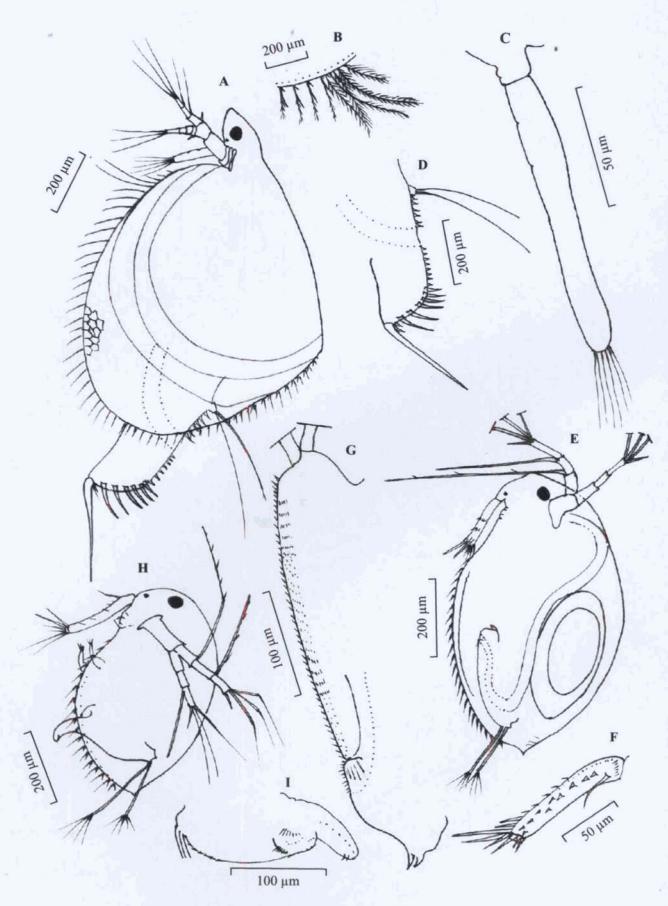


Stage-VII (0.286 mm)



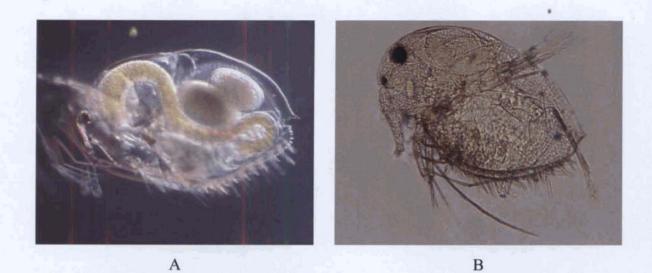
Stage-VIII (0.302 mm).

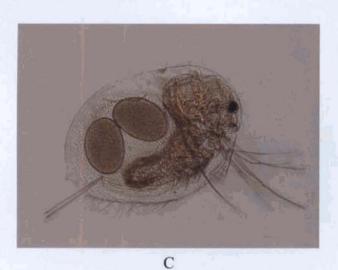
Plate 25

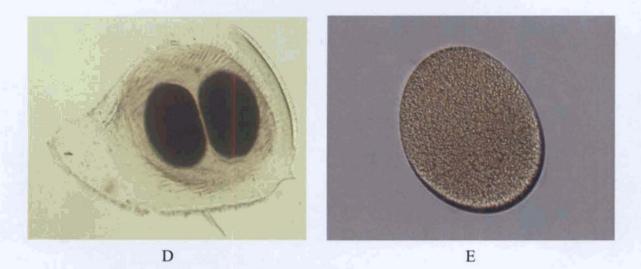


 Ilyocryptus spinifer Herrick Fig. A. Female, B. Anteroventral shell margin, C. Antennule of female, D. Postabdomen of female.
 Macrothrix triserialis (Brady) E. Female, F. Antennule of female.
 G. Postabdomen of female. H. Male, I. Postabdomen of male.

