

LETTER

Species sorting and stoichiometric plasticity control community C:P ratio of first-order aquatic consumers

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Abstract

Ecological stoichiometry has proven to be invaluable for understanding consumer response to changes in resource quality. Although interactions between trophic levels occur at the community level, most studies focus on single consumer species. In contrast to individual species, communities may deal with trophic mismatch not only through elemental plasticity but also through changes in species composition. Here, we show that a community of first-order consumers (e.g. zooplankton) is able to adjust its stoichiometry (C:P) in response to experimentally induced changes in resource quality, but only to a limited extent. Furthermore, using the Price equation framework we show the importance of both elemental plasticity and species sorting. These results illustrate the need for a community perspective in ecological stoichiometry, requiring consideration of species-specific elemental composition, intraspecific elemental plasticity and species turnover.

Keywords

Community C:P, ecological stoichiometry, food quality, C:P ratio, price equation, stoichiometric mismatch.

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INTRODUCTION

Several main drivers of global change, such as eutrophication and increasing atmospheric CO₂-concentrations, result in strong alterations of the amounts and ratios of essential elements available to ecosystems (Falkowski *et al.* 2000; Van De Waal *et al.* 2010). Ecological stoichiometry (Sterner & Elser 2002; Hessen *et al.* 2013) represents a powerful research avenue that may contribute to a better understanding of how such anthropogenic alterations may affect the functioning of ecosystems (Elser *et al.* 2009). Although ratios of elements such as carbon (C), nitrogen (N) and phosphorus (P) vary widely in nature, the elemental composition of organisms is confined more strictly. Organisms tend to be stoichiometrically homeostatic because they are composed of biomolecules with specific elemental ratios (e.g. proteins, lipids, carbohydrates and nucleic acids) and because they need to create a stable internal environment suitable for essential cellular processes (Meunier *et al.* 2014). Nevertheless, primary producers are known to be considerably more flexible in their C:N:P ratios than heterotrophs (Sterner & Elser 2002; Persson *et al.* 2010). Such plasticity may lead to mismatches with the elemental composition of consumers, reduce consumer performance and have implications for population dynamics and food web interactions (Andersen *et al.* 2004; Hall *et al.* 2004; Hessen *et al.* 2013). In addition, high stoichiometric flexibility of producers may affect a variety of stoichiometry-regulated ecosystem functions, such as biogeochemical cycling and carbon sequestration (Mack *et al.* 2004; Dickman *et al.* 2006; Sistla *et al.* 2013). For this reason, there has been a long tradition of research on

aspects of producer stoichiometric plasticity, including its main drivers (Goldman *et al.* 1979; Klausmeier *et al.* 2004) and potential ecosystem consequences (Sterner *et al.* 1997; Dickman *et al.* 2006; van Donk *et al.* 2008; Mette *et al.* 2011; Sardans *et al.* 2012; Sistla & Schimel 2012; Plum *et al.* 2015).

So far, considerably less attention has been given to the potential roles of stoichiometric plasticity of consumers. An important reason for this is that consumers have traditionally been considered as being fixed in their elemental ratios (Andersen & Hessen 1991; Sterner & Elser 2002; Andersen *et al.* 2004). This view has been challenged by studies that demonstrate stoichiometric plasticity of primary consumers when exposed to nutrient-limited food (DeMott & Pape 2005; Persson *et al.* 2010). Such observations have recently sparked an interest in the potential consequences of consumer plasticity for ecosystem functioning. Along with changes in biochemical quality (e.g. composition of fatty acids or sterols), plasticity in the elemental composition of consumers may be an important factor affecting the efficiency of energy transfer to higher food levels (Malzahn *et al.* 2007; Boersma *et al.* 2008; Rowland *et al.* 2015). Such quality-driven bottom-up effects may affect the biomass and productivity of higher trophic levels and ultimately determine important ecosystem services like fish or lobster production (Dickman *et al.* 2008; Schoo *et al.* 2012). Consumers play a central role as nutrient recyclers and have the potential to influence the biomass, stoichiometry and community composition of producers through grazing and via regulation of nutrient supply (Elser *et al.* 2000, 2001; Vanni 2002; Hall 2009). The relative rates at which elements are excreted by consumers are largely dictated

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by the mismatch between the stoichiometric requirements of consumers and the elemental composition of their food source (Sterner 1990; Elser & Urabe 1999). The question of how nutrient-mediated feedback mechanisms between grazers and producers may be altered by elemental plasticity of the consumer communities is still poorly explored empirically, although recent theoretical work has pointed out its potential importance (Mulder & Bowden 2007; Wang *et al.* 2012).

