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Larval development of the Indian mangrove Crab *Psuedosesarma glabrum* (NG, Rani & Nandan, 2017) (Brachyura: Grapsoidea: Sesarmidae)

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Original Article

Abstract

The present study describes the larval development of *Psuedosesarma glabrum* under laboratory conditions. *P. glabrum* is a sesarmid crab inhabiting the intertidal mixed mangrove zones of Vembanad ecosystem, south-west coast of India. Larval phase consists of four zoeal stages and one megalopa before metamorphosis to the first crab stage. Zoea I and zoea II last on an average 4±2 days, zoea III 7±2 days, zoea IV 6±2 days and first megalopa appeared on 29±2 day of hatching. All zoeal and megalopa stages are described and illustrated in detail. This work is the first report on the larval development of *P. glabrum* and larvae are compared with that of other species of *Psuedosesarma*.

Keywords*: Sesarmidae, Pseudosesarma glabrum, zoea, megalopa, larval development*

Introduction

Sesarmid crabs are among the most diverse and important faunal components of mangrove forest, which consist of more than 127 identified and about 30 unidentified species in temperate and tropical regions of the world (Guerao *et al.*, 2004). *Psuedosesarma* is a new genus of family sesarmidae amid a type species *Sesarma edwardsii* De Man, 1887 (Serene and Soh, 1970). The genus *Pseudosesarma* has a carapace slightly shorter than the breadth between the external orbital angles; anterior frontal margin with a shallow but marked median concavity; the anterior border of cheliped with a sub-distal triangular process; the gastric and cardiac region well delimited by a groove; the antero-lateral tooth well marked (Sakai, 1976). Currently, ten species are placed in *Pseudosesarma*: *Pseudosesarma bocourti* (Milne-Edwards, 1869), *Pseudosesarma granosimanum* (Miers, 1880), *Pseudosesarma crassimanum* (De Man, 1887), *Pseudosesarma edwardsii* (De Man, 1887), *Pseudosesarma moeschii* (De Man, 1892), *Pseudosesarma laevimanum* (Zehntner, 1894), *Pseudosesarma modestum* (De Man, 1902), *Pseudosesarma johorense* (Tweedie, 1940), *Pseudosesarma patshuni* (Soh, 1978; Ng *et al.*, 2008) and *Pseudosesarma glabrum* (Ng *et al.*, 2017). *P. glabrum* (Fig. 1) is a tropical-sesarmid crab inhabiting the intertidal mixed mangrove zones along the Vembanad ecosystem (Ramsar site-1214) in Kerala, south-west coast of India.

Studies on larval development have importance in ecological and physiological investigations of a crab species. Larval development of several sesarmid crabs have been described

Fig. 1. *Psuedosesarma glabrum*- Phenotype; Female (carapace width= 12 mm); (A) Dorsal view; (B) Ventral view

by many workers (Terada, 1976; Lago, 1993a; Mia and Shokita 1996, 1997; Cuesta *et al*., 1999). In India, only a few larval studies were reported from the family, *ie Sesarma tetragonum* (Rajabai, 1961); *Sesarma lantum* (Sankoli, 1975); *Sesarma intermedius, Holometopus haematocheir, Chiromantes bidnes* (Baba and Fukuda, 1976); *Sesarma andersoni*, *Sesarma brockii* (Vijayakumar and Kannupandi, 1986; Kannupandi *et al*., 2000), *Neoepisesarma mederi* (Selvakumar *et al.,* 1987), *Sesarma bidens* (Krishnan and Kannupandi,1988), *Sesarma edwardsi* (Kannupandi and Pasupathi, 1994), *Parasesarma plicatum* (Selvakumar, 1999). Among the ten identified species from the genus *Psuedosesarma*, complete larval development has been described only for *P. bocourti* and *P. moeschii* from hatching to megalopa (Guerao *et al*., 2007) and for *P. crassimanum,* only for first zoeal stage (Cuesta *et al*., 2006). The present study gives a detailed description of all zoeal and megalopa stages of *P. glabrum* and is compared with other species of the genus.

