

# Food niches of cyclopoid copepods in eutrophic Plußsee determined by stable isotope analysis

Barbara Santer<sup>1</sup>, Nike Sommerwerk<sup>1, 2</sup> and Jonathan Grey<sup>1, 3</sup>

Max Planck Institute of Limnology, Plön, Germany

With 5 figures

**Abstract:** Feeding studies have suggested interspecific differences in the food sources and trophic position of cyclopoid copepod species. We measured  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of five sympatric cyclopoid copepod species and of bulk POM from a eutrophic lake over a one-year period, to determine if stable isotope derived-data from the field supported the hypotheses from feeding studies. *Cyclops abyssorum* and *C. vicinus* differed significantly in  $\delta^{15}\text{N}$  but not in  $\delta^{13}\text{C}$ , inferring assimilation from similar carbon sources but differentiation in their trophic position. *Cyclops abyssorum*  $\delta^{15}\text{N}$  typically was 1–2‰ higher than *C. vicinus*, suggesting a more predatory feeding mode compared to its congener. The summer species *Mesocyclops leuckarti*, *Thermocyclops oithonoides* and *Diacyclops bicuspidatus* exhibited considerable differences in  $\delta^{13}\text{C}$ . The lower  $\delta^{13}\text{C}$  of the hypolimnetic *D. bicuspidatus* compared to the two epilimnetic species indicates a food niche differentiation by vertical separation. All copepod  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures showed seasonal fluctuations, with low values in winter and high values in summer. The  $\delta^{13}\text{C}$  of seston was generally higher than copepod  $\delta^{13}\text{C}$ . We regard selective feeding from the bulk POM, spatial separation of feeding and assimilation of  $^{13}\text{C}$  depleted food sources responsible for this phenomenon.

**Key words:**  $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ , trophic position, competition, plankton.

## Introduction

Cyclopoid copepods are one of the most important groups within the zooplankton of lakes and often dominate the plankton community for certain periods of the year (SANTER & LAMPERT 1995). Cyclopoid copepods are highly

<sup>1</sup> **Authors' addresses:** Max Planck Institute of Limnology, Post-box 165, 24302 Plön, Germany; E-mail: Santer@mpil-ploen.mpg.de

<sup>2</sup> Angerstraße 36 A, 30539 Hannover, Germany; E-mail: n.sommerwerk@the-blue.net

<sup>3</sup> School of Biological & Chemical Sciences, Queen Mary University of London, Mile End Road, E1 4NS, UK; E-mail: j.grey@qmul.ac.uk

selective feeders (DEMOTT 1986) and since they switch diet during their ontogenetic development, which includes 6 naupliar stages and 6 copepodid stages, potentially they can influence all lower trophic levels. The naupliar stages and early copepodid stages ingest algae and protists; advanced copepodid stages and adults also prey on rotifers, small crustaceans and even fish fry (FRYER 1957, DAVIS 1959, KARABIN 1978, ADRIAN & FROST 1993, BRANDL & FERNANDO 1981, WICKHAM 1995, BRANDL 2005). Not much is known about dietary differentiation between copepod species, but morphological, behavioural or physiological differences between copepod species are assumed to influence their nutrition (HANSEN & SANTER 1995, HOPP & MAIER 2005). The hypolimnetic copepod *Diacyclops bicuspidatus* can tolerate very low oxygen concentrations in thermal stratified lakes (EINSLE 1965) and should have access to food sources potentially unavailable to the epilimnetic species *Mesocyclops leuckarti* or *Thermocyclops oithonoides*. Other co-occurring species seem to differ in the extent to which they utilise algal food sources. Adult *Cyclops abyssorum* are considered carnivorous and even resort to cannibalism when alternative prey items are not available (VAN DEN BOSCH & SANTER 1993), while *Cyclops vicinus* is reported to fulfil its nutritional demands largely with phytoplankton, even when adult (ADRIAN 1991a, b, SANTER 1993, SANTER & VAN DEN BOSCH 1994, HANSEN & SANTER 1995). Such observations suggest considerable variation in cyclopoid copepod trophic status and food niches, which might facilitate coexistence of otherwise ecologically similar species. However, these assumptions are derived largely from observed frequency distributions or from laboratory experiments and extrapolation to copepods living under natural conditions is limited. Feeding experiments are conducted with restricted numbers of food items for simplicity and even if natural food mixtures are used, such experiments often suffer from bottle effects. Ingestion rates and gut content analyses provide only dietary snap-shots, and recalcitrant food items are often over-represented in gut contents, while conversely soft bodied organisms like flagellates are usually under-represented (HART & SANTER 1994).

In contrast, the isotopic composition of a consumer represents an integrated measure of the assimilated diet over time. The natural abundance ratios of  $^{13}\text{C} : ^{12}\text{C}$  and  $^{15}\text{N} : ^{14}\text{N}$  ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  signatures) of organisms are considered to reflect those of their diet with minor enrichments of 0.4‰ for  $\delta^{13}\text{C}$  (MCCUTCHAN et al. 2003), and 2.5‰ for  $\delta^{15}\text{N}$  (VANDERKLIFT & PONSARD 2003), but there is still a degree of uncertainty in interpreting such fractionations, for example with respect to dietary quality (ADAMS & STERNER 2000). Nevertheless, results from a growing number of studies indicate that stable isotope analysis (SIA) can provide information regarding zooplankton dietary carbon source and trophic position (KLING et al. 1992, YOSHIOKA & WADA 1994, ZOHARY et al. 1994, GREY & JONES 1999, SARVALA et al. 2003,

MATTHEWS & MAZUMDER 2003, FEUCHTMAYR et al. 2004, SOMMER et al. 2005) and might even provide a reliable method to detect small dietary differences (GREY et al. 2001, KARLSSON et al. 2004).

In this study, we measured the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of sympatric cyclopoid copepods from a eutrophic lake, to see if stable isotope derived-data supported the hypothesised differing diets and trophic positions suggested with conventional methods.

## Material and methods

Plußsee is a small (14.3 ha), but deep (29.2 m) naturally eutrophic kettle lake in Schleswig-Holstein, northern Germany. The lake is dimictic with an anoxic hypolimnion in summer. Since the mid 1960s, numerous research projects have been carried out in the lake (OVERBECK & CHRÓST 1994, SPAAK & BOERSMA 2006) and some physical and chemical parameters (e. g. temperature, oxygen, chlorophyll and various nutrients) are monitored weekly. The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of the crustacean plankton functional groups were monitored regularly (since 2001) to determine trophic structure and temporal variation within the community (see HARROD & GREY 2006). However, cyclopoid copepods were amalgamated into one trophic functional group for routine monitoring because distinction of species and life-stages is difficult and time-consuming.

