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Metabolism and chemical composition of phyllosoma larvae, with special reference to the tropical rock lobster *Panulirus ornatus* (Decapoda; Palinuridae)

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Running head: Metabolism and chemical composition of tropical phyllosoma

ABSTRACT

Rates of oxygen consumption and ammonia excretion and chemical composition (water, ash and CN composition) were determined throughout larval development, from Stages I-IX, of *Panulirus ornatus* phyllosoma larvae raised in the laboratory. Dry mass (DM) specific oxygen consumption rates varied from 1.4 to 3.3 μl O_2 $(DM)^{-1}$ h^{-1}, showing a pattern of developmental decline. In contrast, specific ammonia excretion rates fluctuated irregularly from 0.02 to 0.067μg NH_4-N $(DM)^{-1}$ h^{-1}, yielding oxygen consumption to ammonia excretion ratios (O:N atomic ratio) as wide as 51-180. With progressive development, ash decreased from 34% to 17% of DM while C composition increased from 33% to 41% of DM. No consistent changing pattern with development was seen for water contents (74-83% of WM), N composition (6.9-9.5% of DM) and C:N ratios (4.0-5.1). Supplemental data of both specific oxygen consumption and ammonia excretion rates of laboratory-raised Stage III *P. homarus* and wild *Parribacus antarcticus* phyllosoma (stage unknown) of this study and literature data on phyllosomas of other lobster species by previous workers were shown to be comparable to those of *P. ornatus* phyllosomas when the differences in DM and temperature were taken into account. No appreciable differences due to the origin of the specimens, whether wild or laboratory-raised, were evident. Comparison of the present results with those of stomatopod larvae and holoplanktonic crustaceans in the same tropical marine habitats revealed that phyllosoma larvae are characterized by somewhat lower DM specific oxygen consumption rates and N composition. The study revealed markedly reduced ammonia excretion rates in phyllosomas, suggesting reduced swimming activity and preferential utilization of dietary protein to somatic growth rather than metabolism.

Keywords: phyllosoma, *Panulirus ornatus*, oxygen consumption, ammonia excretion, O:N ratio, CN composition

1. Introduction

The zoeal form of decapod larvae from the families of Palinuridae (rock or spiny lobsters) and the Scyllaridae (slipper lobsters) is characterized by a glassy transparency, flattened leaf-like shape with 3-5 pairs of extending biramous appendages and classified as “phyllosoma” (leaf-like) larvae. Phyllosomas are planktonic raptorial feeders and undergo sequential molts to metamorphose into pre-juvenile puerulus phase which finally settle on the bottom of coastal waters as juveniles to develop into adults (cf. Phillips and Sustry, 1980; Anger, 2001). Despite a wide tropical and semi-tropical global geographical distribution and a frequent member of zooplankton assemblages in temperate-tropical oceans, phyllosoma metabolism and nutrition have been little studied to date, largely because of difficulties in collecting undamaged specimens from plankton net tows due to the fragile bodies of the phyllosoma larval form.

Information about metabolism (oxygen consumption and ammonia excretion rates) and body chemical composition (water content, ash and carbon and nitrogen composition) have proved to be useful to provide a wide perspective for understanding zooplankton energy demand, metabolic balance and nutritional conditions within their environments (Ikeda et al., 2000; Anger, 2001). For decapod crustacean larvae other than phyllosomas, intensive studies have been reported on many species of wild and laboratory-raised specimens as well as ontogenetic changes in metabolism and body composition (cf. review of Anger, 2001). Knowledge on the biology, physiology and behavior of phyllosoma larvae of lobsters has advanced recently as part of a growing interest to establish appropriate aquaculture methods of this commercially valuable marine resource (Jeffs and Hooker, 2000). Nevertheless, presently available data on the metabolism and chemical composition of phyllosomas are limited to the California

67 spiny lobster *Panulirus interruptus* (Belman and Childress, 1973) and the southern rock
68 lobster *Jasus edwardsii* (Bermudes and Ritar, 2004, 2005; Bermudes et al., 2008; Ritar
69 et al., 2003). However, both *P. interruptus* and *J. edwardsii* phyllosomas are from
70 temperate waters and no data is currently available for species from tropical zones.
71 The tropical rock lobster *Panulirus ornatus* is distributed through the Indo-West
72 Pacific (Holthuis, 1991) and is a target species of fisheries of Australia, South East Asia
73 and Indian Ocean countries. In the Coral Sea, the spawning season of *P. ornatus* is
74 estimated to span from December-April, with a planktonic duration of phyllosomas
75 lasting for 4-6 months (Dennis et al., 2009). *P. ornatus* phyllosoma have been reared
76 through their entire larval phase in the laboratory with a full morphological description
77 of a total of 11 larval stages (Smith et al., 2009b). The aim of this study is to extend the
78 knowledge of metabolism and chemical composition of the phyllosomas of a tropical
79 Palinurid lobster species by comparing sequential larval stages of *P. ornatus* raised in
80 the laboratory, supplemented by those of other tropical lobster species raised in the
81 laboratory and collected in the wild in the Coral Sea. The results are compared with
82 those of wild and morphologically similar stomatopod larvae as well as epipelagic
83 marine zooplankton in an attempt to identify any unique features of phyllosoma larvae
84 as members of the tropical zooplankton community
85
86 **2. Materials and methods**
87 *2.1. Phyllosoma*
88 *P. ornatus* and *P. homarus* phyllosomas were raised from eggs in the tropical
89 aquaculture facilities of Australian Institute of Marine Science (AIMS) in Townsville,
90 Queensland, Australia. Some *P. ornatus* and *Parribacus antatcticus* larvae of

91 unknown stage used in this study were collected from the Coral Sea.

