# Dietary effects on life history traits of riverine *Bosmina*

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## SUMMARY

1. We compared growth, reproduction and life history characteristics of *Bosmina* raised on Ohio River seston versus a unialgal culture (*Scenedesmus acutus*), to assess potential nutritional constraints experienced by riverine populations.
2. *Bosmina* grew well in both treatments during their juvenile stage. Analysis of variance showed that *Bosmina* growth prior to the start of reproduction did not differ significantly between the treatments. After the onset of reproduction *Bosmina* fed on *Scenedesmus* grew faster and exhibited higher fecundity than their counterparts fed river seston.
3. Significant quantities of triacylglycerol (visible lipid droplets) were gradually accumulated in *Bosmina* fed on *Scenedesmus*. Visual lipid ovary indices were positively correlated with adult growth rate and fecundity and negatively correlated with longevity.
4. Biochemical analysis showed that *Scenedesmus* had significantly greater total $\omega3$, $\omega6$ and polyunsaturated fatty acids (PUFA) whereas river seston had more eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA). Thus, in contrast to *Daphnia*, EPA and DHA do not appear to influence *Bosmina* growth or reproductive performance, but linolenic acid or total PUFA may be important.
5. Comparatively weak performance by *Bosmina* feeding on river seston suggest that nutritional constraints may be important despite high particulate organic carbon and low C : N and C : P of river seston.

*Keywords*: cladoceran, fatty acids, Ohio River, *Scenedesmus*, stoichiometry

## Introduction

Zooplankton are common in large rivers throughout the world (Pace, Findlay & Lints, 1992; Thorp *et al.*, 1994; Pourriot & Rougier, 1997; Basu, Kalff & Pinel-Alloul, 2000) and, like their lentic and marine counterparts, serve the important function of linking energetic pathways from primary and bacterial production to higher trophic levels. Their importance in riverine food webs is exemplified by the diverse array of consumers, including native and exotic mussels and fish that derive a measurable fraction of their energetic intake through planktivory (Van Dijk & Van Zanten, 1995; Jack & Thorp, 2000; Dettmers *et al.*, 2001; Jack &

Thorp, 2002). Zooplankton production in rivers is constrained through a variety of mechanisms whose relative importance is determined by seasonal variations in temperature and discharge. During periods of elevated discharge, advective loss and feeding interference by suspended particulates limit zooplankton community development (Pace *et al.*, 1992; Basu & Pick, 1997). As river stage and discharge decline, water temperatures increase and other factors such as food limitation and predation may assume greater importance (Pourriot *et al.*, 1982; Pace *et al.*, 1992; Pourriot & Rougier, 1997; Jack & Thorp, 2002; Guelda *et al.*, in press). Despite these constraints, zooplankton are often abundant, particularly in lowland rivers, with reported densities attaining 100s $L^{-1}$ in the Orinoco and Illinois Rivers (Saunders & Lewis, 1989; Dettmers *et al.*, 2001) and 1000s $L^{-1}$ in the Hudson and St Lawrence Rivers (Pace *et al.*, 1992; Basu *et al.*, 2000).

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There is an extensive literature describing cladoceran growth and reproductive responses to variation in food resources. Most of these studies have focused on *Daphnia* and on lacustrine systems (Black, 1980; Kerfoot, 1980; Goulden, Henry & Tessier, 1982; Urabe, 1991; Schulz & Sterner, 1999). In rivers, small-bodied zooplankton, particularly *Bosmina*, are often numerically dominant (Thorp *et al.*, 1994; Basu *et al.*, 2000; Dettmers *et al.*, 2001). Although *Bosmina* is a cosmopolitan cladoceran genus, found in lakes as well as rivers, we know of no prior studies that quantify life history parameters using natural food resources. Most life history studies on cladocerans have been done using laboratory-grown unialgal cultures; very few have used natural seston (e.g. Urabe, 1991; Ripka, 1998; Spaak, Vanoverbeke & Boersma, 2000; Urabe & Sterner, 2001). *Bosmina* grows and reproduces well on green algae (Kerfoot, 1974; Hanazato & Yasuno, 1985; Urabe, 1991) but their feeding mechanism is more selective than that of *Daphnia*. Behavioural and morphological mechanisms contribute to dietary differences between these two genera (DeMott, 1982). These selective abilities and its smaller body size led DeMott (1982) to suggest that *Bosmina* was a superior competitor to *Daphnia* when food concentrations were low. These attributes may give *Bosmina* a competitive advantage in riverine environments. Selective feeders like *Bosmina* may be able to distinguish among particles of variable food quality (Acharya, Kyle & Elser, 2004a) and thus enjoy a competitive advantage particularly when high mineral turbidities interfere with the grazing of less selective feeders such as *Daphnia*.