Theoretical and experimental studies on the consequences of plasticity in consumer stoichiometry so far have mostly emphasised plasticity of single species in linear food webs (Plum *et al.* 2015). Evaluation of stoichiometric constraints for ecosystem functioning nevertheless requires consideration of stoichiometric plasticity at the level of entire communities. Upscaling from organismal to community level requires consideration of the fact that communities may consist of species with different elemental composition, both constitutively as well as in terms of their plasticity (Andersen & Hessen 1991; Persson *et al.* 2010). Changes in the elemental limitation of the food source may affect the relative performance of species in a community, which by itself may be function of body elemental composition (Danger *et al.* 2008). For example, taxa with high growth rates need to contain high levels of P in order to sustain high rates of protein synthesis via P-rich ribosomes (Elser *et al.* 1996; Main *et al.* 1997). Compared to low P consumer taxa, they are expected to have superior performance when feeding on P-rich food, but suffer stronger fitness reductions when food is P deficient. For this reason the prediction is that increases in the P content of the food source will result in community compositional shifts towards a dominance of fast-growing P-rich consumers, whereas reductions in food P content are expected to result in relative increases in P-poor consumers (Sterner *et al.* 1997; Hall *et al.* 2004; Urabe *et al.* 2010). Stated more generally, along a gradient of environmental change, the stoichiometric response of a trophic level to a change in the stoichiometry of its resources is not only function of the elemental plasticity at the level of its individual constituent species but also of potential shifts in species composition along the gradient (Schade *et al.* 2005; Dickman *et al.* 2006; Plum *et al.* 2015). Consequently, for a given trophic level, pronounced among-species variability in elemental composition may result in a degree of elemental plasticity that is larger than predicted from phenotypic variability at the organismal level alone (see also Danger *et al.* 2008).

Surprisingly, few attempts have been made to combine information on within- and among-species variability in the analysis of community-level elemental ratios. Probably, this is due to lack of suitable datasets and an appropriate analytical framework that allows the simultaneous evaluation of the role of species-level phenotypic responses and community compositional change. One such framework may be found in Price's theorem that was recently extended for use in research on the relationship between biodiversity and ecosystem functioning (e.g. Loreau & Hector 2001; Winfree *et al.* 2015). The extended Price equation (Fox 2006; Fox & Kerr 2012) is a modified version of the original Price equation (Price 1970) and is developed to explain variation in an observed ecosystem function among a pair of sites by species gains, species losses and species' specific context-dependent contributions to

the function. Actually, it can be applied to any additive community property, such as total community C and P content, and be used for the analysis of community elemental ratios (e.g. C:P). As such, the extended Price equation provides a promising tool for the study of the mechanisms underlying changes in stoichiometry at the community level. Here, we demonstrate its value for the analysis of community elemental plasticity with data of zooplankton herbivore communities that were experimentally exposed to a gradient of seston stoichiometry in the absence of predators. In outdoor mesocosms, we created different levels of seston C:P by manipulating P supply and light intensity in a multifactorial design. Our experiment is unique in that we assessed zooplankton community composition simultaneously with the elemental composition of all individual constituent species in each of the experimental treatments. Through application of the Price framework, our approach allows us to address the extent of community-level elemental plasticity and to evaluate the impact of its major underlying drivers, i.e. organismal plasticity, and community assembly processes such as species' losses and gains and changes in the relative abundance of species.

