Limnol. Oceanogr., 56(1), 2011, 403–414

doi:10.4319/lo.2011.56.1.0403

Metabolic rates and carbon budget of early developmental stages of the marine cyclopoid copepod *Oithona davisae*

Rodrigo Almeda,* Miquel Alcaraz, Albert Calbet, and Enric Saiz

Institut de Ciències del Mar (CSIC), Barcelona, Spain

Abstract

The genus *Oithona* has been considered the most abundant and ubiquitous copepod in the world's oceans. However, despite its importance, the metabolism of its developmental stages (nauplii and copepodites), crucial to explain their evolutionary success, is almost unknown. We determined respiration rates, ammonium and phosphate excretion rates, and the net growth efficiencies of early developmental stages of *Oithona davisae* as related to stage, body weight, temperature, and food availability. Respiration and excretion rates increased with increasing body weight and were positively related to temperature and food. Specific respiration rates of nauplii and copepodites varied from 0.11 to 0.55 d^{-1} depending on stage, body weight, temperature, and food availability. Metabolic C : N ratios were higher than 14, indicating lipid-oriented metabolism. Assimilation efficiencies and net growth efficiencies ranged from 65% to 86% and from 23% to 32%, respectively, depending on body weight, stage, and temperature. Assimilation efficiencies and net growth efficiencies estimated using the respiration rates of nauplii with food were 1.7 times higher and 0.6 times lower, respectively, than those calculated using respiration rates of nauplii without food. Therefore, the use of respiration rates measured in filtered seawater led to substantial bias on the estimations of zooplankton carbon budget. *O. davisae* developmental stages exhibited similar assimilation and growth efficiencies but lower carbon-specific respiratory losses than calanoid copepods. Hence, the low metabolic costs of *Oithona* compared with calanoids may be one reason for their success in marine ecosystems.

Among the factors that regulate the success of copepod populations in the oceans, energetic balance is of prime importance. The assessment of metabolic budgets may reveal important differences in the cost of maintenance and in the efficiency of food utilization by different organisms that help to understand their evolutionary success. For this reason, respiration and excretion rates of copepods have been extensively investigated during the last century (for review, Marshall 1973; Ikeda 1985; Ikeda et al. 2001). However, most of the available information stems from studies devoted to late copepod stages while copepod nauplii have traditionally been ignored. Copepod nauplii are the most abundant forms of metazoans on the planet (Fryer 1986) and the main prey of most fish larvae (Last 1980); therefore, their production contributes significantly to the recruitment of commercially important fish species (Castonguay et al. 2008). Given that copepod nauplii may stand for an important fraction of the biomass of metazooplankton in spite of their small size (Calbet et al. 2001) and that the specific metabolic rates are inversely related to body mass (Ikeda 1985; Ikeda et al. 2001), the potential importance of nauplii in the energy flow of marine food webs must be stressed.

Among copepods, the genus *Oithona* has been considered the most abundant and ubiquitous in the world's oceans (Gallienne and Robins 2001). The success of oithonids over calanoids in some marine systems has been attributed, among other reasons, to their comparatively low respiration rates (Lampitt and Gamble 1982; Paffenhöfer 1993; Castellani et al. 2005). However, there is no information regarding the respiration rates of the *Oithona* naupliar stages that allows the corroboration of this hypothesis. Given the importance of the larval stages for the success of population recruitment, understanding the variability and the control exerted by the environmental (temperature, food availability, etc.) and inherent (age, size, etc.) factors on their metabolic activity is of major importance to comprehend the capacity of *Oithona* to exploit diverse marine ecosystems.

The present study will provide basic information concerning the metabolic rates (respiration and excretion rates) of the naupliar and early copepodite stages of the genus *Oithona.* We used as a model the species *Oithona davisae* that commonly inhabits productive embayments, where it can be the most abundant copepod (Uye and Sano 1995). The specific objectives of this study were to determine the respiration and ammonium and phosphate excretion rates of *O. davisae* early developmental stages in relation to body weight, temperature, and food availability, and to estimate the assimilation and net growth efficiencies deduced by combining the present respiration rates and the ingestion and growth rates reported from this and previous studies (Almeda et al. 2010*b*).

Methods

Experimental organisms—The specimens of *O. davisae* were obtained from a laboratory culture maintained in the Institut de Ciències del Mar (CSIC, Barcelona, Spain) since October 2000. *Oithona davisae* was grown in 20-liter Plexiglas tanks, at 20 ± 1°C in a constant-temperature room and under a 12 : 12 light : dark cycle. Copepod cultures were routinely fed a suspension of the heterotrophic dinoflagellate *Oxyrrhis marina* (equivalent spherical

* Corresponding author: ralmeda@icm.csic.es

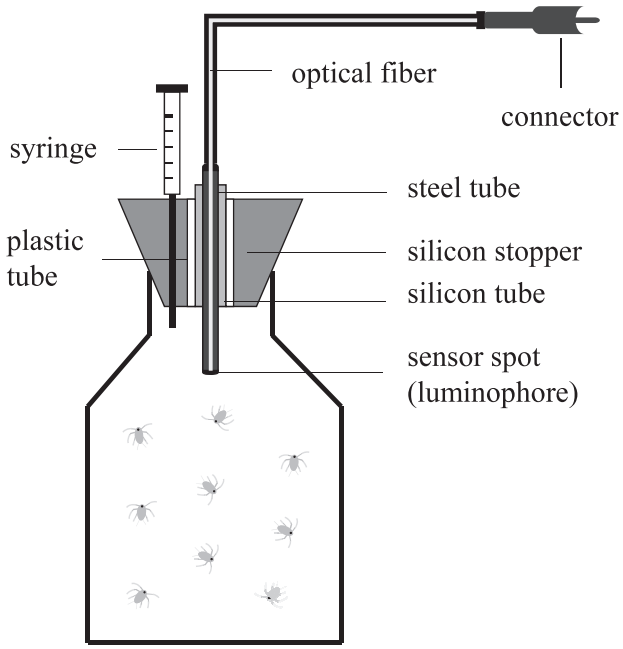

Fig. 1. Schematic representation of incubation chambers used for the estimation of metabolic rates.

diameter [ESD] = 16 μm). In turn, *Oxyrrhis marina* were fed the cryptophyte *Rhodomonas salina* (ESD = 8 μm) grown in f/2 medium. Prey sizes were measured using a Coulter Multisizer III particle counter (Beckman Coulter). To obtain cohorts of nauplii, adults (including egg-bearing females) from the stock culture were removed with a 132-μm-mesh-size sieve and placed in a new tank, where they were fed ad libitum with *Ox. marina* ($>$ 3000 cells mL^{-1}, equivalent to $>$ 660 μg C L^{-1}). After 20 h, adults were separated from the hatched nauplii with a 100-μm mesh. In order to remove the dislodged sacs of eggs, we allowed them to settle to the bottom of the tank, and subsequently the upper water containing only the nauplii were siphoned out to a new tank.

General experimental procedures—We conducted three series of experiments in order to determine the influence of body weight, temperature, and food availability on the metabolic rates of *O. davisae* early developmental stages. In all cases, respiration and excretion rates were estimated by the classical incubation method in closed chambers. These chambers consisted of Pyrex bottles (130-mL total volume) capped by silicon stoppers pierced by the oxygen probes and by a syringe needle to compensate for pressure changes due to small temperature oscillations (Fig. 1).

The respiration rates of copepod nauplii were measured using optical oxygen sensors ("oxygen optodes," Oxygen Dipping Probe DP-PSt3, Presens®), which consist of a fiber optic cable with a distal tip coated with a luminophore. The light emitted from the source (blue light-emitting diode) is transmitted through the fiber optic cable to the luminophore. The intensity, lifetime, and modulation of phase angle of the resulting fluorescence signal depend on the oxygen concentration (Holst et al. 1997). The frequency of oxygen concentration measurements was 1 min. Ammonia (NH_4-N) and phosphate (PO_4-P) concentrations were determined by the reactions of Berthelot and molybdate, respectively (Hansen and Koroleff 1999), using a Double Beam spectrophotometer (VarianCary®). The chemical analyses were made immediately after taking samples.

Table 1. Cohort composition (dominant stages) at the beginning of the experiments. Size (total body length for nauplii and prosome length for copepodites) and estimated body weight (W) of the *Oithona davisae* developmental stages used in the study are also provided. Carbon content was calculated from size using the equation reported in Almeda et al. (2010*a*).

