The early larval stages of phyllosomas of the spiny lobsters *Panulirus echinatus* Smith, 1869 and *P. laevicauda* (Latreille, 1804) (Decapoda: Palinuridae)

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Abstract

The phyllosoma larvae of the spiny lobsters *Panulirus echinatus* Smith, 1869 and *P. laevicauda* (Latreille, 1804) were reared in circular tanks of water recirculation and fed on *Artemia* sp nauplii. Each phyllosoma stage was described in detail and the main morphological structures of the two species were compared. Structurally, the phyllosoma of *P. echinatus* and *P. laevicauda* are similar in the three first initial stages, showing the same number of setae in the exopods of the maxillule, maxilla, 2nd and 3rd maxillipeds and, 1st and 2nd pereiopods. However, the carapace lengths were different, averaging 0.86, 1.18 and 1.29 mm for the I, II and III stages of *P. laevicauda*, respectively, and 1.04, 1.25 and 1.55 mm, for the same stages, respectively, for *P. echinatus*. Other difference between the two species was the presence and size of the coxal and sub-exopodal spines of the 1st and 2nd pereiopods. For *P. laevicauda*, these spines are longer, whereas *P. echinatus* lacks setae. These findings could support further investigations on phyllosoma larvae collected in plankton samples.

Key words: spiny lobster, phyllosoma, morphology, larviculture.

Introduction

The larval development of spiny lobsters of the genus *Panulirus* White 1847, in the open ocean, comprises basically two larval phases: phyllosoma and puerulus. The literature has related a relatively wide range in the duration of the phyllosoma period, ranging from 4-6 months for *P. homarus* (Linnaeus, 1758) and 10-13 months for *P. japonicus* (Von Siebold, 1824) studied by Booth and Phillips (1994). This extended developmental period seems to be the major obstacle in the study of the larval development of lobsters in the laboratory.

Among the species of the genus *Panulirus*, only two sub-tropical species were successfully reared (complete larval development) in the laboratory: *P. japonicus* by Inoue (1978); Kittaka and Kimura (1989) and Yamakawa *et al.* (1989) and *P. longipes* (A. Milne-Edwards, 1868) by Matsuda and Yamakawa (2000). Some attempts were made to cultivate *P. homarus* by Radhakrisnan and Vijayakumaran (1995) and *P. echinatus* by Santiago (2001), but they did not surpass the 10th stage.

In Brazil, the species *P. argus* and *P. laevicauda* are a very important economic resources. Other species, *P. echinatus*, supported for long time an inexpressive capture, but actually, an increase in its production has been observed (Santiago, 2001). Despite the economic value of these species, little is known about the biology of their larvae. Studies clearly demonstrated that more information on the biology, ecology and larval recruitment is necessary (Radhakrisnan and Vijayakumaran, 1995; Santiago, 2001). However, there are many difficulties in the study of phyllosomas collected in the wild due to be not easy the distinction of morphological features of phyllosomas because they are very similar in their initial stages.

Important contributions on the morphological description of the Brazilian species of *Panulirus* were described by Lewis (1951) of the complete development of *P. argus* collected in plankton samples and of the late stages of *P. laevicauda* by Baisre and Ruiz de Quevedo (1982) and *P. echinatus* by Coelho *et al.* (1995). But, phyllosomas in the advanced larval stages have present morphological characteristics not observed in the early larval stages (Matsuda and Yamakawa, 2000).

In the present work, early stages of phyllosomas (I to III stages) of the species *P. echinatus* and *P. laevicauda*, obtained in the laboratory, were described and compared. The results contribute to the identification of spiny lobster larvae collected in zooplankton samples and elucidate some aspects of their biology, mainly, those regarding to their ecology and larval dispersion.

Material and methods

Egg-bearing females of *P. echinatus* were obtained at Iparana Beach, Caucaia, Ceará in October 2001. The individuals were collected by diving in coastal shallow water near the beach. Plastic baskets (10 L) containing local seawater were used to transport the females to the laboratory.