METHODS

Mesocosm experiment

In June of 2014 twenty outdoor mesocosms were filled with tap water (180 L). The experimental design consisted of a 2×2 factorial combination of light and phosphorus (P), with 5 replicates per multifactorial combination. The goal of these experimental manipulations was to create variation in the stoichiometry of primary producers (phytoplankton). The factor levels for light were full day light ('Full light') or shade ('Shade'). Shade was created by placing layers of cloth over the mesocosm which reduced incoming photosynthetic active radiation (PAR) by 70%. The factor levels of P consisted of high (HP) or low (LP) P availability. At day 1, HP mesocosms initially received 71.03 $\mu\text{mol N/L}$ (0.995 mg N/L) and 14.20 $\mu\text{mol P/L}$ (0.44 mg P/L); LP mesocosms received 71.03 $\mu\text{mol N/L}$ and 1.420 $\mu\text{mol P/L}$. This corresponded to initial N:P molar ratios of 5 : 1 in HP and 50 : 1 in the LP mesocosms. Throughout the experiment, 10% of the initial nutrient amounts were added twice a week to compensate for nutrient loss through sedimentation in the system (Hall *et al.* 2004). The experiment ran for a total of 11 weeks from 30-06-2014 to 12-09-2014. The mesocosms were inoculated with phytoplankton and zooplankton from six water bodies on day 6 and day 13, respectively, yielding a total mesocosm volume of 183.5 L. Seston and zooplankton samples were collected at the end of the experiment (day 74). Cladocerans were counted and identified at the species level, copepods at the order level. In addition, we measured the elemental C and P content of the seston and of each individual zooplankton species. For detailed information about experimental set-up, sampling and sample analysis, see Supplement 1.

Calculation of total C, total P and community C:P ratio

We calculated total zooplankton community C and P content in each mesocosm as $C = \sum_i^n \bar{C}_i \cdot N_i$ and $P = \sum_i^n \bar{P}_i \cdot N_i$ with

N_i being the population density and \bar{C}_i and \bar{P}_i being the mean individual C and P content of species i , respectively. The two components were used to calculate the community C:P ratio.

Explaining changes in total community C and P by compositional turnover and elemental plasticity

The main aim of our analysis was to investigate the response of zooplankton community C:P to increasing seston C:P and evaluate if such response was caused by species turnover or by plasticity of species-specific elemental ratios. To this end, we applied the extended version of the Price equation by Fox & Kerr (2012). Application of this equation mathematically partitions the change in any additive community property among two mesocosms into at least three independent components (Fox & Kerr 2012): (1) a 'species richness effect' of gained and lost species ($SRE = SRE_G + SRE_L$) which represents the extent by which the community property has changed as a result of random gain and loss of species, independent of the identity or traits of these species; (2) a 'species composition effect' of gained and lost species ($SCE = SCE_G + SCE_L$) which quantifies the extent by which the community property has changed due to non-random species losses or gains, explicitly taking into account the specific contributions to this property of species that are lost and gained and (3) the 'context dependent effect' (CDE), unlike the SRE and SCE, represents between-mesocosm differences in contributions of species that are present in both mesocosms. CDE itself can be partitioned into three components (Fox 2006): a component resulting from changes in species' abundances between mesocosms (CDE_n), a component resulting from individual species' phenotypic changes (CDE_p) and a component caused by the altered abundance of species that also exhibit phenotypic change (CDE_i). Although the partitioning of the CDE component has received little attention so far, it proves very useful for our study because it allows disentangling the effects of changes in species abundance from phenotypic trait responses on community elemental content and ratios. We regrouped the Price partitions to focus on four components, i.e. net effects of species gains and losses ($SRE+SCE$), contribution of species abundance changes (CDE_n), effect of phenotypic changes (CDE_p) and interaction between abundance and phenotypic change (CDE_i).

Application of the Price equation requires that one mesocosm (the 'comparison') is compared to a reference mesocosm or 'baseline'. In our analyses, we evaluated the responses of community C and P to P reduction within light treatment levels using HP tanks as baseline and LP tanks as comparison. Similarly, responses to increasing light intensity within P treatment levels were studied by taking shaded tanks as baseline and full light tanks as comparison.