### Plate 26

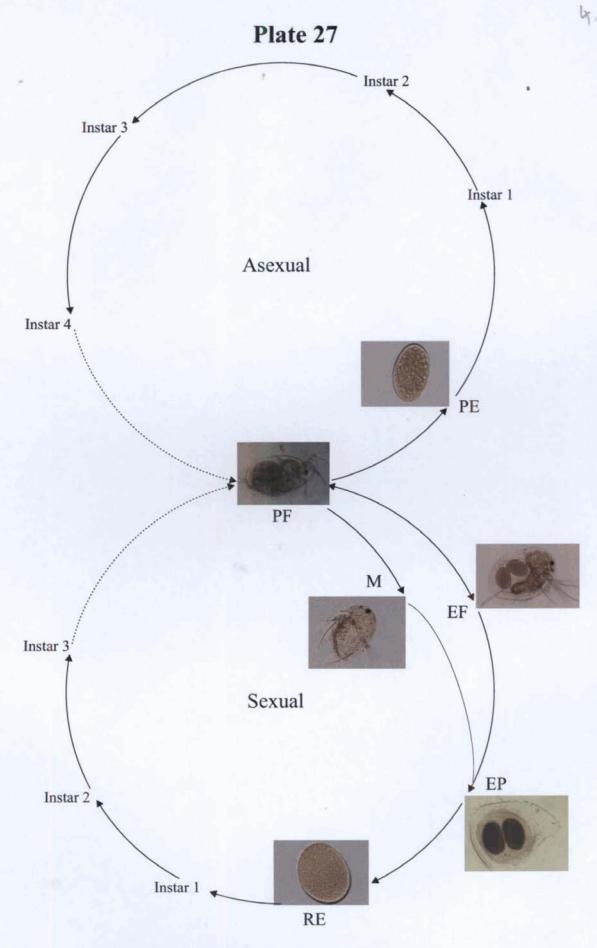






#### Macrothrix triserialis

Fig. A.Parthenogenetic female (0.884 mm), B. Male (0.521 mm), C. Ephippial female (0.770 mm), D. Ephippium (0.452 mm) E. Ephippial egg enlarged (0.216 mm).



Macrothrix triserialis Life cycle

EP-Ephippium EF-Ephippial female M-Male PE-Parthenogenetic egg, PF-Parthenogenetic female RE- Resting egg.

Ą

Plate 28



Stage I (0.176 mm)



Stage III (0.182 mm)



Stage-V (0.216 mm)



Stage II (0.180 mm)



Stage IV (0.184 mm)



Stage-VI ( 0.220 mm)



Stage-VII (0.238 mm)



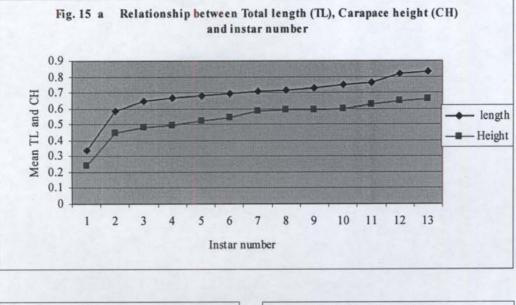
Stage-VIII (0.262 mm)

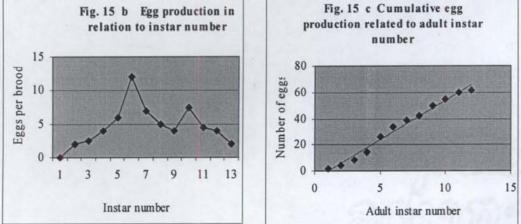


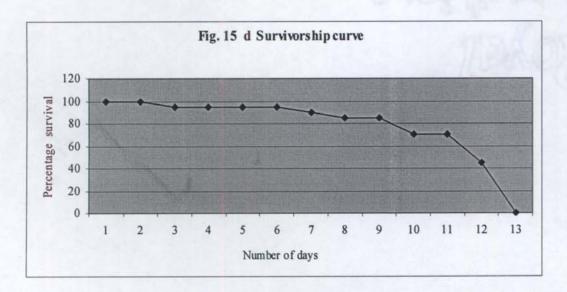
Stage-IX (0.296 mm)