Cohort age (h)	Developmental stage	Length ± SE (μm)	W ± SE (ng C ind.$^{-1}$)
20	NI	84.4±0.6	28.3±0.4
48	NI–NII	93.4±0.8	34.7±0.7
72	NII–NIII	102.2±1.1	42.9±1.0
76	NII–NIII	107.3±1.2	46.9±1.1
83	NIII–NIV	114.5±1.2	54.8±1.2
96	NIV–NV	127.2±1.0	68.4±1.2
180	CII–CIII	210.9±1.5	176.0±2.1

Body weight effect on respiration and excretion rates—Several cohorts of newly hatched nauplii were kept at saturating food conditions ($>$ 3000 cells mL^{-1}, *Ox. marina*) during the time required to reach the desired developmental stage (Table 1). Developmental stages (at least 30 individuals [ind.]) were identified under an inverted microscope (×100) according to Uchima (1979). Prior to the experiments ($\sim$ 30 min), the individuals from each cohort were concentrated using a 37-μm mesh for nauplii and a 60-μm mesh for copepodites, and thoroughly rinsed with autoclaved 0.2-μm-filtered seawater (0.2-μm-FSW) to remove food and other particles. We counted the organisms in each cohort by triplicate under the microscope, and aliquots were added to the experimental bottles (three replicates) to obtain the desired density of experimental organisms (from 10 to 60 ind. mL^{-1}, depending on the developmental stage). The bottles were incubated in darkness at 20°C (± 0.1°C) in temperature-controlled water baths. Three additional bottles with only 0.2-μm-FSW were set as controls. The temperature of the water baths was continuously monitored during the incubation by an external sensor (ENDECO®).

At the end of the incubation, three replicate water samples for ammonium and phosphate analysis were taken from each experimental and control bottle by siphoning with a silicone tube with the submerged tip fitted with a 37-μm-mesh gauze to avoid losing experimental organisms. The remaining water was sieved through a 37-μm mesh and the experimental organisms concentrated and fixed with Lugol's solution. Subsamples of nauplii were counted under an inverted microscope (×40) to estimate their concentration during the incubation, and 50 individuals were measured by image analysis in order to estimate their carbon contents using the equation provided by Almeda et al. (2010*a*). The mortality of nauplii or copepodites at the end of the incubation experiments was always negligible.

Temperature effect on respiration and excretion rates—Oxygen consumption and ammonia and phosphate excretion rates of *O. davisae* nauplii (NIII–NIV) were simultaneously determined at four different temperatures (16°C, 20°C, 24°C, 28°C) in 0.2-μm-FSW. The temperatures used in this experiment are into the temperature range experienced by this species in nature (8.9–28.2°C, Uye and Sano 1995). A cohort of nauplii (NIII–NIV), obtained as described above, was divided into aliquots and conditioned for $\sim$ 2 h at each temperature under food satiating conditions (*Ox. marina*, $>$ 3000 cells mL^{-1}). After acclimation, nauplii were concentrated again using a 37-μm-mesh sieve rinsed in 0.2-μm-FSW. The experimental concentration of nauplii ranged from 90 to 140 ind. mL^{-1} and their length averaged 114.5 $\pm$ 1.2 μm SE (54.8 ng C ind.$^{-1}$). Control and experimental bottles were incubated in water baths at the corresponding temperatures. The water-bath temperatures during the incubations were continuously monitored by an external sensor (ENDECO®). Water samples for ammonia and phosphate analysis and nauplii were collected at the end of the incubation and processed as described above. The effect of temperature on respiration and excretion rates was estimated by the Q_{10} value:

$$Q_{10} = (M_2/M_1)^{10/(T2-T1)} \quad (1)$$

where M_2 and M_1 are the rates of the studied process at temperatures T2 and T1 (in °C), respectively.

Food availability effects on respiration and excretion rates—The experimental setup consisted of measuring the metabolic rates of (1) *O. davisae* nauplii (NII–NIII) with food (*Ox. marina*), (2) nauplii without food (in 0.2-μm-FSW), (3) *Ox. marina* alone, and (4) control bottles with only 0.2-μm-FSW. There were three to four replicates per treatment. The density of nauplii was $\sim$ 15 ind. mL^{-1} on average. We used an *Ox. marina* concentration of 11,000 cells mL^{-1} (2.75 μg C mL^{-1}) to maintain saturating conditions during the incubation. Nauplii were concentrated, counted, and added to the experimental bottles as described before. All these incubations were conducted at constant temperature (20°C). Prior to preparing the food suspensions for the experiment, the stock culture of *Ox. marina* was filtered through a 10-μm mesh to remove cell aggregation and other particles. The culture of *Ox. marina* was not fed the day before the experiments began in order to ensure that all *Rhodomonas salina* were depleted by *Ox. marina*. The absence of *R. salina* in stock bottles was verified by checking with a Coulter Multisizer particle counter. In order to reduce the number of bacteria in the stock cultures, *Ox. marina* was concentrated by exposure to low temperatures (4°C), so that the dinoflagellates settled by reducing their swimming activity. After that, the supernatant was carefully siphoned out and the dinoflagellate resuspended again in filtered seawater; this washing protocol was repeated two times. Prior to the experiments, *Ox. marina* was acclimated to 20°C and their swimming activity checked under an inverted microscope ($\times$200). Incubation conditions and other procedures were conducted as described above, with the exception of the incubation time that was shorter (16 h).

The specific ingestion and growth rates of fed nauplii were simultaneously determined as described below.

Calculation of weight-specific respiration and excretion rates—The oxygen consumption rates were calculated as the slopes of the linear regression equations relating incubation time and dissolved oxygen concentration for each experimental condition. When the decrease in oxygen concentration was not linear along the incubation (Fig. 2A,B), respiration rates were calculated using the linear decrease in oxygen during the first hours of incubation before the change of the slope.

Respiration rates of starving nauplii (incubated without food) and *Ox. marina* bottles were determined after subtracting the oxygen consumption rate in the control bottles with only filtered seawater. The respiration rates of feeding nauplii were calculated after taking into account also the oxygen consumption in the incubation bottles due to *Ox. marina*. In order to do that, the initial and final concentration of *Ox. marina* (cells mL^{-1}) in the bottles with only *Ox. marina* and the bottles with the feeding nauplii (i.e., with *Ox. marina*) were measured with a Coulter Multisizer, and the respective average concentrations during the incubation were determined according to Frost's (1972) equations. Concentrations were converted into carbon units from cell volume according to Pelegri et al. (1999). Carbon-specific respiration rates of *Ox. marina* were calculated in the *Ox. marina* bottles (without nauplii), and then were used to subtract the oxygen consumption by *Ox. marina* in the feeding nauplii bottles using the average concentration of *Ox. marina* in them. The respiration rate of the feeding nauplii was estimated from the remaining oxygen consumption rate divided by the number of nauplii incubated.

Individual excretion rates were calculated as the difference in NH_4-N and PO_4-P concentrations between experimental and control bottles at the end of the incubation divided by the number of nauplii. In the experiments with feeding nauplii, excretion rates were corrected for the presence of *Ox. marina* in the incubations similarly as described above for respiration rates.

Per capita respiration and excretion rates were converted into carbon-specific rates using the carbon content of *Oithona* nauplii as estimated described above. Oxygen consumption rates were transformed into carbon losses using a respiratory quotient of 0.97 (Omori and Ikeda 1984). The C : N (respired C : excreted N), N : P (excreted N : excreted P), and C : P (respired C : excreted P) metabolic ratios (by atoms) were calculated for each experiment in order to determine the stoichiometric composition of metabolic products as well as the type of catabolism (Omori and Ikeda 1984).

In the food effect experiment, the clearance and ingestion rates were estimated according to Frost (1972) and the specific ingestion rates were calculated using the average biomass of nauplii during the incubation. Specific growth rates (G, μg C μg C^{-1} d^{-1}) were calculated as