A single ovigerous female of *P. laevicauda* captured along the northeastern coast of Fortaleza, Ceará in May 2000 was used in the experiment. The capture procedure and transport of the lobster to the laboratory was similar to those referred to above for *P. echinatus*.

Phyllosomas were reared in tanks of water recirculation (10 larvae/liter), made of acrylic (70 cm Ø and 30 cm deep), similar to those used for the culture of many species of spiny lobsters of the family Palinuridae (Kittaka, 1994). In these tanks, free circulation of flow water and total visualization of the floating elements were available. Salinity and temperature were maintained between 33 to 38‰ and 27 to 30°C, respectively.

The larvae in the first stage were fed on *Artemia* sp. Microalgae *Dunaliella viridis* (Teodoresco) were added daily in the culture system. Despite phyllosomas not being phytoplankton feeders, the microalgae were responsible for nitrogenated compound absorption and maintenance of the water quality. Temperature, pH, dissolved ammonium and salinity were rigorously monitored using specific equipments and proper determination kits.

Samples of larvae were taken after each stage molting and fixed in 10% formaline solution and, after 24 hours, preserved in glycerol + alcohol 70% (1:1) solution. Measurements of cephalic shield were made using 10 larvae of each stage. The larvae were dissected under an optical binocular microscope. The illustrations, including appendages, were made using a camera lucida. Terminology used in the descriptions of the larval stages follows: Nishida *et al.* (1990), Lemmens and Knott (1994) and Wolfe and Felgenhauer (1991).

Results

P. laevicauda (Fig. 1, 2)

Stage I
(Fig. 1)

· **Cephalic shield**: Mean length 0.86 mm (range: 0.85-0.92), semicircular, pear- shaped.

· **Eyes**: Ocular peduncle unsegmented.

· **Antennule** (Fig. 1b): Uniramous, unsegmented, longer than antenna, ending in a short spine, 3 long aesthetascs and 2 simple setae.

· **Antenna** (Fig. 1c): Uniramous, unsegmented, ending in a short curved spine and 2-3 simple setae; 2 simple setae in the distal region near to the apex.

· **Maxillule** (Fig. 1d): Coxal endite with 2 long serrulate setae and 1 short simple seta; basal endite with 2 strong serrulate setae distally and 1-2 subdistally; endopod short, unsegmented with 2 simple setae.

· **Maxilla** (Fig. 1e): Basal segment enlarged with 2-3 short setae, distal segment short with 4 long plumose setae.

· **First maxilliped**: A short, uniramous and unsegmented protuberance.

· **Second maxilliped** (Fig. 1f): Endopod 5-segmented, distal segment ending in 1 strong distal and 2 subdistal spines and 1 seta, subdistal segment rounded distally with 5 long serrulate setae, second

segment with 2 short medial setae; exopod absent.

· **Third maxilliped** (Fig. 1a): Endopod well developed, 5-segmented, distal segment with 4 setae distally, subdistal segment with 4 long setae rounding the distal margin and 2 long setae on the subdistal margin; exopod with 3 pairs of long and plumose setae.

· **First and second pereiopods** (Fig. 1g,h): Endopod 5-segmented, completely developed, distal segment ending in a strong spine, dorsal and ventral subexopodal spines elongated, coxal spine long, (disposition of setae as illustrated); exopod with 5 pairs of long and plumose setae.

· **Third pereiopod** (Fig. 1a): Endopod very long, 5-segmented, ending in a strong curved spine, coxal spine present; exopod undeveloped showing as a small unarmed protuberance.

· **Abdomen**: Short, unsegmented with 3 setae and 1 spine at each ramous on the posterior portion.

Figure 1: *P. laevicauda*: a) stage I in ventral view; b) antennule; c) antenna; d) maxillule; e) maxilla; f) 2nd maxilliped; g) 1st pereiopod; h) 2nd pereiopod.
Abbreviations: an1 = antennule; an2 = antenna; mp1 =1st maxilliped; mp2 = 2nd maxilliped; mp3 = 3rd maxilliped; mx2 = maxilla; p1 = 1st pereiopod; p2 = 2nd pereiopod; p3 = 3rd pereiopod; cs = coxal spine; sbs = sub-exopodal spine. Scale bar: 0.5mm.

