Length–weight relationships and chemical content of the planktonic copepods in the Cananéia Lagoon estuarine system, São Paulo, Brazil

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Abstract: An attempt was made to measure body length, dry weight and chemical composition of the principal planktonic copepod species in the Cananéia Lagoon estuarine system, São Paulo, Brazil. Individual dry weight of the copepods increased logarithmically with increasing body size (prosome and total body lengths), and species-specific regression equations of the length-weight relationships were obtained for *Acartia lilljeborgi, A. tonsa, Pseudodiaptomus acutus, Paracalanus crassirostris, P. quasimodo, Temora turbinata, Labidocera fluviatilis, Oithona hebes, O. oswaldocruzi and Euterpina acutifrons.* Carbon, nitrogen and hydrogen content of these copepods was 44.2–46.4%, 10.9–12.1% and 6.7–7.2% of dry weight, respectively.

Key words: Copepoda, length-weight relationships, chemical content, Cananéia Lagoon estuarine system, Brazil

Introduction

Measurement of zooplankton biomass is essential in studies of production ecology of zooplankton. Zooplankton biomass can be measured using various methods, including gravimetric, volumetric and chemical procedures, expressed practically as, for example, settling volume, displacement volume, wet weight, dry weight, ash-free dry weight, organic weight and caloric values (Winberg 1971; Beers 1976; McCauley 1984; Omori & Ikeda 1984; Harris et al. 2000). Among these, chemical procedures that express biomass in terms of carbon or nitrogen content, is most important for understanding the biochemical cycle of elements and energy flow in aquatic food webs.

In determining zooplankton biomass in terms of carbon or nitrogen weight, organisms with a small body size (e.g. copepods) are difficult to separate out from plankton samples that contain other material such as detritus or phytoplankton. Additionally, when material is insufficient for accurate weight determination, biomass cannot be directly determined. However, when regression equations of length-

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weight relationships are obtained for each or all species that occur, their biomass can be facilely calculated from measurements of body length.

Species-specific length-weight relationships and chemical compositions have been determined for diverse planktonic organisms, such as copepods, from various estuarine, lagoonal and neritic waters of the world (e.g. Ikeda 1974; Uye 1982; Chisholm & Roff 1990; Hopcroft et al. 1998). However, until now, there has been no study available from South America. The present study presents species-specific regression equations of length-weight relationships and chemical composition (carbon, nitrogen and hydrogen content) for the principal planktonic copepod species in the Cananéia Lagoon estuarine system, a mangrove-surrounded estuary situated near the southern border of São Paulo State, Brazil.

Materials and Methods

Zooplankton samples were collected several times during the period from February 1995 to January 1996, at a fixed station (25°01'11"S, 47°55'43"W) located in Mar de Cananéia (Fig. 1), by oblique hauls of a plankton net (mesh



Fig. 1. Map showing the sampling station in the Cananéia Lagoon estuarine system.

size: $150 \,\mu\text{m}$). Net samples were immediately preserved in 5–10% formalin-seawater solution.

Copepods were identified by species, stage (C1-6) and sex under a microscope. Length and weight measurements were undertaken for the 10 principal copepod species: 7 species of Calanoida (i.e. Acartia lilljeborgi Giesbrecht, A. tonsa Dana, Pseudodiaptomus acutus [Dahl], Paracalanus crassirostris Dahl, P. quasimodo Bowman, Temora turbinata [Dana] and Labidocera fluviatilis Dahl), 2 species of Cyclopoida (i.e. Oithona hebes Giesbrecht and O. oswaldocruzi Oliveira) and 1 species of Harpacticoida (i.e. Euterpina acutifrons [Dana]). Measurements of body length were done for all copepodite and adult stages (C1-6) of these copepods. Measurements of dry weight of stages C1-6 were made for A. lilljeborgi, A. tonsa, P. acutus, T. turbinata and L. fluviatilis, whereas the weights of stages C3-6 were measured for P. crassirostris, P. quasimodo, O. hebes, O. oswaldocruzi and E. acutifrons since their C1 and C2 were not collected in sufficient amounts to weigh.

Both prosome and total body lengths were measured using an eyepiece micrometer. For adults of *A. lilljeborgi*, which possess a pair of large spines on the posterior part of the prosome, these spines were not included in the prosome length. For adult females of *P. acutus*, which possess asymmetric furca, the measurement of body length was done regarding the longer one. For *P. acutus*, *P. crassirostris*, *P. quasimodo*, *T. turbinata* and *E. acutifrons*, the bodies of which often bend, measurements of several body parts were made to obtain their body lengths.

