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ORIGINAL ARTICLE

Altered complementary feeding strategies of the consumers *Hydrobia ulvae* and *Idotea emarginata* via passive selectivity

N. Aberle · A. M. Malzahn · J. Grey · H. Hillebrand · K. H. Wiltshire

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Abstract This study aimed to identify differences in selectivity, foraging behaviour and complementary feeding of two benthic consumers (the isopod *Idotea emarginata* and the snail *Hydrobia ulvae*) using traditional cell counting as an indicator for algal biomass reduction and stable isotope labelling to detect differences in assimilation and digestion. We hypothesized that even when active feeding preferences of food components are not apparent, passive selectivity via mechanisms such as food assimilation and digestion can be of relevance. Algal biomass was reduced to a similar degree by the grazers independently from grazer and prey combinations without any indication for an active choice of food components. However, the isotope labelling approach indicated that passive selectivity can alter complementary feeding strategies, as we detected shifts in feeding preferences in relation to food quantity and competition. Thus, stable isotope labelling of food components opens up new perspectives in community ecology, allowing assessment of such complex mechanisms as passive selectivity, complementary feeding and competition.

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Keywords Benthic microalgae · Isotope fractionation · Resource partitioning · Microphytobenthos · Stable isotope enrichment

Introduction

Numerous studies have addressed selectivity and competition for microalgae between benthic consumers in aquatic habitats (Steinman et al. 1987; Blanchard 1990; Sommer 1997). This is not surprising since benthic microalgae are considered to be the major food source for benthic consumers in the euphotic zone (Sumner and McIntire 1982; Underwood and Thomas 1990; Hillebrand et al. 2002) and thus, their abundance and species composition are crucial factors for studies in aquatic community ecology.

Complementary foraging strategies and selective resource use of competitors that allow coexistence are aspects which are gaining recognition for studies on grazer–microalgae interaction studies (Fenchel 1975; Hillebrand et al. 2002; Aberle et al. 2005). Wilson et al. (1999) demonstrated that complementary feeding facilitates coexistence and that this is an aspect which commonly regulates consumer–resource systems. In this context the grazers' ability to choose actively between food components (further referred to as "active selectivity") is an important feature that can be significant in influencing grazer–microalgae interactions and is a major force in structuring benthic microalgae community composition (Jaschinski et al. 2008). In addition, passive feeding preferences dependent on differential assimilation and digestion of prey items has also attracted interest and there is evidence to suggest a predominance of passive feeding preferences in grazer–microalgae interactions in contrast to active processes (Steinman 1996; Aberle et al. 2005).

We hypothesize that consumers change their feeding behaviour in the presence of competitors. Following optimum foraging theory it is likely that passive mechanisms like the optimization of assimilation and digestion efficiency are first to be altered before costly behavioural changes were made if two competitors compete for the same food source.

To test this hypothesis we used stable isotope labelling of different food sources in a set of grazing experiments to detect selectivity and complementary feeding strategies of two invertebrate consumers. In recent years, stable isotope labelling approaches have increasingly been used in order to quantify flux processes (Levin et al. 1999; Aberle and Witte 2003), to differentiate between different food sources (Herman et al. 2000) or to detect feeding selectivity (Aberle et al. 2005).

The chain-forming, bentho-pelagic diatom *Fragilaria islandica* and the single-celled, benthic diatom *Nitzschia thermalis* were chosen as food items as both algae occur frequently in benthic habitats along the coasts of Europe and thus represent potential food sources for benthic consumers. In addition, their distinct morphologically differences (chain-forming versus single celled) allowed us to account for potentially different feeding habits of the consumers. Both, the isopod *Idotea emarginata* (Fabricius, 1793) and the gastropod *Hydrobia ulvae* (Pennant, 1777) served as model organisms representing two common, co-occurring benthic consumers which are abundant herbivores in the littoral zones of temperate coastal marine habitats all over Europe. The diet of isopods of the genus *Idotea* spp. and of the snail species *H. ulvae* consists of macro- and microalgae as well as detritus, and selectivity patterns for different algal taxa are known to occur (Blanchard et al. 2000; Sommer 2000; Hagerthey et al. 2002; Orav-Kotta and Kotta 2004). In addition, an overlap in trophic niches is expected as both consumers are known to feed on a variety of food sources, e.g. microalgae and detritus (Franke and Janke 1998; Blanchard et al. 2000; Schanz et al. 2002; Hagerthey et al. 2002).