$$G = [\ln(W_2/W_1)]/t \quad (2)$$

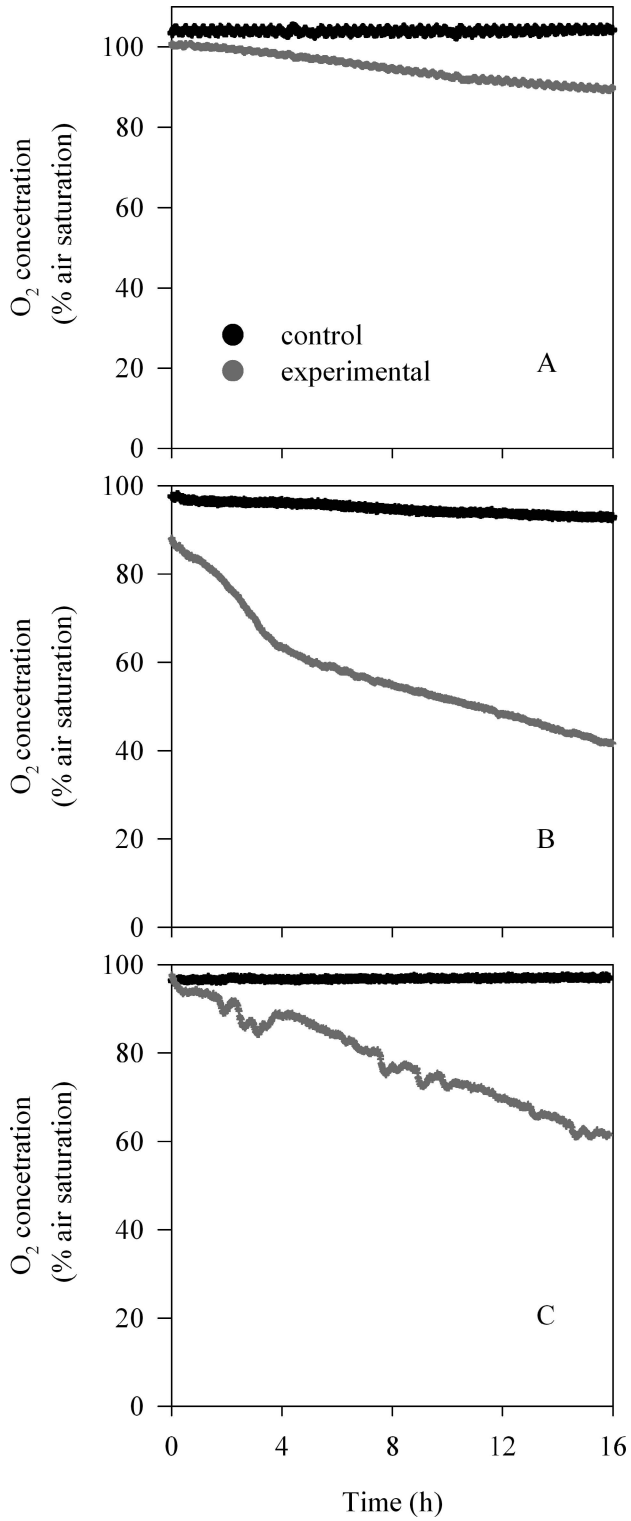

Fig. 2. Some illustrative examples of the temporal evolution of oxygen concentration in the experimental chambers under different experimental conditions. Control chambers are bottles with only 0.2-μm-FSW, and experimental chambers correspond to (A) nauplii incubated in filtered seawater at 20°C, (B) nauplii incubated in filtered seawater at 28°C, and (C) nauplii incubated with food (*Oxyrrhis marina*) at 20°C.

where t is the duration of incubation (d), and W_1 and W_2 are the initial and final carbon content of the nauplii, respectively. Carbon content of nauplii was estimated as described above.

Metabolic balance: assimilation and net growth efficiencies—The carbon budget corresponding to the larval stages of *Oithona davisae* was calculated by combining the metabolic rates and the feeding and growth rates determined in the present study with the growth and ingestion rates under saturating food conditions estimated according to Almeda et al. (2010*b*).

The assimilation efficiency (AE), i.e., the percentage of ingested food that is digested, can be expressed as

$$AE = \left(G + R_C^*\right)/I \times 100 \quad (3)$$

where G is the carbon-specific growth rates (μg C μg C^{-1} d^{-1}), R_C* is the carbon-specific respiration rates (μg C μg C^{-1} d^{-1}), and I is the carbon-specific ingestion rates (μg C μg C^{-1} d^{-1}) under food saturation conditions.

The net growth efficiency (NGE), i.e., the percentage of assimilated food converted into growth, was calculated as

$$NGE = G/\left(G + R_C^*\right) \times 100 \quad (4)$$

When carbon-specific respiration rates were measured in filtered seawater (R_C), the expected carbon-specific respiration rates under food saturation conditions (R_C*) were estimated using the correction factor obtained in the food effect experiment (*see* below).

Results

Effects of body weight and stage on respiration and excretion rates—Respiration rates increased potentially with increasing body weight from ~ 0.015 μL O_2 ind.$^{-1}$ d^{-1} in early nauplii to ~ 0.07 μL O_2 ind.$^{-1}$ d^{-1} in copepodites (Fig. 3A). Under similar conditions (at 20°C and without food), weight-specific respiration rates declined with increasing body weight and stage from ~ 0.29 to 0.18 d^{-1} (Table 2). Ammonium and phosphate excretion rates (nmol ind.$^{-1}$ d^{-1}) were potentially related to body weight (Fig. 3B,C). In contrast to weight-specific respiration rates, weight-specific excretion rates did not follow a clear pattern in relation to body weight or stage (Table 2). Under similar conditions (at 20°C, without food, and incubation time > 20 h), the C : N and C : P metabolic ratios tended to decrease with increasing body weight and stage; in contrast, N : P ratios were positively related to body weight and stage, increasing from 9 to 21 (Table 2).

Effect of temperature on respiration and excretion rates—Respiration rates of nauplii NIII–NV (54.8 ng C ind.$^{-1}$) followed an exponential increase as a function of temperature (Fig. 4A), except for the experiments at 16°C, which rendered lower values (*see* Discussion). Weight-specific respiration rates increased from 0.10 d^{-1} at 16°C to 0.35 d^{-1} at 28°C (Table 2). The calculated Q_{10} values for respiration decreased as increasing temperature (Q_{10} [16–19.3°C] = 6.5; Q_{10} [19.3–24°C] = 2.0; Q_{10} [24–28°C] = 1.7)

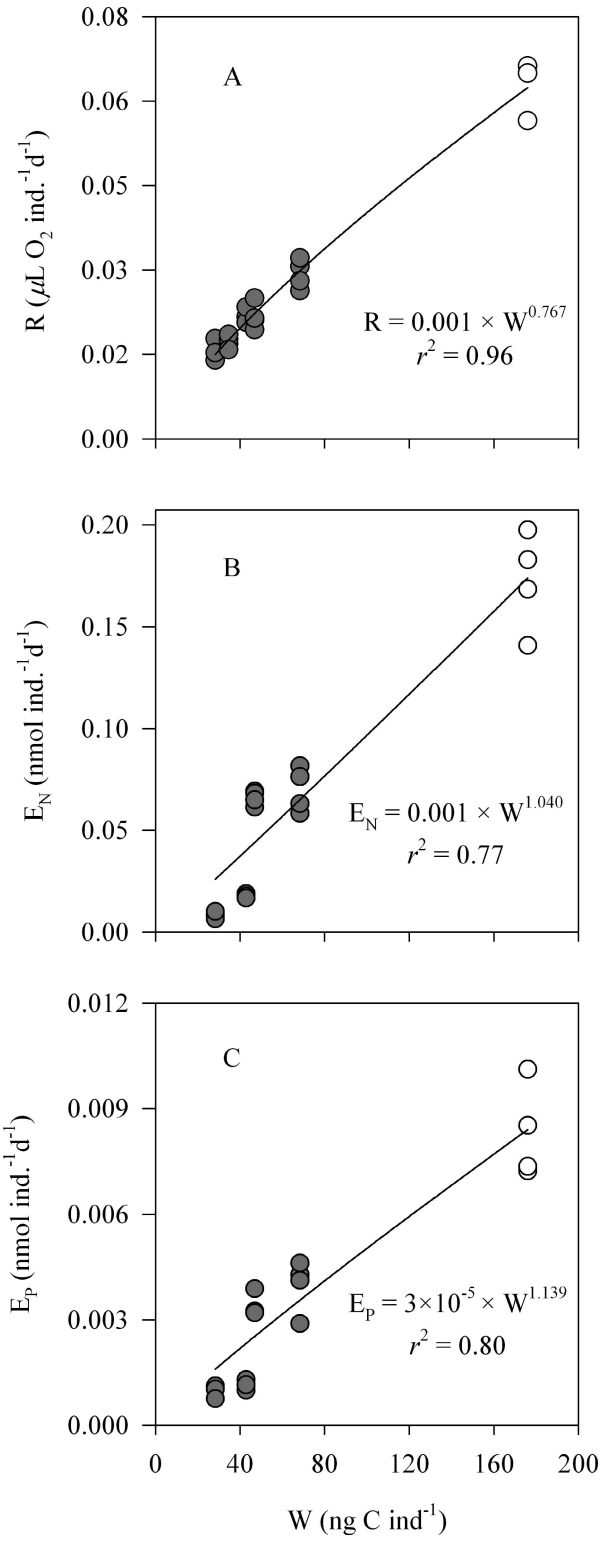

Fig. 3. Relationships between carbon body weight and (A) respiration rate, (B) ammonium excretion rate, and (C) phosphate excretion rate for *Oithona davisae* larval developmental stages incubated in filtered seawater at 20°C. Filled circles correspond to naupliar stages and open circles to copepodites. The continuous lines correspond to the allometric functions fitted to the data. W is the body weight (ng C ind.$^{-1}$), R is the respiration rate (μL O_2 ind.$^{-1}$ d^{-1}), E_N is the ammonium excretion rate (nmol NH_4-N ind.$^{-1}$d^{-1}), E_P is the phosphate excretion rate (nmol PO_4-P ind.$^{-1}$ d^{-1}), and r^2 is the coefficient of determination.

and rendered a value of 2.64 considering the total range of temperature (16–28°C). Both ammonium and phosphate excretion rates were fitted to an exponential model in the temperature range of 19.3°C to 28°C (Fig. 4B,C); as for the respiration rates, excretion at 16°C, particularly for phosphate, was much lower than expected and excluded from the fitting. Weight-specific ammonium and phosphate excretion rates increased from 0.0011 to 0.0094 μmol NH_4-N μmol C^{-1} d^{-1} and from 0.00010 to 0.00038 μmol PO_4-P μmol C^{-1} d^{-1} at, respectively, 16°C and 28°C. The Q_{10} for ammonium excretion decreased with increasing temperature (Q_{10} [16–19.3°C] = 70.8; Q_{10} [19.3–24°C] = 3.6; Q_{10} [24–28 °C] = 1.7). The Q_{10} for phosphate excretion were close to 1 at high temperatures (Q_{10} [16–19.3°C] = 34.8; Q_{10} [19.3–24°C] = 1.1; Q_{10} [24–28 °C] = 1.2). The C : N metabolic ratio was significantly higher at 16°C (ANOVA, Tukey test, $F_{3,8}$ = 27.9, $p < 0.01$), whereas no significant differences were found among metabolic C : N ratios at 19.3°C, 24.5°C, and 28°C. The N : P metabolic ratio increased with increasing temperature and C : P metabolic ratio did not follow any clear pattern with temperature (Table 2).