For measurements of dry weight, undamaged specimens were picked up under a stereoscopic microscope. Lots consisting of 3 to ca. 2000 indiv., depending on species, stage and sex, were obtained to attain a minimum dry weight of ca. 100 μ g. The animals were rinsed with a small amount (ca. 1–2 ml) of distilled water and placed onto pre-weighed glass-fiber filters (Whatman GF/C, 25 mm in diameter) or small aluminum foil dishes (ca. 20 mm in diameter). Then, these were dried at 60°C for 24 h in an electric oven, cooled in a desiccator at room temperature, and their dry weights were measured several times using an electronic microbalance (Sauter, Model D81) until reaching a constant weight.

I assumed no weight loss during preservation, since there was no significant difference in the dry weight of copepodites and adults of *A. lilljeborgi* between live animals and specimens preserved in formalin-seawater solution for 1-12months.

For determining chemical composition, samples that consisted of a single species containing all stages were processed following the same methods used for dry weight. Then their chemical (carbon, nitrogen and hydrogen) composition was determined using a CHN analyzer (Perkin Elmer, Elemental Analyzer 2400CHN).

The relationship between body length $(L, \mu m)$ and dry weight $(W, \mu g)$ of a copepod can be expressed as:

 $W = aL^b$

where a and b are constants. The equation is fitted by conversion to logarithms of base 10, and the linearized equation,

$$\log_{10} W = \log_{10} a + b \log_{10} L$$

was solved by the least squares method.

Results

Dry weight

Dry weight of each species increased exponentially with developmental stage (Fig. 2). Females in C4–6 were generally larger and heavier than males. Stage-to-stage increments of weight of each species were constant for most stages. However, dry weight of *P. crassirostris* decreased from C5 to C6 in the males. *Euterpina acutifrons* was found to have dimorphism in the males of C4–6, and the weights of the small males of C4–6 were constant.

Length-weight relationships

Individual dry weight increased logarithmically with increasing prosome and total body lengths, and species-specific regression equations of length-weight relationships were obtained with a slope of approximately 3 in each case (Figs 3, 4). All of these 329 plots were combined in order to obtain regression equations for all copepod species ana-



Fig. 2. Dry weight of the developmental stages C1-6 of 10 principal copepod species in the Cananéia Lagoon estuarine system. Dry weight is expressed as mean (\oplus)±SD (vertical bars), respectively.

lyzed in the present study. There were highly significant correlations between body lengths and dry weight (DW, μ g), expressed as:

DW=8.487×10⁻⁸ PL^{2.693} (
$$r$$
=0.943, p <0.0001)
DW=6.522×10⁻⁹ BL^{2.959} (r =0.954, p <0.0001)

where PL and BL were prosome and total body length (μm) , respectively.

Chemical composition

Chemical composition (carbon, nitrogen and hydrogen content), expressed as percentage of dry weight, of 10 principal copepod species are presented in Table 1. Carbon, nitrogen and hydrogen content did not differ among species and were 44.2-46.4% (mean±SD: $45.6\pm0.7\%$), 10.9–

12.1% (11.5 \pm 0.4%) and 6.7–7.2% (6.9 \pm 0.1%), respectively.

Discussion

There was no significant difference in the dry weight of Acartia lilljeborgi between live animals and specimens preserved in formalin-seawater solution for 1-12 months. Similarly, for the copepods Acartia spp., Centropages velificatus, Temora turbinata and Undinula vulgaris collected in Kingston Harbour, Jamaica, Chisholm & Roff (1990) found no significant loss in individual weight between recently collected living animals and specimens preserved in 10% formalin for 10 months. On the other hand, for marine copepods in temperate, subboreal and boreal waters, several studies have shown that the preservation of animals with formalin could cause a significant loss (25% or more) in dry weight (Durbin & Durbin 1978; Williams & Robins 1982; Giguère et al. 1989). The weight loss is attributed principally to loss in proteins and/or lipids stored in the body (Hopkins 1968; Durbin & Durbin 1978; Omori 1978; Salonen & Sarvala 1980). Proteins are the major component in the protoplasm of zooplankton (e.g. copepods, decapods, euphausiids, mysids, chaetognaths) with a high degree of variation among species (Raymont et al. 1964; Raymont 1983), and are the dominant metabolic reserve of zooplankton in waters with constant food supplies such as tropical estuarine environments (Madhupratap et al. 1979). In practice, the proteins of organisms preserved with formaldehyde (formalin) are difficult to lyse and decompose, due to cross-linkage between protein molecules. The lipids are frequently concentrated in large oil droplets, which are a prominent feature of copepods inhabiting boreal, subboreal and temperate waters (Ikeda 1974; Hirakawa & Imamura 1993). In the present study, however, oil droplets were not found in any copepods, i.e. neither live animals nor specimens preserved in formalin-seawater solution, as similarly observed for marine copepods inhabiting tropical and subtropical waters such as Kingston, Jamaica (Chisholm & Roff 1990) and São Sebastião Channel, São Paulo, Brazil (De La Rocha 1998). Additionally, there was no evidence of lipid leakage at the surface of plankton samples stored in formalin-seawater solution for 12 months after collection. Therefore, in the present study, there would be no significant loss in dry weight for most of the copepods preserved with formalin, and this can be attributed mainly to the small amount of lipids stored in their bodies.