The seston of lakes and rivers varies seasonally in elemental (Sterner, Elser & Hessen, 1992; Urabe, 1993) and biochemical (Arts, Robarts & Evans, 1997; Kreeger *et al.*, 1997) composition. This variation arises from variable contributions of allochthonous organic matter and from changes in the species composition and physiological status of the algal component (Kilham *et al.*, 1997). Seston properties shown to be important for consumers include phosphorus content (Schulz & Sterner, 1999; Acharya *et al.*, 2004a) and the abundance and composition of fatty acids (Müller-Navarra, 1993, 1995a,b; Brett & Müller-Navarra, 1997). Fatty acids (FA) that are thought to be important for zooplankton nutrition include eicosapentaenoic acid (EPA; 20:5ω3), docosahexaenoic acid (DHA; 22:6ω3) and linoleic acid (Ahlgren *et al.*, 1990; Ahlgren,

Gustafsson & Boberg, 1992; Müller-Navarra, 1995a,b; Weers, Seiwertsen & Gulati, 1997; Ahlgren, Zeipel & Gustafsson, 1998). Biochemical analysis of natural seston is very limited (Arts *et al.*, 1997; Kreeger *et al.*, 1997), and the essential FA requirements of *Bosmina* remain unknown. Therefore, the applicability of these findings to riverine food resources and zooplankton communities is poorly understood.

One of the challenges in nutritional studies is to relate laboratory-feeding experiments to growth and reproductive performance in natural environments. Growth rate is a critical parameter for most animals, as it affects age at first reproduction, adult body size (predation risk), and other aspects of life history (Arendt, 1997). However, assessment of performance at an early stage of the life cycle may be misleading especially for cladocerans because of their differential allocation to growth and reproduction after the onset of maturity. Longer experiments allow the collection of detailed information on life history traits such as development time, age and size at maturation, brood size, size of the neonates, and survivorship and can thus provide a more complete picture of performance at the individual and population level (Urabe & Sterner, 2001).

In this study, we examined the growth and life history characteristics (body size, age at maturation, fecundity, neonate size, and survival rate) of *Bosmina* isolated from the Ohio River. To our knowledge, this is the first published life history analyses for riverine *Bosmina*. Life history traits were compared between two food treatments, river seston and a unialgal culture of *Scenedesmus acutus* (Meyen). Treatment effects on short-term growth rates and long-term growth and fecundity were related to stoichiometric (C : P and C : N) and biochemical (form and abundance of fatty acids) properties of the food resources.

## Methods

### Collection and culture

*Bosmina* (*sinobosmina*) *freyi* (formerly *Bosmina longirostris* complex; O. F. Müller; DeMelo & Hebert, 1994) was collected from the Ohio River, near the midpoint of the McAlpine navigation pool (Ohio River Kilometer 933). This site is near the University of Louisville's Ohio River Experimental Station where prior work has characterised plankton communities

(Sellers & Bukaveckas, 2003; Koch, Guelda & Bukaveckas, 2004; Guelda et al., in press). Water residence time within the McAlpine Pool is <2 days during high flow (typically January to April) but may be as long as 30 days when the river is at a base flow (usually July to October; Sellers & Bukaveckas, 2003). Gravid *Bosmina* were collected in early May and kept overnight in 63 μm-filtered river water supplemented with a small amount of cultured *Scenedesmus*. The next day 60–70 neonates were isolated and grown individually in 25 mL scintillation vials. The vials were filled with fresh river water (collected every other day and filtered through 63 μm mesh) and capped to avoid trapping animals in the surface tension. Second generation neonates born from these animals at third or fourth clutches (within a 12 h period) were used for the life history experiments.

## Life history experiments

We examined the effects of two diet types (a unialgal laboratory culture and river seston particles <35 μm) on a range of life history characteristics (growth rate, body size, age at maturation, fecundity, neonate size, and longevity). For the river seston treatment, filtered river water (63 μm mesh) was poured into a large trash can and stored overnight in the dark (with aeration). Before using, the water was filtered through 35 μm mesh to remove large filaments which would not be ingested by *Bosmina* and which may cause feeding interference or toxic effects (*Anabaena* occurs in the river during summer months). For the unialgal treatment, river water was filtered to remove all seston >0.45 μm (GF/F filter) and supplemented with the green alga *S. acutus* to achieve a food density of 1.5 mg C L$^{-1}$. Previous studies by DeMott (1982, 1986), Urabe (1991) and Schulz & Sterner (1999) have shown that *Bosmina* does well in green algae such as *Scenedesmus* and *Chlamydomonous*, and these cladocerans are not known to prefer bacteria over algae (DeMott, 1982; Urabe, 1990). The *S. acutus* was grown in a chemostat on filtered (0.45 μm GF/F) and autoclaved river water modified to contain at least 1000 μM N and 50 μM P (molar N : P ratio 20) in batch cultures.