Material and methods

Wild ovigerous females of *P. glabrum* (16 ± 2 mm carapace width and 14 ± 2 mm carapace length) were collected along the mangrove swamps in the northern Vembanad ecosystem (9°52′1.42′′N, 76°18′54.97′′E), south-west coast of India and brought to the animal rearing facility of Department of Marine Biology, Microbiology and Biochemistry, School of Marine Sciences, Cochin University of Science and Technology, India. 5 crabs were reared in separate glass tanks (0.5m x 0.5m) containing filtered sea water maintained at a salinity of 25 ppt and temperature of 26 ± 1^{0}C. The crabs were fed with fresh leaves of *Rhizophora mucronata* and *Avicennia officinalis* and water exchanged partially for removal of waste materials. The average incubation period lasted for 27 ± 2 days for all crabs and newly hatched larvae were transferred to glass beakers of 500 ml capacity (25/ litre) for mass culture. Individual larvae were also reared in test tubes (50 ml capacity) to characterize developmental stages. 25 ml of sea water used for 50 ml test tubes and 25% of water replaced twice a day. Seawater with 25 ppt salinity and temperature of 26 ± 1^{0}C was maintaained. Larvae were subjected to a 12 h light: 12 h dark artificial light regime. A mixed algal diet of *Nannochloropsis salina* (10x10^6 cells/ml), *Chaetoceros calcitrans* (2.85x10^{6}cells/ml), *Tetraselmis gracilis* (7 x 10^6 cells/ml) and the rotifer *Brachionus rotundiformes* (10-15 ind./ml) were offered once in a day. Larvae of each developmental stage were fixed and preserved in 4% formaldehyde. All measurements were made by an ocular micrometer and were based on measurements of 10 individuals per stage. The carapace length (CL) for zoeal stages was measured from the base of the rostrum to the most posterior margin; carapace width (CW) was the greatest distance across the carapace; rostro-dorsal length (RDL) from the tip of the rostral spine to the tip of the dorsal spine; antennal exopod length (AEL) from the base of the antennal exopod to the distal margin; protopodal process length (PPL) from base of the antennal exopod to the tip of the protopodal process; telson length (TL) from a line across the anterior margin to the posterior margin of the telson; furcal length (FL) is an imaginary line across the base of the outer seta on the posterior margin of the telson to the furcal tip (Clark *et al*., 1998). Appendages of each stage were dissected carefully using fine needles with the help of stereo microscope. Drawings of the entire larvae and dissected appendages were made using a stereo and phase contrast microscope with camera lucida (NIKON-YIDT-JAPAN).

Results

Larval development of *P. glabrum* consist of four zoeal stages and one megalopa stage; the latter metamorphosed to crab stage (Crab Instar-I). Zoea I and II stages took 4 ± 2 days each, zoea III 7 ± 2 days, the zoea IV 6 ± 2 days and megalopa 29 ± 2 days. The morphometric details and duration of different zoeal stages are presented in Table 1.

Table 1. Zoeal stages with morphometric details of Indian mangrove crab *Pseudosesarma glabrum* (Mean length and standard deviation in mm).

Zoeal stages (Duration in days)	RDL (mm)	CL (mm)	AEL (mm)	PPL (mm)	FL (mm)	TL (mm)
Zoea-I (4 ± 2 d)	0.45 ± 0.07	0.37 ± 0.04	0.07 ± 0.02	0.12 ± 0.01	0.15 ± 0.02	0.27 ± 0.01
Zoea-II (4 ± 2 d)	0.60 ± 0.15	0.45 ± 0.06	0.12 ± 0.01	0.15 ±0.01	0.19 ± 0.05	0.30 ± 0.03
Zoea-III (7 ± 2 d)	0.90 ± 0.11	0.57 ± 0.05	0.15 ± 0.01	0.18 ± 0.02	0.30 ± 0.03	0.37 ± 0.04
Zoea-IV (6 ± 2 d)	1.12 ± 0.04	0.67 ± 0.02	0.18 ± 0.02	0.22 ± 0.04	0.37 ± 0.01	0.45 ± 0.03

RDL, Rostro-Dorsal Length; CL, Carapace Length; AEL, Antennal Exopod Length; PPL, Protopodal Process Length; FL, Furcal Length ; TL, Telson Length

Descriptions of zoeal stages are given below; first zoeal stage is given in detail, and in the subsequent stages, only the differences and further development in appendages or setation are given.

Pseudosesarma glabrum (Ng *et al*., 2017)

Zoea-I

(Size: RDL: 0.45 mm; CL: 0.37 mm)

Morphology – Carapace (Fig. 2a): globose, smooth without tubercles, dorsal spine well developed and posteriorly curved, rostral spine straight, similar in length to dorsal spine, lateral spines absent, eyes are sessile.

1st antennae/antennule (Fig. 3A-a): uniramous, endopod absent, exopod unsegmented with 1terminal setae and 3 terminal aesthetascs.

2nd antennae (Fig. 3B-a): biramous, have an equal length of rostral spine and bearing two unequal rows, protopodal process with one row of 6-7 spines of different sizes, exopod elongate more than half of the protopod length with one large and one small terminal setae along with small terminal spines.

Mandible: Incisor and molar processes well developed and endopod palp absent.

1st maxillae/maxillule (Fig. 4A-a): endopod two segmented, one seta, on the proximal segment; one sub terminal seta and 4 terminal setae, exopod and epipod setae absent, coxal endite with 6 setae and basal endite with 5 setae.