Methods

Stable isotope labelling

Prior to the experiment, the diatoms *F. islandica* and *N. thermalis* were cultured at 15°C in artificial seawater amended with f/2 medium (Guillard and Lorenzen 1972). The axenic *F. islandica* cultures contained 30% $NaH^{13}CO_2$ (99 atm.%; Chemotrade Leipzig), whereas 30% $NH_4\ ^{15}NO_3$ (95 atm.%; Chemotrade Leipzig) was added to the cultures of *N. thermalis*. The algae were cultivated in 500 ml Erlenmeyer flasks under a 16 h light:8 h dark regime for 10 days.

Culture conditions of invertebrates

In the preliminary stage of the experiment, the invertebrates were cultured separately under natural temperature and light conditions, using tanks with running seawater stocked with macroalgal thalli and associated flora from the Helgoland rocky shore. Twenty-four hours before the experiment the grazers were sorted by size class and held without any food at 15°C.

Experimental design

Experiments with *H. ulvae* (shell height 1 mm) and *I. emarginata* (total length 3 mm) were conducted using four different treatments: (1) a control treatment without grazers (C), (2) single-grazer treatments with *I. emarginata* (I), (3) single-grazer treatments with *H. ulvae* (H), (4) a combined-grazer treatment (HI).

Each treatment was replicated four times and the entire experimental set-up was duplicated to allow independent sampling for two different incubation times (d1 = one day; d2 = two days). After one day half of the setup was harvested and after the second day the rest was harvested.

Incubations of short time period were chosen in order to detect natural feeding preferences immediately and to avoid adaptive feeding that might occur over time.

Erlenmeyer flasks (300 ml) served as experimental units and were filled with 100 ml filtered (0.2 µm) and autoclaved North Sea water. Each culture flask in each treatment was inoculated with a mixed, labelled algal solution containing 6 ml of *N. thermalis* (small cells, single-celled algae; 36,000 cells ml^{-1}) and 20 ml of *F. islandica* (large cells, chain-forming colonies; 2,500 cells ml^{-1}). The different initial volumes of algal solution ensured a comparable biovolume of each algal species in the treatments as the biovolume of a single cell of *N. thermalis* was 200 μm^3 and that of *F. islandica* was four times higher (800 μm^3).

Algae were allowed to settle before grazers were added. Grazer addition followed a supplementary design whereby the grazer biomass in each treatment was constant. The number of individuals added to each experimental unit was calculated from their individual dry weights, i.e. (1) eight *I. emarginata* with 5.2 mg total dry weight in treatment I, (2) 24 *H. ulvae* with 5.1 mg total dry weight in treatment H, (3) four *I. emarginata* and 12 *H. ulvae* with 5.2 mg total dry weight in the mixed-grazer units HI.

Sample processing

The animals were picked live from the flasks, washed with distilled water and oven dried at 60°C for 24 h. Snail body

tissues were removed from their shells after treating with 1 M HCl-solution.

To collect faecal pellets for the measurement of ^{13}C and ^{15}N egested by the animals, the remaining suspension was sieved through a 50 µm-gauze and the sieve-residues collected on a pre-combusted GF/F-filter. The residues were checked under a binocular microscope to ensure that only faecal pellets were retained on the filters. Faecal pellet material from all four replicates of each treatment was pooled to obtain sufficient material for stable-isotope analyses (approx. 0.3–0.4 mg).

For the determination of cell numbers and biovolume, 10 ml of the algal suspension were transferred into brown-glass bottles and preserved with Lugol's solution.

Prior to counting, the bottles were mixed gently and 10 ml of samples were transferred immediately to Utermöhl counting chambers (total volume 10 ml). After settlement of the sample for 24 h, algal cells were counted under an inverted microscope and converted to biovolume following the methods of Hillebrand et al. (1999). Grazing rate was calculated separately for each diatom species, based on differences in their biovolume. The implication of this method is that each herbivore has two different feeding rates, one for each diatom taxon. Grazing rate per hour was calculated from the difference between the gross growth rate $\mu = (\ln V_c - \ln V_0) \times h^{-1}$) and the net growth rate $r = (\ln V_{gr} - V_0) \times h^{-1}$ (V_c = biovolume of controls; V_0 = biovolume at start; V_{gr} = biovolume of grazer treatments all at the end of the experiment).