To obtain an overview of cyclopoid species inhabiting the pelagic zone of the lake, we determined their seasonal abundances over a one-year period (2004–2005), as well as their vertical distribution. Three replicate vertical tows through the water column (0–25 m) were taken with a 100  $\mu\text{m}$  net (opening diameter 9 cm) weekly or every other week (with few exceptions due to logistic problems or ice cover) close to the deepest point of the lake. The contents of the hauls were combined in a 250 ml plastic vessel and preserved with formalin (4% final concentration). Sub-samples were taken after the zooplankton had been filtered over a 100  $\mu\text{m}$  net and rinsed with tap water to remove the formalin. Only copepodid stages were counted, the early instars (nauplii) were not considered since they cannot be distinguished. Copepods were determined to species level and abundances were calculated as individuals per  $\text{m}^2$  of lake surface area.

To examine the vertical distribution of the copepods throughout the water column, water was pumped from discrete depths in vertical intervals of 1 m (50 l of water per depth) over a 100  $\mu\text{m}$  net and samples were preserved as above for later counting.

Copepods for SIA had been sampled from the lake previously in October and November 2001 (SOMMERWERK 2002) and between July and September 2003. These samples were analysed to examine whether copepod species exhibited different  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  before a regular sampling program was initiated. From April 2004 to May 2005, zooplankton samples for SIA were taken weekly or every other week with a 250  $\mu\text{m}$  net. In the laboratory, sub-samples were taken and a few drops of carbonated water added to narcotise the copepods. This was necessary to distinguish between species and life-stages under a binocular microscope. Adult females without eggs were removed from the sample with a pipette, measured from the upper end of the cephalo-

thorax to the lower end of the furcal rami, and placed into 250 ml glass beakers (one copepod species per beaker) filled with membrane filtered lake water. We focussed on adult females for size reasons and because most feeding experiments use this life stage. The  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  of males and copepodids (fourth and fifth stage) were measured only three times (*Cyclops abyssorum*) or four times (*C. vicinus*) between December 2004 and January 2005.

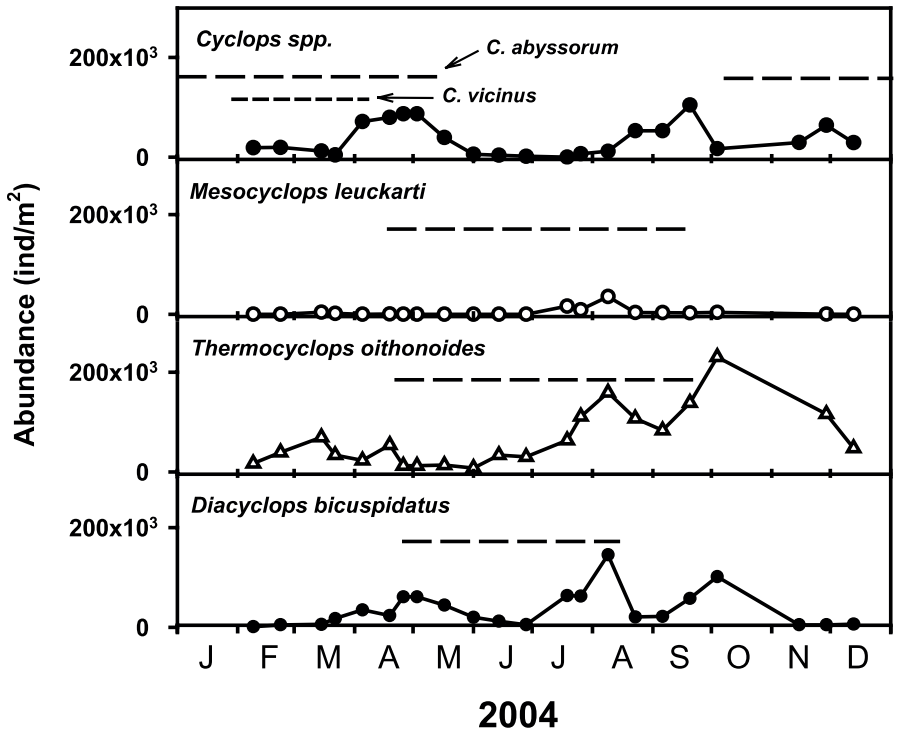
The copepods were maintained for 24 h to allow for gut evacuation (FEUCHTMAYR & GREY 2003). After rinsing with filtered water through a 250  $\mu\text{m}$  mesh to remove faecal material, the copepods were pipetted onto a Whatman GF/C filter and oven dried (55 °C) for 24 h. The number of individuals per sample varied depending on the species size and abundance. Approximately 0.5 mg of dry zooplankton tissue was packed into tin capsules for subsequent analysis by continuous flow isotope ratio mass spectrometry (Eurovector elemental analyser coupled to an IsoPrime Mass Spectrometer, Micromass) as described by FEUCHTMAYR & GREY (2003). SIA of bulk particulate organic matter (POM) was performed in autumn 2001 and in spring and summer 2004. Thirty litres of water sampled from 0.5 m depth were filtered through a 55  $\mu\text{m}$  mesh to remove large zooplankton. POM was concentrated by continuous flow centrifugation and freeze-dried. Approximately 2 mg of dried material was analysed for  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ .

Samples were analysed in triplicate whenever sufficient material was available. The averages were calculated for each sampling date and tested for differences between species with One Way Repeated Measures Analysis of Variance (RM ANOVA) or, if the test for normality failed, with non-parametric Friedman RM ANOVA on Ranks, using the software Sigmastat 3.1 (©Systat Software, Inc., USA).

## Results

Eight cyclopid copepod species were found during the sampling period in the pelagic zone of the Plußsee. *C. abyssorum* (♀ 1.9–2.3 mm) occurred between August and May and reproduced between October and May. *C. vicinus* (♀ 1.6–2.3 mm) was found from September to May, females with eggs were observed mainly in February and March (Fig. 1). Both copepod species were most abundant in spring and late summer and were found as diapausing copepodids in the sediment in June and July. *C. bohater* and an unidentified species, probably *C. strenuus*, were occasionally present in small numbers, but it would have been too time consuming to distinguish all copepodid stages of these four morphologically similar species. The vertical distribution of *C. abyssorum* and *C. vicinus* did not differ. In November 2004 their occurrence was restricted to the oxic zone, which extended to 9 m depth (data not shown); in April 2005 they occurred in the upper layer of the lake where the high chlorophyll values indicated high algal biomass (Fig. 2).

*Mesocyclops leuckarti* (♀ 1.0–1.3 mm) and *Thermocyclops oithonoides* (♀ 0.7–1.0 mm) the “summer species” in this lake, reproduced between April and



**Fig. 1.** Seasonal abundance of cyclopoid copepods (ind./m<sup>2</sup> lake surface) in Plußsee. Ice prevented sampling in January. The four species of the genus *Cyclops* were amalgamated (*C. abyssorum* was by far the most frequent species), since distinction of copepodid stages is very time consuming in this group. The dotted lines indicate the periods of reproduction.