For each diet treatment, 60 neonates were isolated and transferred to 25-mL vials (one individual per vial). An additional 25 neonates (<12 h) were put aside to estimate initial body size. Food and media

were replenished every 24 h by pipette transferring of *Bosmina* from each vial. Experimental vials were maintained under 14 : 10 h light : dark cycle and a temperature of 25°C. All the experimental vials were gently rotated twice a day to re-suspend settled particulates. Every 48 h three animals from each treatment were randomly selected for image analyses and were discarded after taking the photographs. This was repeated until we ran out of the animals. For growth rate estimation, initial and final body area measurements (from image analyses) were converted to body dry mass (see below) for calculation of growth rate (μ day$^{-1}$) as:

$$\mu = \frac{\ln(\text{final weight/initial weight})}{\text{Day(s)}}$$

A separate set of six individuals per diet treatment was isolated for quantification of the reproductive output and longevity. Each day all the animals were hand transferred to new media and spent media was checked for new offspring. Once the reproduction started, five to eight neonates were pooled randomly from each clutch for image capturing at birth. Body size data were collected for 22 days, after which time few animals remained. Video images were also analysed for visual lipid-ovary index (Goulden & Henry, 1981; Tessier & Goulden, 1982).
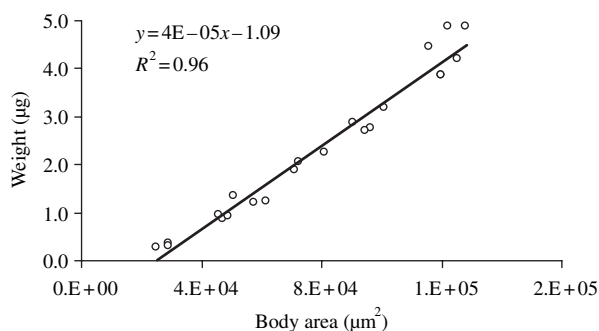
## Weight-area regression

Simultaneous with the experiments just described, a second experiment was performed to establish the relationship between body area (from image analysis) and body mass. From the large cohort of *Bosmina* neonates (<12 h old collected in a similar fashion as described above), three replicates of 50 animals each were removed for initial measurements and the rest were placed in 250 mL jars containing filtered (35 μm GF/F) Ohio River water plus *Scenedesmus* supplement. There were 20 jars in total with 30–50 animals in each jar. The body area of animals was measured by taking lateral images of each animal and then calculating its area (Acharya et al., 2004a) using Spot Advanced software Version 4.0.6 (Diagnostic Instruments Inc., Sterling Heights, MI, U.S.A.). The animals were then placed on a small (1 cm × 2 cm) 20 μm Nitex cutout which was dried in an oven at 60°C for 24 h before the weight measurement. After weighing, all the animals were removed and the Nitex was

checked under the dissecting microscope to make sure nothing was left behind. The Nitex was re-weighed immediately and the dry weight of the animals was calculated. From the remaining cohort, three replicates of 30–50 animals were sampled every 24 h for 6 days to obtain images and dry weight measurements. The animals remaining in the 250 mL jars were transferred to new food and media every 24 h by filtering each bottle through a cylinder with Nitex mesh at the bottom. During incubation, jars were gently agitated several times a day to prevent the settling of the food. The dry weight and body area measurements were used to derive a regression for estimating growth of animals used in the life history analyses (described above). Because of the large number of animals required for weight measurements, each replicate represents a group of 30–50 animals photographed individually but weighed together. Three replicate measurements from each day (0–6) yielded a total of 21 determinations. The dry weight–body area regression was linear ($P < 0.0001$ and $R^2 > 0.90$; Fig. 1).

*Sample analyses*

Algal samples and river seston samples collected on filters were dried in an oven at 60°C and held in a desiccator until analysed. Carbon (C) and nitrogen (N) content was determined using a Perkin-Elmer model 2400 elemental analyser (Perkin Elmer Inc., Wellsley, MA, U.S.A.) and P content using persulphate oxidation followed by the acid molybdate technique (APHA, 1998). Samples for fatty acids were collected on precombusted GF/F and were kept at −80 °C. Extraction and methylation were performed according to Kattner & Fricke (1986). Fatty acid composition

was analysed using a gas chromatograph (Hewlett-Packard 6890, Agilent Technologies, Palo Alto, CA, U.S.A.). Fatty acid peaks were compared with retention times of standard mixes and checked with gas chromatograph equipped with mass spectrometer. Samples for C, N, P and particulate organic carbon (POC) were collected in alternate days and fatty acid data were obtained at days 10 and 25 of the experiment.