Stable isotope analyses

Individual *I. emarginata* were weighed into tin cups, whereas two or three individual *H. ulvae* were pooled to obtain a sufficient mass of nitrogen for analyses. Tin cups were oxidised in a Carlo Erba NA 1500 elemental analyser coupled to a Micromass IsoPrime continuous-flow isotope ratio mass spectrometer. Isotope ratios are expressed using the standard delta notation ($\delta^{13}C$, $\delta^{15}N$) given in per mil (‰). The reference materials used were atmospheric nitrogen and, for carbon, a secondary standard of known relation to the international standard of Vienna Pee Dee belemnite. Repeat analyses of an internal standard resulted in typical precision and accuracy of <0.2‰ for $\delta^{13}C$ and <0.4‰ for $\delta^{15}N$. Uptake of ^{13}C (and similarly ^{15}N) by the herbivores was calculated as excess above background and is expressed as specific uptake $\Delta\delta^{13}C$ ($\Delta\delta^{13}C = \delta^{13}C_{sample} - \delta^{13}C_{background}$). Thus, prior to the labelling experiment, background (natural abundance) isotope signatures of each grazer species were measured to substitute into the calculation of specific uptake. A selectivity index (Q) was defined as the quotient $\Delta\delta^{13}C/\Delta\delta^{15}N$, which expresses the relative uptake of ^{13}C compared to the uptake of ^{15}N.

Statistical analyses

To test for a significant impact of herbivores on algal biomass, a full-factorial ANOVA was used. Independent factors comprised time (d1, d2) and treatment (C, H, I, HI). The uptake by both grazers were analysed separately using a full-factorial ANOVA with the dependent variables ^{13}C- or ^{15}N-uptake and the independent factors time (d1, d2) and species combination (single, mixed). No transformation was necessary for the ^{13}C- and ^{15}N-uptake data as the variances showed no significant deviation from homogeneity. We tested for a relationship between the biomass-specific grazing rate and ^{13}C- and ^{15}N-uptake using linear regression analysis. In addition, an ANOVA on selectivity was performed using the untransformed dependent variable Q ($^{13}C/^{15}N$) and the independent factors time (d1, d2) and species combination (single, mixed). As for ^{13}C- and ^{15}N-uptake both grazers were analysed separately.

Results

Algal biovolume

Both grazers reduced the biovolume of the two algal species significantly throughout the incubation (Table 1). As for *N. thermalis*, the biovolume of *F. islandica* increased in the controls, but showed a significant decline in all grazer treatments (Fig. 1a; $P < 0.05$; Table 1). Grazer presence reduced the biovolume of *F. islandica* to almost zero already after one day while grazing on *N. thermalis* led to a reduction of 55–87% (day 1) and on day 2 by 72–91% (day 2) in algal biomass. No significant difference occurred between grazer species. Grazing rates on *F. islandica* ranged from 0.14 to 0.34 μm^2 biovolume h^{-1} and from 0.09 to 0.16 μm^3 biovolume h^{-1} for *N. thermalis*.

Isotope signatures of cultured algae

At the beginning of the in situ labelling experiment the isotope signatures of the labelled cultures exhibited isotope signatures of 11,248‰ ($\delta^{13}C$) and 4.7‰ ($\delta^{15}N$) for *F. islandica* while *N. thermalis* cultures showed values of −17.6‰ ($\delta^{13}C$) and 47,750‰ ($\delta^{15}N$).

Background isotope signatures of invertebrates

Similar natural isotope compositions were detected for both grazers. Values for *H. ulvae* $\delta^{13}C$ were slightly lower (mean −20.0 ± 0.3‰) than those of *I. emarginata* (−16.7 ± 0.3‰), whereas $\delta^{15}N$ values of *H. ulvae* (13.5 ± 0.6‰) and *I. emarginata* (13.0 ± 0.3‰) were isotopically indistinct.