92 General procedures for laboratory culturing of *P. ornatus* and *P. homarus* are
93 described elsewhere (Smith et al. 2009b). Briefly, for *P. ornatus*, a batch of
94 phyllosoma larvae were obtained from a gravid female collected from the field and
95 raised at a constant temperature of 28^{o}C. Newly hatched *Artemia* nauplii were
96 provided as the sole diet for early larvae with juvenile *Artemia* supplemented by fresh
97 blue mussel (*Mytilus edulus*) gonad given for post-Stage IV phyllosomas. Under these
98 conditions, the intermolt period for each phyllosoma molt took about one week. The
99 body size (total length) of the larvae at a given stage in captivity was similar to those of
100 wild larvae at the same developmental stage (Smith et al., 2009b), suggesting adequacy
101 of food provided for larval growth. Almost synchronous molting in early larvae
102 became gradually asynchronous with development. During this study, development of
103 the larvae in the batch cultures was monitored and post-molt larvae were sorted and
104 combined as a new batch with similar molt stage. In this way, the larvae at one day
105 post-molt of each stage were used consistently throughout the study.

106 Wild phyllosoma larvae of *P. ornatus* and *P. antarcticus* were collected during the
107 cruise of R.V. Cape Ferguson to Osprey Reef, Coral Sea during 5-22 May 2010.
108 Samplings were made at night with a modified Issac-Kidd midwater trawl net (mouth
109 opening: 2.2 m × 2.4 m) with a solid cod-end to minimize physical damage of the larvae
110 collected. The net was towed at approximately 2 knots through the surface water. In
111 addition to phyllosomas, wild stomatopod alima larvae which have a transparent body
112 similar to that of phyllosomas yet exhibiting much faster swimming behavior were
113 collected and used in the present experiments for comparative purpose.

114 Seawater supplied from the Seawater Precinct of AMIS (land laboratory

115 experiments) or collected from 2 m depth at each sampling site with 10 liter Niskin
116 bottles (shipboard experiments) was filtered through GF/F filters, well oxygenated and
117 used for the following experiment.
118 *2.2. Metabolic measurements*
119 Experiments were started 0900 hours for laboratory raised specimens or within
120 1 to 2 hours after the collection for wild specimens. Oxygen consumption and
121 ammonia excretion rates were measured simultaneously by a sealed-chamber method
122 (cf. Ikeda et al., 2000). The specimens were briefly rinsed 3-4 times with filtered
123 seawater and a single individual, or a batch of 50-100 specimens, were transferred to
124 glass bottles (100, 300 or 500 ml capacity depending on the size of specimens) filled
125 with filtered seawater. Experiments were designed to end before the oxygen saturation
126 in experimental bottles declined >70% since oxygen consumption rates is reported to be
127 dependent on oxygen saturation of ambient water in *P. interruptus* phyllosoma larvae
128 (Belman and Childress, 1973). Control bottles without specimens were prepared
129 concurrently. Experiments were run in the dark for 3-12 h in a constant temperature
130 room of which temperature was adjusted to 28^{o}C on land, or *in situ* temperature (28^{o}C)
131 by immersing glass bottles in an incubation tank on the deck through which surface
132 seawater overflowing. During the experiments on land, both experimental and control
133 bottles were placed on a grazing wheel which rotates at 0.5 rpm to prevent settling the
134 specimens on the bottom of bottles. The gazing wheel was not used for the
135 experiments at sea. At the ends of experiments, duplicate 15 and 10 ml water samples
136 were siphoned out for the measurements of dissolved oxygen and ammonia, respectively.
137 Dissolved oxygen and ammonia were determined by the Winkler titration method and
138 the phenol-hypochlorite method, respectively (Strickland and Parsons, 1972).

Specimens left in experimental bottles were rinsed briefly with a small amount of distilled water, blotted on a filter paper to remove water adhering to the body and weighed (wet mass, WM). At sea, the specimens were stored at -20˚C, and the frozen specimens were weighed (WM) in the land laboratory after the cruise.

2.3. Chemical composition

In the land laboratory, fresh or frozen specimens were placed in an electric oven and dried at 60^{o}C overnight to estimate dry mass (DM) and water content. The dried specimens were pooled by stage or species then finely ground with a ceramic mortar and pestle. Powdered samples were used for analysis of CN composition with an elemental analyzer (TruSpec CN Determinator, LECO Corp. USA) using ethylenediaminetetraacetic acid (EDTA) as a standard. For ash determination, weighed fractions of powdered samples were incinerated at 450˚C overnight and reweighed. Measurements were replicated 2-4 times for CN composition and 2 times for ash. From replicate determinations of the same sample, the precision (coefficient of variation of these measurements were 3% for C and N and 6% for ash. Water content was expressed as percent of WM, whereas the contents of ash, carbon and nitrogen were expressed as percent of DM.

3. Results

3.1. Oxygen consumption, ammonia excretion and O:N ratio

Of the total of 11 larval stages of *P. ornatus* phyllosoma, sufficient number of individuals was available for Stages I, II, IV, V, VI, VIII and IX from four broods (A-D)(Table 1). The data obtained from these seven stages were pooled for the analysis of developmental pattern of *P. ornatus* phyllosoma. Body mass increased

163 exponentially from 0.041 to 6.27 mg DM through the Stages I-IX (Table 1, Fig. 1A).
164 In parallel with the increase in body mass, oxygen consumption and ammonia excretion
165 rates increased rapidly from 0.10 to 8.26 μl O_2 (individual)$^{-1}$ h^{-1} and from 0.00076 to
166 0.216μg NH_4–N (individual)$^{-1}$ h^{-1}, respectively. The rapid increase in both rates per
167 individual modulated in terms of the rates per mg body DM (=specific rates). Specific
168 oxygen consumption rates varied from 1.4 to 3.3 μl O_2 (DM)$^{-1}$ h^{-1}across stages and
169 between-stage differences were significant (*F*-test, $p < 0.001$), showing an overall
170 pattern of decline toward late stages (Fig.1B). Between-stage differences were highly
171 significant for specific ammonia excretion rates which varied from 0.02 to 0.07μg
172 NH_4-N (DM)$^{-1}$ h^{-1} (*F*-test, $p < 0.001$), but no consistent trend with development was
173 detected (Fig. 1C). Between-stage differences in O:N ratios were also significant
174 (*F*-test, $p < 0.001$).
175 Oxygen consumption and ammonia excretion rates of Stage III *P. homarus* larvae
176 weighing 0.186 mgDM were 0.44 μl O_2 (individual)$^{-1}$ h^{-1} and 0.017 μg NH_4–N
177 (individual)$^{-1}$ h^{-1} , respectively, resulting a O:N ratio of 32 (Table 1). We could not
178 identify the developmental stage of *P. antatcticus* larvae, of which body mass varied
179 from 5.4 to 52.3 mgDM (mean: 20.6). The ranges of oxygen consumption, ammonia
180 excretion and resultant O:N ratios of *P. antatcticus* larvae were 6.7-29.3 μl O_2
181 (individual)$^{-1}$ h^{-1} (mean: 13.2), 0.08-1.02 μg NH_4–N (individual)$^{-1}$ h^{-1} (0.46) and 16-125
182 (56), respectively (Table 1). Stomatopod alima larva with similar body mass to that of
183 *P. antarcticus* consumed oxygen (16.6-42.0 μl O_2 (individual)$^{-1}$ h^{-1}, mean: 26.9) and
184 excreted ammonia (0.83-3.12 μg NH_4–N (individual)$^{-1}$ h^{-1}, mean: 2.19) much faster than
185 *P. antarcticus*, yielding lower O:N ratios (10-29, mean: 17).
186 In order to examine between-species differences in specific rates of oxygen