Macrothrix triserialis Embryonic development

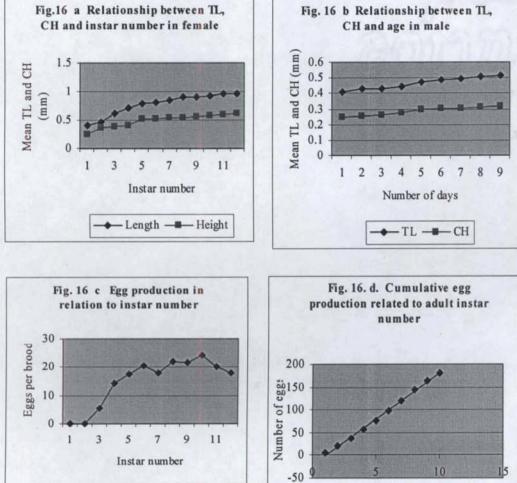
Ilyocryptus spinifer



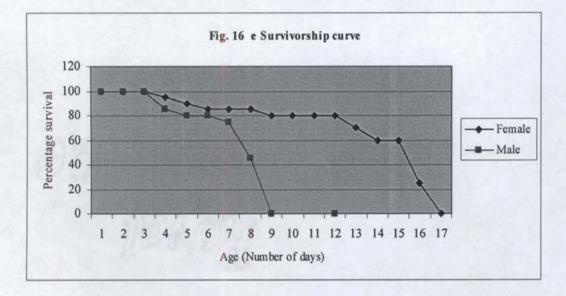


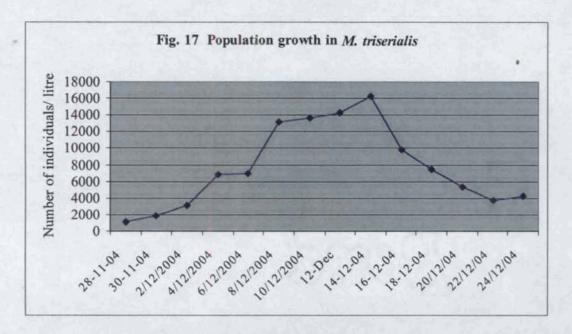


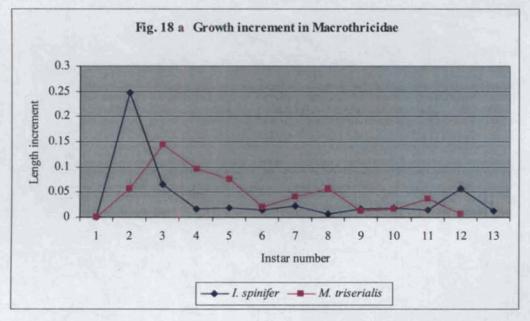
#### Macrothrix triserialis

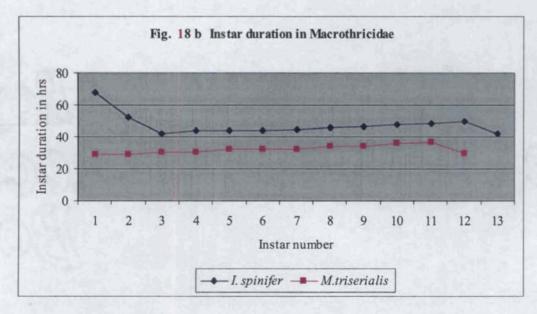


Adult instar number









### Family Chydoridae Stebbing, 1902

### 5.8 Alona pulchella King, 1853

*Alona pulchella*, a common littoral species which occurs in ponds, paddy fields, streams and reservoirs (Idris, 1983). This species was reported first in India by Petkovski (1966) from Gujarat. Subsequent reports are from Rajasthan (Nayar, 1971); West Bengal (Sharma, 1978); Tamil Nadu (Michael and Sharma, 1988) and Andhra Pradesh, (Durga Prasad and Padmavathy, 2003a).

#### 5. 8. 1. External Morphology

#### Parthenogenetic female (Plate 29. Fig. A)

Body oval. The valves punctuate, without longitudinal striations, posterodorsal and posteroventral corner rounded; ventral margin with series of setae (Plate 30. Fig. H). Eye distinct; ocellus slightly smaller than the eye, situated half way between eye and tip of the rostrum. Antennules small, not reaching the tip of the rostrum (Plate 30. Fig. F). Labrum with convex anterior margin (Plate 30. Fig. G). Postabdomen with parallel dorsal and ventral edges; with 9 anal teeth, lateral setae arranged in 7 fascicles, distalmost seta being the longest; claw with basal spine, about  $1/3^{rd}$  the length of claw (Plate 30. Fig. I). Mean size:  $0.438 \times 0.258$  mm.

Male (Plate 29. Fig. B)

Smaller than female. Valves with almost parallel dorsal and ventral margins, posterodorsal and posteroventral corner rounded. Antennules stout and broad extending beyond the apex of rostrum with sensory setae (Plate 30. Fig. J), First thoracic leg with a copulatory hook. Postabdomen slightly narrowing distally, with groups of lateral setae; claw with long basal spine. Mean size:  $0.350 \times 0.225$  mm

#### Ephippial female (Plate 29. Fig. C)

Ephippial female similar to parthenogenetic female; but with distinct black pigmentation. Ephippium single, attached to carapace with single egg placed in its upper half and shed along with a major portion of the valve (Plate 29. Fig. D). Egg single, oval and brown coloured. Mean size of ephippial female:  $0.475 \times 0.291$  mm. Mean size of ephippium: 0.421 mm.