September, between October and March the majority of life-stages observed were diapausing C4 stages (both species are known to undergo a winter-diapause). The smallest copepod species, *T. oithonoides*, was the most abundant, whereas the co-occurring *M. leuckarti* was rare (Fig. 1). Both copepods inhabited the epilimnion in summer when the lake was stratified (Fig. 3). However, *T. oithonoides* exhibited a population maximum just below that of *M. leuckarti*. *Diacyclops bicuspidatus* (♀ 1.0–1.4 mm) was also found between February and October in the pelagic zone, but we do not know where this copepod spent the winter. This species was restricted to the deeper zones with the majority of the population in the hypolimnion between 6 and 7 m at oxygen concentrations < 1 mg/l. An analysis of the day-night distribution in July 2005 revealed that the copepod does not undergo vertical migration. Approximately 90% of the population (copepodids and adults) was found in the hypolimnion below 4 m depth, both at noon and at midnight (data not shown).

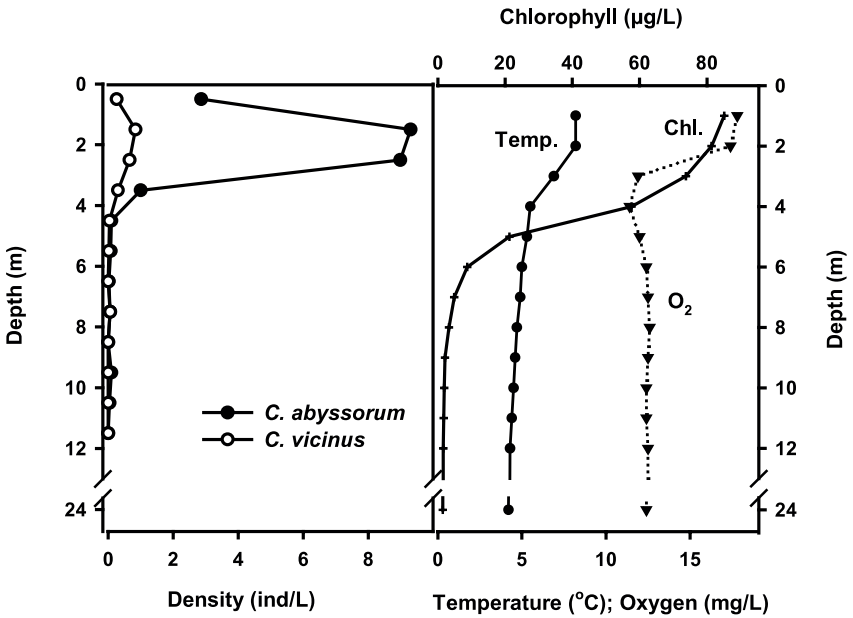


Fig. 2. Vertical distribution of *Cyclops vicinus* and *C. abyssorum* (left panel), temperature, oxygen, and chlorophyll (right panel) in April 2004 in Plußsee.

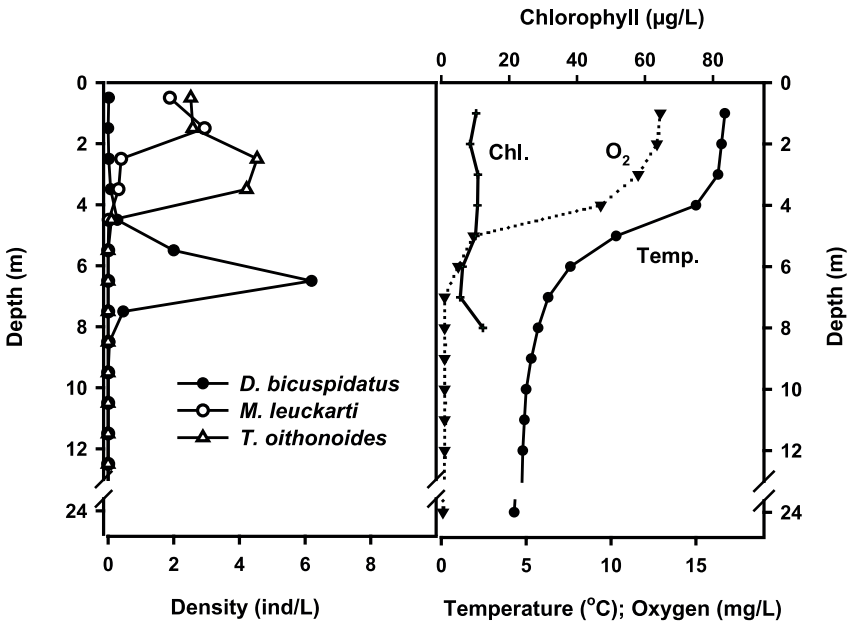
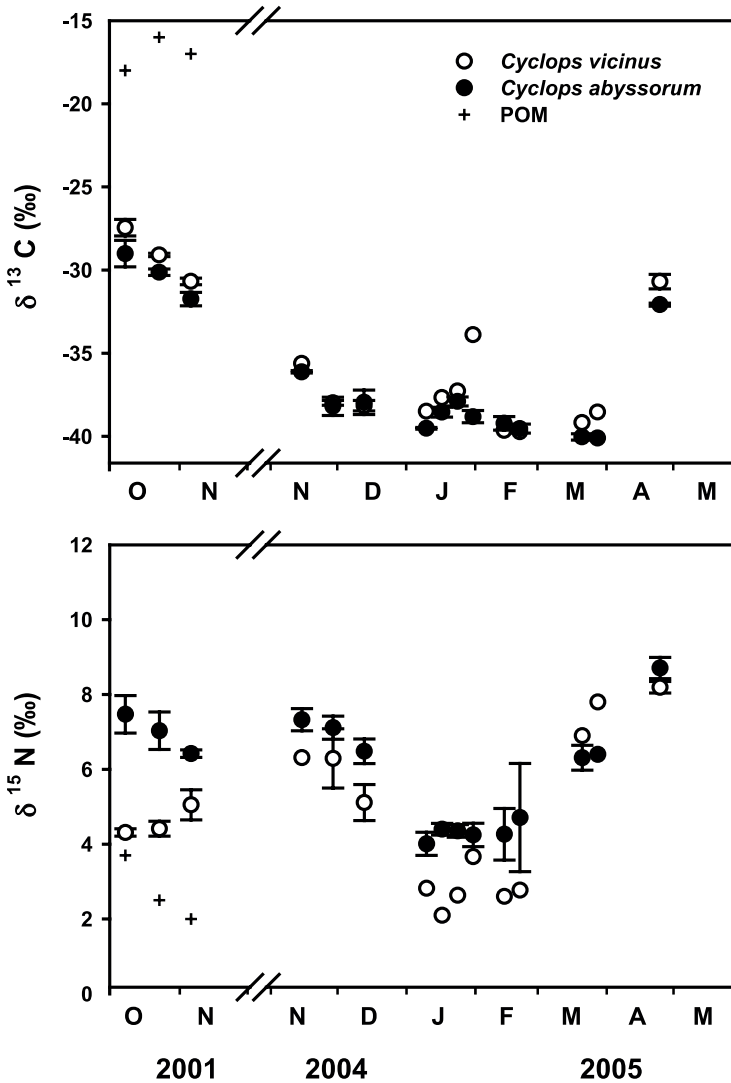
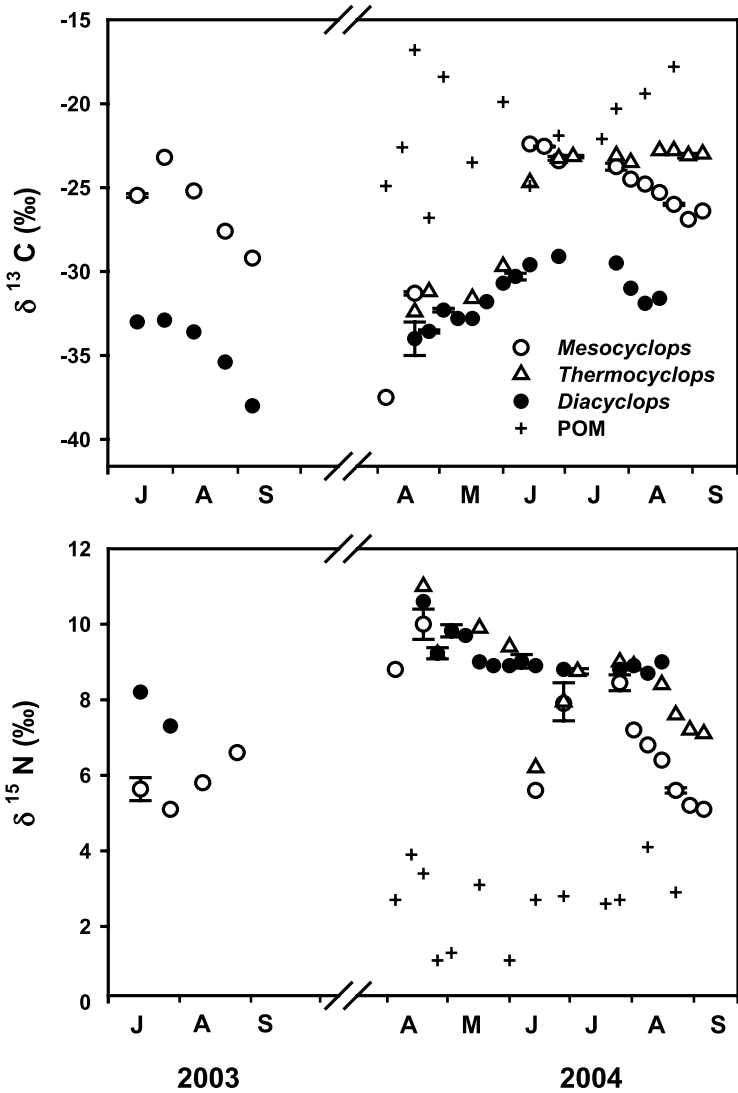