Evaluating the impact of compositional turnover and plasticity on changes in community elemental ratios

Changes in total C and P content equal the sum of their Price components ($\Delta C = \sum_j \Delta C_{pj}$; resp. $\Delta P = \sum_j \Delta P_{pj}$), where ΔC_{pj} and ΔP_{pj} refer to changes in community C and P due to the individual Price component p_j . Direct application of the Price

equation to community C:P is not possible because the partitions of a ratio do not add up. Nevertheless, we can evaluate the relative effect of each Price component on the community C:P ratio separately, which requires the assumption of all else being equal. For each Price component p_j the community content of C and P in the comparison can be estimated as $C_{comp,p_j} = C_{base} + \Delta C_{pj}$ and $P_{comp,p_j} = P_{base} + \Delta P_{pj}$, where C_{base} and P_{base} represent the community C and P content in the baseline. Hence, the expected unique effect of Price component p_j on the community C:P in the comparison site can be evaluated as $\Delta C:P_{pj} = C_{comp,p_j}/P_{comp,p_j} - C_{base}/P_{base}$. Note that ΔC_{pj} or ΔP_{pj} of $SRE+SCE$ and CDE_n values can each be affected by changes in the relative abundance of its species as well as by total community abundance. $\Delta C:P_{pj}$ values, however, are not affected by a change in total abundance as C and P content are tied together within the individual organisms making up the community. Thus, increasing total abundance without changing species abundances relative to one another would increase both ΔC_{pj} and ΔP_{pj} proportionately and thereby cancel out effects in $\Delta C:P_{pj}$ values.

Statistical analyses

Seston and zooplankton community biomass and C:P differences were analysed with a full-factorial generalised linear model ($\alpha = 0.05$) with a gamma distribution and a log-link function to deal with heteroscedasticity. Post hoc comparisons of treatments were carried out using Tukey contrasts, applying a correction for multiple comparisons (Westfall 1997). Differences between zooplankton taxa in C:P were assessed using a K-sample permutation test with 99999 Monte-Carlo resamplings. The relationship between seston C:P and zooplankton community C:P was analysed using a nonlinear logistic model. Community shifts in relative abundance due to experimental treatments were tested with a distance-based redundancy analysis (dbRDA, Legendre & Anderson 1999) using Odum's percentage difference dissimilarity. For any pair of contrasting treatments we tested whether Price components were significantly different from the components calculated among random combinations of replicates within these treatments using a two-sample location test (further details given in Supplement 1). All analyses were carried out in R (3.2.1) using a modified code of Winfree *et al.* (2015) and the packages coin, vegan and ggplot2 (Hothorn *et al.* 2006; Ginestet 2011; Oksanen *et al.* 2015). All R code is archived in the Dryad depository belonging to this article.

RESULTS

Our manipulations of phosphorus and light availability strongly influenced the C:P ratio and biomass of seston (Fig. 1, Table S2.1), with response patterns being robust throughout the experiment (Fig. S3.1). Seston C:P increased with P reduction and light availability (Fig. 1). The increase in seston C:P with light was stronger under conditions of low compared to high P availability: under HP conditions, molar seston C:P increased from 135 to 189 with increasing light, whereas under LP conditions, seston C:P increased from 295 to 677. Seston biomass increased with increasing light