#### 5.8.2 Reproduction

Males and Ephippial females were produced in the laboratory culture throughout the period of study. However the parthenogenetic females dominated the culture. The events in the life cycle of parthenogenetic female are described below.

#### Life cycle of parthenogenetic female

The population obtained in the collection from Site 6 comprised of a good number of adult parthenogenetic females and ephippial females. The

adult females were found to bear only 2 eggs. The egg bearing females were sorted out for the laboratory culture.

The population developed in the laboratory culture comprised asexually reproducing females, ephippia bearing females and males. A good number of males and ephippial females appeared in the culture throughout the period of study. This indicates the frequent occurrence of sexual reproduction in this species. However, the parthenogenetic females dominated the culture. The life cycle of *A. pulchella* is described below.

#### **Pre-adult instar**

The neonates produced from parthenogenetic females had a mean birth size (SaB) of  $0.291 \times 0.175$  mm. Both the first and second moulting occurred in a uniform duration of 23.5 hrs. The total pre-adult instar duration was 47.0 hrs and mean PID was 23.5 hrs.

#### Attainment of maturity

The ovary was clearly seen at  $26\pm1$  hrs during the 2<sup>nd</sup> instar. They started to bear eggs after completion of 2<sup>nd</sup> moult at 47.0 hrs; and hence the AFR was 1.96 days. The SFR was  $0.425 \times 0.250$  mm.

#### Egg production

The eggs were deposited into brood pouch after completion of two moults. The eggs measured a mean size of  $0.166 \times 0.096$  mm. During this primiparous instar (3<sup>rd</sup> instar) egg production started with an average of

2.0 eggs/ brood. The primiparous instar was completed in 26.5 hrs and the FGT is calculated as 73.5 hrs.

During the subsequent instars there was a steady production of 2.0 eggs/ brood. This trend of egg production continued throughout the life span upto the 12<sup>th</sup> instar (Table 34). The female underwent moulting towards the end of each instar.

#### Fecundity

The relationship of egg production with instar number is represented in Fig. 19 b. A constant clutch size of 2 eggs/ brood was produced throughout the early adult instars with 1.45 eggs/ day of adult life. The cumulative number of eggs produced ( $\Sigma mx$ ) during the entire life span was 20.0 (Table 34). Ten broods were produced during the entire life.

The  $\Sigma$ mx is linearly correlated with instar number Fig. 19 c. The rate of egg production (REP) of *A. pulchella* is calculated as 1.923.

#### 5.8.3 Growth

The first pre-adult neonates had a mean TL of 0.325 mm and second instar had TL of 0.350 mm. Primiparous stage was attained during the  $3^{rd}$  instar when the mean TL was 0.425 mm. The maximum mean TL of 0.525 mm was attained at the end of  $12^{th}$  instar (Table 35).

The mean CH during the first pre-adult instar was 0.183 mm and attained CH of 0.204 mm at second instar. They attained a mean CH of 0.250 mm during the primiparous condition. Maximum CH was attained in 12<sup>th</sup>

instar with CH of 0.328 mm (Table 35). During the life span each individual has undergone two pre-adult and ten adult moults.

The relationship between TL, CH and instar number of *A. pulchella* has been represented in Fig. 19 a. The correlation coefficients of the life history characters are given in Table 36, which shows positive correlation between TL and CH (r = 0.996).

The increment of TL and CH during each instar is given in Table 35. Maximum growth increment recorded during the life cycle was in 3<sup>rd</sup> instar with TL of 21.42% and CH of 22.54% respectively. The most significant decrease in growth increment occurred after 7<sup>th</sup> instar.

#### 5.8.4 Life Span and Survivorship

The survivorship curve (Fig. 19 d) indicates the relationship of age (days) and percentage survival of *A. pulchella*.

The rate of survival was higher near the age of maturity and declined further a few days after maturity. The maximum life span (Lmax) observed during the present study was 15.75 days.

## 5.9 Oxyurella singalensis (Daday, 1898)

Oxyurella singalensis is a littoral cladoceran inhabiting in all types of freshwater habitats especially ponds, paddy fields and marshes. The name Oxyurella was coined by Dybowski and Grochowski (1894). The first description of this species is by Daday (1898).