Fig. 3. Vertical distribution of *Mesocyclops leuckarti*, *Thermocyclops oithonoides* and *Diacyclops bicuspidatus* (left panel), temperature, oxygen and chlorophyll (right panel) in June 2004 in Plußsee. Chlorophyll was only measured to 8 m water depth.



**Fig. 4.** Temporal pattern of mean ( $\pm 1$  SD) carbon (upper panel) and nitrogen (lower panel) isotope signatures of female *C. vicinus* and *C. abyssorum* and of particulate organic matter (POM  $< 55 \mu\text{m}$ ) in Plußsee in autumn 2001 (after SOMMERWERK 2002) and from November 2004 until May 2005. Where no error bars are present, only a single sample was analysed.

The results of the stable isotope analyses revealed differences between species. Female *C. abyssorum* and *C. vicinus* (Fig. 4) differed significantly in  $\delta^{15}\text{N}$  (RM ANOVA,  $F_{(1, 11)} = 9.158$ ,  $p = 0.012$ , for 2004–2005), but not in  $\delta^{13}\text{C}$  (Friedman RM ANOVA, Chi-square = 3.000,  $p = 0.146$ , same period). *C. abys-*



**Fig. 5.** Temporal pattern of mean ( $\pm 1$  SD) carbon (upper panel) and nitrogen (lower panel) isotope signatures of female *Mesocyclops leuckarti*, *Thermocyclops oithonoides* and *Diacyclops bicuspidatus* and of particulate organic matter (POM < 55  $\mu\text{m}$ ) in Plußsee in summer 2003 and between April and September 2004. Where no error bars are present, only a single sample was analysed.

*sorum*  $\delta^{15}\text{N}$  was typically higher than that of *C. vicinus* throughout the year except for two sampling dates in spring 2005. The two species did not differ in their C to N ratio (RM ANOVA,  $F_{(1, 11)} = 0.711$ ,  $p = 0.417$ , for 2004–2005). The same  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  pattern between the two species was found in a pre-



liminary sample set from autumn 2001 (SOMMERWERK 2002). In both species males did not differ significantly in  $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ , or C/N ratio from females (RM ANOVAs, 3 comparisons). Male *C. abyssorum* exhibited higher  $\delta^{15}\text{N}$  values ( $6.7 \pm 0.5$ ,  $n = 3$ ) relative to male *C. vicinus* ( $5.2 \pm 0.7$ ,  $n = 4$ ), the difference was, however, not significant (RM ANOVA,  $F_{(1,3)} = 1.399$ ,  $p = 0.447$ , for Dec 2004–Jan 2005).