consumption and ammonia excretion, the data of Stages I-IX *P. ornatus*, Stage III *P. homarus*, and the mixed stages of *P. antarcticus* phyllosomas and stomatopod alima larvae were plotted against DM (Figs. 2, 3). A regression line (referred to as the"ornatus" line hereafter) and its 95% confidence interval (CI) belt were calculated for Stages I-IX (for specific oxygen consumption rates) or Stages IV-IX *P. ornatus* phyllosomas (for specific ammonia excretion rates and O:N ratios). Since the regression of specific ammonia excretion rates and O:N ratios on DM was not significant ($p > 0.05$), the "ornatus" line was represented by the means and its 95%CI. Previous oxygen consumption data of Stage I *P. interruptus* phyllosoma determined at 17.3^{o}C (Belman and Childress, 1973), and oxygen consumption/ammonia excretion data of unidentified species at 28^{o}C (Ikeda, 1974) and Stages I-V *J. edwardsii* phyllosomas at 18^{o}C (Bermudes et al., 2008), as showed in Table 1, were also plotted in Figs. 2 and 3. The oxygen consumption/ammonia excretion data on Stages I-III *J. edwardsii* phyllosomas of Bermudes and Ritar (2004, 2005) are not shown as these data are well represented by those of Stages I-V *J. edwardsii* of Bermudes et al. (2008). In order to make direct comparison with the present data possible, the previous data of Belman and Childress (1973) and Bermudes et al. (2009) were converted to the data at 28^{o}C by using a $Q_{10} = 2$, which was derived from statistical analysis of comprehensive oxygen consumption and ammonia excretion data of epipelagic marine zooplankton from world oceans (Ikeda, 1985).

3.2. Water, ash and elemental composition

Across Stages I-IX *P. ornatus* larvae, between-stage differences were significant for water content, ash, C and N composition (*F*-test, $p < 0.03$) but not for C:N ratio (*F*-test, $p > 0.25$). With regard to changing patterns with development, water

content changed irregularly (75-83%, Fig. 4A), but ash decreased consistently from 34 to 17-18% (Table 2, Fig. 4B). C and N composition ranged from 33 to 42%, and from 6.9 to 9.5%, respectively (Table 2), and significant developmental increase was detected for C but not N (Figs. 4C, D). C and N composition and C:N ratios of wild Stages VIII/XI larvae from the field fell well within the ranges of those raised in the laboratory but ash (15%) of the former was slightly less than the latter (17-34%, Table 2).

Supplemental data of water contents, C and N composition and C:N ratios of Stage III *P. homarus* and *P. antatcticus* phyllosomas overlap those of *P. ornatus* phyllosomas (Table 2). Ash content (18%) of Stage III *P. homarus* phyllosoma fell within the range of those (17-34%) of Stages I-IX *P. ornatus* phyllosomas but *P. antatcticus* phyllosomas (15%) was not.

4. Discussion

4.1. Metabolic comparison

Larval development of decapods is accompanied by a rapid increase in body mass, with the progressive decline in their specific metabolic rates with development being termed the “ontogenetic decline” and has been reported in several species (cf. review of Anger, 2001). As a divergent pattern from developmental decline, near constant specific ammonia excretion rates have been noted during the development of zoea I through megalopa of *Carcinus maenas* (Harms et al., 1994). For phyllosoma larvae, Bermudes et al. (2008) observed this ontogenetic decline in specific rates of oxygen consumption and ammonia excretion by Stages I-V *J. edwardsii* phyllosomas. The present results on Stages I-IX *P. ornatus* phyllosomas showed the same ontogenetic pattern in specific oxygen consumption rates but not specific ammonia excretion rates

235 (Figs. 1B, C). As a measure of metabolic rates, oxygen consumption rates represent
236 total metabolism, while ammonia excretion rates represent protein-oriented metabolism
237 (cf. Ikeda et al., 2000).
238 Several lines of evidence indicate that the very low specific ammonia excretion
239 rates of Stage I *P. ornatus* larvae are due to preferential allocation of dietary protein to
240 somatic growth of the larvae rather than to metabolism. Phylllosoma larvae begin
241 feeding immediately after hatching, with little dependence on egg yolk reserves
242 (Johnston and Ritar, 2001), their diet is high in protein (*Artemia* nauplii in this study,
243 and possible zooplankton in the field, cf. Ritar et al., 2003), their body composition is
244 highly proteinaceous (see "Chemical composition" section below) and their digestive
245 enzymes have high protease activity (Johnston et al., 2003). The different
246 developmental patterns seen in specific ammonia excretion rates of early phyllosoma
247 larvae of temperate *J. edwardsii* and tropical *P. ornatus* may reflect species-specific
248 differences in nutritional functions of early larval phase.
249 The phyllosoma data of *P. homarus* and *P. antarcticus* of the present study, and *P.*
250 *interruptus, J. edwardsii* and the unidentified species by the previous workers fall
251 within or close to the 95% CI belt of "ornatus" line (Figs. 2, 3) , indicating no
252 significant between-species differences within phyllosomas when differences in body
253 mass and temperature are taken into account. No marked deviation of the data of wild
254 phyllosomas of *P. antarcticus* and the unidentified species from those of other lobster
255 larvae raised in the laboratory is evident (Figs. 2, 3). Specific oxygen consumption
256 data of stomatopod alima larvae, which are more active swimmers than phyllosomas, lie
257 reasonably above the "ornatus" line.
258 From global model of epipelagic marine zooplankton in which metabolic rates