Table 1 Grazing on *N. thermalis* + *F. islandica*

	df	MS	*F* ratio	*P* level
Grazer effect on *N. thermalis*				
Time	1	6.8167E + 13	183.45	0.0000
Treatment	3	1.7503E + 14	471.05	0.0000
Time × treatment	3	7.0903E + 13	190.81	0.0000
Error	24	3.7158E + 11		
Grazer effect on *F. islandica*				
Time	1	1.1060E + 14	38.014	0.0000
Treatment	3	4.8699E + 14	167.38	0.0000
Time × Treatment	3	1.1309E + 14	38.867	0.0000
Error	24	2.9096E + 12		

Results of a full factorial ANOVA for total algal biovolume, with time (d1, d2) and treatment (C, H, I, HI) as independent factors, and total biovolume as dependent variable

Uptake of ^{13}C and ^{15}N

The $\Delta\delta^{13}$C of *H. ulvae* showed no significant variations over time and between species combination whereas for *I. emarginata* a significant effect of species combination was shown (Fig. 2a; $P < 0.05$; Table 2a). Mean specific uptake of *N. thermalis* led to ^{15}N-enrichment of both grazers and $\Delta\delta^{15}$N-values of *H. ulvae* and *I. emarginata* showed no significant effect of time and species combination (Fig. 2b; Table 2b). No correlation was found between biomass-specific grazing rates and stable isotope-uptake, either for different treatments or for incubation time. The only positive correlation was found on day 2: between ^{13}C-uptake and the biomass-specific grazing rate ($P < 0.05$).

Faecal pellets

Stable isotope analyses of faecal material revealed distinctive signatures for the pellets of both grazer species. All signatures from the faecal pellet material showed a higher degree of enrichment on the second day of the experiment when compared to the first day (Fig. 3). Faecal pellets of *I. emarginata* in the single-grazer treatments (I) were enriched in ^{15}N- and ^{13}C, although, to a three times lesser extent than those of *H. ulvae*, as a single grazer (H). An intermediate degree of enrichment was found in the combined grazer treatments (HI).

Selectivity *Q*

The selectivity index *Q* ($\Delta\delta^{13}$C/$\Delta\delta^{15}$N) showed similar values for *I. emarginata* and *H. ulvae* (Fig. 4; Table 3). *I. emarginata* showed a significant decline in *Q* between day 1 and day 2 ($P = 0.05$), both in the single- and in the mixed-grazer treatments. Thus, a significant change in feeding preferences from day 1 to day 2 was detected for the isopod species, with a higher uptake of *F. islandica* at the beginning of the experiment. Differences in selectivity over time and between the single- and mixed-grazer treatments of *H. ulvae* were not detected (Fig. 4; Table 3). In contrast, the *Q*-values for *I. emarginata* differed significantly between the single- and mixed-grazer treatments ($P < 0.05$; Fig. 4; Table 3). After the first and after the

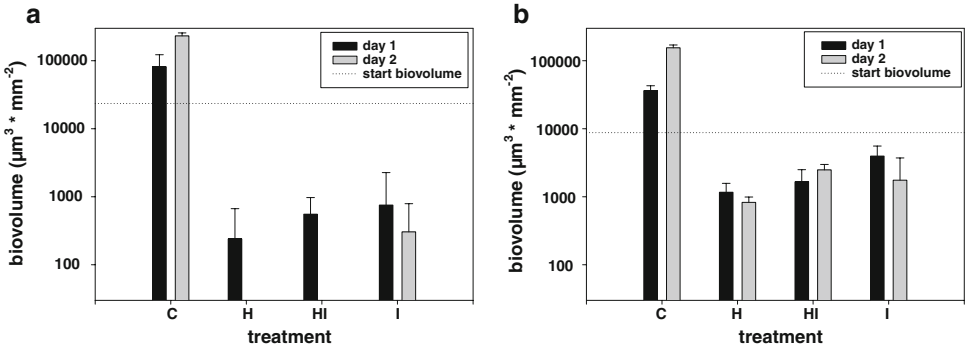

Fig. 1 Biovolume (mean ± SE) of (**a**) *F. islandica* and (**b**) *N. thermalis* in control (*C*), single-grazer with *H. ulvae* (*H*) and *I. emarginata* (*I*), and mixed grazer treatments (*HI*) on day 1 and day 2 of incubation