2nd maxillae/maxilla (Fig. 4B-a): coxal endite bilobed with 5+3 setae, basal endite bilobed with 4+3 setae, endopod bilobed and unsegmented (2+3 setae), scaphognathite with 4 marginal setae.

1st maxilliped (Fig. 5A-a): coxa with 1 setae, basis with 9-10 medial setae arranged on the inner side and a mat of long dorso basal microtrichiae on the outer side, Endopod 5 segmented with 2, 2, 1, 2, 4 ± 1; third segment with a mat of dorsal microtrichiae, exopod two segmented, distal segment with 4 long plumose natatory setae.

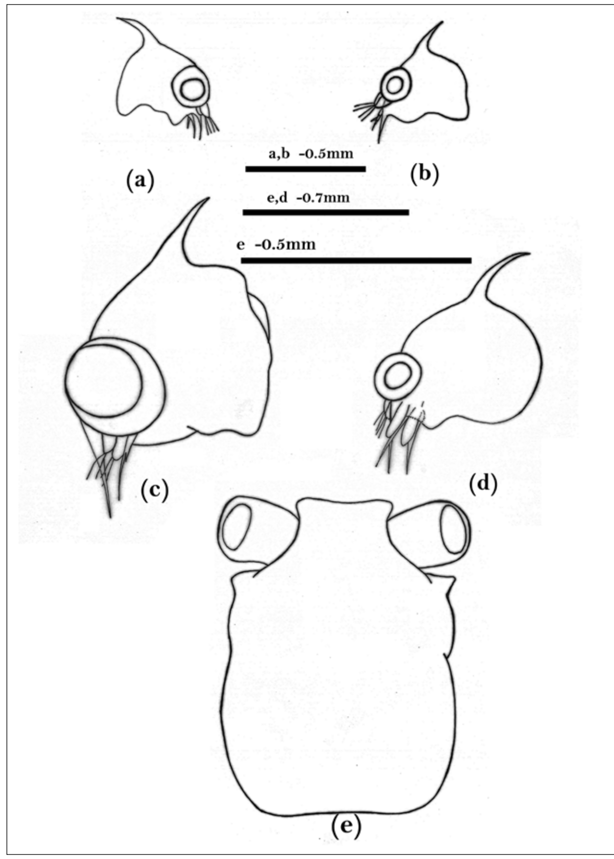

Fig. 2. *Psuedosesarma glabrum*- Carapace (10X), lateral view: a: zoea I; b: Zoea II; c: zoea III; d: zoea IV; Carapace dorsal view, e: megalopa

2nd maxilliped (Fig. 5B-a): coxa without setae, basis with 4 medial setae arranged 1+1+1+1, exopod two segmented, distal segment with 4 long plumose natatory setae, endopod 3 segmented with 0, 1, 5 setae.

3rd maxilliped: absent.

Pereiopods: absent

Pleon (Fig. 6A-a): 5 somites, 2nd and 3rd somites have a pair of dorsolateral processes, 3rd to 5th somites have posterolateral