were expressed as a function of body mass and habitat temperature (Ikeda, 1985), regression lines (referred as “general zooplankton” line hereafter) of specific oxygen consumption and ammonia excretion rates on DM at 28^{o}C were computed and compared with the “ornatus” line (Figs. 2, 3). Common to specific oxygen consumption and ammonia excretion rates, the “general zooplankton” line lies close to the upper 95% CI belt of the “ornatus” line in which all phyllosoma data are contained. Within the DM ranges studied, specific oxygen consumption rates and specific ammonia excretion rates of phyllosomas are calculated as half and one-tenth to half, respectively, of the average of those of “general zooplankton”.

4.2. O:N ratios

The O:N ratios of Stages I-IX *P. ornatus* larvae, with ratios 51-180 (Fig. 1D) are suggestive of lipid/carbohydrate-oriented metabolism (Mayzaud and Conover, 1988; Ikeda et al., 2000). Ritar et al. (2003) compared protein, carbohydrate and lipid composition of Stages I-VI *Jasus edwardsii* larvae before and after starvation for 15-74d and found that while protein and carbohydrate decreased slightly, lipid declined >50% of the initial value. Thus, the present results derived from O:N ratios and Ritar et al.’s (2003) results based on proximate composition of starved specimens are consistent in that protein is not the dominant metabolic substrate for food-deprived phyllosoma larvae.

A pattern of developmental decline in the O:N ratio of Stages I-IX *P. ornatus* larvae observed in this study contrasts that of results of *J. edwardsii* (Bermudes et al., 2008) but are consistent with American lobster (*Homarus americanus*) larvae (Sasaki et al., 1986) and the decapod *Carcinus maenas* larvae (zoea I through megalopa)(Harms et al., 1994). O:N ratios have been demonstrated to vary in response to molt phase (pre-,

284 inter- or post-molt), but molt phases causing maximum or minimum O:N ratios are not
285 the same between species (Anger, 2001). The effect of molt phase of the larvae on the
286 pattern of developmental changes in O:N ratios is considered to be minimal for
287 *Panulirus ornata* in this study since specimens where 1-day post-molt and hence each
288 specimen would be in the equivalent physiological state of the molt cycle (see
289 "Materials and methods" section). However, the standardization of the larvae by age
290 or molt stage could not be made for wild *Parribacus* phyllosomas.
291 With exceptions of two anomalous data (too high value of Stage I *P. ornatus* of
292 this study and too low value of the unidentified species (Ikeda, 1974)), the O:N ratios of
293 Stages IV-IX *P. ornatus*, Stage III *P. homarus* and *P. antatcticus* (mixed stage) larvae of
294 this study, and Stages I-III *J. edwardsii* larvae (Bermudes et al., 2008) shown to fall
295 within or close to the 95% CI belt of the "ornatus" line (Fig. 5). Further comparison of
296 the "ornatus" line with the "general zooplankton" line indicate that while the two lines
297 are near parallel of each other but the former is 4 times greater than that of the latter. It
298 is noted that the O:N ratios of stomatopod alima larvae are lower than those predicted
299 from the "ornatus" line but fit to the "general zooplankton" line, implying equivalent
300 importance of protein catabolism to that in general zooplankton. Overall, the results
301 indicate protein catabolism in phyllosomas is less important than other members of
302 tropical marine zooplankton.
303
304 *4.3. Chemical composition*
305 Across all developmental stages of *P. ornatus* phyllosoma development, from
306 Stage I to IX, the highest value of water contents (83%) and ash (34%) and the lowest
307 ones of N (6.9%) and C (33%) were recorded on Stage I phyllosomas (Table 2). The

only comparable data available from Belman and Childress (1973) reported a much higher ash (49%) and lower N composition (5.4%) for Stage I *Panulirus interruptus* larvae hatched in the laboratory (Table 2). A higher water content and ash, and lower C and N composition of Stage I larvae are suggestive of exhaustion of internal organic matter, including protein, for metabolism during its embryonic development before hatching. The observation that phyllosomas begin feeding immediately post-hatching has been reported on *J. edwardsii* (Johnston et al., 2003) and *P. ornatus* (Smith et al., 2009a), and indicate that phyllosomas are not lecithotrophic larvae. Increases in water content and ash content (=decrease in energy content per DM), accompanied with decreases in C and N composition, has been observed in several decapods larvae which have been starved (Anger and Dawirs, 1982; Harms et al., 1994). Body C:N ratios have been shown to be an insensitive index of nutritional conditions of the decapods larvae (Anger and Dawirs, 1982; Harms et al., 1994), as is also the case for *P. ornatus* in this study (Fig. 2). Inconsistency in the water contents and ash in Stages I-IV of captive reared *P. ornatus* (Table 2) may partly be reflected variations in egg quality, and hence nutritional conditions, of the larvae from different females and spawnings.

As phyllosomas are a common group of zooplankton occurring from tropical seas, it is interesting to compare the grand means of water content, C and N composition, C:N ratio and ash with those of other zooplankton groups in the same habitats to evaluate unique features, if any, of them (Table 2). The Stage I data was omitted from the calculation of the grand means because of its anomalous features mentioned above. Compared with stomatopod alima larvae, which exhibit the same body transparency, but are much more active swimmers, phyllosomas have lower C and N composition but higher ash contents though the significant test is not amenable due to limited sample

size (N = 2). Further comparison of phyllosoma data with those of copepods (Ikeda, 1974; Morris and Hopkins, 1983) and euphausiids/decapods (Ikeda unpublished) from tropical waters revealed that the former contain consistently lower N (9% versus 11-12%) and higher C:N ratios (4.5 versus 3.6-3.9) and ash (19% versus 7-15%) than the former (Mann-Whitney U-test at $p < 0.05$ or 0.01). Water contents of phyllosomas were higher than that of euphausiids/decapods (78% versus 71%) but lower than that of copepods (78% versus 82%). Since the major source of N is protein (Gnaiger and Bitterlich, 1984), lower N composition of phyllosomas suggests lower protein (=muscle mass) in the body and, perhaps, reduced locomotive activity compared to other crustacean plankton. Higher ash contents in phyllosomas are indicative of lower proportion of metabolically active components (organic matter) in the body. These interpretations are in concert with somewhat lower DM specific oxygen consumption rates of phyllosomas as compared with epipelagic marine zooplankton (cf. Fig. 2).