## Results

*Food resources*

Cultured *Scenedesmus* had a biomass C : P ratio of 124.4 (±2.1) and C : N ratio of 14.8 (±2.0). C : P and C : N of the filtered river seston at the beginning of the experiments were similar to that of the *Scenedesmus* (116.6 ± 10.5, 13.5 ± 2.38, respectively). The C : N : P of river seston varied only slightly (C : P, approximately 100–150; C : N, approximately 12–15) during the length of the experiment. Total POC of the filtered seston was 27 mg C L$^{-1}$ at the start of the experiment and ranged between 10 and 30 mg C L$^{-1}$ depending on flow conditions in the river. River seston samples collected near the mid-point of the experiment were analysed for algal community composition. Diatoms were the most abundant algal group (80%; Table 1) as is typical for the Ohio River at this time of year (R. Tucker, pers. comm.). Green algae and *Euglenoids* accounted for 7 and 6% (respectively) of



**Fig. 1** Relationships between *Bosmina freyi* body dry weight (μg) versus body area (μm$^2$).

**Table 1** Results of cell counts for (a) total algae, and (b) green algae for Ohio River seston. (Data courtesy of R. Tucker, Louisville Water Company)

| | Count (units mL$^{-1}$) | Percentage |
|---|---|---|
| (a) Algal types | | |
| *Cyanobacteria* | 34 | 1.0 |
| *Chrysophyte* | 11 | 0.3 |
| Diatom | 2915 | 83.0 |
| *Euglenoid* | 218 | 6.2 |
| Green algae | 252 | 7.2 |
| *Chryptophyte* | 80 | 2.3 |
| Total | 3512 | 100.0 |
| (b) Green algal types | | |
| *Ankistrodesmus* | 126.2 | 50.0 |
| *Chlorella* | 23.0 | 9.1 |
| *Scenedesmus* | 80.3 | 31.8 |
| *Staurastrum* | 11.5 | 4.5 |
| *Actinastrum* | 11.5 | 4.5 |
| Total | 252.5 | 100.0 |

algal cell densities. The unialgal and seston food treatments differed with respect to the types and concentrations of fatty acids present. Among the unsaturated fatty acids, 20:5ω3 (EPA) and 22:6ω3 (DHA) were 10 and 15 times greater (respectively) in river seston than in *Scenedesmus* (Table 2). However, *Scenedesmus* had nearly 50 times more 18:2ω6 (linoleic acid) and greater than seven times more 18:3ω3 (linolenic acid) than Ohio River seston. Analysis of variance showed that *Scenedesmus* had significantly higher total -ω3, -ω6 and sum of all polyunsaturated fatty acids (PUFA, ANOVA: $P < 0.001$) than Ohio River seston.

**Table 2** Fatty acid (FA) profiles of Ohio River seston and laboratory cultured *Scenedesmus acutus* at days 10 and 25 of the experiment. 10:0, 11:0, 12:0, 13:0, 15:1, 18:2w6 trans, 20:2w6, 20:3w3, 23:0, 22:4w6 and 22:3w3 FAs were not detected (nd) in both food types.

| | Ohio River seston | | *Scenedesmus acutus* | |
| --- | --- | --- | --- | --- |
| FA | Rep 1 (µg mg$^{-1}$ DWC) | Rep 2 (µg mg$^{-1}$ DWC) | Rep 1 (µg mg$^{-1}$ DWC) | Rep 2 (µg mg$^{-1}$ DWC) |
| 14:0 | 1.15 | 0.90 | 1.75 | 1.56 |
| 14:1 | 0.02 | 0.02 | 0.00 | 0.00 |
| 15:0 | 0.12 | 0.13 | 0.12 | 0.12 |
| 16:0 | 3.85 | 4.70 | 22.40 | 21.90 |
| 16:1 | 1.59 | 1.69 | 0.66 | 0.65 |
| 17:0 | 0.06 | 0.07 | 0.27 | 0.27 |
| 17:1 | 0.10 | 0.08 | 1.18 | 1.07 |
| 18:0 | 1.66 | 2.53 | 3.05 | 3.09 |
| 18:1w9cis | 0.59 | 0.69 | 29.81 | 28.94 |
| 18:1w9trans | 0.29 | 0.34 | 1.39 | 1.42 |
| 18:2w6cis | 0.23 | 0.30 | 10.04 | 9.69 |
| 18:3w6 | 0.08 | 0.10 | 0.49 | 0.49 |
| 19:0 | 0.02 | 0.03 | nd | nd |
| 18:3w3 | 0.67 | 0.72 | 5.74 | 5.46 |
| 18:4w3 | 0.57 | 0.60 | 1.54 | 1.43 |
| 20:0 | 0.07 | 0.09 | 0.20 | 0.19 |
| 20:1 | 0.03 | 0.03 | 0.64 | 0.62 |
| 20:3w6 | 0.01 | 0.03 | 0.21 | 0.21 |
| 21:0 | 1.85 | 1.85 | 6.38 | 8.51 |
| 20:4w6 | 0.18 | 0.22 | 0.54 | 0.48 |
| 20:5w3 | 2.74 | 2.86 | 0.25 | 0.24 |
| 22:0 | 0.06 | 0.06 | 0.34 | 0.31 |
| 22:1w9 | 0.06 | 0.06 | 0.12 | 0.10 |
| 22:2w6 | nd | 0.26 | nd | nd |
| 22:5w3 | 0.04 | 0.06 | nd | nd |
| 24:0w0 | 0.11 | 0.11 | 0.16 | 0.16 |
| 22:6w3 | 0.39 | 0.46 | 0.12 | 0.10 |
| 24:1w0 | 0.13 | 0.16 | 0.07 | 0.06 |
| Total FA | 16.68 | 19.16 | 87.47 | 87.07 |
| Sum of ω3 FA | 4.41 | 4.71 | 7.65 | 7.23 |
| Sum of ω6 FA | 0.50 | 0.90 | 11.28 | 10.87 |

DWC is dry weight carbon.