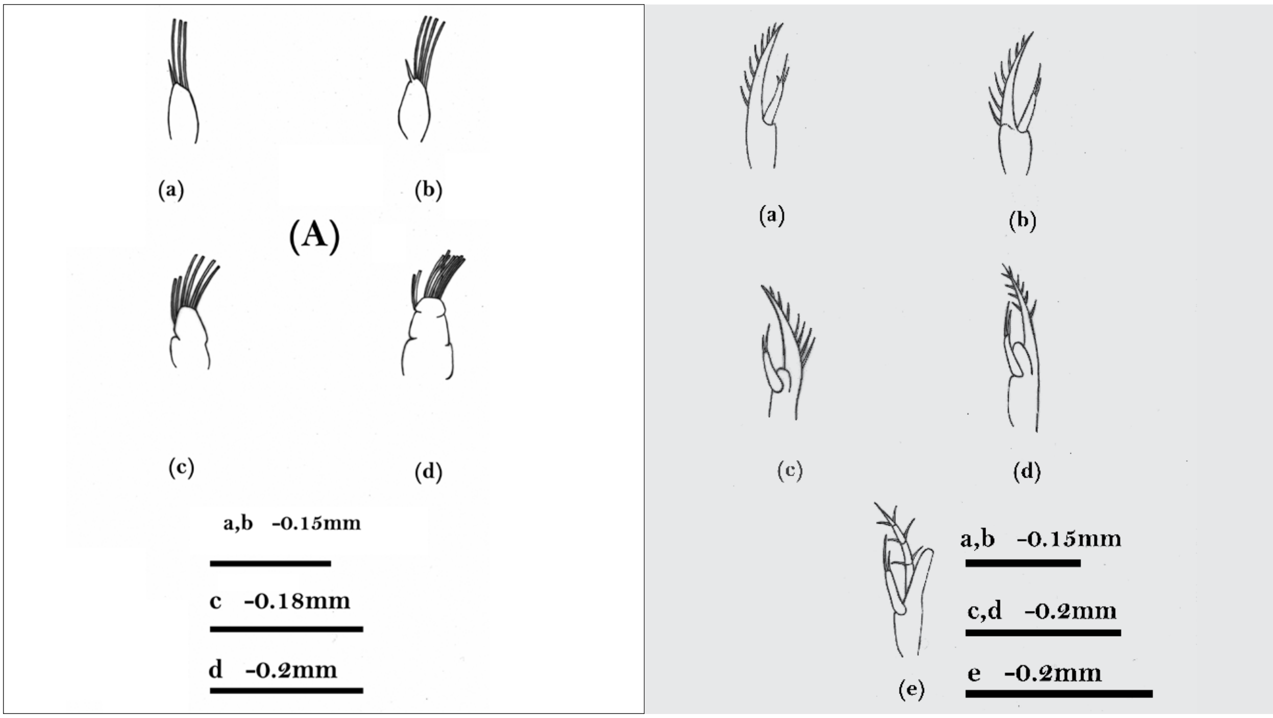

Fig. 3. *Psuedosesarma glabrum*- (A) Antennule (10X): a: zoea I; b: zoea II; c: zoea IV; d: megalopa; (B) Antennae (10X): a: zoea I; b: zoea II; c: zoea III; d: zoea IV; e: megalopa

processes and posterolateral processes on 5th somite is very long and slightly curved, pleopods absent; 2-5 somites with a pair of posterodorsal setae.

Telson: bifurcated, 3 pairs of serrulate setae on inner margin, furca long with two rows of spines and outer part without spine, only scattered minute spinules.

Zoea-II

(Size: RDL: 0.60 mm; CL: 0.45 mm)

1st antennae/antennule (Fig. 3A-b): uniramous, exopod with 4 terminal aesthetascs and 1setae.

2nd antennae (Fig. 3B-b): minute endopod bud is present and exopod spines are absent.

2nd maxillae/maxilla (Fig. 4B-b): scaphognathite with 5-6 marginal setae.

1st maxilliped (Fig. 5A-b): exopod distal segment with 6 long plumose natatory setae.

2nd maxilliped (Fig. 5B-b): exopod distal segment with 6 long plumose natatory setae.

Pereiopods: absent.

Pleon (Fig. 6A-b): 1st somite with a long mid dorsal seta and 2-5 somites with a pair of posterodorsal setae, pleopods absent.

Telson: similar to the previous stage.

Zoea-III

(Size: RDL: 0.90 mm; CL: 0.57 mm)

2nd antennae (Fig. 3B-c): Endopod bud elongated, not as long as exopod

1st maxillae/maxillule (Fig. 4A-b): epipod setae present; endopod 2 segmented, 1 proximal seta, 2 subterminal and 4 terminal setae.

2nd maxillae/maxilla (Fig. 4B-c): scaphognathite with 9-10 marginal setae, basal endite with 5+5 setae.

1st maxilliped (Fig. 5A-c): 2nd and 4th segments of endopod with one additional seta, exopod distal segment with 8 long plumose natatory setae.

2nd maxilliped (Fig. 5B-c): endopod 3 segmented with 0, 1, 6 setae, exopod distal segment with 8 long plumose natatory setae.