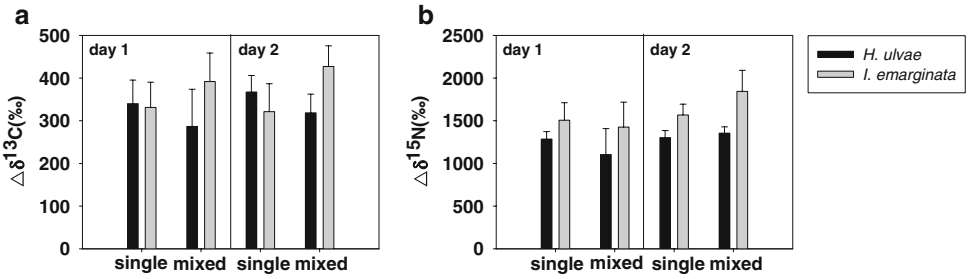

Fig. 2 **a** $\Delta\delta^{13}$C (mean ± SE) and **b** $\Delta\delta^{15}$N (mean ± SE) of *H. ulvae* and *I. emarginata* in the single- and mixed-grazer treatments after day 1 and day 2 of the incubation

Table 2 ^{13}C and ^{15}N-uptake by (a) *H. ulvae* and (b) *I. emarginata*

	df	MS	*F* ratio	*P* level
(a) *H. ulvae*				
^{13}C-uptake				
Time	1	6859	1.1951	0.2825
Species combination	1	13,900	2.4220	0.1295
Time × species combination	1	11	0.0019	0.9653
Error	32	5,739		
^{15}N-uptake				
Time	1	115,397	2.2530	0.1432
Species combination	1	14,243	0.2781	0.6016
Time × species combination	1	81,194	1.5852	0.2171
Error	32	51,219		
(b) *I. emarginata*				
^{13}C-uptake				
Time	1	972	0.1184	0.7327
Species combination	1	74,874	9.1134	0.0045
Time × species combination	1	6,338	0.7715	0.3853
Error	38	8,216		
^{15}N-uptake				
Time	1	510,630	3.0833	0.0872
Species combination	1	156,080	0.9425	0.3378
Time × species combination	1	362,286	2.1876	0.1474
Error	38	165,610		

Results of a full factorial ANOVA for tracer uptake, with time (d1, d2) and species combination (single, mixed) as independent factors and total ^{13}C- or ^{15}N-uptake as dependent variables

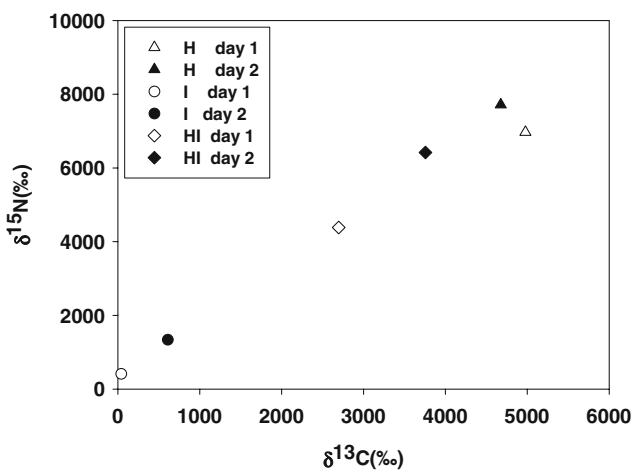

Fig. 3 δ^{13}C and δ^{15}N of faecal pellets (single measurements) in single-grazer (*H* and *I*) and mixed grazer treatments (*HI*) on day 1 and day 2 of the incubation

second day of the incubation, *I. emarginata* alone showed lower *Q*-values than those of the isopods in the mixed-grazer treatments.