In contrast to the winter species, the summer species exhibited considerable difference in  $\delta^{13}\text{C}$  (Fig. 5). In the preliminary sample set from summer 2003, when only two species had been analysed, epilimnetic *M. leuckarti* were between 7.5 and 9.7‰  $^{13}\text{C}$ -enriched relative to the hypolimnetic *D. bicuspidatus*. The two species again exhibited clear differences in  $\delta^{13}\text{C}$  in summer 2004 (5.8–7.2‰). During spring 2004, insufficient individuals of *M. leuckarti* were found for SIA. *T. oithonoides* was abundant during spring 2004 and exhibited minor  $^{13}\text{C}$ -enrichment relative to *D. bicuspidatus*. In June, when the lake had stratified and a severe hypolimnetic oxygen depletion had established (see also HARROD & GREY 2006) *T. oithonoides*  $\delta^{13}\text{C}$  had shifted and was similar to that of *M. leuckarti*. Indeed, no significant difference was observed between these two epilimnetic species in summer ( $p = 0.537$ ), while *D. bicuspidatus* differed significantly from both species during the same period (RM ANOVA,  $F_{(2,4)} = 74.013$ ,  $p < 0.001$ , Holm-Sidak post hoc test for pairwise multiple comparisons). *T. oithonoides* exhibited slightly higher  $\delta^{15}\text{N}$  (< 1‰) relative to the other two species in April and May, no consistent pattern was seen in June and July, but *T. oithonoides*  $\delta^{15}\text{N}$  was around 2‰ higher than *M. leuckarti*  $\delta^{15}\text{N}$  in August and September.

Copepod  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  exhibited seasonal fluctuation (Fig. 4);  $\delta^{13}\text{C}$  was low in winter with a minimum of  $-40$ ‰ for *C. abyssorum* in March, in spring,  $\delta^{13}\text{C}$  values typically increased, with the epilimnetic cyclopoids reaching  $-23$ ‰ in summer (Fig. 5). Copepod  $\delta^{15}\text{N}$  exhibited a similar trend. In winter the values ranged between 2 and 5‰, then values tended to increase in spring but the highest values were recorded in April (10–11‰).

Seston was generally  $^{13}\text{C}$ -enriched relative to copepods (Figs 4, 5). The POM  $\delta^{13}\text{C}$  was 10–15‰ higher than that of the two *Cyclops* species in autumn 2001. In spring and summer 2004, epilimnetic POM  $\delta^{13}\text{C}$  fluctuated between  $-25$ ‰ and  $-17$ ‰ and was, except for one sampling event (14<sup>th</sup> of June), also  $^{13}\text{C}$ -enriched relative to *M. leuckarti* and *T. oithonoides*, and the hypolimnetic *D. bicuspidatus*. The POM  $\delta^{15}\text{N}$  was less variable than  $\delta^{13}\text{C}$  (1–4‰) and lower in  $^{15}\text{N}$  compared to the copepods.

## Discussion

As found already in other studies (e. g. JONES et al. 1999, GREY et al. 2000, 2001), copepods were typically  $^{13}\text{C}$ -depleted relative to bulk POM. Such car-

bon isotopic depletion relative to smaller size fractions does not conform to postulated enrichment between trophic levels. However, this appears to be a general feature in lakes (DEL GIORGIO & FRANCE 1996) and has been attributed to lipid-accumulation, spatial separation of feeding or selective feeding from the bulk POM used as a putative food source comprising myriad components which are actually isotopically distinct (GREY et al. 2001, PEL et al. 2004, MATTHEWS & MAZUMDER 2005 a). Our bulk POM was only collected from the surface waters and was unlikely to be representative of food available in different water depths. Meta- and hypolimnetic microplankton can be  $^{13}\text{C}$ -depleted relative to epilimnetic microplankton due to the incorporation of respiratory carbon (DEL GIORGIO & FRANCE 1996 and references therein, LEHMANN et al. 2004). Species like the hypolimnetic *D. bicuspidatus* and also juveniles of other copepods (nauplii) that may serve as prey for advanced cyclopoid stages are known to utilize deep food patches (ADRIAN & SCHIPOLOWSKI 2003). Thus, spatial separation of feeding may account for some of the isotopic discrepancy (see also below). Lipid accumulation was not considered in the current study primarily because lipid-free mixed zooplankton  $\delta^{13}\text{C}$  typically differed by  $<1\%$  compared to uncorrected mixed zooplankton  $\delta^{13}\text{C}$  in Plußsee, and only after the ice-break was the difference was higher (up to  $8\%$  for a short period, GREY unpubl. data). Therefore, although we cannot rule out lipid accumulation explaining some of the variation in our data, we feel it is unlikely to account for the consistent difference in  $\delta^{13}\text{C}$  between the copepods and POM we observed throughout the summer period.

There appears to be food niche differentiation between the sympatric species *C. abyssorum* and *C. vicinus* and between *M. leuckarti*, *T. oithonoides* and *D. bicuspidatus*. The lack of significant difference between *C. abyssorum* and *C. vicinus*  $\delta^{13}\text{C}$  values throughout the year indicates assimilation from similar carbon sources. However, the relatively consistent difference in  $\delta^{15}\text{N}$  signatures of the two species suggests that they differ in trophic status within the lake and that this is maintained throughout the season. The  $^{15}\text{N}$ -enrichment of *C. abyssorum* in contrast to *C. vicinus* indicates a more predatory feeding mode, or conversely, more omnivory by the latter. The degree of separation ( $1\text{--}2\%$ ) was usually less than is generally accepted as indicative of a trophic level increase (VANDERKLIFT & PONSARD 2003), and thus, might be due to a partial overlap of their food sources. The short period when *C. vicinus* was  $^{15}\text{N}$ -enriched relative to *C. abyssorum* arose immediately following ice-break which is a very dynamic period in Plußsee resulting in a rapid shift in zooplankton isotope signatures in response to the first spring bloom (see HARROD & GREY 2006, and discussion below). This has parallels to upwelling events and rapid assimilation of new nutrients into phytoplankton and some zooplankton in Lake Tanganyika causing disparity in trophic levels delineated from  $\delta^{15}\text{N}$  (O'REILLY et al. 2002). We suppose that *C. vicinus* was quicker to

respond to the changing phytoplankton isotopic signature via direct assimilation; *C. abyssorum* tracked the phytoplankton signature eventually but with more of a lag due to indirect assimilation via intermediate prey. Thus, our stable isotope data give some support to different feeding behaviours of these two sympatric species under natural conditions, which previously have been hypothesised from laboratory experiments (ADRIAN 1991 a, b, VAN DEN BOSCH & SANTER 1993, SANTER & VAN DEN BOSCH 1994, HANSEN & SANTER 1995) and from recently reported differences in the morphological structures of the mouthparts (HOPP & MAIER 2005).