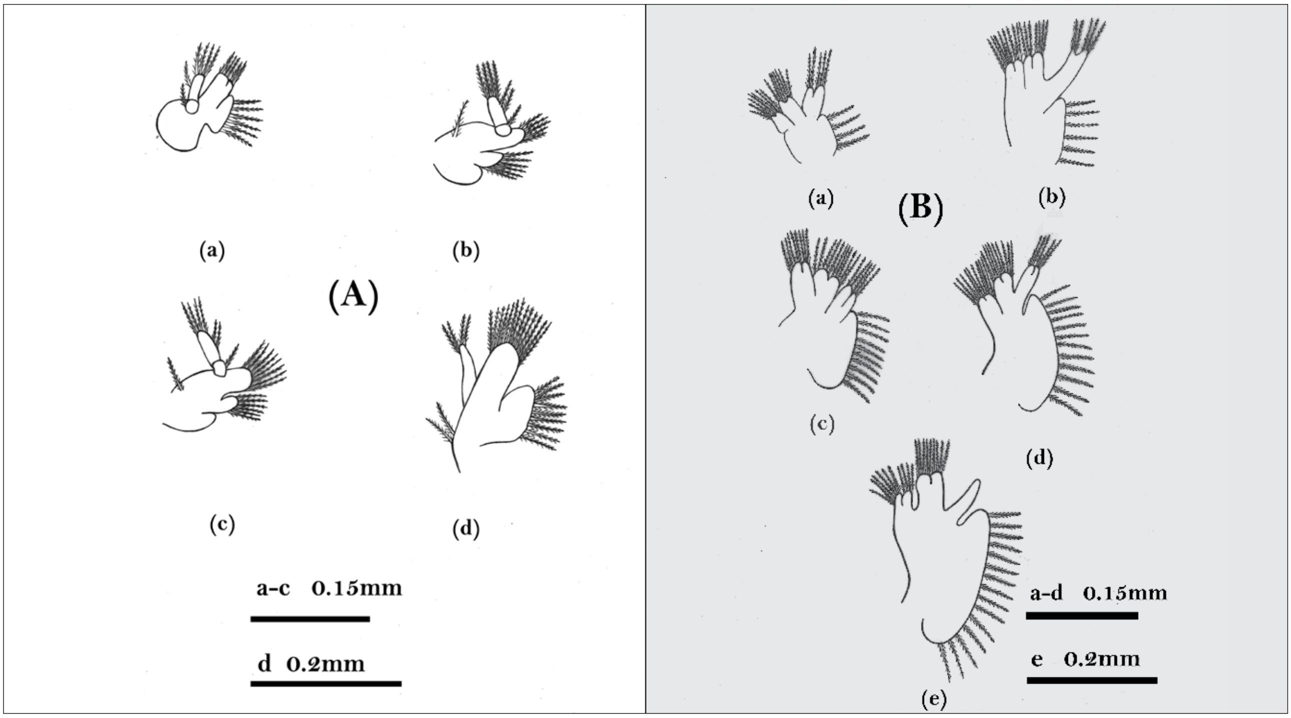

Fig. 4. *Psuedosesarma glabrum*- (A) Maxillule (20X): a: zoea I; b: zoea III; c: zoea IV; d: megalopa; (B) Maxilla (20X): a: zoea I; b: zoea II; c: zoea III; d: zoea IV; e: megalopa