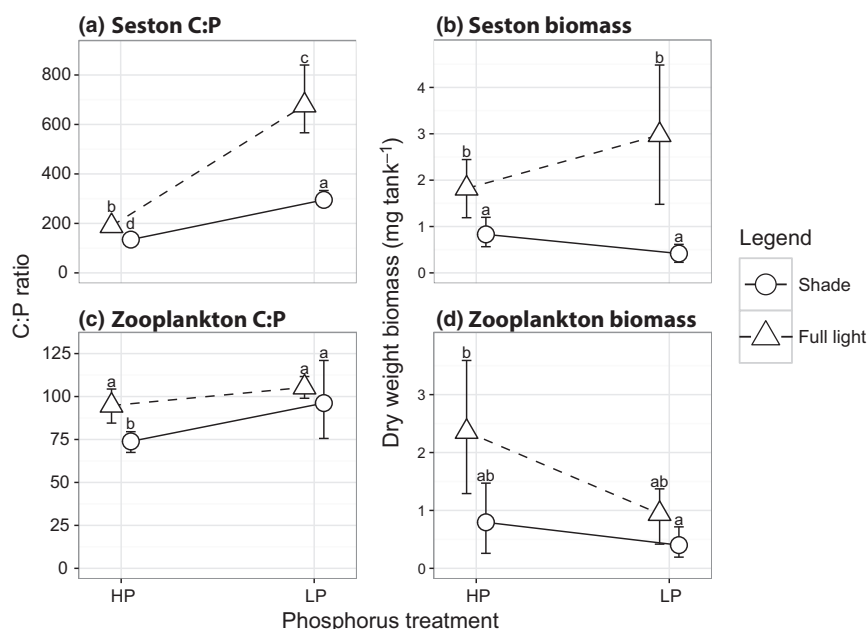


Figure 1 Reaction norm plots representing responses of seston (a), and zooplankton (c) C:P ratios and seston (b) and zooplankton (d) total tank biomass (mg dryweight) to phosphorus addition (HP, LP) and light treatments (triangles, dotted line: full light; circles, continuous line: shaded). Error bars indicate 97.5% confidence limits. Letters indicate significant pairwise differences.

availability but showed no significant response to P availability (Table S2.1). The phytoplankton communities were primarily composed of small cells (0–2 and 2–30 μm) of eukaryote phytoplankton and showed no significant differences in functional group composition between treatments (Fig. S3.4).

Community-level zooplankton C:P increased with decreasing P availability and increased with increasing light availability (Fig. 1; Table S2.1). Zooplankton C:P responded in a nonlinear fashion to variation in seston C:P (Fig. 2). Starting from low levels, zooplankton C:P levels increased with increasing seston C:P, but levelled off beyond a seston C:P of 270 (Nonlinear logistic least square regression: R^2 adj = 18.3%, Asym = 101.9, Xmid = 69.3, scal = 58.4, Table S2.4). A shift from low to high light under HP conditions resulted in a 29% increment of seston C:P (from 135 to 189; Fig. 1a) and a 22% gain of the mean zooplankton C:P (from 74 to 95; Fig. 1c). A decrease in P availability under shaded conditions caused a 54% augmentation of seston C:P (from 135 to 295) and a 22% increase in zooplankton C:P (i.e. from 74 to 95). In contrast, although seston C:P increased 3.6-fold (from 189 to 677) in response to a decrease in P availability under full light conditions, zooplankton C:P increased only 10% (from 95 to 105). Similarly, a 2.3-fold increase in seston (from 295 to 664) with increased light intensity under LP conditions was followed by a zooplankton C:P raise of 10% only (from 95 to 105). Zooplankton community biomass responded positively to increases in P and light availability (Fig. 1d). Under conditions of full light, the increase in seston C:P associated with reduced P availability coincided with a strong decline in zooplankton biomass.

Experimental treatment combinations for which significant shifts in zooplankton community C:P were observed (i.e. from

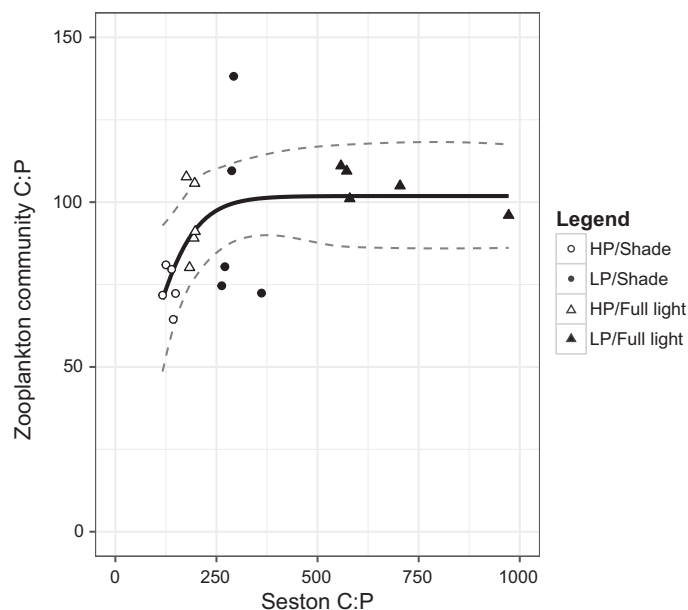


Figure 2 Response of community zooplankton C:P ratio to variation in the seston C:P ratio. The continuous line represents a nonlinear logistic regression curve with its 97.5% confidence interval (dashed lines).