In India, this species is reported first by Sharma (1978) from West Bengal and by Michael and Sharma (1988) from Kerala. A study on the biology of this littoral species is lacking, and hence the present study of the life history of male and parthenogenetic female has been made.

#### 5. 9. 1 External Morphology

#### Parthenogenetic female (Plate 31. Fig. B)

Body evenly rounded, posterior margin convex, maximum height in the middle. Head shield broad in the middle, narrowly rounded anteriorly and broadly rounded posteriorly. Rostrum blunt. Ocellus smaller than eye, situated closer to the eye than to the apex of rostrum. Plate of labrum rounded with slightly acute apex (Plate 30. Fig. A). Valves with almost straight ventral margins, with a series of setae and setules along the posteroventral corner. Antennules short, not reaching the apex of rostrum. Antennae with 3-segmented dorsal and ventral ramus. Antennary setation: (0-0-3)/(1-1-3). Postabdomen slightly tapering distally, with 12 sharply pointed anal denticles which decrease in size proximally; distinct preanal and postanal corners, distal corner rounded; claw long, curved dorsally, setules on the concave surface; basal spine long about  $\frac{1}{2}$  the length of claw and an additional small spine at the base of the claw (Plate 30. Fig. B). Mean size:  $0.733 \times 0.475$  mm.

#### Male (Plate 31. Fig. C)

Valves with rounded posterodorsal and posteroventral corner; ventral margin with convexity in the middle; short marginal setae before convexity and longer setae behind it (Plate 30. Fig C). The first thoracic leg bears a curved blunt hook (Plate 30. Fig. D). Postabdomen almost uniformly wide, but slightly narrowed distally; anal spines confined mostly to rounded dorsal end (Plate 30. Fig. E). Mean size:  $0.583 \times 0.341$  mm.

#### Ephippial female (Plate 31. Fig. D)

The ephippial female in external appearance resemble parthenogenetic females. The shell does not bulge outward. The ephippium is transparent, light yellow coloured. The egg single, dark brown in colour, and occupied almost at the centre of ephippium surrounded by a foamy mass. Ephippium cast of along with a major portion of the carapace (Plate 31. Fig. E). Some of the ephippium was cast off without eggs within it (Plate 31. Fig. F). Mean size of female: 0.670 mm. Size of ephippium: 0.570 mm.

#### 5.9.2 Reproduction

Males were present throughout the period, while the ephippial females made their appearance only occasionally. However, the parthenogenetic females dominated the culture. The important events in the life cycle of male and female are given below.

#### Life cycle of Male

The males of *O. singalensis* were reported first from India by Michael and Sharma (1988). The males were produced in the laboratory throughout the period of present study. Twenty neonates of less than 12 hrs of age were sorted out and reared individually for life cycle studies following the methodology cited in section 3. 2. 3.

The male neonates had a mean birth size (SaB) of  $0.437 \times 0.269$  mm. The first moulting was at 96.0 hrs after birth. There was no moulting further and they attained maturity. The sexually mature individuals could be recognized by their yellow coloured testis. After the first moulting they attained mean TL of 0.516 mm and CH of 0.320 mm (Table 40). The maximum percentage increment of CH was observed on 4<sup>th</sup> day with 6.67%. The size increments decreased subsequently (Table 40).

The relationship between TL and CH are represented in Fig. 20 b. Maximum size attained during the study was  $0.560 \times 0.344$  mm on  $23^{rd}$  day. The mean life span of male is calculated as 9.37 days while the maximum life span observed during the present study was 22.79 days.

#### Life cycle of parthenogenetic female

The population developed during the laboratory culture comprised asexually reproducing females, ephippia bearing females and males. The parthenogenetic reproduction occurred throughout the period. The features characteristic of the life cycle is given as follows.

#### **Pre-adult instar**

The neonates produced from the parthenogenetic females had a birth size (SaB) of  $0.492 \times 0.318$  mm. The first moulting occurred after duration of 36.0 hrs; while the second moulting occurred at an interval of 38.5 hrs. The total pre-adult instar duration of *O. singalensis* was 74.5 hrs. The mean duration (PID) is 37.25 hours.

#### Attainment of maturity

The ovary was conspicuous at  $40\pm2$  hrs towards the beginning of  $2^{nd}$  instar (Plate 31. Fig. A). However, they attained maturity after the second moult and started to bear eggs at 74.5 hrs (Table 37), and hence the AFR was 3.10 days. They attained a mean SFR of 0.544×0.400 mm during  $3^{rd}$  instar.