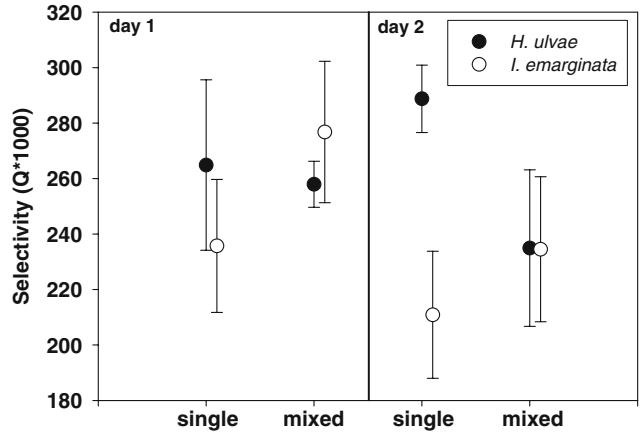

Fig. 4 Selectivity index *Q* ($\Delta\delta^{13}$C/$\Delta\delta^{15}$N × 1,000) of *H. ulvae* and *I. emarginata* in single-grazer (*H* and *I*) and mixed grazer treatments (*HI*) on the first (day 1) and the second day (day 2) of incubation (mean ± SE)

Table 3 Selectivity *Q*

	df	MS	*F* ratio	*P* level
Q for *I. emarginata*				
Time	1	11,225	4.8639	0.0335
Species combination	1	11,993	5.1964	0.0283
Time × species combination	1	725	0.3140	0.5785
Error	38	2,308		
Q for *H. ulvae*				
Time	1	44	0.028	0.8671
Species combination	1	4905	3.200	0.0831
Time × species combination	1	4,081	2.662	0.1126
Error	32	1,533		

Results of a full-factorial ANOVA for selectivity with time (d1, d2) and species combination (single, mixed) as independent factors and *Q* ($\Delta\delta^{13}$C/$\Delta\delta^{15}$N) as dependent variable

Discussion

Algal biovolume

A decrease in cell number of microphytobenthic or epiphytic communities in the presence of invertebrate grazers is a well-known phenomenon which has been detected in numerous studies (Sommer 1997; Hillebrand et al. 2000; Schanz et al. 2002; Jaschinski et al. 2008). In our experiment, both algal species were grazed to a similar degree by the studied grazers indicating that no active choice of food components took place.

Uptake of ^{13}C and ^{15}N

The $\Delta\delta^{13}$C and $\Delta\delta^{15}$N values for both invertebrates indicated a rapid uptake of ^{13}C and ^{15}N from the labelled

F. islandica and *N. thermalis*. In general it is agreed that the fractionation of ^{13}C between trophic levels is much smaller than for ^{15}N (Peterson and Fry 1987). Fractionation of carbon and nitrogen derive from a preference for the lighter isotope during assimilation, protein synthesis as well as during excretion and catabolic processes (Macko et al. 1982; Ponsard and Averbuch 1999) and consequently, the consumers' tissues show higher stable isotope ratios then their diets. The accumulation of carbon and nitrogen heavy isotopes greatly exceeded the trophic fractionation usually found for natural isotope studies which can be explained by the high level of enrichment of the *F. islandica* and *N. thermalis* cultures ingested.

The changes in feeding preference that we observed were based on tracer uptake, rather than algal biomass determination and provide evidence that passive selection can occur even if active selection does not. Since algal biovolumes declined in the single- and mixed-grazer treatments, it is assumed that differential uptake of algal material resulted from different digestion efficiencies, e.g. by taking up predominately lipids from their diets while excreting unnecessary components such as cell walls. Our data confirm that the actual abundance of grazed algal cells did not automatically reflect the actual amount of digested material (see also Underwood and Thomas 1990; Brendelberger 1997a).