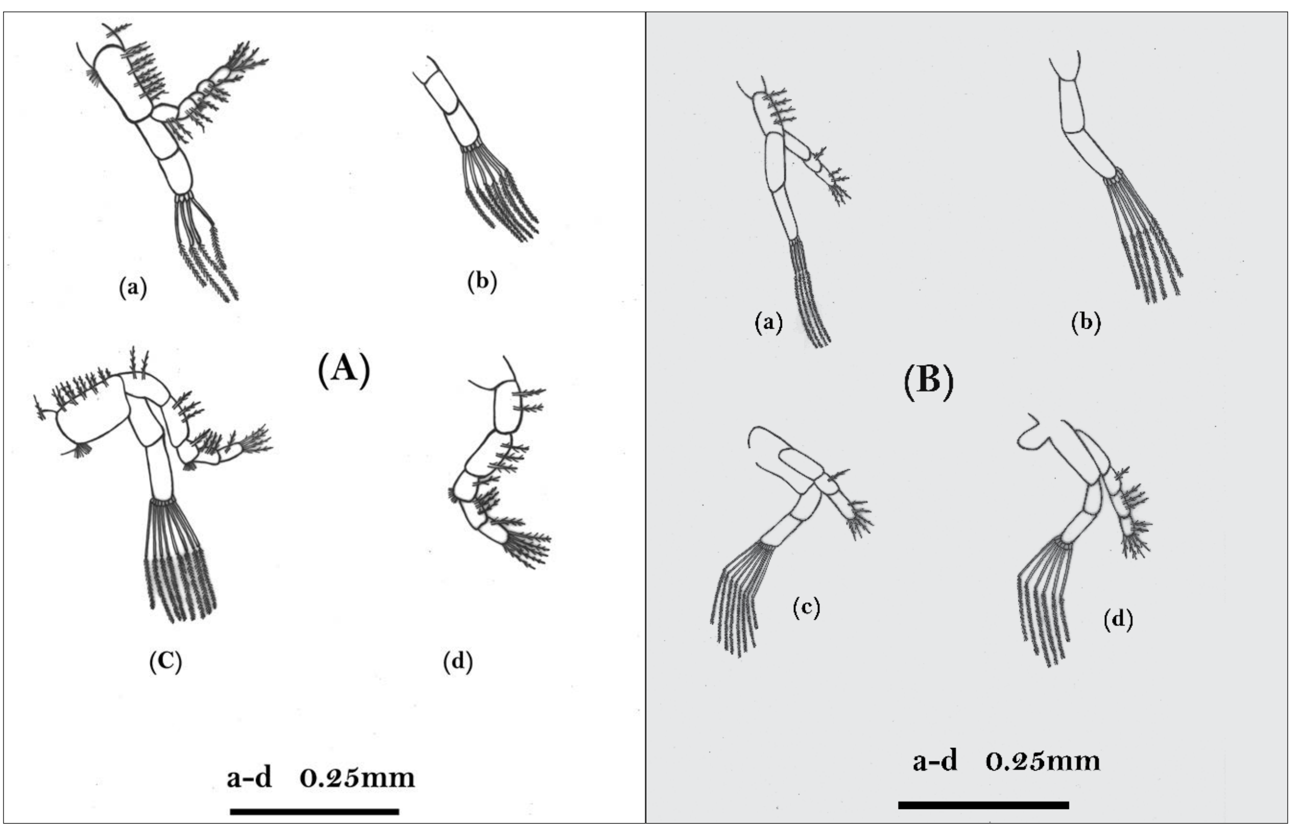

Fig. 5. *Psuedosesarma glabrum*- (A) First maxilliped (20X): a: zoea I; b: zoea II; c: zoea III; d: zoea IV; (B) Second maxilliped (20X): a: zoea I; b: zoea II; c: zoea III; d: megalopa

Pereiopods: unsegmented, first pereiopod (cheliped) is bilobed in structure.

Pleon (Fig. 6A-c): 6th somite is formed, tip of posterolateral processes of 5th somite is reaching posterior margin of 6th somite, pleopod buds are present as buds on somites 2-5, endopods absent.

Telson: outer part of the furca with two, minute scale like spines.

Zoea-IV

(Size: RDL: 1.12 mm; CL: 0.67 mm)

1st antennae/antennule (Fig. 3A-c): Exopod is partially two segmented with 2 sub terminal and 4 terminal aesthetascs.

2nd antennae (Fig. 3B-d): protopod bearing two unequal rows of 3-5 spines. endopod is elongated, reaching half of protopodal processes.

1st maxillae/maxillule (Fig. 4A-c): coxal endite with 6-7 setae and basial endite with 10 setae.

2nd maxillae/maxilla (Fig. 4B-d): scaphognathite with 11-12 marginal setae, bilobed basal endite with 6+5 setae, bilobed coxal endite with 6+5 setae.

1st maxilliped (Fig. 5A-d): Segments 5 with one additional seta, exopod distal segment with 8 long plumose natatory setae.

2nd maxilliped: exopod distal segment with 8 long plumose natatory setae.

Pereiopods: bilobed cheliped and 2-5 pereiopods are slightly segmented.

Pleon (Fig. 6A-d): pleopod buds are well developed, endopod buds formed; 1st somite with 3 long mid dorsal setae