#### Egg production

The eggs were deposited into the brood pouch within 15.0 minutes after the second moult. During this primiparous instar ( $3^{rd}$  instar) egg production started with 2.0 eggs/ brood. Soon after deposition into brood pouch the eggs attained a triangular shape. The primiparous instar was completed in a mean duration of 40.2 hrs and the first generation time (FGT) is calculated as 114.7 hrs.

During the succeeding instars each clutch comprised of 2.0 eggs/ brood. This steady egg production continued until 15<sup>th</sup> instar without any peak (Fig. 20 c). They continued moulting and egg production until the penultimate instar (16<sup>th</sup>). However, there was no egg production in the last two instars. The relationship of instar duration and instar number is represented in Fig. 22.

#### Fecundity

The relationship of egg production with instar number is represented in (Fig. 20 c). The females produced a constant number of eggs throughout the adult instars with 0.70 eggs/day of adult life.

The cumulative number of eggs produced ( $\Sigma$ mx) during the entire life span was 26.0 (Table 37). Thirteen broods were produced during the entire life with an average of 2.0 eggs /brood. The cumulative number of eggs produced  $\Sigma$ mx is linearly correlated with instar number (Fig. 20 d). The rate of egg production (REP) of *O. singalensis* is 1.848.

#### 5.9.3 Growth

The first pre-adult neonates had a mean TL of 0.504 mm and  $2^{nd}$  instar had TL of 0.520 mm. Primiparous stage was attained during the  $3^{rd}$  instar when the mean TL was 0.544 mm. The maximum mean TL of 0.824 mm was attained at  $14^{th}$  instar, and no increase further upto  $17^{th}$  instar (Table 38).

The mean CH during the first pre-adult instar was 0.328 mm and attained CH of 0.344 mm at 2<sup>nd</sup> instar. A mean CH of 0.400 mm was attained during the primiparous condition. The maximum CH was attained in 14<sup>th</sup> instar with a mean CH of 0.536 mm and no increase further (Table 38). During the life span each individual has undergone two pre-adult and fifteen adult moults.

The size increments during each instar are represented in Table 38. Maximum growth increment recorded during the life cycle was with TL (23.52%) and CH (16.27%). The relationship between TL, CH and instar number of *O. singalensis* female is represented in Fig. 20 a. The correlation coefficients of the life history characters are given in Table 39.

#### 5. 9. 4 Embryonic Development

The stages of embryonic development of *O. singalensis* are represented in Plate 32. The most conspicuous features of the developmental stages are given below.

Stage I: This stage is recognized by the presence of oval egg with yellow colour. Mean duration: 1.5 hrs. Mean size: 0.223 mm

**Stage II:** This stage is recognized by the presence of triangular-shaped egg with dark-brown colour. Mean duration: 3.0 hrs. Mean size: 0.20 mm.

**Stage III:** The embryo shows two distinct zones. The outer area becomes transparent while the inner area is granular with a yellow coloured fat globule. Mean duration: 8.4 hrs. Mean size: 0.234 mm.

Stage IV: The head lobe starts to develop. The yolk and fat globules get concentrated in the middle. Mean duration: 6.0 hrs. Mean size: 0.256 mm.

**Stage V:** During this stage the head lobe become more distinct. The cellular divisions are clearly visible. Mean duration: 5.6 hrs. Mean size: 0.268 mm.

Stage VI: The cellular divisions of anterior region are completed with the development of rudiments of head and antennae. Mean duration: 3.5 hrs. Mean size: 0.286 mm.

Stage VII: This stage could be recognized by the presence of eye, antennules and antennae. Mean duration: 9.5 hrs. Mean size: 0.294 mm

**Stage VIII:** During this stage the development of head and postabdomen is completed. The eye and ocellus become more conspicuous, thoracic legs are developed and the embryo resembled the adult in external morphology. Mean duration: 2.5 hr. Mean size: 0.326 mm.