Effect of food availability on respiration and excretion rates—The average specific respiration rates of *Ox. marina* at 20°C was 0.27 d^{-1}, whereas specific ammonium and phosphate excretion rates of *Ox. marina* were 2.37 and 0.69 nmol μg C^{-1} d^{-1}, respectively. These values were used as described in the Methods section to correct the values obtained in incubations with feeding copepods. Feeding activity significantly increased the respiration rates of *O. davisae* nauplii by a factor of 2.3 in comparison with those estimated without food (ANOVA, $F_{1,6}$ = 11.5, $p < 0.05$, Fig. 5A; Table 2). Similarly, the ammonium and phosphate excretion rates increased by a factor of 1.4 and 11.0, respectively (ANOVA, $F_{1,6}$ = 19.5, $p < 0.01$ and $F_{1,6}$ = 75.0, $p < 0.01$, respectively; Fig. 5B,C; Table 2). The C : N metabolic ratio for feeding nauplii was significantly higher than for those without food (ANOVA, $F_{1,6}$ = 6.6, $p < 0.05$; Table 2); on the contrary, N : P and C : P metabolic ratios were significantly lower for fed than for unfed animals (ANOVA, $F_{1,6}$ = 621.0, $p < 0.01$ and $F_{1,6}$ = 163.8, $p < 0.01$, respectively; Table 2).

Starvation effects were evident in experimental treatments without food because the decrease in oxygen concentration was not constant along the incubation time and depended on temperature (Fig. 2A,B). On the contrary, the rate of oxygen consumption corresponding to nauplii feeding on *Ox. marina* was nearly constant (Fig. 2C). Excretion rates of unfed nauplii were negatively related to incubation time (Table 2). Ammonium and phosphate excretion rates at 20°C decreased from 17 to ~ 3 nmol NH_4-N μmol C_{Zoo}^{-1} d^{-1}, and from ~ 0.86 to 0.41 nmol PO_4-P μmol C_{Zoo}^{-1} d^{-1}, respectively, with increasing incubation time from 16 to 25 h (Table 2).

Table 2. Average values of weight-specific respiration rates (R_C, μmol C μmol C_{Zoo}^{-1} d^{-1}), ammonia excretion rates (E_N, μmol NH_4-N μmol C_{Zoo}^{-1} d^{-1}), phosphate excretion rates (E_P, μmol PO_4-P μmol C_{Zoo}^{-1} d^{-1}), and metabolic quotients (C : N, N : P, C : P, by atoms) of different larval stages of *Oithona davisae*. W, carbon body weight (ng C $ind.^{-1}$); T, temperature of incubation (°C); t, incubation time (h); SE, standard error. All metabolic rates are from incubations conducted with filtered seawater, with the exception of those values indicated with an asterisk that were carried out with a suspension of *Oxyrrhis marina* as food (*see* Methods, Food effect experiment).

Experiment	W	Stage	T	t	R_C (±SE)	E_N (±SE)	E_P (±SE)	C : N (±SE)	N : P (±SE)	C : P (±SE)
Body weight effect	28.3	NI	20.0	25.3	0.288(0.021)	0.00352(0.00045)	0.00041(0.00005)	84(9)	9(2)	718(86)
	34.7	NI–NII	20.0	20.3	0.258(0.008)	—	—	—	—	—
	42.9	NII–NIII	20.0	23.3	0.265(0.009)	0.00498(0.00018)	0.00032(0.00002)	53(3)	16(1)	832(55)
	68.4	NIV–NV	20.0	21.4	0.222(0.008)	0.01226(0.00094)	0.00070(0.00009)	19(1)	18(1)	328(39)
	176.0	CII–CIII	20.0	20.1	0.185(0.008)	0.01176(0.00084)	0.00057(0.00005)	17(2)	21(0.5)	320(41)
Temperature effect	54.8	NIII–NIV	16.0	20.2	0.109(0.011)	0.00110(0.00013)	0.00010(0.00002)	101(11)	11(1)	1108(176)
	54.8	NIII–NIV	19.3	20.0	0.201(0.007)	0.00467(0.00014)	0.00033(0.00002)	43(2)	14(0.3)	604(22)
	54.8	NIII–NIV	24.5	19.8	0.290(0.011)	0.00775(0.00044)	0.00035(0.00002)	38(4)	22(0.4)	845(74)
	54.8	NIII–NIV	28.0	19.6	0.349(0.013)	0.00945(0.00036)	0.00038(0.00002)	37(3)	25(0.3)	938(77)
Food effect	46.9	NII–NIII	20.0	16.2	0.240(0.013)	0.01685(0.00045)	0.00086(0.00004)	14(0.5)	20(1)	278(6)
	46.9	NII–NIII	20.0	16.2	*0.553(0.090)	*0.02418(0.00160)	*0.00955(0.00100)	23(3)	3(0.3)	62(16)

The ingestion and clearance rates (avg. ± SE) of nauplii NII–NIII (46.9 ng C $ind.^{-1}$) were 243 ± 10 cells $ind.^{-1}$ d^{-1} and 0.023 ± 0.001 mL $ind.^{-1}$ d^{-1}, respectively. Specific ingestion and growth rates (d^{-1}) were 1.09 ± 0.05 d^{-1} and 0.22 ± 0.03 d^{-1}, respectively (Table 3).

Metabolic balance: assimilation and net growth efficiencies—For the calculation of AE and NGE, carbon-specific respiration rates under food saturation conditions (R_C*, μg C μg C^{-1} d^{-1}) were estimated as follows:

$$R_C^* = R_C \times 2.3 \quad (5)$$

where R_C is the carbon-specific respiration rates measured with filtered seawater (μg C μg C^{-1} d^{-1}) and 2.3 is the correction factor obtained in the food effect experiment. AE ranged from 65% to 86% and NGE varied from 23% to 32% depending on body weight, stage, and temperature (Table 3). The AE was higher in early nauplii (NI–NIII) than in later stages (NIV–CIII), and the NGE decreased with increasing body size and stage (Table 3). AE tended to increase with increasing temperature (Table 3). With the exception of the value at 16°C, the NGE kept nearly constant as related to temperature (Table 3). Figure 6 shows the carbon budget of nauplii in the food effect experiment where ingestion, growth, and respiration rates were measured simultaneously under food saturation conditions. According to this carbon budget, the carbon losses corresponding to egestion and sloping feeding would represent ~ 15 ng C d^{-1} (~ 28% of ingested C, Fig. 6).

Discussion

Effect of body weight and stage—Metabolic rates seem to follow general scaling laws (Peters 1983), and examples for allometric scaling in biological systems can be found from the cell level to the ecosystem level (West and Brown 2005). The question about the exact value of the power exponent was reopened in recent years and nowadays metabolic rates in many ectotherms are widely accepted to follow a three-quarters power law to body mass (power exponent = 0.75, Peters 1983; West and Brown 2005). Similar relationships between metabolic rates and body weight have been also found in copepods (Ikeda et al. 2001). However, these relationships commonly exclude naupliar developmental stages (Ikeda 1985; Ikeda et al. 2001). In the case of *Oithona davisae*, respiration rates of their developmental stages conform to the general three-quarters power law scaling to body mass (Fig. 3A, power exponent = 0.77). Surprisingly, excretion rates as a function of body weight in our experiments showed a power exponent ≥ 1, probably due to the very low rates recorded for the smallest nauplii. We cannot discard possible artifacts to explain such low values, for instance a strong reduction of excretion rates after 24 h of starvation.