Telson: similar to the previous stage

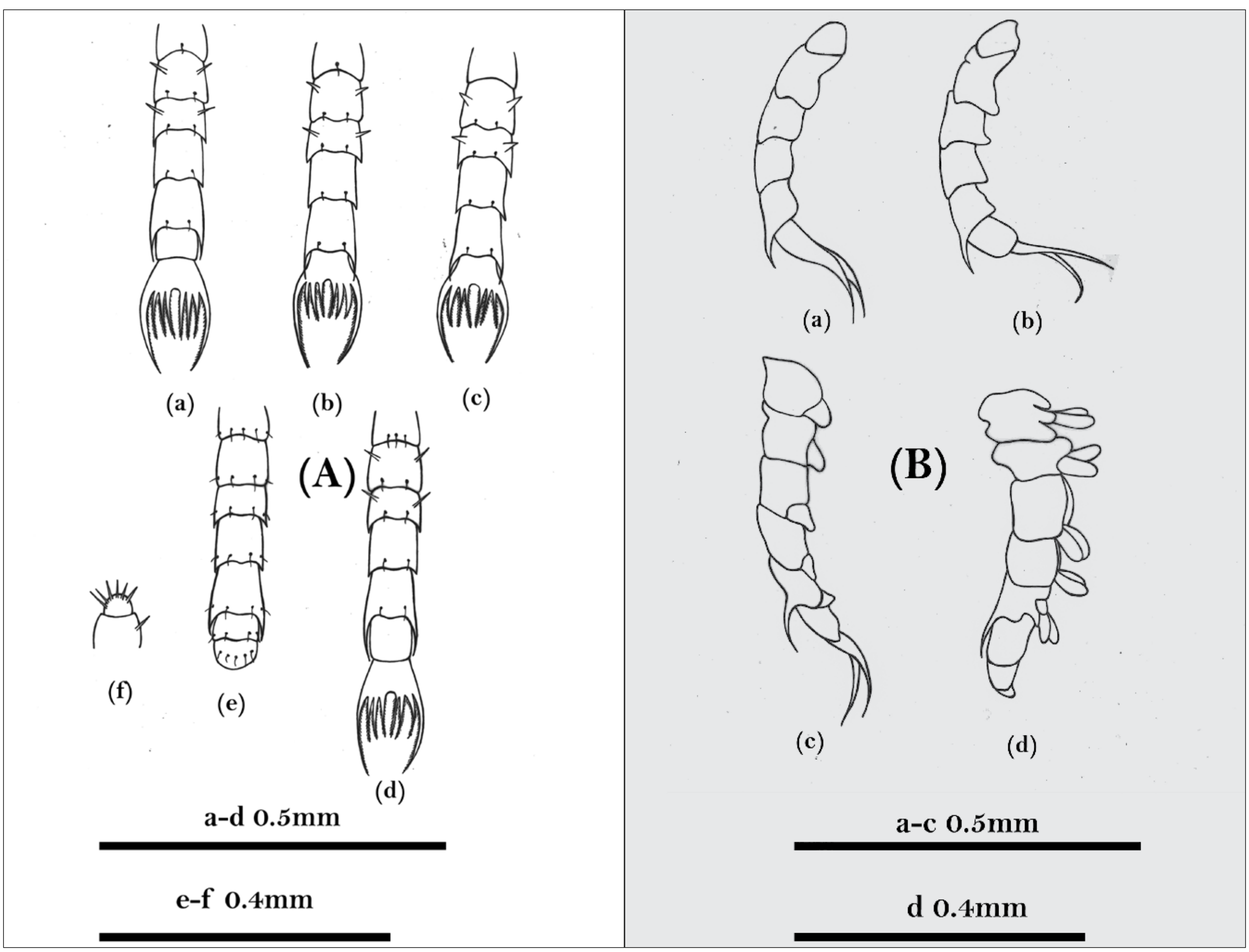

Fig. 6. *Psuedosesarma glabrum*- (A) Pleon, dorsal view (20X): a: zoea I; b: zoea II; c: zoea III; d: zoea IV; e: megalopa; f: uropod of megalopa; (B) Pleon, lateral view (20X): a: zoea I; b: zoea III; c: zoea IV; d: megalopa

Megalopa

(Size: RDL: 1.20 mm; CL: 0.72 mm)

Carapace (Fig. 2e): longer than broad, rostrum ventrally deflected with the medial cleft.

1st antennae/antennule (Fig. 3A-d): endopod is absent; exopod is partially three segmented with 2 sub terminal and 6 terminal aesthetascs.

2nd antennae (Fig. 3B-e) antennal peduncle 3 segmented with 2, 2, 2 setae respectively.

1st maxillae/maxillule (Fig. 4A-d): coxal endite with 8-9 setae and basial endite with 12-15 setae; endopod unsegmented with 4 setae; 2 setae present on epipod region.

2nd maxillae/maxilla (Fig. 4B-e): scaphognathite with 12-15 marginal setae, bilobed coxal endite with 8+4 setae, endopod unsegmented without setae.

2nd maxilliped (Fig. 5B-d): epipod rudimentary; coxa and basis not differentiated without setae; endopod 4 segmented with 0, 1, 3, 6 setae respectively; exopod 2 segmented, proximal segment with 1 seta, distal segment with 6 plumose setae.

Pleon (Fig. 6A-e): with 6 somites; somite 3-5 with posterolateral processes, somite 2-5 each with pair of biramous pleopod.

Telson: sub square shape with 5-6 dorsal and 1 ventral setae; uropod (Fig. 6A-f) two segmented with 1, 6 setae.

Discussion

About 80% of the 6900 described species of Brachyura pass through biphasic life cycles. Internal fertilization is followed by embryogenesis inside a semipermeable egg membrane. Following hatching, crab larvae join the planktonic community and grow through several morphologically similar zoeal stages, each stage showing an increase in the number and complexity of swimming and feeding appendages (Castro *et al.*, 2015). The number of larval stages may vary between species, but higher taxonomic groupings typically show characteristic patterns; for example, only two stages in Majidae, three in Hymenosomatidae, four or five in the majority of brachyuran families, four to six in most Portunidae, and up to eight in Grapsidae (Ingle, 1992; Cuesta *et al.*, 2011). The zoeal stages of family sesarmidae ranging from two to five (Lago, 1993b; Anger, 1995) before moulting to the megalopa. In this family, the number of zoeal stages throughout the entire course of development differs according to the genera. It has been known that there is intraspecific variation in the number of zoeal stages in some sesarmid species. Four stages are more frequent in sesarmid species and are observed in other species such as: *Aratus pisonii, Armases angustipes, A. cinereum, A. ricordi, Bresedium brevipes, Parasesarma catenata, P. plicatum*, *Perisesarma bidens*, and *Sesarma lanatum*. Overall, the zoeal stages of *P. glabrum* consist of four zoeal stages and one megalopa, which is very similar with *P. bocourti* and *P. moeschii* reported by Guero *et al.* (2007) and closely related to *Perisesarma fasciatum* (Guero *et al.*, 2004).