Stable isotope data obtained for *H. ulvae* and *I. emarginata* in the single-grazer treatments showed very similar ^{13}C- and ^{15}N-uptakes. These results indicate that as long as there is no co-occurring grazer present, both invertebrates showed similar assimilation and digestion efficiencies to both microalgal species. Although it is known that microalgal cell sizes, structures and morphotypes can influence digestive pathways (Moore 1975; Underwood and Thomas 1990), such size- or morphotype-dependant effects were not detected in the single-grazer treatments of our experiment. However, in the mixed-grazer treatments *I. emarginata* in general showed a higher uptake of the labelled algal material as *H. ulvae*. Many studies in community ecology have tried to analyse the effects of complementary feeding and coexistence on the resource use of competitors (Rossi et al. 1983; Wilson et al. 1999). In our study, interspecific competition appeared to induce a shift in assimilation efficiency leading to a stronger uptake of the algal material by *I. emarginata*. A possible explanation for the change in resource use by *I. emarginata* in the presence of *H. ulvae* could be related to an increased enzymatic activity in the case of coexistence. In this context a common phenomenon is the interchange of digestive enzymes between species as a result of coprophagy (Brendelberger 1997a). If faecal pellets of coexisting species are used as supplementary food sources, the enzyme activities of the consumers can be stimulated (Brendelberger 1997b). *I. emarginata* may have taken up faecal material together with associated bacteria and their enzymes from the coexisting *H. ulvae*, and the supplementary enzymes might have led to a more efficient uptake by *I. emarginata*. Thus, it can be speculated that complementary feeding as a result from passive selectivity took place. Furthermore, this assumption is supported by the fact that, in terms of ^{13}C-uptake, there were not only inter-specific differences between the assimilation and digestion efficiencies of *I. emarginata* and *H. ulvae* in the mixed-grazer treatments, but that there were also intra-specific differences between *I. emarginata* from the single- and the mixed-grazer treatments. Therefore, passive selectivity as a result of higher uptake efficiencies may occur between both grazer species even when active selectivity patterns are not detectable from biovolume data. Thus, the combination of traditional biomass estimates with stable isotope labelling techniques seems to be a promising approach for the detection of passive selectivity and complementary feeding strategies.

Faecal pellets

Mean faecal pellet $\delta^{15}N$ from each treatment reached a value of 4,540‰ and for $\delta^{13}C$ a mean of 2,793‰ was detected. When the $\Delta\delta^{13}C$ and $\Delta\delta^{15}N$ values for the animals are compared to the isotope composition of their faecal pellets, it is apparent that substantial ^{15}N- and ^{13}C-accumulation occurred in the faecal pellets indicating a strong fractionation towards the lighter isotopes during gastrointestinal assimilation. Gorokhova and Hansson (1999) reported similar results for mysid shrimps, pointing at an accumulation of the heavy isotopes ^{13}C and ^{15}N in their faeces. Another possible explanation for the high accumulation of ^{15}N and ^{13}C could be the result of differential digestion, e.g. by taking up essential components from their diets while excreting unessential ones. However, the proportion of heavy isotopes accumulated during assimilation and protein synthesis was still higher than in natural isotope studies due to the high initial label of the algal diets.

Both heavy isotopes accumulated in the faeces of the invertebrate species to different degrees, suggesting different magnitudes of kinetic isotope fractionation during chemical and biochemical reactions of *H. ulvae* and *I. emarginata*. In general, gastrointestinal assimilation is considered to be the first step in trophic fractionation (Gorokhova and Hansson 1999) and isotopic composition of faecal material therefore can provide valuable dietary information which would otherwise be overlooked.

Even though the causative factors are difficult to define, the inter-specific variability in isotope fractionation is becoming more widely recognised (Gannes et al. 1997). Few experimental studies address such variations. However, differences in isotope discrimination are known to

occur between algal groups and species (Burkhardt et al. 1999; Needoba et al. 2003) and studies on natural stable isotope signatures of metazoans have shown that fractionation can be variable as well as species-specific (Macko et al. 1982; Vander Zanden and Rasmussen 2001; Post 2002; Aberle and Malzahn 2007). Possible explanations for species-specific discrimination of heavier isotopes include differences in metabolic processes (assimilation, excretion), different levels of isotope enrichment, the diets' and consumers' nutrient content as well as the degree of starvation (Gorokhova and Hansson 1999; Adams and Sterner 2000; Vanderklift and Ponsard 2003). Our results suggest that the gastropod *H. ulvae* discriminated against the heavier isotopes more strongly than the isopod *I. emarginata*, thus providing further support for species-specific fractionation processes (Vander Zanden and Rasmussen 2001; Post 2002).