The metabolic quotients depend on the metabolic substrate of the animal (proteins, carbohydrates, or lipids; reviewed in Omori and Ikeda 1984). According to Ikeda (1974), an O : N metabolic ratio of 24 (C : N = 12) indicates that protein and lipid are metabolized in equal quantities at

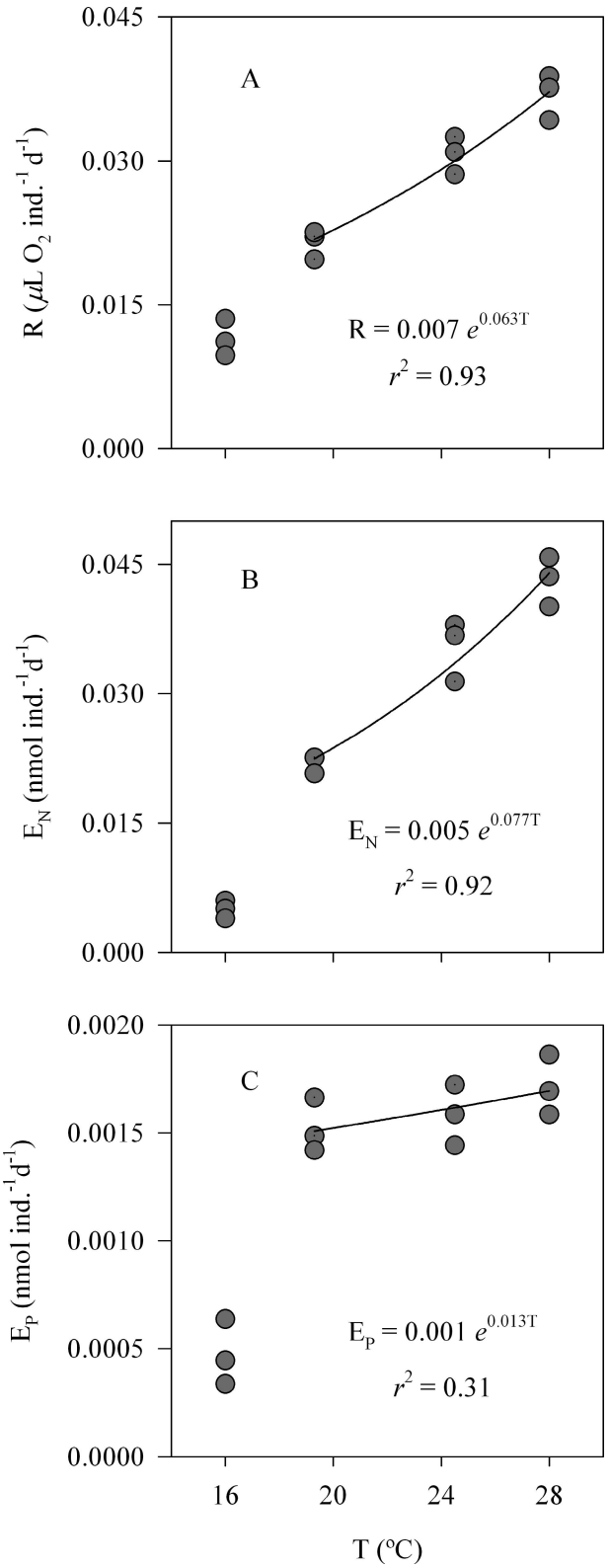

Fig. 4. Relationships between temperature and (A) respiration rates, (B) ammonium excretion rate, and (C) phosphate excretion rate for *Oithona davisae* nauplii (NIII–NIV) incubated in filtered seawater. The continuous lines correspond to the fitted exponential functions for temperatures between 19.3°C and 28°C. T is the temperature (°C), R is the respiration rate (μL O_2 ind.$^{-1}$ d^{-1}), E_N is the ammonium excretion rate (nmol NH_4-N ind.$^{-1}$ d^{-1}), E_P is the phosphate excretion rate (nmol PO_4-P ind.$^{-1}$ d^{-1}), and r^2 is coefficient of correlation.

the same time, whereas an O : N metabolic ratio < 24 (C : N < 12) indicates protein-oriented metabolism, and a ratio > 24 (C : N > 12) indicates lipid-oriented metabolism (Omori and Ikeda 1984). The C : N metabolic ratios of *O. davisae* developmental stages were higher than 12 (range 14–101, Table 2), hence indicating a lipid-based metabolism. The decrease in C : N metabolic ratios during the development of *O. davisae* may be a consequence of the differences in biochemical composition between stages. Early copepod nauplii usually have significant amounts of lipid reserves from yolk. Almeda et al. (2010*a*) provided evidence that C : N composition ratio of *O. davisae* declined along their development, probably because early nauplii exhibit higher lipid reserves (remains of yolk) than later stages. This C : N composition pattern is partly reflected by the decrease in C : N metabolic ratios along the development (Table 2, body size effect experiment). The low N : P ratios are also characteristic of a lipid-based metabolism (Ikeda 1977; Omori and Ikeda 1984). However, differences on incubation time and starvation effects between experiments could mask some of these effects, as indicated by the relatively low C : N metabolic ratios obtained in short incubations (~ 16 h, Table 2).

The effects of body size on the AE and NGE of zooplankton are unclear in the literature and different patterns have been reported depending on the species (Conover 1966, 1978) and developmental stage (Vidal 1980). Ontogenetic differences in AE and NGE of copepods may reflect differences in size-specific rates of anabolic and catabolic processes (Vidal 1980). At a given temperature (20°C) and similar conditions (body weight effect, Table 3), the NGE of *O. davisae* larvae appeared inversely related to body weight following the trend observed in other copepods (Vidal 1980) and confirming the global pattern of NGE of marine copepods reported by Ikeda et al. (2001).

Effect of temperature on respiration and excretion rates—For poikilotherms like copepods one would expect metabolic rates to be higher at increasing temperature because of the dependence of biochemical kinetics on temperature (Arrhenius law). In our experiments respiration rates of *O. davisae* nauplii followed that law, in agreement with previous observations for calanoids (Ikeda et al. 2001) and adult stages of other oithonids (Castellani et al. 2005). This contrasts with Hiromi et al. (1988), who found no effect of temperature on respiration activity of adult stages of *O. davisae* over a wide temperature range (5–30°C). Nevertheless, some copepods, mainly estuarine species, have been reported to exhibit the ability of dampening changes in their metabolic rates (homeostasis) in spite of temperature changes (low Q_{10}) as an adaptation to rapid temperature fluctuations (Gaudy et al. 2000).

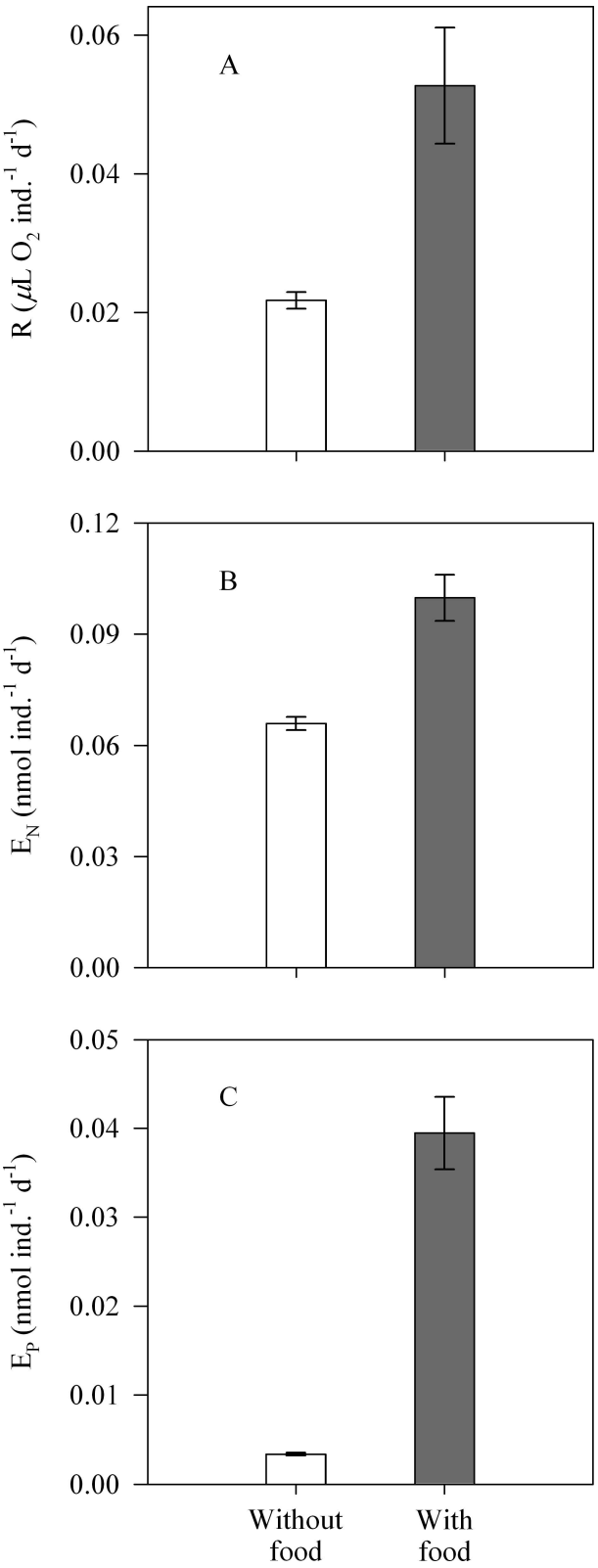

Fig. 5. Effect of food concentration on (A) respiration rates, (B) ammonium excretion rates, and (C) phosphate excretion rates of *Oithona davisae* nauplii (NII–NIII) incubated at 20°C. R is the respiration rate (μL O_2 ind.$^{-1}$ d^{-1}), E_N is the ammonium excretion rate (nmol NH_4-N ind.$^{-1}$ d^{-1}), and E_P is the phosphate excretion rate (nmol PO_4-P ind.$^{-1}$ d^{-1}). The columns represent the mean value of four replicates and the error bars the corresponding standard error (SE).