The  $\delta^{13}\text{C}$  of *M. leuckarti*, *T. oithonoides* and *D. bicuspidatus* reflect food niche differentiation by vertical separation in summer. *M. leuckarti* was typically found in the upper epilimnetic waters of the lake where the majority of primary production has been observed (MEFFERT & OVERBECK 1994) and consequently exhibited  $\delta^{13}\text{C}$  values commonly reported for highly productive lakes (GREY et al. 2000). *D. bicuspidatus* is known to tolerate very low oxygen concentrations (EINSLE 1965), and was generally found in Plußsee just below the oxycline (KRAMBECK et al. 1994). This zone is characterised by high concentrations of phytoplankton, especially cryptophytes (SANTER & GREY, unpubl. data), and ciliates and heterotrophic nanoflagellates grazing on the bacterial production, which is equal to the bacterial production in the epilimnion in summer (MEIER & RECK 1994). Both, cryptophytes and protists are suitable food sources for cyclopoid copepods (HANSEN & SANTER 1995, WICKHAM 1995, JÜRGENS et al. 1996) and due to their uptake of  $^{13}\text{C}$ -depleted respired carbon from deeper water layers, are likely to be  $^{13}\text{C}$ -depleted relative to epilimnetic microplankton (DEL GIORGIO & FRANCE, 1996 and references therein). Sporadic measurements of POM taken from different depths in Plußsee in 2001 (SOMMERWERK 2002) and once in June 2004 (0–3 m:  $-22\text{‰}$ , 6–9 m:  $-24\text{‰}$ ) exhibit vertical variation of  $\delta^{13}\text{C}$ . This has also been shown in other lakes (e. g. Lake Lugano, BERNASCONI et al. 1997). In conjunction, the water column oxic boundary is also an active site of methane-oxidation in Plußsee emanating from the anoxic sediments and hypolimnetic water (OBERHAUSER-NEHLS et al. 1994, NUSSLEIN & CONRAD 2000). Ingestion and assimilation of food derived from  $^{13}\text{C}$ -depleted respired  $\text{CO}_2$  and/or methane (BASTVIKEN et al. 2003) most likely accounts for the relatively low  $\delta^{13}\text{C}$  of the hypolimnetic *D. bicuspidatus*, compared to the epilimnetic species. The similar  $\delta^{13}\text{C}$  values of the three copepod species in April 2004 (prior to stratification) suggests that either there is little dietary differentiation between them when the water column is well mixed or that their food items all exhibited similar  $\delta^{13}\text{C}$  due to homogenisation of the inorganic carbon pool. Increasing oxygen deficiency within the hypolimnetic zone in late spring limited the vertical distribution of *T. oithonoides*; in June and July,  $\delta^{13}\text{C}$  values suggest that both *T. oithonoides* and *M. leuckarti* feed mainly in the epilimnion. The higher

$\delta^{15}\text{N}$  of *T. oithonoides* compared to *M. leuckarti* might indicate more predatory feeding, but since their  $\delta^{13}\text{C}$  differed so markedly especially in late summer, then differences in  $\delta^{15}\text{N}$  may simply reflect different baselines. The spatial separation of *D. bicuspidatus* from where we collected our POM prevents us from estimating trophic position of this copepod.

The seasonal fluctuation of the carbon signatures observed in both cyclopoid copepods groups discussed in the current study, and also observed for calanoid copepods and cladocerans throughout 2005 (HARROD & GREY, 2006) is a common feature of productive lakes (see data from Lake Kinneret – ZOHARY et al. 1994, and Lake Suwa – YOSHIOKA et al. 1994). Phytoplankton  $\delta^{13}\text{C}$  depends upon  $\text{CO}_2$  availability, the  $\delta^{13}\text{C}$  of the dissolved inorganic carbon source and cell physiology during  $\text{CO}_2$  uptake. Thus, during summer, phytoplankton with high growth rates in warm surface waters may rapidly utilise  $\text{CO}_2$  in the water column resulting in influx of atmospheric  $\text{CO}_2$  to make up the deficit. Associated high pH affects carbon speciation as well, so the majority of phytoplankton is often  $^{13}\text{C}$ -enriched (RAU et al. 1992). The high production and extent of heterotrophy in the lower water column and associated recycling of carbon via respiration tends to result in more  $^{13}\text{C}$ -depleted values when the water column overturns, and the respired carbon signal tends to dominate over-winter when phytoplankton production is low (MEILI et al. 1996). The amplitude of the zooplankton  $\delta^{13}\text{C}$  seasonal variation in Plußsee is likely exacerbated by small dietary contributions from methanotrophic biomass over-winter, and the high phytoplankton productivity during summer. Interannual variation in zooplankton  $\delta^{15}\text{N}$  is reported from several lakes (MATTHEWS & MAZUMDER 2005 b, PERGA & GERDEAUX 2005); in Plußsee, high  $\delta^{15}\text{N}$  values in summer and low values in winter were observed in adult cyclopoids (this study), in cladocerans and calanoid copepods, but surprisingly not in the cyclopoid bulk samples (HARROD & GREY 2006). This discrepancy might be caused by selective algal feeding of juvenile *Cyclops* stages that were very abundant in the zooplankton bulk samples in spring. The limited POM  $\delta^{15}\text{N}$  data make it difficult to determine whether the seasonal fluctuation in zooplankton  $\delta^{15}\text{N}$  was due to trophic variation (i. e. summer copepods being more predatory compared to winter species) or to baseline variations. We feel it is more probably the latter since cyclical oscillations in seasonal baseline data are frequently observed in productive lakes (BERNASCONI et al. 1997, ZOHARY et al. 1994).

We consider the interspecific differences in trophic position and carbon sources of the sympatric copepod species the most interesting and important result of our study. Food niche differentiation is regarded an important factor in the structuring of zooplankton assemblages (GUISANDE et al. 2003 and references cited therein). However food sources might often be too similar to detect species-specific feeding differences with conventional methods (TURNER

1991) and even if the same material is ingested, it might be digested and assimilated to differing degrees (HART & SANTER 1994). Thus, only highly sensitive methods like SIA or amino acid analysis seem to be appropriate to detect small dietary differences in nature (GUISANDE et al. 2003, PEL et al. 2004) but of course rely upon the isotopic signatures of all the putative food sources being characterised concurrently. Food niche differentiation between copepod species should be especially important as a mechanism to reduce competition when low POC values indicate food limitation, for example in winter or after the clear water phase in early summer (SOMMER et al. 1986). In contrast to calanoid copepods and cladocerans that are restricted to a rather narrow food spectrum, cyclopoid copepods are able to utilise a much broader food spectrum; they probably switch opportunistically between food sources (crustaceans and rotifers to protists, phytoplankton and even bacterial aggregations and detritus) as availability changes within their habitat. This probably contributes considerably to their success in temperate lakes.

## Acknowledgements

We thank S. DEMBECK, H. DEIWICK and H. PAUL-WARDENGA for lake sampling, S. DEMBECK and H. BUHTZ for technical assistance, and C. HARROD for help with data analysis. We are especially grateful to WINFRIED LAMPERT for generous support and creative freedom. It was a great time.