Stage II

(Fig. 2a)

· **Cephalic shield**: Mean length 1.18 mm (range: 1.15-1.22 mm).

· **Eyes**: Ocular peduncle segmented.

· **First and second pereiopods**: Exopod with 6 pairs of long and plumose setae.

· **Third pereiopod**: Exopod a little more developed compared to the first stage, lacking setae.

Stage III

(Fig. 2b)

· **Cephalic shield**: Mean length 1.29 mm (range: 1.27-1.33 mm).

· **Third maxilliped**: Exopod with 4 pairs of long and plumose setae.

· **First and second pereiopods**: Exopod with 7 pairs of long and plumose setae.

· **Third pereiopod**: Exopod well developed compared to previous stages with 3 pairs of long and plumose setae.

· **Fourth pereiopod**: Present as unsegmented biramous bud.

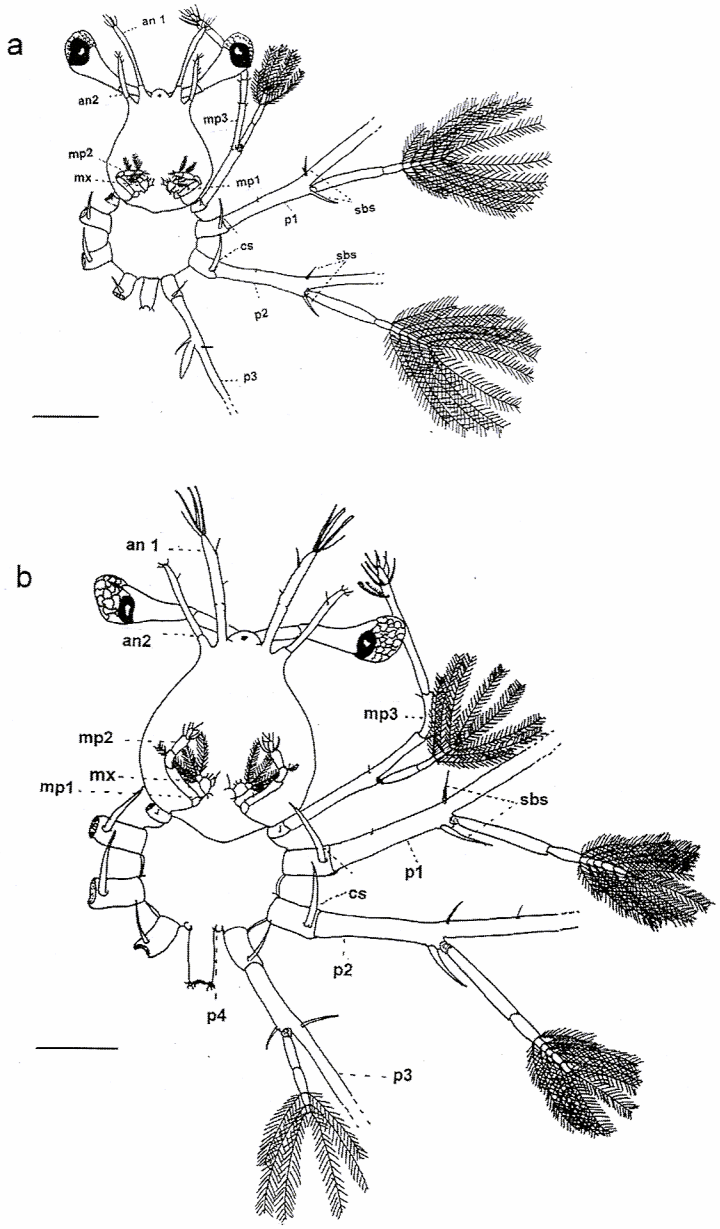

Figure 2: *Panulirus laevicauda* (in ventral view); a) stage II; b) stage III.
Abbreviations: an1 = antennule; an2 = antenna; mp1 =1st maxilliped; mp2 = 2nd maxilliped; mp3 = 3rd maxilliped; mx = maxilla; p1 = 1st pereiopod; p2 = 2nd pereiopod; p3 = 3rd pereiopod; p4 = 4th pereiopod; cs = coxal spine; sbs = sub-exopodal spine. Scale bar: 0.5mm.