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Figure captions

Fig. 1. (A) Body mass, (b) specific oxygen consumption rates, (C) specific ammonia excretion rates and (D) metabolic O:N ratios of Stage I-IX *Panulirus ornatus* phyllosoma larvae. The best fit regression of the variables on the stage number was sought by applying linear, exponential or power models, and only significant regression lines were superimposed. ND = no data.

Fig. 2. Relationship between specific oxygen consumption rates at 28^{o}C of and body dry mass of phyllosoma larvae of *Panulirus ornatus* (this study), *Panulinus homarus* (this study), *Parribacus antarcticus* (this study), *Panulirus interruptus* (Belman and Childress, 1976), *Jasus edwardsii* (Bermudes et al., 2008) and an unidentified species (Ikeda , 1974). Note that all data fall within or close to the 95%CI belt of “ornatus” line (hatched lines) of this study. The figure includes the data of stomatopod alima larvae (this study) and “general zooplankton” line (solid line) predicted from global model of epipelagic marine zooplankton at 28^{o}C (Ikeda, 1985) for comparison. See text, for details.

Fig. 3. Relationship between specific ammonia excretion rates at 28^{o}C and body dry mass of *Panulirus ornatus* (this study), *Panulinus homarus* (this study), *Parribacus antarcticus* (this study), *Jasus edwardsii* (Bermudes et al., 2008) and an unidentified species (Ikeda, 1974). Note that all data fall within or close to the 95%CI belt of “ornatus” line (hatched lines) of this study. The figure includes the data of stomatopod alima larvae (this study) and “general zooplankton” line predicted from global model of epipelagic marine zooplankton at 28^{o}C (Ikeda, 1985) for comparison. For symbols of lobsters, see Fig. 2.

Fig. 4. (A) Water content, (B) ash, (C) C composition, (D) N composition and ((E) C:N ratios of Stage I-IX *Panulirus ornatus* phyllosoma larvae. The best fit regression of the variables on the stage number was sought by applying linear, exponential or power models, and only significant regression lines were superimposed.

Fig. 5. Relationship between O:N ratios and body dry mass of *Panulirus ornatus* a(this study), *Panulinus homarus* (this study), *Parribacus antarcticus* (this study), *Jasus edwardsii* larvae (Bermudes et al., 2008) and an unidentified species (Ikeda, 1974). Note that with two exceptions of Stage I *P. ornatus* (circled) and the unidentified species these data fall within or close to the 95%CI belt of “ornatus” line (hatched lines) of this study. The figure includes the data of stomatopod alima larvae (this study) and “general zooplankton” line (solid line) predicted from global model of epipelagic marine zooplankton at 28^{o}C (Ikeda, 1985) for comparison. For symbols of lobsters, see Fig. 2.

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Table 1. Body mass, oxygen consumption and ammonia excretion rates and O:N ratios of phyllosoma larvae of tropical rock and slipper lobsters of this study and other lobsters including unidentified species by previous workers. A-D for *P. ornatus* denotes different broods. The data of previous workers were converted to the rate at 28°C by using a Q_{10}=2. The data on stomatopod alima larvae are included for comparison. Means ± SD, with the number of replicates in parenthesis and range (only for mixed stages of *Parribacus antarcticus*). ND=no data.

Species	Source	Phyllosoma Stage	Expt T (°C)	Body mass (mgDM individual^{-1})	Oxygen consumption rate (µlO_2 individual^{-1}h^{-1})	Ammonia excretion rate (µgN individual^{-1}h^{-1})	O:N ratio, (by atoms)	Reference
Panulirus ornatus	Lab-raised (A)	I	28	0.041 ± 0.002 (6)	0.10 ± 0.01 (6)	0.00076 ± 0.00023 (6)	180 ± 49 (6)	This study
	Lab-raised (A)	II	28	0.079 ± 0.003 (6)	0.24 ± 0.01 (6)	ND	ND	This study
	Lab-raised (B)	IV	28	0.297 ± 0.012 (6)	0.98 ± 0.04 (6)	0.020 ± 0.003 (6)	62.6 ± 10.3 (6)	This study
	Lab-raised (A)	V	28	1.19 ± 0.07 (6)	2.08 ± 0.43 (6)	0.032 ± 0.016 (6)	93.6 ± 34.6 (6)	This study
	Lab-raised (A)	VI	28	1.52 ± 0.04 (6)	2.20 ± 0.70 (6)	0.079 ± 0.077 (6)	60.0 ± 33.5 (6)	This study
	Lab-raised (C)	VIII	28	3.98 ± 0.74 (8)	6.30 ± 1.15 (8)	0.167 ± 0.063 (8)	50.8 ± 12.1 (8)	This study
	Lab-raised (D)	IX	28	6.27 ± 0.78 (10)	8.26 ± 1.51 (10)	0.216 ± 0.065 (10)	53.5 ± 22.9 (10)	This study
Panulirus homarus	Lab-raised	III	28	0.186 ± 0.0129 (6)	0.44 ± 0.07 (6)	0.017 ± 0.003 (6)	31.6 ± 5.0 (6)	This study
Parribacus antarcticus	Wild	mixed	28	20.6 ± 18.4 (5) (range: 5.4-52.3)	13.2 ± 9.3 (5) (6.7-29.3)	0.458 ± 0.342 (5) (0.075-1.02)	56.4 ± 48.6 (5) (16.0-125)	This study
Panulirus interruptus	Lab-raised	I	17.3	0.065	0.073 (=0.154 at 28°C)	ND	ND	Belman & Childress (1973)
Jasus edwardsii	Lab-raised	I	18	0.069 ± 0.005	0.077* (=0.154 at 28°C)	0.0031* (=0.0062 at 28°C)	31.3	Bermudes et al. (2008)
	Lab-raised	II	18	0.129 ± 0.005	0.104* (=0.207 at 28°C)	0.0027* (=0.0054 at 28°C)	48.0	Bermudes et al. (2008)
	Lab-raised	III	18	0.275 ± 0.080	0.234* (=0.468 at 28°C)	0.0050* (=0.0100 at 28°C)	58.5	Bermudes et al. (2008)
	Lab-raised	V	18	0.502 ± 0.024	0.348* (=0.700 at 28°C)	0.0042* (=0.0084 at 28°C)	103	Bermudes et al. (2008)
unidentified	Wild	?	28	91.5 (1)	39.2	5.02 (1)	9.7 (1)	Ikeda (1974)
Stomatopod alima larvae	Wild		28	20.0 ± 7.26 (6)	26.9 ± 10.3 (6)	2.19 ± 0.87 (6)	16.7 ± 6.7 (6)	This study