HP to LP in the shade and from Shade to Full light in HP) were partitioned using the Price equation (Figs 3 and 4, respectively). For both comparisons, we detected an increase in zooplankton community C:P of around 22 (Figs 3c and Fig. 4c,f. Total response). With a reduction of P supply under shaded conditions, the net effect of species losses and gains resulted in reduced community C and P (Fig. 3; SRE + SCE), but both effects cancelled each other out and were therefore

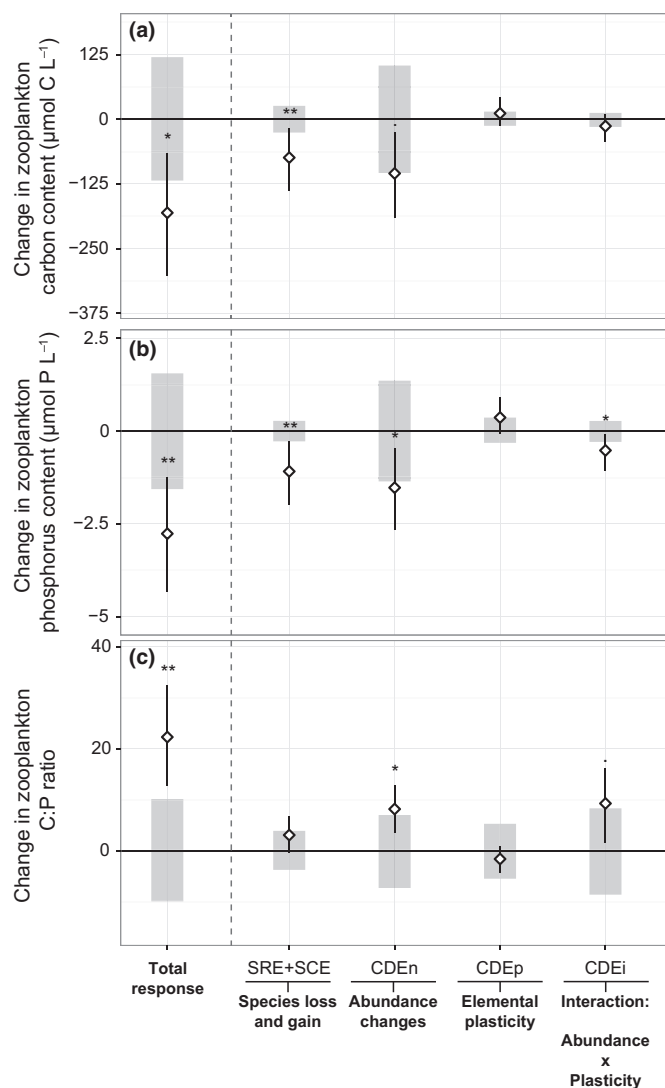


Figure 3 Price partitions of changes in the zooplankton community elemental content between HP and LP treatments under shaded conditions (baseline condition: HP/Shade treatment; comparison condition: LP/Shade treatment) and their effects on community elemental ratio. (a) Community C content; (b) community P content; (c) community C:P elemental ratio. For a detailed explanation on the individual Price partitions (SRE+SCE, CDE_n, CDE_p and CDE_i), we refer to Supplement 1. Error bars represent 97.5% confidence limits. Grey bars represent the zero centred null distribution of values obtained by random permutations within treatments. Significance levels were assessed using a permutation-based location test: *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$, $P < 0.10$.

inconsequential for zooplankton community C:P. Declines in total abundance of resident species resulted in strong reductions of both community C and P content (Fig. 3; CDE_n). Here, changes in the relative abundances of species caused a proportionally stronger reduction in P than C content, and had a significant, positive impact on community C:P (Fig. 3c). Plastic responses of species to a reduction in P availability were relatively weak (Fig. S4.1) and had no influence on community C:P (Fig. 3; CDE_p), although a marginally significant effect of the CDE_i component (i.e. the interaction between