P. echinatus (Figs. 3, 4)

Stage I

(Fig. 3)

· **Cephalic shield**: Mean length 1.04 mm (range: 1.00-1.07 mm), semicircular, pear like in shape.

· **Eyes**: Ocular peduncle unsegmented.

· **Antennule** (Fig. 3b): Uniramous, unsegmented, longer than antenna, ending in a short spine, 3 long aesthetascs and 2 simple setae.

· **Antenna** (Fig. 3c): Uniramous, 2-incompletely segmented, ending in a short curved spine and 2-3 simple setae, 2 simple setae in the distal region near to apex.

· **Maxillule** (Fig. 3d): Coxal endite with 2 long serrulate setae and 1 short simple seta; basal endite with 2 strong serrulate setae distally and 1-2 subdistally; endopod short, unsegmented with 2 simple setae.

· **Maxilla** (Fig. 3e): Basal segment enlarged with 2-3 short setae, distal segment short with 4 long plumose setae.

· **First maxilliped**: Small, uniramous and unsegmented protuberance.

· **Second maxilliped** (Fig. 3f): Endopod 5-segmented, distal segment ending in a strong distal and 2 subdistal spines and 1 seta, subdistal segment rounded distally with 5 long serrulate setae, second segment with 2-3 short setae; exopod absent.

· **Third maxilliped** (Fig. 3a): Endopod well developed, 5-segmented, segment distal with 3 simple setae on the distal margin, subdistal segment with 4-5 setae rounding the distal margin and 2 setae on the subdistal margin; exopod with 3 pairs of long and plumose setae.

· **First and second pereiopods** (Fig. 3g, h): Endopod 5-segmented, completely developed, distal segment ending in a strong spine, dorsal and ventral subexopodal spines absent, coxal spine present (disposition of setae as illustrated); exopod with 5 pairs of long and plumose setae.

· **Third pereiopod:** Endopod very long, 5-segmented, ending in a strong curved spine, coxal spine present; exopod undeveloped showing a small unarmed protuberance.

· **Abdomen:** Short, unsegmented with 3 setae and 1 spine at each ramous on the posterior portion.

Stage II

(Fig. 4a)

· **Cephalic shield**: Mean length 1.25 mm (range: 1.18-1.30 mm).

· **Eyes**: Ocular peduncle segmented.

· **First and second pereiopods**: Exopod with 6 pairs of long and plumose setae.

· **Third pereiopod**: Exopod a little more developed compared to the first stage, lacking setae.

Stage III

(Fig. 4b)

· **Cephalic shield**: Mean length 1.55 mm (range: 1.38-1.74 mm).

· **Third maxilliped**: Exopod with 4 pairs of long and plumose setae.

· **First and second pereiopods**: Exopod with 7 pairs of long and plumose setae.

· **Third pereiopod**: Exopod well developed compared to previous stages with 3 pairs of long and plumose setae.

· **Fourth pereiopod**: As an unsegmented biramous bud.

Discussion

Phyllosoma larvae of the *P. laevicauda* and *P. echinatus* were reared succeeding to the 3rd stage in recirculation tanks. Samples of collected individuals during the experiment were sufficient to describe the larval development from the I to III stages of each observed species.

Similarities were observed among the morphological structures in the number of setae for Palinuridae species described in the present work (Table I) and those reported in the literature for *P. penicillatus*,

(Olivier, 1791) *P. longipes* and *Panulirus* sp. by Coutures (2000), *P. inflatus* by Johnson and Knight (1966), *P. japonicus* by Inoue (1978, 1981), *P. homarus* by Radhakrishnan and Vijayakumaran (1995) and *P. longipes* by Matsuda and Yamakawa (2000). Although the number and disposition of exopodal setae in the second maxilla, third maxilliped and pereiopods are a distinctive characteristc of *Panulirus* genus such features, unfortunately, are not appropriated for proceeding morphological distinctions among species.