### Growth and reproduction

Animals grew well in both treatments (Fig. 2a). Growth rates during the early life stage (0–6 days) were remarkably similar between the two treatments but there was a significant difference in the long-term growth rates (Table 3; Fig. 2b). After their first clutch (6–8 days), *Bosmina* fed *Scenedesmus* grew at a faster rate than those in the seston treatment (Tukey HSD test for postmaturity growth, $P < 0.0001$). Food type also affected the brood size and the number of days between clutches (Fig. 3a,b). Individuals feeding on *Scenedesmus* typically had shorter intervals between clutches throughout their reproductive life than those feeding on river seston. *Bosmina* fed river seston matured at a larger size (approximately 2.5 µg) than those fed *Scenedesmus* (<2.0 µg) and took longer to reach reproductive stage (seston = 7.5 days, *Scenedesmus* = 5.7 days). Analysis of variance (clutch number nested within treatment, ANOVA, $P < 0.001$) indicated
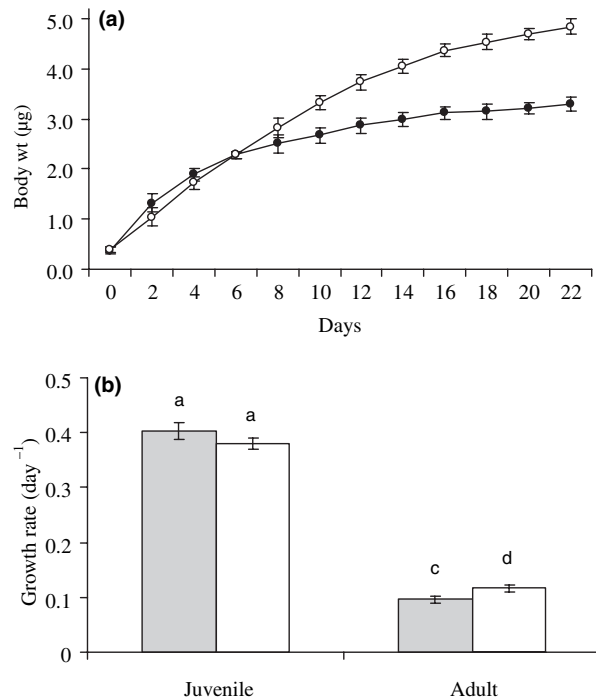


**Fig. 2** (a) Time sequence of change in body weight (µg), and (b) juvenile (short-term) and adult (long-term) average growth rates of *Bosmina freyi* in the *Scenedesmus* and river seston food treatments. Open symbols (○) and un-shaded bars are *Scenedesmus* treatment, and closed symbols (●) and shaded bars are river seston treatment. Error bars are standard error of the mean. Small letters over bars are results of statistical analysis (Tukey HSD).
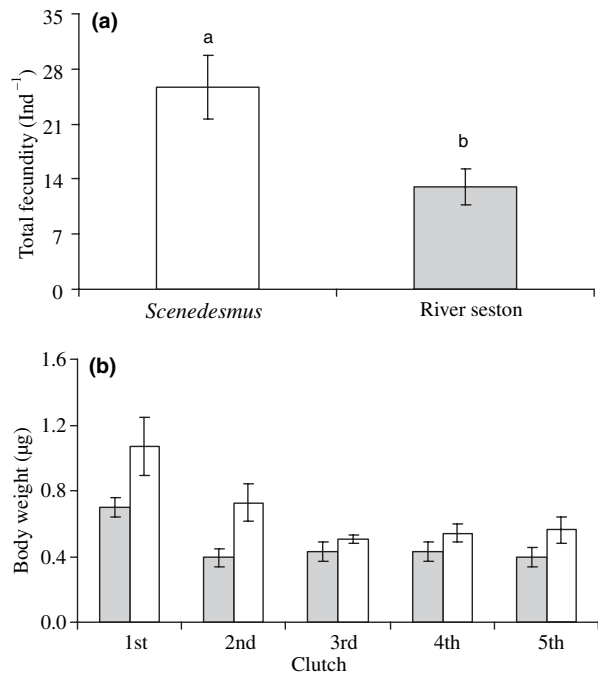
**Table 3** Results of analysis of variance (ANOVA) for average body sizes (weight, μg) and average growth rates (day$^{-1}$) of *Bosmina* fed *Scenedesmus* versus river seston at ages 2–22 days

| Analysis of variance | P-values | |
|---|---|---|
| | Weight | Growth rate |
| 2 | 0.047 | NS |
| 4 | NS | NS |
| 6 | NS | NS |
| 8 | NS | 0.044 |
| 10 | 0.031 | 0.027 |
| 12 | 0.019 | 0.042 |
| 14 | 0.028 | 0.043 |
| 16 | 0.002 | 0.020 |
| 18 | 0.010 | 0.030 |
| 20 | 0.011 | 0.034 |
| 22 | 0.010 | 0.021 |