## References

- ADAMS, T. S. & STERNER, R. W. (2000): The effect of dietary nitrogen on trophic level  $^{15}\text{N}$  enrichment. – Limnol. Oceanogr. **45**: 601–607.
- ADRIAN, R. (1991 a): Filtering and feeding rates of cyclopoid copepods feeding on phytoplankton. – Hydrobiologia **210**: 217–223.
- (1991 b): The feeding behaviour of *Cyclops kolensis* and *C. vicinus* (Crustacea, Copepoda). – Verh. Internat. Verein. Limnol. **24**: 2852–2863.
- ADRIAN, R. & FROST, T. M. (1993): Omnivory in cyclopoid copepods: comparison of algae and invertebrates as food for three, differently sized species. – J. Plankton Res. **15**: 643–658.
- ADRIAN, R. & SCHIPOLOWSKI, T. (2003): Bacterial and protozoan mass accumulation in the deep chlorophyll maximum of a mesotrophic lake. – Arch. Hydrobiol. **157**: 27–46.
- BASTVIKEN, D., EJLERTSSON, J., SUNDH, I. & TRANVIK, L. (2003): Methane as a source of carbon and energy for lake pelagic food webs. – Ecology **84**: 969–981.
- BERNASCONI, S. M., BARBIERI, A. & SIMONA, M. (1997): Carbon and nitrogen isotope variations in sedimenting organic matter in Lake Lugano. – Limnol. Oceanogr. **42**: 1755–1765.
- BRANDL, Z. (2005): Freshwater copepods and rotifers: predators and their prey. – Hydrobiologia **546**: 475–489.
- BRANDL, Z. & FERNANDO, C. H. (1981): The impact of predation by cyclopoid copepods on zooplankton. – Verh. Internat. Verein. Limnol. **21**: 1573–1577.

- DAVIS, C. C. (1959): Damage to fish fry by cyclopoid copepods. – *The Ohio J. Sci.* **59**: 101.
- DEL GIORGIO, P. A. & FRANCE, R. L. (1996): Ecosystem-specific patterns in the relationship between zooplankton and POM or microplankton  $\delta^{13}\text{C}$ . – *Limnol. Oceanogr.* **41**: 359–365.
- DEMOTT, W. R. (1986): The role of taste in food selection by freshwater zooplankton. – *Oecologia* **69**: 334–340.
- EINSELE, U. (1965): Ökologische Studien an einer pelagisch lebenden Population von *Diacyclops bicuspidatus* (Crust. Cop.). – *Gewässer und Abwässer* **39**: 40.
- FEUCHTMAYR, H. & GREY, J. (2003): Effect of preparation and preservation procedures on carbon and nitrogen stable isotope determinations from zooplankton. – *Rapid Commun. Mass Spectrom.* **17**: 2605–2610.
- FEUCHTMAYR, H., ZÖLLNER, E., SANTER, B., SOMMER, U. & GREY, J. (2004): Zooplankton interactions in an enclosure experiment: insights from stable isotope analyses. – *Freshwat. Biol.* **49**: 1495–1504.
- FRYER, G. (1957): The food of some freshwater cycloids and its ecological significance. – *J. Anim. Ecol.* **26**: 263–286.
- GREY, J. & JONES, R. I. (1999): Carbon stable isotopes reveal complex trophic interactions in lake plankton. – *Rapid Commun. Mass Spectrom.* **13**: 1311–1314.
- GREY, J., JONES, R. I. & SLEEP, D. (2000): Stable Isotope analysis of the origins of zooplankton carbon in lakes of differing trophic state. – *Oecologia* **123**: 232–240.
- – – (2001): Seasonal changes in the importance of the source of organic matter to the diet of zooplankton in Loch Ness, as indicated by stable isotope analysis. – *Limnol. Oceanogr.* **46**: 505–513.
- GUISANDE, C., BARTUMEUS, F., VENTURA, M. & CATALAN, J. (2003): Role of food partitioning in structuring the zooplankton community in mountain lakes. – *Oecologia* **136**: 627–634.
- HANSEN, A. M. & SANTER, B. (1995): The influence of food resources on the development, survival and reproduction of the two cyclopoid copepods: *Cyclops vicinus* and *Mesocyclops leuckarti*. – *J. Plankton Res.* **17**: 631–646.
- HARROD, C. & GREY, J. (2006): Isotopic variation complicates analysis of trophic relations within the fish community of Plußsee: a small, deep, stratifying lake. – *Arch. Hydrobiol.* **167**: 289–299, this issue.
- HART, R. C. & SANTER, B. (1994): Nutritional suitability of some uni-algal diets for freshwater calanoids: unexpected inadequacies of commonly used edible greens and others. – *Freshwat. Biol.* **31**: 109–116.
- HOPP, U. & MAIER, G. (2005): Implication of the feeding limb morphology for herbivorous feeding in some freshwater cyclopoid copepods. – *Freshwat. Biol.* **50**: 742–747.
- JONES, R. I., GREY, J., SLEEP, D. & ARVOLA, L. (1999): Stable isotope analysis of zooplankton carbon nutrition in humic lakes. – *Oikos* **86**: 97–104.
- JÜRGENS, K., WICKHAM, S., ROTHHAUPT, K. O. & SANTER, B. (1996): Feeding rates of macro- and microzooplankton on heterotrophic nanoflagellates. – *Limnol. Oceanogr.* **41**: 1833–1839.
- KARABIN, A. (1978): The pressure of pelagic predators of the genus *Mesocyclops* (copepoda, crustacea) on small zooplankton. – *Ekologia Polska* **26**: 241–257.