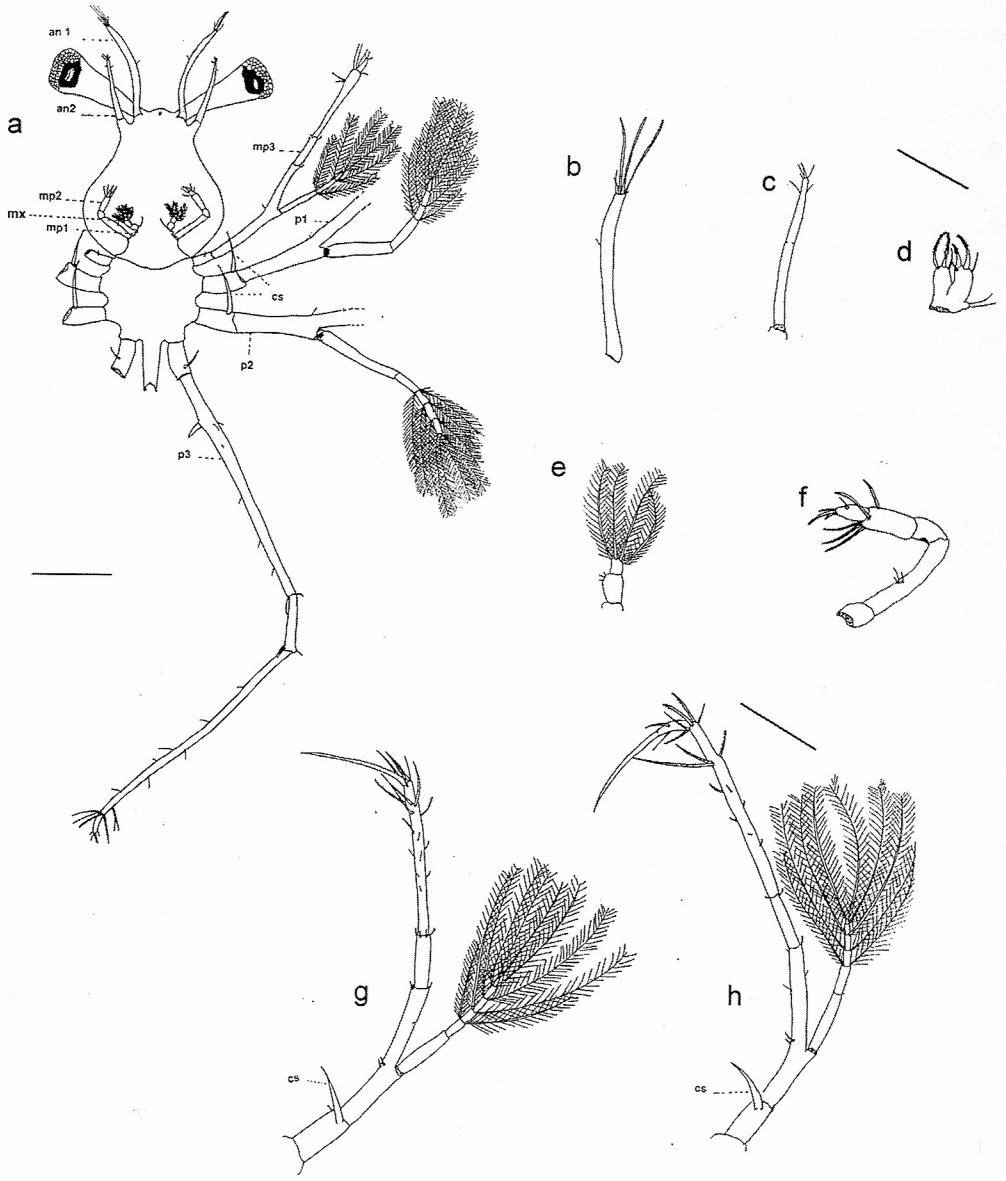

Figure 3: *P. echinatus*: a) stage I in ventral view; b) antennule; c) antenna; d) maxillule; e) maxilla; f) 2nd maxilliped; g) 1st pereiopod; h) 2nd pereiopod.
Abbreviations: an1 = antennule; an2 = antenna; mp1 =1st maxilliped; mp2 = 2nd maxilliped; mp3 = 3rd maxilliped; mx2 = maxilla; p1 = 1st pereiopod; p2 = 2nd pereiopod; p3 = 3rd pereiopod; cs = coxal spine. Scale bar: 0.5mm.

Other characteristics such as the different length of the carapace could be consistent information for larval distinction as observed in the Table I. However, variations within species may occur in the phyllosoma according to changes in any environmental factors or feeding conditions. These facts do not suggest the use of this kind of data adequated for phyllosoma identification. Matsuda and Yamakawa (2000) reported the similar impossibility in demonstrating morphological differences between cultured larvae of the closed related species *P. longipes* and *P. japonicus* based on the characteristics above described (cephalic shield lenght and setal number).

Controversely, the phyllosomas of Palinuridae may be separated into two distinct groups according to the presence (group A) or the absence (group B) of exopodal spines (Table II) (Gurney, 1936; Baisre and Ruiz de Quevedo, 1982; McWilliam, 1995; Coutures, 2000). According to the obtained results *P. laevicauda* phyllosoma presented the coxal and sub-exopodal spines elongated and well developed, whereas *P. echinatus* larvae showed only the coxal spines placing these species in different groups.

Alterations in the presence of coxal and sub-exopodal spines of the maxillipeds and pereiopods may occur during the development of phyllosomas. In the case of *P. argus* and *P. echinatus*, coxal spines in the pereiopods of late larval stages were absent (Lewis, 1951; Coelho *et al.*, 1995). On the other hand, the coxal and subexopodal spines still present in the late stage of *P. laevicauda* as assumed by Baisre and Ruiz de Quevedo (1982). These morphological features are useful to differentiate morphologically *P. argus* and *P. echinatus* from *P. laevicauda* species even in diverse phyllosomal ages. However, the early stage of *P. echinatus* is very similar to the phyllosoma of *P. argus* described by Lewis (1951), which suggest the use of biochemical and molecular methods to supports the morphological differentiation of *P. argus* and *P. echinatus* species.