474 *read from their Figs. 2 and 3 (data of the specimens placed in the "dark")

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Table 2. Water content, ash, C and N composition, C:N ratio and ash of phyllosoma larvae of the tropical rock lobster and slipper lobster of this study and other lobsters by previous workers. Values are mean ± SD with the number of replicates in parenthesis for water contents, and means of 2-4 replicates for C and N, and 2 replicates for ash. The data of stomatopod alima larvae, copepods and euphausiids/decapods from tropical waters are included for comparison with grand means of phyllosomas (Mann-Whitey U-test). The italic value in parenthesis of copepods and euphausiids/decapods' data represents the number of species studied. ND = no data. *; $p < 0.05$, ** : $p < 0.01$.

Species	Source	Phyllosoma stage	Water(% of WM)	C(% of DM)	N(% of DM)	C:N (by mass)	Ash(% of DM)	Reference
Decapod phyllosoma larvae								
Panulirus ornatus	Lab-raised	I	82.8 ± 1.3 (4)	32.5	6.9	4.8	34.2	This study
	Lab-raised	II	73.9 ± 1.2 (6)	39.8	9.5	4.2	20.8	This study
	Lab-raised	IV	81.9 ± 1.1 (6)	35.5	9.0	4.0	29.3	This study
	Lab-raised	V	75.4 ± 1.4 (6)	42.0	8.5	5.1	17.0	This study
	Lab-raised	VI	77.9 ± 1.6 (6)	40.5	9.3	4.3	17.6	This study
	Lab-raised	VIII	79.2 ± 4.2 (6)	40.9	9.4	4.4	18.5	This study
	Lab-raised	IX	77.4 ± 1.8 (10)	40.9	9.0	4.5	18.4	This study
	Wild	VIII,XI	ND	41.3	8.4	5.0	15.0	This study
Panulirus homarus	Lab-raised	III	80.2 ± 4.4 (6)	40.1	8.9	4.6	18.1	This study
Parribacus antarcticus	Wild	mixed	74.6 ± 1.2 (4)	39.8	8.8	4.6	14.8	This study
Panulirus interruptus	Lab-raised	I	ND	ND	5.4	ND	49.0	Belman & Childress (1973)
Grand mean (excluding Stage I data)			77.6 ± 2.8 (8)	40.1 ± 1.9 (9)	9.0 ± 0.4 (9)	4.5 ± 0.4 (9)	18.8 ± 4.3 (9)	
Stomatopod alima larvae	Wild		ND	45.1	9.6	4.7	14.7	This study
Copepods	Wild		ND	42.5** ± 1.5 (*8*)	11.0** ± 1.0 (*8*)	3.9** ± 0.3 (*8*)	ND	Ikeda (1974)
	Wild		81.6** ± 1.9 (7)	ND	ND	ND	6.8** ± 1.4 (7)	Morris & Hopkins (1983)
Euphausiids/decapods	Wild		71.3* ± 5.2 (*5*)	41.6^{NS} ± 1.1 (*5*)	11.7** ± 0.7 (*5*)	3.6** ± 0.1 (*5*)	14.5* ± 1.5 (*5*)	Ikeda (unpubl.)