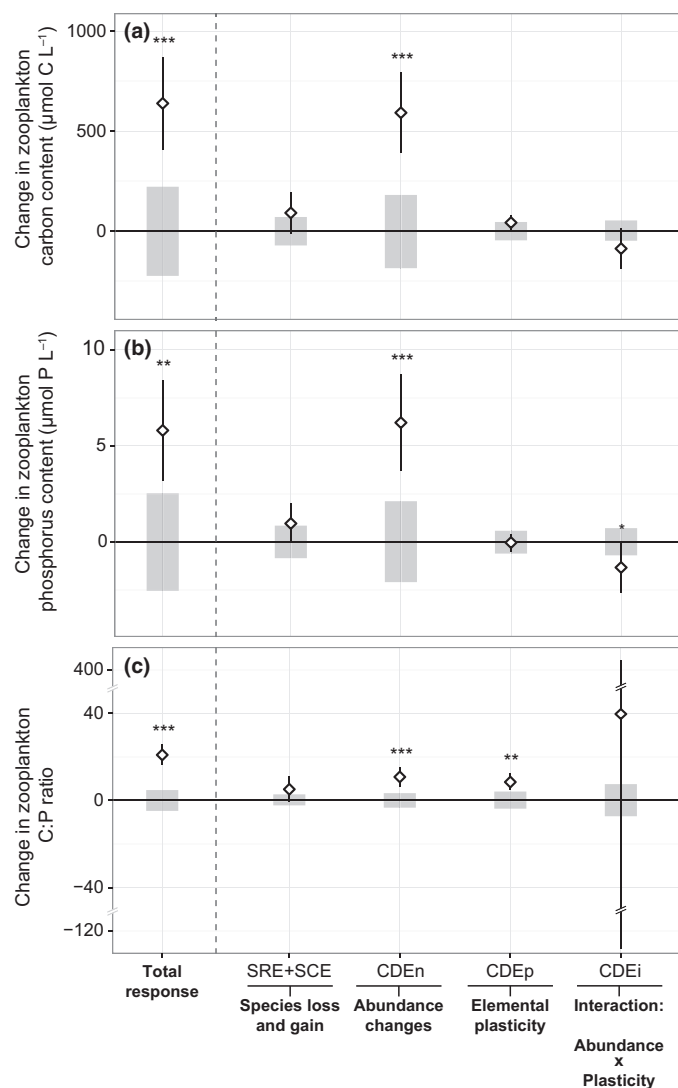


Figure 4 Price partitions of changes in the zooplankton community elemental content between Shade and Full light under HP conditions (baseline condition: Shade/HP; comparison condition: Full light/HP) and their effects on community elemental ratio. (a) Community C content; (b) community P content; (c) community C:P elemental ratio. For a detailed explanation on the individual Price partitions (SRE+SCE, CDE_n, CDE_p and CDE_i), we refer to Supplement 1. Error bars represent 97.5% confidence limits. Grey bars represent the zero centred null distribution of values obtained by random permutations within treatments. Significance levels were assessed using a permutation-based location test: *** $P < 0.001$, ** $P < 0.01$, * $P < 0.05$, $P < 0.10$.

CDE_p and CDE_n) suggests that a change in the abundance of some plastic species may have influenced the community C:P.

With an increase in light intensity under HP conditions, the combined effect of species losses and gains did not influence community C and P (Fig. 4; SRE+SCE), although the gain component of SRE was found to have a positive contribution when considered in isolation (Figure S3.3, Supplement 3). Total zooplankton abundance increased strongly and resulted in augmented community C and P content (Fig. 4a,b; CDE_n). A significant, positive impact of the CDE_n component on community C:P indicates a compositional shift towards more

C-rich species. Plasticity (CDE_p) had a significant impact on community C:P, where species per capita C content increased disproportionately compared to per capita P content (Fig. 4).