NS indicates not significant.
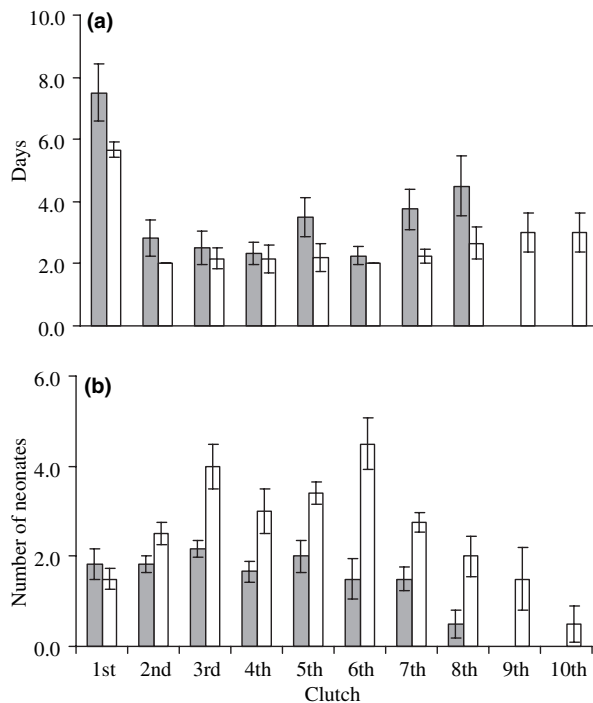


**Fig. 3** (a) Number of days between the clutches and (b) fecundity (number of neonates born) for *Bosmina freyi* on *Scenedesmus* and river seston food treatments. Error bars are standard error of the mean.



**Fig. 4** Mean (a) total average fecundity per individual and (b) body weight (μg) of neonates (<12 h) of *Bosmina freyi* at *Scenedesmus* and river seston food treatments. Un-shaded bars are *Scenedesmus* and shaded bars are river seston treatments. Error bars are standard error of the mean. Small letters over bars are results of statistical analysis (Tukey HSD).

that there was a significant difference in fecundity between the two food treatments. Tukey HSD pairwise comparison test showed that *Bosmina* fed *Scenedesmus* had a higher number of offspring in each clutch (except first clutch) and higher total fecundity per individual (approximately 25 versus approxi-

mately 14 neonates; Fig. 4a). Also, river seston treatment animals stopped their reproductive activity earlier (approximately eighth clutch) than those on *Scenedesmus* (approximately 10th). First and second clutch neonates born from mothers feeding on *Scenedesmus* had significantly larger body sizes than those feeding on seston. However, neonates from all other clutches did not differ in their body sizes between treatments (Fig. 4b, Tukey HSD pairwise comparison clutches first and second; $P < 0.01$, third to fifth; $P > 0.1$). Although *Bosmina* fed *Scenedesmus* produced more offspring and grew faster, their overall survivorship was significantly lower than those feeding on seston (Fig. 5; ANOVA $P < 0.001$).

Lipid ovary indices (visual oil droplets outside and inside the ovary) differed between the two food treatments (Fig. 6). *Bosmina* neonates in both treatments started with statistically similar lipid reserves but after days 6–7 (onset of reproduction), individuals fed *Scenedesmus* maintained a higher lipid index than those in the river seston treatment (pairwise comparison Tukey HSD, $P < 0.0001$). *Bosmina* fed seston had
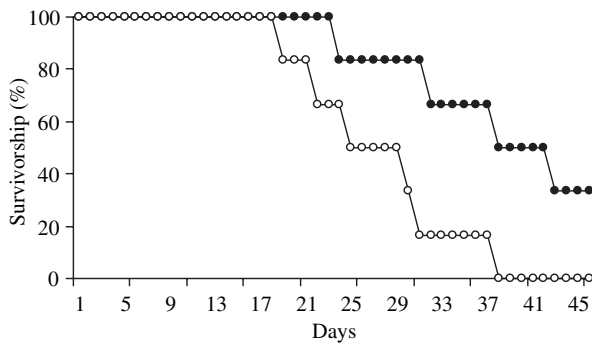
**Fig. 5** Survival rate of *Bosmina freyi* on *Scenedesmus* and river seston food treatments. Open symbols (○) are *Scenedesmus* and close symbols (●) are river seston treatments.
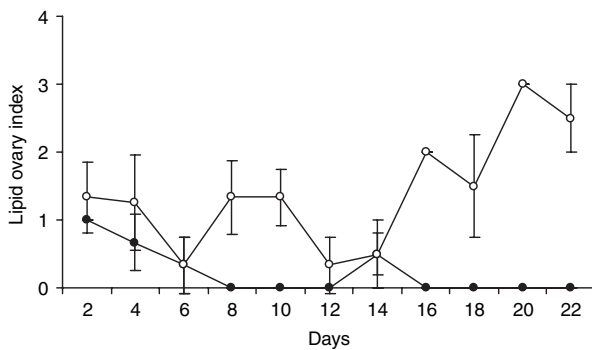