We have noticed, however, that in our experiments, the lowest temperature (16°C) rendered very low metabolic rates, likely reflecting the thermophilic character of this species (Uye and Sano 1995). *O. davisae* is a perennial species but its population density varies remarkably with season. This species is very scarce during winter and spring (when temperatures are < 20°C), whereas it is very abundant during the warm seasons, with temperatures between 20°C and 28°C. Hence, 16°C might be considered as suboptimal temperature for this species. With the exception of our lowest experimental temperature, the Q_{10} values obtained for respiration and ammonium excretion (~ 2.5) are within the range reported for other copepods (Ikeda et al. 2001; Castellani et al. 2005). Phosphate excretion rates showed a less clear pattern. Because the main component of yolk-sack lipovitellin of crustacean is phosphatidylcholine (Lee et al. 2006), the exhaustion of this reserve under starvation conditions and at higher temperatures may result in low phosphate excretion rates. The effect of temperature on the metabolic ratios of *O. davisae* also reflected effects of starvation. The C : N metabolic ratios decreased and the N : P increased as a function of temperature, indicating a protein-oriented metabolism at higher temperature, the result of a higher degree of starvation after the exhaustion of lipid reserves (Mayzaud and Conover 1988).

The effects of temperature on assimilation efficiencies of zooplankton are controversial in the literature, with reports of both positive and negative as well as the lack of effects (Conover 1966; Chervin 1978). According to the global pattern of NGE of epipelagic marine copepods reported by Ikeda et al. (2001), the NGE is expected to decrease with increasing temperature. However, for a given species, the effects of temperature on NGE may differ depending on the developmental stage (Vidal 1980) and the range of thermal tolerance of species (Iguchi and Ikeda 2005). For *O. davisae* nauplii, with the exception of the value at 16°C, the NGE kept nearly constant as related to temperature. According to this, the allocation of assimilated materials of *O. davisae* nauplii did not vary with a change in temperature, reflecting similar temperature dependence between physiological processes. It is supported by previous results showing that ingestion and growth rates (Almeda et al. 2010*b*) have quite similar Q_{10} values than respiration rates at similar temperature range.

Effect of food availability—Body size and temperature are considered to be the main factors influencing zooplankton metabolic rates. In their review, Ikeda et al. (2001) concluded that body weight and temperature explained 93–96% of the variance in respiration rates and $\sim$ 84% and 87% of the ammonium and phosphate excretion rates. However, it is important to note that most of the previous measurements of metabolic rates of

Table 3. Assimilation efficiencies (AE, %) and net growth efficiencies (NGE, %) of *Oithona davisae* developmental stages. W, carbon body weight; T, temperature (°C); I, carbon-specific ingestion rates (μg C μg C^{-1} d^{-1}); G, carbon-specific growth rates (μg C μg C^{-1} d^{-1}); R_C, specific respiration rates with filtered seawater (μg C μg C^{-1} d^{-1}); R_C*, specific respiration rates under food satiating concentrations (μg C μg C^{-1} d^{-1}). I and G were calculated from Almeda et al. (2010*b*) except those from the "Food effect experiment" that were estimated from this study. Note that AE and NGE calculations were based on R_C* (*see* Methods).

Experiment	W	Stage	T	I	G	R_C	R_C*	AE	NGE
Body weight effect	28.3	NI	20.0	1.13	0.31	0.29	0.67	86	32
	34.7	NI–NII	20.0	1.08	0.28	0.26	0.60	81	32
	42.9	NII–NIII	20.0	1.03	0.25	0.27	0.62	85	29
	68.4	NIV–NV	20.0	0.92	0.20	0.22	0.51	77	28
	176.0	CII–CIII	20.0	0.74	0.13	0.19	0.44	77	23
Temperature effect	54.8	NIII–NIV	16.0	0.53	0.09	0.11	0.25	65	26
	54.8	NIII–NIV	19.3	0.87	0.19	0.20	0.46	75	29
	54.8	NIII–NIV	24.5	1.29	0.29	0.29	0.67	74	30
	54.8	NIII–NIV	28.0	1.36	0.35	0.35	0.81	85	30
Food effect	46.9	NII–NIII	20.0	1.09	0.23	0.24	0.55	72	29

copepods were obtained in the absence of food. Food availability appears to be the main factor driving copepod feeding rates in the field (Saiz and Calbet 2007) and, consequently, it must influence their metabolism. An increase in metabolic rates in fed animals has been previously reported for calanoid copepods (Kiørboe et al. 1985). In the case of oithonids, respiration rates of fed adult stages of *O. davisae* were between 1.4 and 2.8 times higher than for starved animals (Nakata and Nakane 1987; Hiromi 1994) in clear agreement with our results for naupliar stages. The observed increase of respiration rates in association with feeding activity is commonly referred to as "specific dynamic action" (SDA) and it is attributed to the cost of biosynthesis of new tissue from ingested food (Kiørboe et al. 1985). However, besides SDA, the increase on swimming and feeding activity under the presence of food may result in an increase of energetic expenditure (Paffenhöfer 1993, 2006). The respiration rates under starved conditions have been considered somewhere between basal and routine metabolism (Prosser 1973; Ikeda et al. 2001), whereas the respiration rates in the presence of food may be closer to active metabolism. The use of filtered seawater simplifies the experiments from a technical viewpoint, but it may result in an important underestimation of copepod metabolic rates when extrapolated to field conditions. Moreover, not only the presence–absence of food is relevant for the correct estimation of the respiration rates of copepods. The quantity and quality of food are also important factors (Conover 1966; Ikeda 1977) and, therefore, it should be considered for a better understanding of copepod metabolism in nature. Similarly to respiration rates, zooplankton excretion rates are positively affected by the presence of food (Takahashi and Ikeda 1975; Ikeda 1977). In *O. davisae*, the increase in ammonium excretion of fed nauplii was lower than previous reports for other zooplankton (factors of ~ 2 to 6, Takahashi and Ikeda 1975). However, the increase on phosphate excretion of fed nauplii was particularly high and it may be an artifact produced by the loss and fragmentation of food during the ingestion (sloppy feeding, Saba et al. 2009).

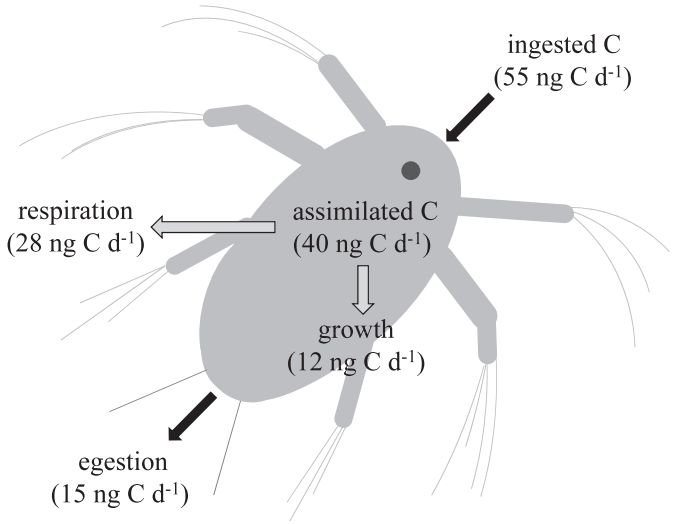

Fig. 6. Carbon budget of *Oithona davisae* nauplii (NII–NIII) at 20°C and under saturating food conditions (*Oxyrrhis marina*). Egestion was calculated as the difference between ingested and assimilated carbon biomass. Carbon budget was calculated using the average carbon biomass of nauplii during the incubation (50.7 ng C ind.$^{-1}$).

Laboratory experiments under controlled conditions are a fundamental tool for understanding the effects of environmental variables on activity of marine zooplankton. Nonetheless, caution is required when extrapolating laboratory results to the field (e.g., crowding effects, lack of turbulence, etc.). Long incubation time in experiments with filtered seawater may result in starvation effects that translate into a progressive decrease in the metabolic rates of copepods (Mayzaud 1973, 1976; Ikeda 1977). In the case of *O. davisae* nauplii, starvation effects were evident in all experiments with filtered seawater since the decrease in oxygen consumption was nonlinear during the incubation (Fig. 5), in agreement with previous reports for other zooplankton (Mayzaud 1976; Kiørboe et al. 1985). The observed changes in respiration rates were temperature dependent and may be related to changes in metabolic substrates under starving conditions (Conover 1978).