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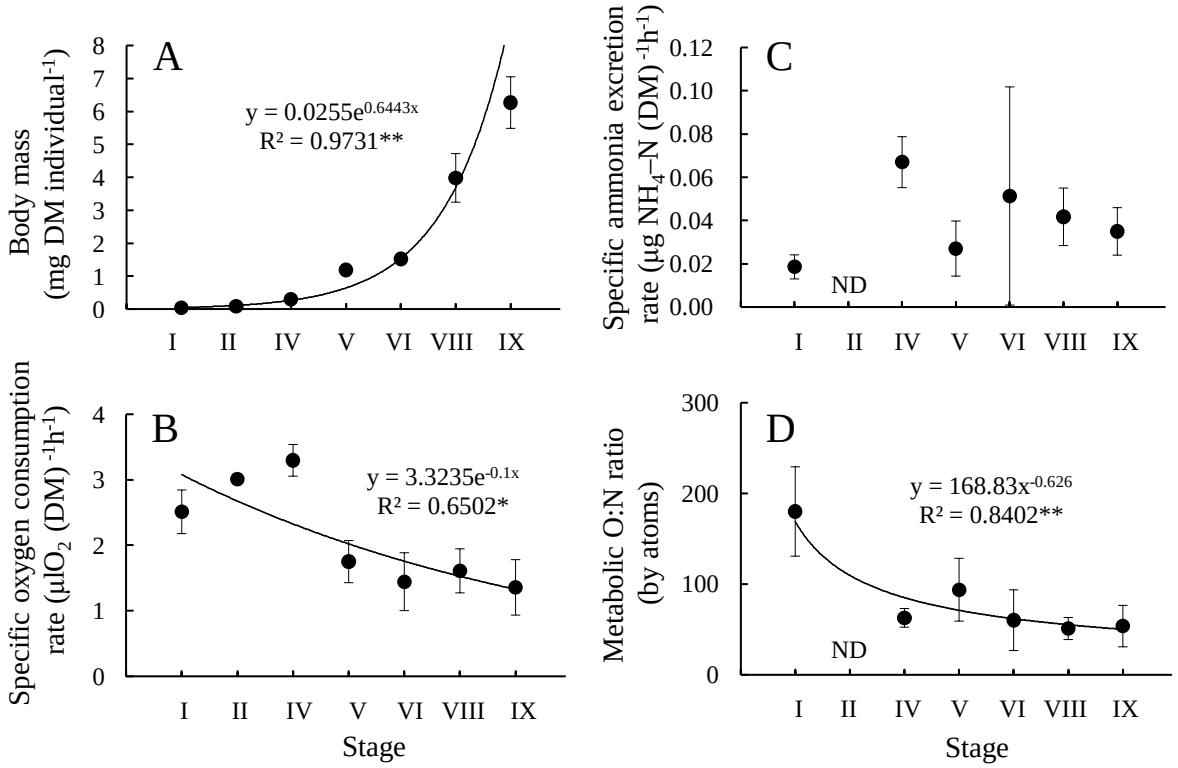
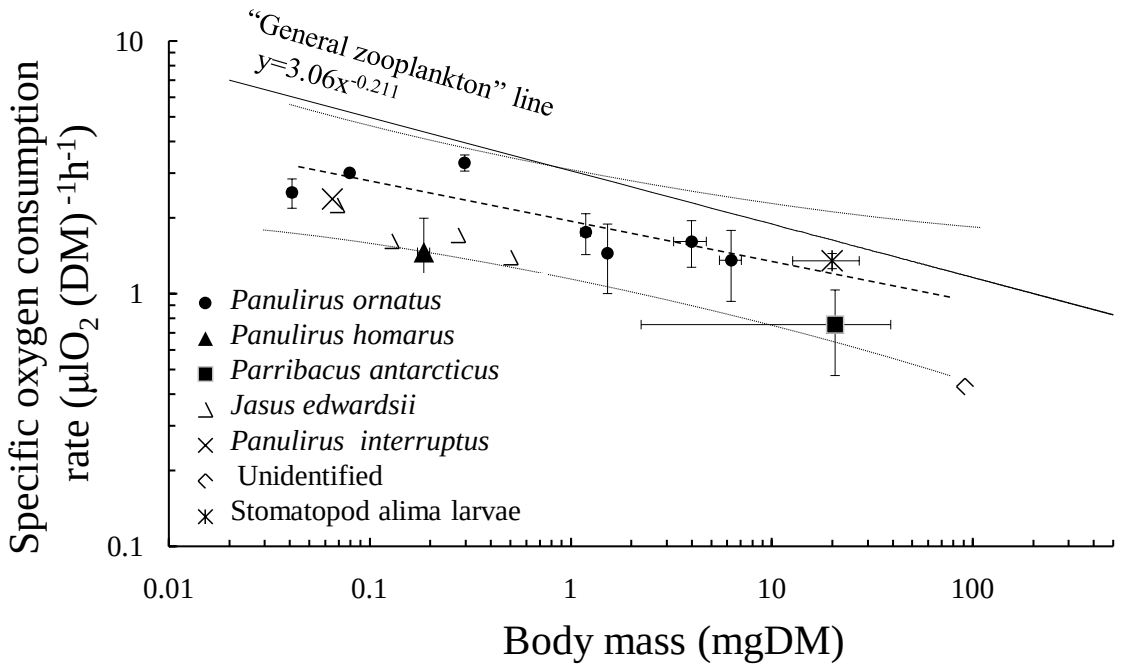


Ikeda et al. Fig. 1



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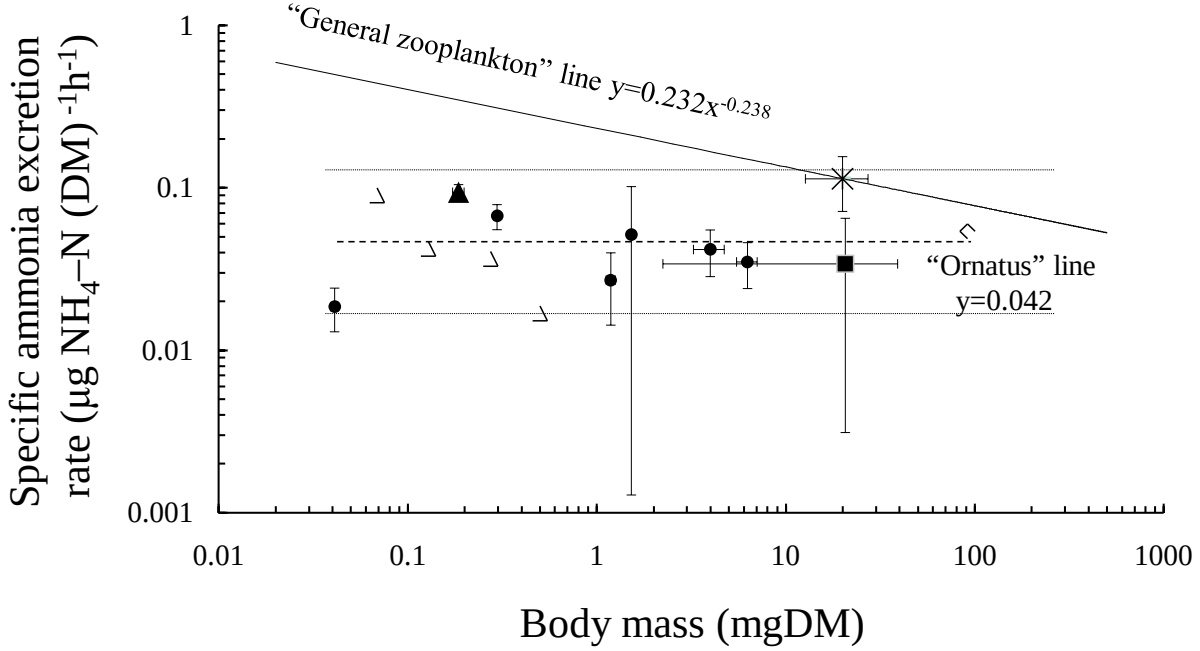