Selectivity *Q*

Interpretation of $\Delta\delta^{13}$C and $\Delta\delta^{15}$N values requires care since the initial degree of isotope enrichment of the two algal species was different. This makes it difficult to draw direct comparisons between ^{13}C- and ^{15}N-uptakes and to treat the values derived from isotope uptake as absolute values. Thus, direct comparisons should only be drawn between treatments rather than between the δ^{13}C and δ^{15}N values. In order to avoid inaccurate direct comparisons between δ^{13}C and δ^{15}N signatures, we applied the selectivity index *Q* which represents a ratio between both signatures and evaluates the relative importance of each diatom species in the diet of each grazer. Thus, relative shifts in preference can be determined which overcome difficulties inherent in comparing δ^{13}C values directly with δ^{15}N values. It was not possible to apply two-end-member-mixing models (see e.g. Vander Zanden and Rasmussen 2001; Post 2002) as our experiment was too short to assume a complete turnover of the animal tissues and an isotopic equilibrium between the consumer and its diet. Moreover, gut contents were included in the analyses, thus preventing the use of such a model. Thus, we used the selectivity index *Q* instead, even though it does not incorporate a fractionation factor.

For the isopod *I. emarginata* in the single grazer treatments a significant effect of time was detected, indicating that this species consumed a higher percentage of *F. islandica* during the first day of incubation and switched to a *N. thermalis*-based diet on the second day. The shift from one food source to the other in case of *I. emarginata* can be explained by changes in relative amounts of each alga available, and the increased effort in consuming *F. islandica* compared to *N. thermalis*. Since the biovolume of each algal species had already declined significantly by day 1 it seems likely that consuming the more uniformly distributed single-celled diatom *N. thermalis* was a better feeding strategy than having to scavenge actively to find the few remaining colonies of *F. islandica*. Thus, as long as large amounts of different algae were available, *I. emarginata* actively chose the preferred prey species and as soon as food became limited a rather unselective but more efficient feeding strategy was chosen. The correlation between food concentration and selectivity is a well-known phenomenon in planktonic systems (DeMott 1995; Boenigk et al. 2002) and in line with optimum foraging theory (MacArthur and Pianka 1966). Thus, we assume that concentration-dependent shifts in preference have led to a switch in resource use of *I. emarginata* in our experiment.

In addition, on the second day of the experiment *I. emarginata* showed different feeding preferences as a single grazer than in the presence of its competitor *H. ulvae* (mixed grazer treatment). As a single grazer *I. emarginata* showed a preference for *N. thermalis* while when both invertebrates had to share food sources, *I. emarginata* consumed a higher share of *F. islandica*. Thus, the presence of *H. ulvae* induced a shift in resource use of *I. emarginata*, indicating complementary feeding behaviour.

Many studies in community ecology have investigated the effect of coexistence on the resource use of competitors and demonstrated that complementary feeding facilitates coexistence (Ricklefs and Schluter 1993; Wilson et al. 1999). In our study interspecific competition appeared to induce a passive shift in feeding preference and in resource use by *I. emarginata* in the presence of *H. ulvae*. These results are similar to investigations on freshwater grazer–microalgae interactions published by Aberle et al. (2005) and it can be speculated that this phenomenon is related to the adaptive potential of digestive enzymes. As already pointed out in the section on "^{13}C- and ^{15}N-uptake", the shift in feeding preference of *I. emarginata* might be related to an increase in enzymatic activity as a result of taking up faecal pellet material from coexisting species which enables an interchange of digestive enzymes between species (Brendelberger 1997b). Thus, the potential consumption of faecal pellets, associated bacteria and their enzymes from the coexisting *H. ulvae* might have led to a more efficient uptake of *F. islandica*-cells by *I. emarginata*.

The observed changes in resource use and the ability of consumers to develop complementary feeding strategies in case of coexistence were based on tracer uptake rather than algal biomass determination. Our data on ^{13}C- and ^{15}N-uptake as well as on the selectivity index *Q* support findings showing that the degrees to which microalgae are consumed by a grazer do not automatically reflect the actual amount of digested material (Underwood and Thomas 1990; Brendelberger 1997a). By using this isotope labelling approach we were able to provide evidence that consumer–

resource systems not only depend on active foraging activities but that passive mechanisms such as assimilation and digestion can affect a systems' community ecology considerably. Thus, analyses of the mechanisms that regulate feeding preferences, complementary feeding and species coexistence should always consider the system as a whole by including not only the active choice of food components but also passive selectivity.

Conclusions

By using differential labelling of algal food with stable isotopes, we were able to detect differences in active and passive selectivity of two co-occurring species. Active choice of food items as a result of morphological or size-dependent features of algal prey was not observed. In contrast, passive feeding preferences were shown, thus, revealing new mechanisms that can alter complementary feeding strategies and coexistence.

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