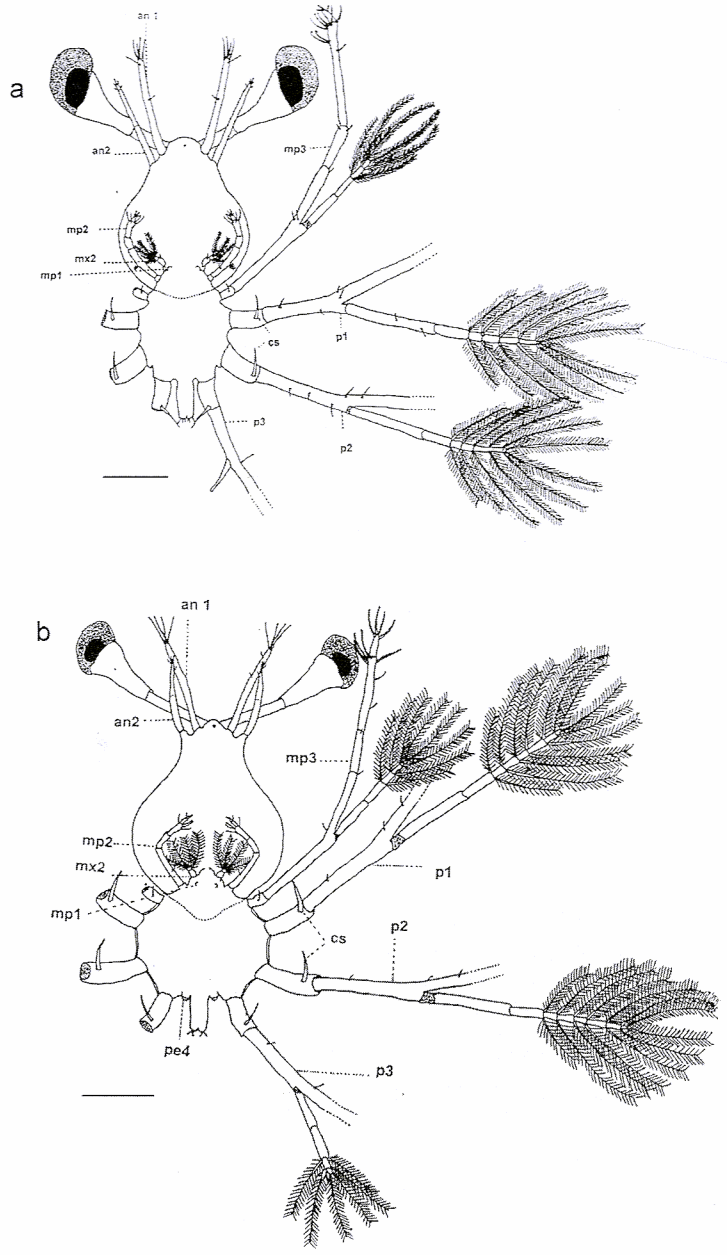

Figure 4: *P. echinatus* (in ventral view): a) stage II; b) stage III.
Abbreviations: an1 = antennule; an2 = antenna; mp1 =1st maxilliped; mp2 = 2nd maxilliped; mp3 = 3rd maxilliped; mx = maxilla; p1 = 1st pereiopod; p2 = 2nd pereiopod; p3 = 3rd pereiopod; p4 = 4th pereiopod; cs = coxal spine. Scale bar: 0.5mm.

Table I: Morphological comparisons among the first stage of *P. laevicauda* and *P. echinatus* observed in the present study and other *Panulirus* species.

Species	Author	Average cephalic shield (mm)	Maxilla (distal seg.)	Number of pair of setae in the exopod					
				3rd Mp	1st Pe	2nd Pe	3rd Pe	4th Pe	5th Pe
P. echinatus	Present work	1.04	4	3	5	5	0	-	-
P. laevicauda	Present work	0.86	4	3	5	5	0	-	-
P. penicillatus	Coutures, 2000	1.07	4	3	5	5	0	-	-
P. longipes femoristriga	Coutures, 2000	0.91	4	3	5	5	0	-	-
Panulirus sp.	Coutures, 2000	0.90	4	3	5	5	0	-	-
P. japonicus	Inoue, 1978, 1981	0.89	4	3	5	5	0	-	-
P. longipes	Matsuda and Yamakawa, 2000	1.02	4	3	5	5	0	-	-
P. inflatus	Johnson and Knight, 1966	0.67	4	3	5	5	0	-	-
P. argus	Lewis, 1951	*	4	*	*	*	*	-	-

* not described by the author
Mp = Maxilliped, Pe.= Pereiopod, Distal seg. = Distal segment

Table II: Morphological comparisons among the spines present in the pereiopods of the first stage for *P. laevicauda* and *P. echinatus* and other *Panulirus* species.

Species	Author	Coxal spines	Ventral Sub-exopodal spines	Dorsal Sub-exopodal spines
P. laevicauda	Present description	present	present	present
P. echinatus	Present description	present	absent	absent
P. penicillatus	Coutures, 2000	present	absent	absent
P. longipes femoristriga	Coutures, 2000	present	present	present
Panulirus sp.	Coutures, 2000	present	absent	absent
P. homarus	Radhakrishnan and Vijayakumaran, 1995	present*	present*	absent*
P. argus	Lewis, 1951	present	absent	absent
P. japonicus	Inoue, 1978 and 1981	present	absent	absent
P. longipes	Matsuda and Yamakawa, 2000	present	absent	absent

*Not described but showed in the illustration by the author

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