#### **Release of neonates**

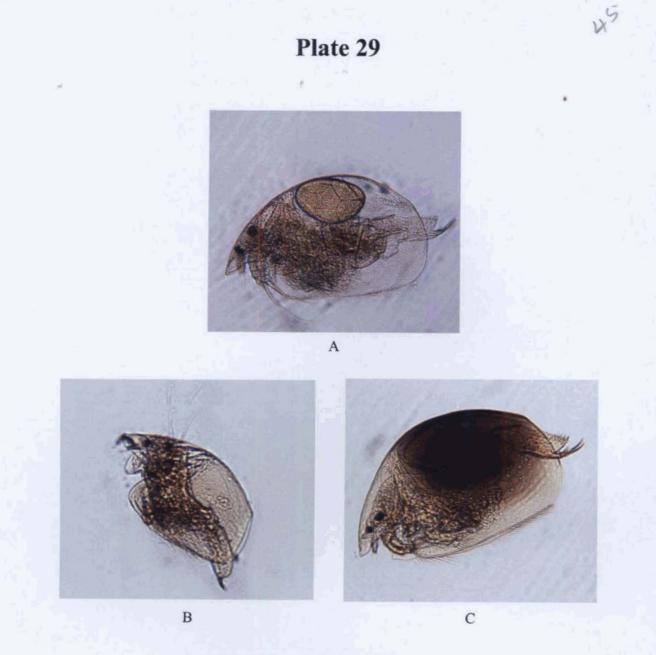
Embryonic development of primiparous instar of *O. singalensis* was completed in a mean duration of 40.0 hrs. At the end of each adult instar two neonates were released from the brood pouch of the mother by the movement of of female postabdomen followed by moulting.

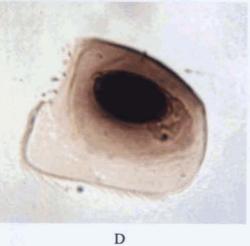
#### Moulting

The moulting was completed in two steps. First, the carapace is separated from the body at the region of head shield. This is followed by the separation of the exoskeleton of the head shield and postabdomen. The old carapace is shed as exuvium which gets detached from the animal and sinks to the bottom.

#### 5. 9. 5 Life span and Survivorship

Fig. 20. e represents the survivorship curve of *O. singalensis*. The mean life span ( $\Sigma$ lx) of female is 17.14 days, while the maximum life span (Lmax) observed in the present study was 40.0 days (Table 38).



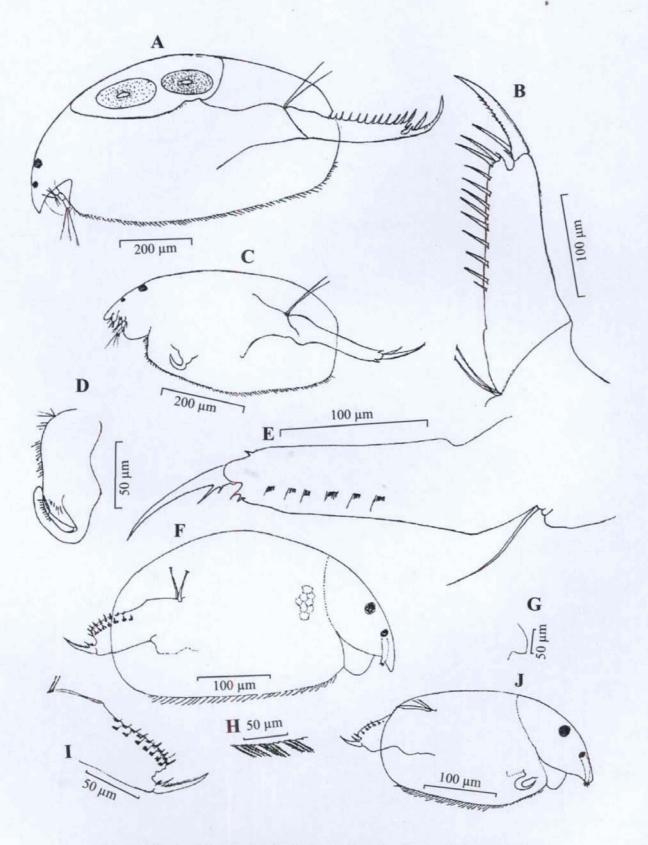


### Alona pulchella

Fig. A. Parthenogenetic female (0.443 mm), B. Male (0.348 mm), C. Ephippial female (0.468 mm), D. Ephippium ( 0.426 mm).

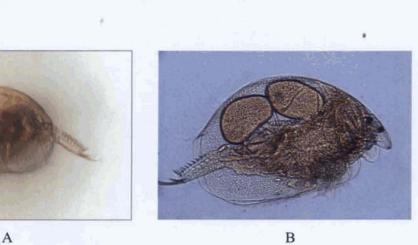
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X



 Oxyurella singalensis (Daday) Fig.A. Female. B. Postabdomen of female, C. Male, D. 1<sup>st</sup> Thoracic leg, E. Postabdomen of male.
 Alona pulchella King F. Female. G. Plate of labrum. H. Posteroventral margin of shell, I. Postabdomen of female, J. Male.