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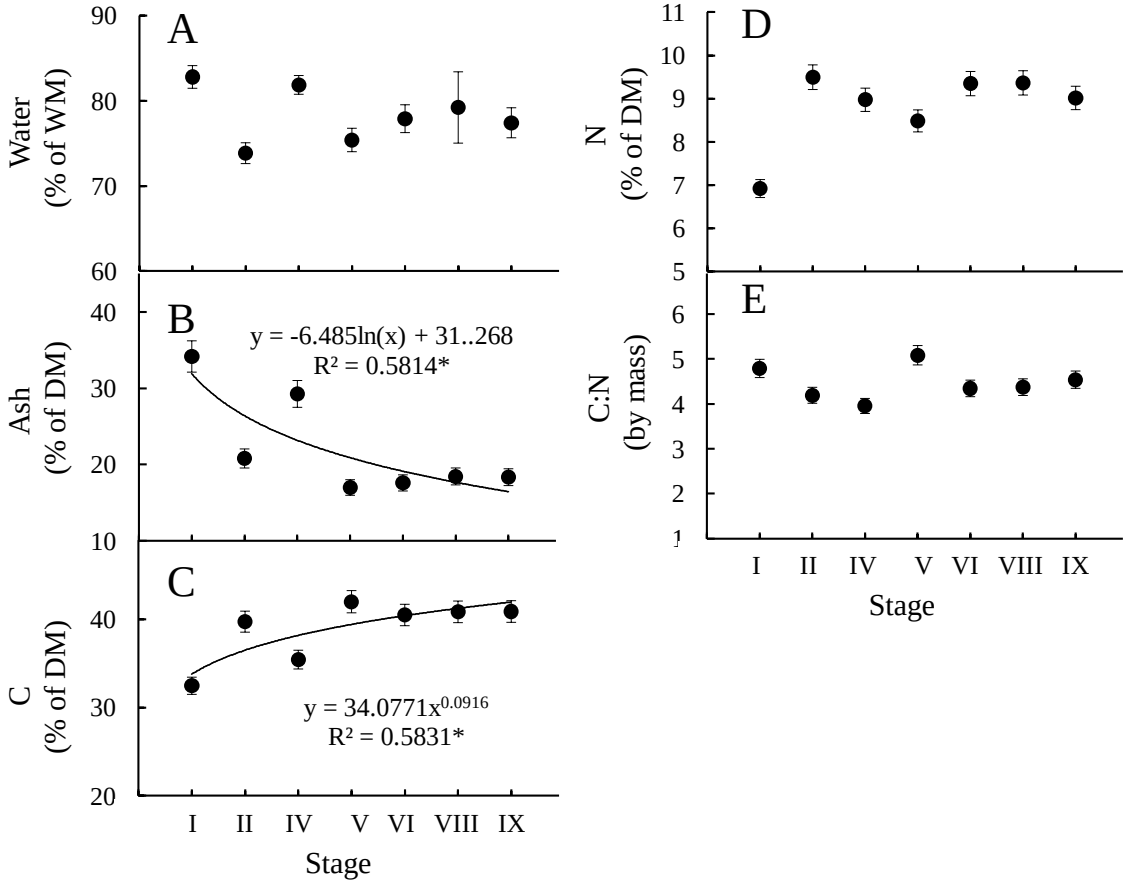

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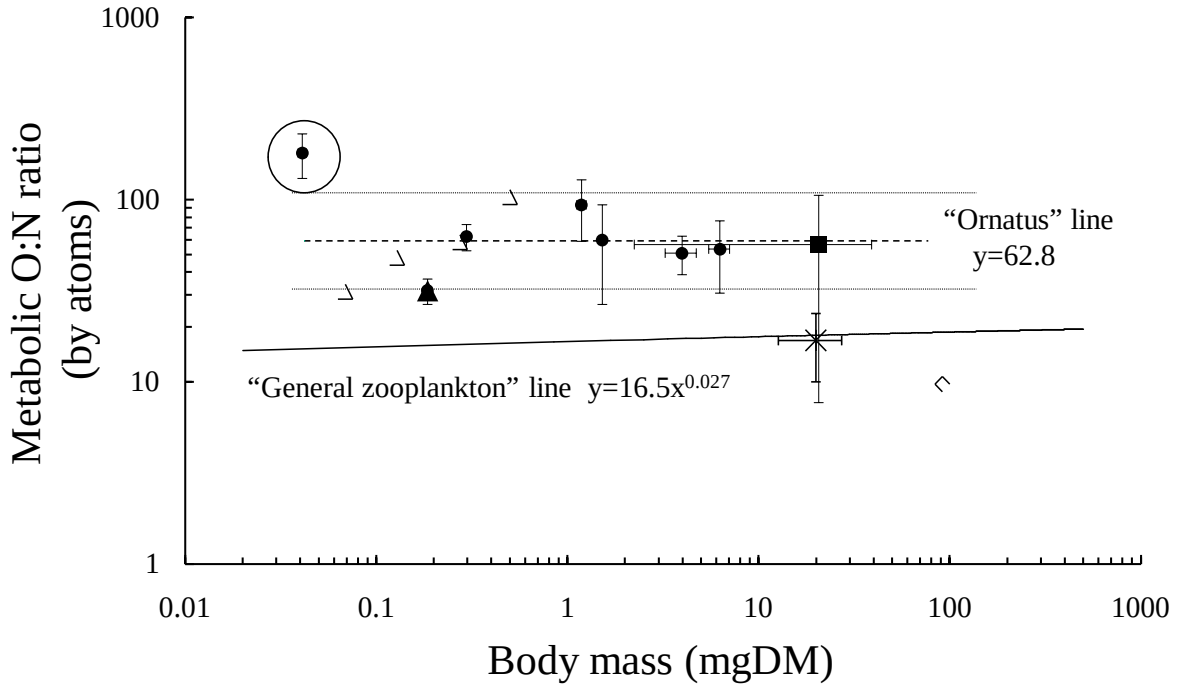

Ikeda et al. Fig. 5