**Fig. 6** Patterns of visible lipid/energy reserves for *Bosmina freyi* on *Scenedesmus* and river seston food treatments. Open symbols (○) are *Scenedesmus* and close symbols (●) are river seston treatments.

no or very little visible lipid reserves after the onset of reproduction. Consequently, neonates born from mothers from the seston treatment had less visible lipid reserves in their body than those born from mothers fed *Scenedesmus*.

## Discussion

Life history traits of river *Bosmina* were generally similar to previously published accounts of lake *Bosmina* feeding under similar resource conditions. Urabe (1991) showed that *Bosmina* from Lake Okutama (Japan) continuously increased their carapace length from birth to 20 days when food concentrations exceeded 0.1 mg C L$^{-1}$. Our data show that they not only increase carapace length, but also continue to gain weight. Schulz & Sterner (1999) showed a similar trend in *Bosmina* (*leideri*) for early

growth stages (to day 7). Bothar (1986, 1987) also reported a high growth rate of riverine *Bosmina* (River Danube, Hungary). The mean brood size in our *Scenedesmus* treatment was similar to that of the phosphorus rich, high quantity treatment of Schulz & Sterner (1999) and the 0.25 mg C L$^{-1}$ treatment of Urabe (1991); however Urabe had slightly higher mean brood size (approximately nine versus six per clutch) at 2.5 mg C L$^{-1}$. Similarly, Bothar (1986) observed 8–10 eggs per ovigerous female in early spring in the River Danube. The maturation age of the animals in our *Scenedesmus* treatments was similar to the high food concentration treatment of Urabe (1991). Urabe's food was comprised of *Scenedesmus* with a small amount of *Chlorella* and flagellates whereas Schulz and Sterner used *S. acutus* (as in our study). Overall, our findings on riverine *Bosmina* are consistent with the previously published life histories in lake and river environments.

Life history traits of cladocerans are known to change according to resource quality or quantity (Urabe & Sterner, 2001) and this view is further supported by our experiments. There were a number of differences in life history traits between *Bosmina* that consumed laboratory-cultured *Scenedesmus* and those fed river seston. Animals performed equally well in both food types prior to the onset of reproduction but thereafter, *Bosmina* fed *Scenedesmus* exhibited greater growth and fecundity. For animals like *Bosmina* which continue to grow after maturation long-term growth and fecundity data may reveal important information not seen in short-term experiments. Had we based our assessment of dietary effects on juvenile growth rates we would have erroneously concluded that animals performed equally well on the two food types. Our data showed that differences in growth rates between the two treatments became apparent after the onset of reproduction. In a prior study, we found that *Daphnia galeata* fed uniform or mixed particle diets with equivalent stoichiometric ratios did not exhibit differences in juvenile growth rates and early reproductive output whereas *Daphnia pulicaria* did (Acharya *et al.*, 2004a). It is arguable that in this and similar studies (e.g. Schulz & Sterner, 1999), measurement of growth and reproduction beyond 7–8 days might have revealed differences among food treatments. Therefore, we recommend that long-term growth and fecundity should be evaluated in studies attempting to link animal per-

formance with population dynamics. The longer duration of our experiments also revealed that animals which produced more and grew faster during their peak reproductive period died earlier (*Scenedesmus* treatment) whereas animals that grew slowly and had a lower reproductive output survived longer (seston treatment). Becker & Boersma (2003) reported similar findings where *Daphnia magna* reproducing more and early had a lower survival rate than those reproducing late and slow. Other researchers have reported that animals growing faster and putting more energy in reproduction tend to have a lower life expectancy (Bell, 1984; Martinez *et al.*, 1994; Rose, Warne & Lim, 2000; Lemke & Benke, 2003). Their argument is supported by our data and emphasises the advantage of assessing life history over the lifespan of the individual.

Reduced performance of adult *Bosmina* feeding on river seston suggests that food resources were insufficient to meet requirements for growth as well as reproduction. In the following section, we consider constraints on growth and fecundity imposed by carbon (energy), phosphorus, and fatty acids.