Although some authors reported higher metabolic rates at the beginning of incubations as likely artifacts due to handling stress, our data show that oxygen consumption of fed nauplii was linear along the incubation. The respiration rates of marine invertebrates are also affected by the concentration of dissolved oxygen in the water (Prosser 1973). For some copepods, a rapid decrease in the respiration rates is observed when the O_2 concentration decreases to below a critical point (Marshall 1973). In the case of fed *O. davisae* nauplii, we observed a linear decrease in oxygen concentration until 60% (Fig. 2C), indicating that the changes of unfed nauplii respiration rates above this concentration are related to starvation and not to the decrease in oxygen concentration. The possibility of detecting changes in the rate of oxygen consumption during the incubation, either as a consequence of starvation or by a limitation in the substrate (oxygen concentration), is a clear advantage of continuous measurement systems (as optodes) as compared to the classical Winkler bottle method.

Excretion rates of zooplankton decrease along the incubation time under starvation conditions (Mayzaud 1973), in agreement with our observations for *O. davisae* nauplii. As mentioned above, excretion rates under starvation will depend on the biochemical composition of the organism, those with high lipid contents being less susceptible to starvation in short incubation time (Mayzaud 1976). We were not able to correct the starvation effects on excretion rates due to the lack of continuous monitoring of ammonium and phosphate concentrations that could allow establishing the relationships between excretion rates and incubation time.

The increase in C : N metabolic rates observed for fed *O. davisae* nauplii indicates a slightly higher lipid catabolism, likely due to food used in our experiments (*Ox. marina*) that has a high content and high capacity of producing essential lipids that are efficiently supplied to copepods by trophic transfer (Veloza et al. 2006). However, the low N : P and C : P ratio when food was available may be the result of the overestimation of phosphate excretion rates mentioned above.

The AE and NGEs of zooplankton vary depending on food concentration and food quality (Paffenhöfer and Köster 2005). The use of respiration rates in filtered seawater for calculating metabolic balance may result in important bias on the estimation of AE and NGE. For *O. davisae* developmental stages, the AE and NGE calculated using respiration rates in filtered seawater would range from 38% to 53% and from 41% to 52%, respectively. Therefore, AE and NGE estimated using the respiration rates of nauplii with food were 1.7 times higher and 0.6 times lower, respectively, than those calculated using respiration rates of nauplii without food.

Metabolic requirements and NGEs of O. davisae *early developmental stages compared with other copepods—Oithona* is able to grow well both in low and high food concentration environments (Calbet and Agustí 1999), whereas calanoid broadcasters are frequently dominant only in high productive areas or seasonally during the phytoplankton blooms (Peterson 1998). Besides other factors, differences in thermal tolerance, prey preferences, and sensory systems efficiency may explain the differences in distribution and abundance among *Oithona* species. However, it has been proposed that the common feature that explains the success of oithonids in marine environments is their low metabolic requirements compared with calanoids (Lampitt and Gamble 1982; Paffenhöfer 1993; Castellani et al. 2005). Although it is difficult to compare respiration rates between naupliar stages of oithonids and calanoids due to the scarcity of data (Klekowski et al. 1977; Köster et al. 2008), they appear to be significantly lower. When respiration rates (R) of nauplii of the calanoid copepod *Eucalanus pileatus* ($R = 0.297\ d^{-1}$, body weight [W] = 2 μg C ind.$^{-1}$, Köster et al. 2008), using the same methodology and under fed conditions, are corrected for size ($R/W^{0.75}$) and temperature ($Q_{10} = 2.5$), specific respiration rates of *O. davisae* happen to be > 5 times lower than those for *E. pileatus* nauplii. In addition, respiration rates of *O. davisae* nauplii (at 20°C) were on average ~ 2.2 times lower than the values predicted by the equation provided by Ikeda et al. (2001) for determining respiration rates of calanoid copepods (adults and copepodites) as a function of body weight and temperature. This is in accordance with the fact that *O. davisae* nauplii, as well as adult stages of other *Oithona* spp., have much lower ingestion and growth rates than similarly size copepods (Lampitt 1978; Paffenhöfer 1993; Saiz and Calbet 2007). These differences between the metabolic rates of *Oithona* and calanoid copepods are not accompanied by differences in the AE and NGE. The majority of AE values reported here for *O. davisae* developmental stages fall within the range of common values observed in calanoid copepods feeding on heterotrophic prey (between 70% and 90%, Conover 1978). Similarly, NGE of *O. davisae* larvae were within the range of the common values reported for adult stages of calanoid copepods (21–54%, Ikeda et al. 2001). Therefore, *O. davisae* developmental stages exhibit quite similar food conversion efficiency compared to adult calanoid copepod and copepodites.

A likely explanation about the reasons of the low metabolic requirements of *Oithona* as compared to calanoids could be the differences in swimming and feeding behavior (Paffenhöfer 1993). In contrast to most calanoids, oithonids (nauplii and adults) move with occasional leaps and rely on detecting prey by hydromechanical signals (ambush feeders, Paffenhöfer 1993; Svensen and Kiørboe 2000; Paffenhöfer and Mazzocchi 2002). As an example, in the absence of prey, *Oithona davisae* nauplii spent 98% of their time sinking, unmotile, and only under the presence of motile prey increase the jumping frequency (Henriksen et al. 2007). This feeding and swimming strategy of *Oithona* is hypothesized to be more energetic–efficient than that of most calanoids (Paffenhöfer 1993).

In summary, two important conclusions can be highlighted from this work. First, as a general methodological aspect in zooplankton physiology, the importance of measuring respiration rates with food. Respiration rates without food result in a significant underestimation of copepod metabolic rates and, in addition, the use of respiration rates measured in filtered seawater led to

important errors in the estimations of assimilation and net growth efficiencies. Therefore, measurements of zooplankton respiration rates with food are required for a correct evaluation of the importance of metazoan zooplankton in marine carbon fluxes. Second, particularly for the studied copepod genus, our study shows that the AEs and growth efficiencies of *Oithona* nauplii are similar to those exhibited by similarly sized calanoids, whereas their specific respiratory losses are comparatively lower; our results, therefore, support the hypothesis that the wide distribution and high degree of ecological success of *Oithona* in marine ecosystems is in part explained by their low metabolic losses.

Acknowledgments

We thank E. Velasco and P. Jiménez for their help in maintaining the algae and copepod cultures. This work was funded by the Spanish Ministry of Science and Innovation (MICINN) through a Ph.D. fellowship to R.A. (BES-2005-7491) and the research projects CTM2004-02775 and Intramural-200630I226 to A.C., CTM2007-60052 to E.S., and CTM2006-12344 to M.A.

References

Almeda, R., M. Alcaraz, A. Calbet, L. Yebra, and E. Saiz. 2010*a*. Effect of temperature and food concentration on survival, development and growth rates of naupliar stages of *Oithona davisae* (Copepoda, Cyclopoida). Mar. Ecol. Prog. Ser. **410:** 97–109, doi:10.3354/meps08625

———, C. B. Augustin, M. Alcaraz, A. Calbet, and E. Saiz. 2010*b*. Feeding rates and gross growth efficiencies of larval developmental stages of *Oithona davisae* (Copepoda, Cyclopoida). J. Exp. Mar. Biol. Ecol. **387:** 24–35, doi:10.1016/j.jembe.2010.03.002

Calbet, A., and S. Agustí. 1999. Latitudinal changes of copepod egg production rates in Atlantic waters: Temperature and food availability as the main driving factors. Mar. Ecol. Prog. Ser. **181:** 155–162, doi:10.3354/meps181155

———, S. Garrido, E. Saiz, A. Alcaraz, and C. M. Duarte. 2001. Annual zooplankton succession in coastal NW Mediterranean waters: The importance of the smaller size fractions. J. Plankton Res. **23:** 319–331, doi:10.1093/plankt/23.3.319

Castellani, C., C. Robinson, T. Smith, and R. S. Lampitt. 2005. Temperature affects respiration rate of *Oithona similis*. Mar. Ecol. Prog. Ser. **285:** 129–135, doi:10.3354/meps285129

Castonguay, M., S. Plourde, D. Robert, J. A. Runge, and L. Fortier. 2008. Copepod production drives recruitment in a marine fish. Can. J. Fish. Aquat. Sci. **65:** 1528–1531, doi:10.1139/F08-126

Chervin, M. B. 1978. Assimilation of particulate organic carbon by estuarine and coastal copepods. Mar. Biol. **49:** 265–275, doi:10.1007/BF00391139

Conover, R. J. 1966. Assimilation of organic matter by zooplankton. Limnol. Oceanogr. **11:** 338–345, doi:10.4319/lo.1966.11.3.0338

———. 1978. Transformation of organic matter, p. 221–500. *In* O. Kinne [ed.], Marine ecology, v. 4. Wiley and Sons.

Frost, B. W. 1972. Effects of size and concentration of food particles on the feeding behaviour of the marine planktonic copepod *Calanus pacificus*. Limnol. Oceanogr. **17:** 805–815, doi:10.4319/lo.1972.17.6.0805

Fryer, G. 1986. Structure, function and behaviour, and the elucidation of evolution in copepods and other crustaceans. Syllogeus **58:** 150–157.