# Plate 31





С



D



#### Oxyurella singalensis

Fig. A. Pre-adult (0.524 mm), B. Parthenogenetic female (0.735 mm), C. Male (0.580 mm),
 D. Ephippial female (0.677 mm), E. Ephippium with egg (0.539 mm)
 F. Ephippium without egg (0.548 mm).

# Plate 32



Stage-I ( 0.226 mm)



Stage-II (0.230 mm)



Stage-III (0.232 mm)



Stage-IV (0.258 mm)



Stage-V ( 0.269 mm)



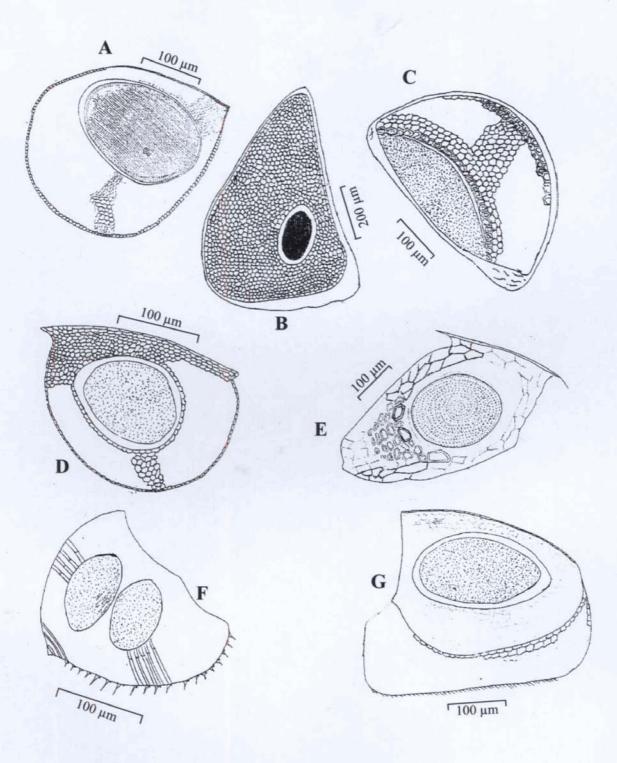
Stage-VI (0.285 mm)



Stage-VII (0.298 mm)



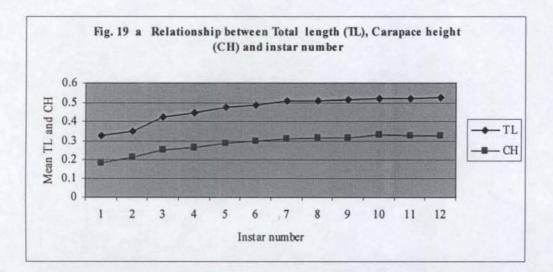
Stage VIII (0.340 mm)

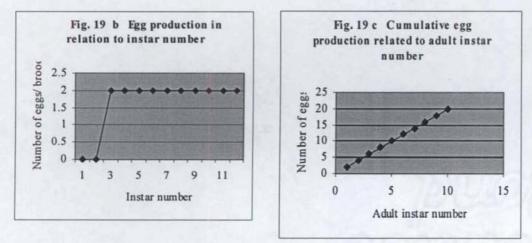


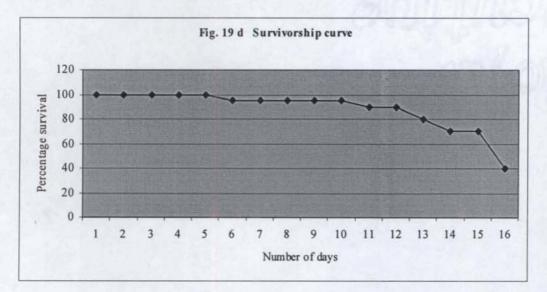
#### **Ephippial Morphology**

Fig. A. Ceriodaphnia cornuta, B. Simocephalus serrulatus, C. Scapholeberis kingi, D. Moina brachiata, E. Moinodaphnia macleayi, F. Macrothrix triserialis, G. Alona pulchella (In Figs. A, C, D, F and G ornamentation is shown only partly)

### Alona pulchella







### Oxyurella singalensis