Although determination of chemical composition was done only on the specimens preserved in formalin-seawater solution for 1–12 months, carbon, nitrogen and hydrogen content values obtained in the present study were similar to those obtained for the same species in former studies in which live animals were utilized for determining chemical composition, being ca. 45, 10–15 and 7% of dry weight, respectively (Omori 1978; Hirota 1981; Uye 1982; Ambler 1985; Abdel-Moati et al. 1993). The regression equations



Fig. 3. Log length-log weight relationships for 8 copepod species from the Cananéia Lagoon estuarine system.



Fig. 4. Log length-log weight relationships for 2 copepod species from the Caranéia Lagoon estuarine system.

Table 1. Carbon, nitrogen and hydrogen content, expressed as percentage (mean \pm SD) of dry weight, of 10 principal copepod species in the Cananéia Lagoon estuarine system. *n*: number of observations.

Species	Carbon	Nitrogen	Hydrogen	n
Acartia lilljeborgi	45.33±0.09	11.71±0.03	6.72±0.05	2
Acartia tonsa	44.21 ± 0.08	11.35±0.01	6.78±0.11	2
Pseudodiaptomus acutus	46.11±0.04	11.64±0.00	7.05 ± 0.02	2
Paracalanus crassirostris	46.26±0.01	10.90±0.01	7.03±0.04	2
Paracalanus quasimodo	45.56±0.21	11.26±0.03	6.90±0.05	2
Temora turbinata	44.57 ± 0.02	11.60 ± 0.01	6.79±0.03	2
Labidocera fluviatilis	45.21±0.04	12.11±0.06	6.94±0.01	2
Oithona hebes	46.11±0.05	11.69±0.04	7.02 ± 0.02	2
Oithona oswaldocruzi	46.37 ± 0.06	10.96±0.05	7.16±0.03	2
Euterpina acutifrons	46.04 ± 0.14	11.30 ± 0.02	7.01 ± 0.00	2
Total (mean)	45.58±0.73	11.45±0.36	6.94±0.14	2

of length-weight relationships obtained in the present study are similar to those obtained for the same species in former studies in which live animals were used for measurements of dry weight (Nassogne 1972; Durbin et al. 1983; Kiørboe et al. 1985; Chisholm & Roff 1990; Cataletto & Fonda Umani 1994; Thompson et al. 1994): the slope of the linearized regression was approximately 3, and that is empirically acceptable for copepods (Omori & Ikeda 1984).

The length-weight relationships and chemical content of marine copepods can vary, depending not only on species, developmental stage and sex, but also on season and geographic location (Omori 1969; Ikeda 1974; Durbin & Durbin 1978; Ambler 1985; Cataletto & Fonda Umani 1994; Viitasalo et al. 1995). This can be explained primarily by variations in temperature, food availability and the physiological state of the animals (Ikeda 1974; Durbin & Durbin 1978; Tanskanen 1994). In the present study, the variations in length-weight relationship and chemical content are masked since these were based on the analysis of materials collected in different seasons of the year. However. I assume that the seasonal variation in length-weight relationship and chemical content of small-sized estuarine and neritic copepods in tropical and subtropical waters would not be large since the seasonal variation in temperature and food availability in tropical and subtropical waters is much smaller than that in temperate, subboreal and boreal waters: the annual variation in water temperature and chlorophyll-a concentration in the Cananéia Lagoon estuarine system during the study period was 18.6-29.4°C and $1.3-20.4 \,\mu g \, l^{-1}$, respectively (Ara 1998, 2001).

The regression equations of the relationships between body lengths and dry weight obtained in the present study are useful for biomass estimates, which are fundamental for copepod production ecology. Unfortunately, in the present study regression equations were not obtained for all of the copepod species occurring in the Cananéia Lagoon estuarine system. However, species-specific and combined regression equations can be utilized to estimate biomass for the juvenile stages (C1-2) of Paracalanus crassirostris, P. quasimodo, Oithona hebes, O. oswaldocruzi and Euterpina acutifrons since the shapes of these copepodite stages are like miniatures of their adults. In general, the shape of most free-swimming copepod species are similar to each other, although here I do not deal with detailed morphological characteristics that taxonomists focus on. However, for rough estimates for marine copepods, the regression equations obtained in the present study are applicable not only to other species whose species-specific length-weight relationships were not obtained, but also to other regions in Brazilian waters because the copepod species analyzed in the present study are very common in estuarine, lagoonal and neritic waters along the coast of Brazil. From these results, the biomass of copepods in terms of dry and chemical (carbon, nitrogen and hydrogen) weight can be facilely calculated from measurements of body length and abundance.

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