Carapace with short rostrum and a relatively slender and curved dorsal spine of zoea is the key factor for identification of sesarmid family. Other characters of this family listed by Cuesta *et al.* (2006) are: 1) carapace without lateral spine; 2) zoea I with a pair of anterodorsal setae; 3) antennal exopod of zoea I with terminal small spines and setae of different size, exopod half the length of the protopod and protopod with well-developed spines distributed in two rows, normally with unequal number of spines; maxillar endopod bilobed with 2+3 setae; 5) first maxilliped basis with 2+2+3+3 setae, endopod setation 2, 2, 1, 2, 5 in the first zoea; 6) Second maxilliped basis with 1+1+1+1 setae; endopod setation 0, 1, 6; 7) pleon of first zoeal stage with 5 somites and last zoeal stage with 6 somites, dorso lateral processes only on somite 2 and 3, in last stage somite 1 presents 3 middorsal setae; 8) telson with 3 serrulate setae on posterior margin through zoeal development, furcal arms with two dorsal rows of spinules of varying size.

Carapace of *P. glabrum* is globose, which is common in all *Psuedosesarma* species and similar with the type species *S. edwardsii* (Kannupandi and Pasupathi, 1994). From the published works, few differences in setation of mouth parts are apparent in zoea I of different species of this family. Lateral carapace spines are absent in all zoeal stages. Rostral spine is well developed and posterorly curved in *P. glabrum,* as in *P. bocourti* and *P. moeschii*, while *P. crassimanum*'s is almost straight (Cuesta *et al.*, 2006). Rostral spine straight in species of *Psuedosesarma* where it is very short and down turned in *S. edwardsii.* Protopodal processes of antennae have one row of 6-7 spines in *Psuedosesarma* and it is gradually decreased during metamorphosis. This may be a character of this genus, whereas in all other sesarmidae, it is gradually increased up to megalopa. Zoeal stages of all the endopod of the maxilla possess 2-4 setae. Here a slight variation was observed from other genus in the case of number of setae (1 seta on proximal segment, 2 subterminal setae and 4 terminal setae).

Seta on the coxa of first maxilliped was observed in all the zoeal stages of genus *Psuedosesarma*, and also reported in *P. fasciatum* (Lanchester, 1900) whereas it is absent in *Neosarmatium indicum* (Islam *et al.*, 2002) indicating that, the pattern of setation shows considerable variations in other sesarmid species. A mat of dorso-microtrichiae on third segment of endopod was also observed in all zoeal stages of known *Psuedosesarma* species. The endopod of maxilla in zoeal stages of all sesarmid species possess 2-4 setae, however in *P. glabrum* seven setae were recorded; one setae on proximal segment, two subterminal setae and four terminal setae.

The first two zoeal stages of *P. glabrum* which possess 5 abdominal somites become 6 in zoea III and IV. The presence of very long and curved posterolateral processes of abdominal somite 5, compared to more anterior ones, has so far only been described for genus *Psuedosesarma*. The presence of these distinct processes may constitute a synapomorphy at generic level (Guero *et al*., 2007). Minute scale like spines on the outer margin of the telson furcal arms have been described for zoeal stages of sesarmidae. These spines are only present from zoea III of *P. glabrum*. These were not observed in zoea I of *P. crassimanum* and zoea I to zoea II of *P. moeschii* and *P. bocourti*.

In this genus, descriptions of megalopae are available for only two species, *P. moeschii* and *P. bocourti* (Guero *et al*., 2007). The morphology of the megalopal stage is similar to the known species of the family sesarmidae: 1) carapace longer than broad, rostrum ventrally deflected with the medial cleft; 2) antennules endopod is absent; 3) antennal peduncle 3 segmented; 4) maxillary scaphognathite with less than 40 marginal plumose setae and with 2 anterior and 1 posterior lateral setae; 5) first maxilliped epipod with 4-5, but always less than 7 setae; 6) second maxilliped without fully developed epipod; 7) pleopod endopods with two cincinuli; 8) uropod with setation 1, 56 (Cuesta *et al*., 2006).

The only remarkable difference is observed in the period of metamorphosis from one zoeal stage to another. The development at duration for zoea I and zoea II took 4±2 days each, zoea III 7±2 days, zoea IV 6±2 days and megalopa 6±2 days. The first megalopae appeared on 29±2 days of hatching. It may be due to the influence of any of the physical parameters provided in the laboratory condition or could be the result of malnutrition. The reason for this can be clarified only by further studies on the influence of the physical parameters on the larval development of *P. glabrum*.

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