Rivers typically exhibit high POC concentrations because of particulate materials which are maintained in suspension by advective forces. Riverine POC is predominantly comprised of allochthonous carbon from terrestrial sources and is assumed to be of low food quality (Thorp & Delong, 2002). Prior studies of the Ohio River have shown that algae constitute about 10% of the total POC (Sellers & Bukaveckas, 2003; Bukaveckas *et al.*, in press). POC concentrations during our experiments always exceeded 10 mg L$^{-1}$ and therefore we assume that the algal C fraction was in excess of 1 mg L$^{-1}$. Diatoms were the dominant component of the algal fraction (>80%; Table 1) but because of their generally larger size may not constitute a significant portion of *Bosmina* food. Green algae constituted approximately 7% and if we assume that these represent the edible fraction, then our estimate of algal C resources is in excess of threshold concentrations for *Bosmina* reported by Guelda *et al.* (in press; 40 μg L$^{-1}$) and Gliwicz (1990; 70 μg L$^{-1}$). A trial experiment in conjunction with this study estimated threshold values based on *Scenedesmus* of 50 μg L$^{-1}$ (K. Acharya, unpubl. data). As the edible algal C fraction exceeded threshold values, we feel it is unlikely that *Bosmina* feeding on river seston suffered carbon (energy) limitation during our experiments

even if their food intake was restricted to the green algal fraction of POC. We cannot discount the possibility that ingestion rates were affected by higher concentrations of total suspended matter in the seston treatment. However, the selective feeding abilities of *Bosmina* (DeMott, 1982, 1986) and removal of particulates greater than 35 μm would minimise the likelihood of feeding interference.

Nutritive content of food resources (particularly phosphorus) may account for differences in growth and fecundity between food treatments. Neonates have a high percentage of phosphorus per unit mass compared with adults especially when they are born from well-fed mothers. For example, *D. pulicaria* and *galeata* neonates had 20–100% more P content than adults (Acharya, Kyle & Elser, 2004b). The *Daphnia* threshold C : P is 300 (Urabe & Watanabe, 1992) above which *Daphnia* suffer P limitation. This number for *Bosmina* is probably much higher because of their lower body P content (Hessen & Lyche, 1991; Schulz & Sterner, 1999). C : P ratios of both river seston and *Scenedesmus* were below 300 (molar) and therefore we think it unlikely that *Bosmina* suffered P limitation during our experiments.

Zooplankton have also been shown to suffer from insufficient essential fatty acids in their food (Ahlgren *et al.*, 1990; Müller-Navarra, 1995a; DeMott & Müller-Navarra, 1997; Weers & Gulati, 1997). We observed that *Bosmina* neonates at the start of the experiments had similar lipid reserves in both treatments but as they grew older, body lipids began to decline in animals feeding on seston. Bosmina born from mothers rich in lipid are more likely to be born with higher lipid reserves (Goulden & Henry, 1981). Therefore it is possible that neonates born from well-fed mothers are less likely to be limited (lack of sufficient availability) by biochemicals early in their growth but their growth and fecundity become diminished when they exhaust their maternal contributions.

It is possible that *Bosmina* fed *Scenedesmus* in our experiments sustained higher growth and fecundity because of higher fatty acid content in their food. Fatty acid analysis showed that *Scenedesmus* had more lenolenic acid, total -ω3, -ω6 and overall PUFA but less EPA and DHA than Ohio River seston. There is general agreement that EPA is the most important PUFA likely to limit *Daphnia* growth and reproduction (Müller-Navarra, 1995a; Brett & Müller-Navarra,

1997; Gulati & DeMott, 1997) because it plays an important role in cellular metabolism, as a precursor of eicosanoids (i.e. prostaglandins), and as a part of cell membranes (Brett & Müller-Navarra, 1997). However, some *Daphnia* species may be able to convert EPA from lenolenic acid (e.g. chain elongation) such that direct intake of EPA may not be necessary (Weers *et al.*, 1997). The *Scenedesmus* in our experiments had higher linolenic acid, total -ω3 and -ω6 and lower EPA than river seston. Thus, our data supports the earlier observations (DeMott & Müller-Navarra, 1997; Weers *et al.*, 1997) on *D. galeata* which showed that algae with low EPA but high linolenic acid might not be a poor food source. We know of no prior studies that have identified specific fatty acids which may be important for *Bosmina*.

## Conclusions

Results from our experiments suggest that *Bosmina* raised on unialgal cultures out-perform those raised on river seston even when algal C content and bulk nutritive values (C : N : P) are similar. Furthermore, we observed that long-term growth and fecundity may reveal important information not seen in the short-term experiments when assessing dietary effects on growth and reproduction for *Bosmina*. We observed that food quality effects differed among juveniles and adults and may be related to maintenance of lipid reserves. Gradual accumulation of triacylglycerol (visible lipid droplets) and improved performance on *Scenedesmus* may indicate that either lenolenic acid (18:3ω3) or total PUFA was an important dietary component for riverine *Bosmina*. Our results show that food quality affects the life history parameters of riverine *Bosmina* and that dietary constraints may be an important factor determining population success in riverine environments. Our results support a prior study using population-based approaches (mesocosm experiments) that showed Bosmina and cyclopoids in the Ohio River experienced food limitation during periods of low POC concentrations (Guelda *et al.*, in press).

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