- KARLSSON, J., JONSSON, M., MEILI, M. & JANSSON, J. (2004):  $\delta^{15}\text{N}$  of zooplankton species in subarctic lakes in northern Sweden: effects of diet and trophic fractionation. – *Freshwat. Biol.* **49**: 526–534.
- KLING, G. W., FRY, B. & O'BRIEN, W. J. (1992): Stable isotopes and planktonic trophic structure in Arctic lakes. – *Ecology* **73**: 561–566.
- KRAMBECK, H.-J., ALBRECHT, D., HICKEL, B., HOFMANN, W. & ARZBACH, H.-H. (1994): Limnology of the Plußsee. – In: OVERBECK, J. & CHRÓST, R. J. (eds): *Ecological Studies: Microbial Ecology of Lake Plußsee*. – Springer, New York, pp. 1–23.
- LEHMANN, M. F., BERNASCONI, S. M., BARBIERI, A., SIMONA, M. & MCKENZIE, J. A. (2004): Interannual variation of the isotopic composition of sedimenting organic carbon and nitrogen in Lake Lugano: A long-term sediment trap study. – *Limnol. Oceanogr.* **49**: 839–849.
- MATTHEWS, B. & MAZUMDER, A. (2003): Compositional and interlake variability of zooplankton affect baseline stable isotope signatures. – *Limnol. Oceanogr.* **48**: 1977–1987.
- (2005 a): Temporal variation in body composition (C:N) helps explain seasonal patterns of zooplankton  $\delta^{13}\text{C}$ . – *Freshwat. Biol.* **50**: 502–515.
  - (2005 b): Consequences of large temporal variability of zooplankton  $\delta^{15}\text{N}$  for modeling fish trophic position and variation. – *Limnol. Oceanogr.* **50**: 1404–1414.
- MCCUTCHAN, J. H. Jr., LEWIS, W. M. Jr., KENDALL, C. & MCGRATH, C. C. (2003): Variation in trophic shift for stable isotope ratios of carbon, nitrogen, and sulfur. – *Oikos* **102**: 378–390.
- MEFFERT, M.-E. & OVERBECK, J. (1994): Primary production, photosynthesis, and chlorophyll a in the Plußsee. – In: OVERBECK, J. & CHRÓST, R. J. (eds): *Ecological Studies: Microbial Ecology of Lake Plußsee*. – Springer, New York, pp. 63–80.
- MEIER, B. G. & RECK, E. (1994): Nanoflagellate and ciliate grazing on bacteria. *Microbial Ecology of Lake Plußsee*. – In: OVERBECK, J. & CHRÓST, R. J. (eds): *Ecological Studies: Microbial Ecology of Lake Plußsee*. – Springer, New York, pp. 251–269.
- MEILI, M., KLING, G. W., FRY, B., BELL, R. T. & AHLGREN, I. (1996): Sources and partitioning of organic matter in a pelagic microbial food web inferred from the isotopic composition ( $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$ ). – *Arch. Hydrobiol. Spec. Iss. Adv. Limnol.* **48**: 53–61.
- NÜSSLEIN, B. & CONRAD, R. (2000): Methane production in eutrophic Lake Plußsee: seasonal change, temperature effect and metabolic processes in the profundal sediment. – *Arch. Hydrobiol.* **149**: 597–623.
- OBERHAEUSER-NEHLS, R., ANAGNOSTIDIS, K. & OVERBECK, J. (1994): Phototrophic bacteria in the Plußsee: Ecology of the sulfuretum. – In: OVERBECK, J. & CHRÓST, R. J. (eds): *Ecological Studies: Microbial Ecology of Lake Plußsee*. – Springer, New York, pp. 287–325.
- O'REILLY, C. M., HECKY, R. E., COHEN, A. S. & PLISNIER, P.-D. (2002): Interpreting stable isotopes in food webs: recognizing the role of time averaging at different trophic levels. – *Limnol. Oceanogr.* **47**: 306–309.
- OVERBECK, J. & CHRÓST, R. J. (Editors) (1994): *Microbial Ecology of Lake Plußsee*. – *Ecological Studies* **105**, Springer, New York, pp. 1–392.

- PEL, R., FLORIS, V. & HOOGVELD, H. (2004): Analysis of planktonic community structure and trophic interactions using refined isotopic signatures determined by combining fluorescence-activated cell sorting and isotope-ratio mass spectrometry. – *Freshwat. Biol.* **49**: 546–562.
- PERGA, M. E. & GERDEAUX, D. (2005): 'Are fish what they eat' all year round? – *Oecologia* **144**: 598–606.
- RAU, G. H., TAKAHASHI, T., DES MARAIS, D. J., REPETA, D. J. & MARTIN, H. (1992): The relationship between  $\delta^{13}\text{C}$  of organic matter and  $\text{CO}_2$  (aq) in ocean surface water: Data from a JGOFS site in the northeast Atlantic Ocean and a model. – *Geochim. Cosmochim. Acta* **56**: 1413–1419.
- SANTER, B. (1993): Potential importance of algae in the diet of adult *Cyclops vicinus*. – *Freshwat. Biol.* **30**: 269–278.
- SANTER, B. & LAMPERT, W. (1995): Summer diapause in cyclopoid copepods: adaptive response to a food bottleneck? – *J. Anim. Ecol.* **64**: 600–613.
- SANTER, B. & VAN DEN BOSCH, F. (1994): Herbivorous nutrition of *Cyclops vicinus*: the effect of a pure algal diet on feeding, development, reproduction and life cycle. – *J. Plankton Res.* **16**: 171–195.
- SARVALA, J., BADENDE, S., CHITAMWEBWA, D., JUVONEN, P., MWAPE, L., MÖLSÄ, H., MULIMBWA, N., TARVAINEN, M. & VUORIO, K. (2003): Size-fractionated  $\delta^{15}\text{N}$  and  $\delta^{13}\text{C}$  isotope ratios elucidate the role of the microbial food web in the pelagial of Lake Tanganyika. – *Aquat. Ecosyst. Health & Manage.* **6**: 241–250.
- SOMMER, U., GLIWICZ, Z. M., LAMPERT, W. & DUNCAN, A. (1986): The Plankton ecology group model of seasonal succession of planktonic events in fresh waters. – *Arch. Hydrobiol.* **106**: 433–472.
- SOMMER, F., SAAGE, A., SANTER, B., HANSEN, T. & SOMMER, U. (2005): Linking foraging strategies of marine calanoid copepods to pattern of nitrogen stable isotope signatures in a mesocosm study. – *Mar. Ecol. Progr. Ser.* **286**: 99–106.
- SOMMERWERK, N. (2002): Stabile Isotopenanalyse ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) von drei Copepodenspezies als Indikator trophischer Interaktion. – Diploma Thesis, Ludwig-Maximilians-University, Munich, Germany, 77 pp.
- SPAACK, P. & BOERSMA, M. (2006): Predator mediated coexistence of hybrid and parental taxa. – *Arch. Hydrobiol.* **167**: 55–76, this issue.
- TURNER, J. T. (1991): Zooplankton feeding ecology: Do co-occurring copepods compete for the same food. – *Rev. Aquat. Sci.* **5**: 101–195.
- VAN DEN BOSCH, F. & SANTER, B. (1993): Cannibalism in *Cyclops abyssorum*. – *Oikos* **67**: 19–28.
- VANDERKLIFT, M. A. & PONSARD, S. (2003): Sources of variation in consumer-diet  $\delta^{15}\text{N}$  enrichment: a meta-analysis. – *Oecologia* **136**: 169–182.
- WICKHAM, S. A. (1995): *Cyclops* predation on ciliates: species-specific differences and functional responses. – *J. Plankton Res.* **17**: 1633–1640.
- YOSHIOKA, T. & WADA, E. (1994): A stable isotope study on seasonal food web dynamics in a eutrophic lake. – *Ecology* **75**: 835–846.
- ZOHARY, T., EREZ, J., GOPHEN, M., BERMAN-FRANK, I. & STILLER, M. (1994): Seasonality of stable carbon isotopes within the pelagic food web of Lake Kinneret. – *Limnol. Oceanogr.* **39**: 1030–1043.