Gallienne, C. P., and D. B. Robins. 2001. Is *Oithona* the most important copepod in the world's oceans? J. Plankton Res. **23:** 1421–1432, doi:10.1093/plankt/23.12.1421

Gaudy, R., G. Cervetto, and M. Pagano. 2000. Comparison of the metabolism of *Acartia clause* and *A. tonsa*: Influence of temperature and salinity. J. Exp. Mar. Biol. Ecol. **247:** 51–63, doi:10.1016/S0022-0981(00)00139-8

Hansen, H. P., and F. Koroleff. 1999. Determination of nutrients, p. 159–228. *In* K. Grasshoff, K. Kremling, and M. Ehrhardt [eds.], Methods of seawater analysis. Wiley-VCH.

Henriksen, C. I., E. Saiz, A. Calbet, and B. W. Hansen. 2007. Feeding activity and swimming patterns of *Acartia grani* and *Oithona davisae* nauplii in the presence of motile and non-motile prey. Mar. Ecol. Prog. Ser. **331:** 119–129, doi:10.3354/meps331119

Hiromi, J. 1994. Further studies on respiration of the small planktonic copepod *Oithona davisae* with special reference to the effect of feeding. Bull. Coll. Agric. Vet. Med. Nihon Univ. **51:** 149–153.

———, T. Nagata, and S. Kadota. 1988. Respiration of the small planktonic copepod *Oithona davisae* at different temperatures. Bull. Plankton Soc. Jpn. **35:** 143–148.

Holst, G., R. N. Glud, M. Kühl, and I. Klimant. 1997. A microoptode array for fine-scale measurement of oxygen distribution. Sens. Actuators B Chem. **38–39:** 122–129, doi:10.1016/S0925-4005(97)80181-5

Iguchi, N., and T. Ikeda. 2005. Effects of temperature on metabolism, growth and growth efficiency of *Thysanoessa longipes* (Crustacea:Euphausiacea) in the Japan Sea. J. Plankton Res. **27:** 1–10, doi:10.1093/plankt/fbh146

Ikeda, T. 1974. Nutritional ecology of marine zooplankton. Mem. Fac. Fish. Hokkaido Univ. **22:** 1–97.

———. 1977. The effect of laboratory conditions on the extrapolation of experimental measurements to the ecology of marine zooplankton. IV. Changes in respiration and excretion rates of boreal zooplankton species maintained under fed and starved conditions. Mar. Biol. **41:** 241–252, doi:10.1007/BF00394910

———. 1985. Metabolic rates of epipelagic marine zooplankton as a function of body mass and temperature. Mar. Biol. **85:** 1–11, doi:10.1007/BF00396409

———, Y. Kanno, K. Ozaki, and A. Shinada. 2001. Metabolic rates of epipelagic marine copepods as a function of body mass and temperature. Mar. Biol. **139:** 587–596.

Kiørboe, T., F. Mohlenberg, and K. Hamburger. 1985. Bioenergetics of the planktonic copepod *Acartia tonsa*: Relation between feeding, egg production and respiration, and composition of specific dynamic action. Mar. Ecol. Prog. Ser. **26:** 85–97, doi:10.3354/meps026085

Klekowski, R. Z., I. V. Kukina, and N. I. Tumanseva. 1977. Respiration in the microzooplankton of the equatorial upwellings in the eastern Pacific Ocean. Pol. Arch. Hydrobiol. (suppl.) **24:** 467–489.

Köster, M., C. Krause, and G. A. Paffenhöfer. 2008. Time-series measurements of oxygen consumption of copepod nauplii. Mar. Ecol. Prog. Ser. **353:** 157–164, doi:10.3354/meps07185

Lampitt, R. S. 1978. Carnivorous feeding by a small marine copepod. Limnol. Oceanogr. **23:** 1228–1231, doi:10.4319/lo.1978.23.6.1228

———, and J. C. Gamble. 1982. Diet and respiration of the small planktonic marine copepod *Oithona nana*. Mar. Biol. **66:** 185–190, doi:10.1007/BF00397192

Last, J. M. 1980. The food of twenty species of fish larvae in the west-central North Sea. Fish. Res. Tech. Rep. 60. MAFF Directorate of Fisheries Research, Lowestoft.

Lee, R. F., W. Hagen, and G. Kattner. 2006. Lipid storage in marine zooplankton. Mar. Ecol. Prog. Ser. **307:** 273–306, doi:10.3354/meps307273

Marshall, S. M. 1973. Respiration and feeding in copepods. Adv. Mar. Biol. **11:** 57–120, doi:10.1016/S0065-2881(08)60268-0

Mayzaud, P. 1973. Respiration and nitrogen excretion of zooplankton. II. Studies of the metabolic characteristics of starved animals. Mar. Biol. **21:** 19–28, doi:10.1007/BF00351188

———. 1976. Respiration and nitrogen excretion of zooplankton: IV. The influence of starvation on the metabolism and the biochemical composition of some species. Mar. Biol. **37:** 47–58, doi:10.1007/BF00386778

———, and R. J. Conover. 1988. O : N ratio as a tool to describe zooplankton metabolism. Mar. Ecol. Prog. Ser. **45:** 289–302, doi:10.3354/meps045289

Nakata, K., and T. Nakane. 1987. Respiration of plankton in the Mikawa Bay. Pollut. Contr. **22:** 281–294.

Omori, M., and T. Ikeda. 1984. Methods in marine zooplankton ecology. John Wiley & Sons.

Paffenhöfer, G. A. 1993. On the ecology of marine cyclopoid copepods (Crustacea, Copepoda). J. Plankton Res. **15:** 37–55, doi:10.1093/plankt/15.1.37

———. 2006. Oxygen consumption in relation to motion in marine planktonic copepods. Mar. Ecol. Prog. Ser. **317:** 187–192, doi:10.3354/meps317187

———, and M. Köster. 2005. Digestion of diatoms by planktonic copepods and doliolids. Mar. Ecol. Prog. Ser. **297:** 303–310, doi:10.3354/meps297303

———, and M. G. Mazzocchi. 2002. On some aspects of the behaviour of *Oithona plumifera* (Copepoda: Cyclopoida). J. Plankton Res. **24:** 129–135, doi:10.1093/plankt/24.2.129

Pelegri, S., J. Dolan, and F. Rassoulzadegan. 1999. Use of high temperature catalytic oxidation (HTCO) to measure carbon content of microorganisms. Aquat. Microb. Ecol. **16:** 273–280, doi:10.3354/ame016273

Peters, R. H. 1983. Ecological implications of body size. Cambridge Univ. Press.

Peterson, W. 1998. Life cycle strategies of copepods in coastal upwelling zones. J. Mar. Syst. **15:** 313–326, doi:10.1016/S0924-7963(97)00082-1

Prosser, C. L. 1973. Oxygen: Respiration and metabolism, p. 165–211. *In* C. L. Prosser [ed.], Comparative animal physiology. W. B. Saunders.

Saba, G. K., D. K. Steinberg, and D. A. Bronk. 2009. Effects of diet on release of dissolved organic and inorganic nutrients by the copepod *Acartia tonsa*. Mar. Ecol. Prog. Ser. **386:** 147–161, doi:10.3354/meps08070

Saiz, E., and A. Calbet. 2007. Scaling of feeding in marine calanoid copepods. Limnol. Oceanogr. **52:** 668–675, doi:10.4319/lo.2007.52.2.0668

Svensen, C., and T. Kiørboe. 2000. Remote prey detection in *Oithona similis*: Hydromechanical versus chemical cues. J. Plankton Res. **22:** 1155–1166, doi:10.1093/plankt/22.6.1155

Takahashi, M., and T. Ikeda. 1975. Excretion of ammonia and inorganic phosphorus by *Euphausia pacifica* and *Metridia paeifiea* at different concentrations of phytoplankton. J. Fish. Res. Board Can. **32:** 2189–2195.

Uchima, M. 1979. Morphological observation of developmental stages in *Oithona brevicornis* (Copepoda, Cyclopoida). Bull. Plankton Soc. Jpn. **26:** 59–76.

Uye, S., and K. Sano. 1995. Seasonal reproductive biology of the small cyclopoid copepod *Oithona davisae* in a temperate eutrophic inlet. Mar. Ecol. Prog. Ser. **118:** 121–128, doi:10.3354/meps118121

Veloza, A. J., F. L. E. Chu, and K. W. Tang. 2006. Trophic modification of essential fatty acids by heterotrophic protists and its effects on the fatty acid composition of the copepod *Acartia tonsa.* Mar. Biol. **148:** 779–788, doi:10.1007/s00227-005-0123-1

Vidal, J. 1980. Physioecology of zooplankton. IV. Effects of phytoplankton concentration, temperature, and body size on the net production efficiency of *Calanus pacificus*. Mar. Biol. **56:** 203–211, doi:10.1007/BF00645344

West, G. B., and J. H. Brown. 2005. The origin of allometric scaling laws in biology from genomes to ecosystems: Towards a quantitative unifying theory of biological structure and organization. J. Exp. Biol. **208:** 1575–1592, doi:10.1242/jeb.01589

Associate editor: Mikhail V. Zubkov

Received: 01 June 2010
Accepted: 19 October 2010
Amended: 08 November 2010