The importance of community compositional shifts underlying zooplankton community C:P changes as revealed by Price partitioning is confirmed by additional analyses. The C:P ratio of species showed constitutive differences across treatments, with mean values ranging from 75 (*Daphnia galeata*) to 128 (*Chydorus sphaericus*, *Scapholeberis mucronata*) (Fig. 5; Table S2.2). Furthermore, dbRDA analyses revealed significant shifts in zooplankton composition but only for the treatment combinations for which significant responses in zooplankton community C:P ratios were observed (Table S2.3), i.e. the HP-LP comparison in shade (Fig. 6a: R^2 adj = 13.8%, F : 2.446; P = 0.039) and the shade–full light comparison at HP (Fig. 6b: R^2 adj = 14.8%, F : 2.561; P = 0.038). This pattern seemed to be mainly driven by the response of *D. galeata* to treatments associated with high seston C:P (see Fig. S4.1 and Table S.1). We observed an overall low responsiveness of body elemental composition of individual species to experimental treatments (Fig. S4.1).

DISCUSSION

The question, to what extent elemental plasticity of consumers may affect food web interactions and ecosystem functions, has so far mainly been based on an evaluation of the phenotypic response of single species in simple linear food webs (Plum et al. 2015). In natural systems, however, consumer communities may consist of multiple species differing in elemental composition. As a result, along gradients of changing relative resource supply, community compositional shifts may result in community plasticity greater than that expected by the plasticity of individual species alone (Danger et al. 2008). This

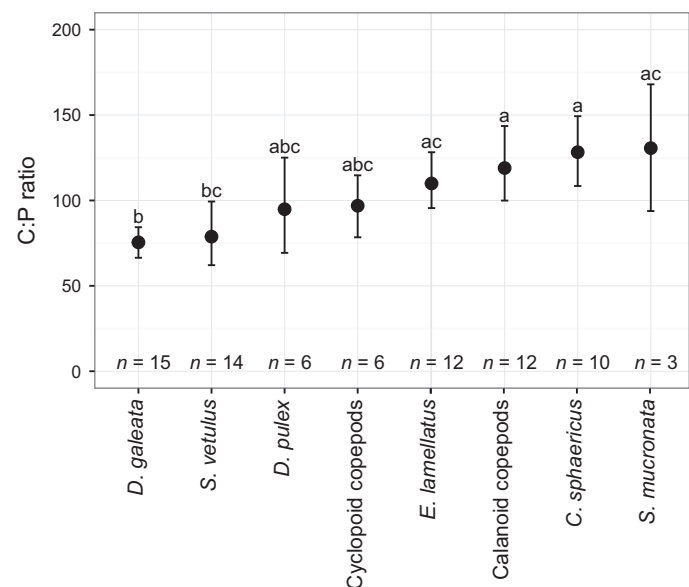


Figure 5 Mean C:P ratio of different zooplankton taxa calculated across treatments. Error bars represent 97.5% confidence limits around the mean. Letters indicate significant pairwise differences.

idea is confirmed by the results of our Price-based analysis which demonstrated a relatively large impact of compositional shifts on community C:P (CDE_n components in Figs 3c and 4c) relative to phenotypic elemental plasticity of its constituent species (CDE_p components in Figs 3c and 4c). Indeed, species were found to differ in body elemental composition (Fig. 5) and multivariate analysis demonstrated consistent shifts in community composition across seston C:P gradients (Fig. 6).

Our results support the prediction of ecological stoichiometry, in that shifts towards food sources with high P should result in compositional shifts towards a dominance of fast-growing, P-rich consumers, whereas reductions in food P content should result in a dominance of P-poor consumers that are less sensitive to P limitation (Sternner et al. 1997). The P-rich taxon *Daphnia galeata*, for example, responded nicely according to that prediction and dominated in the treatment combination with lowest seston C:P (i.e. under low light with high nutrient concentrations), while its relative abundance decreased in the treatment combinations with higher seston C:P. However, no significant trends in the relative abundance of other taxa were found. A similar observation was made by Hall et al. (2004) who also reported a dominance of *Daphnia* at the lowest seston C:P levels, but were unable to relate elemental composition of other taxa with their abundance response along experimental resource supply gradients. One explanation for this is that the relative performance of zooplankton along a food C:P gradient is determined by more traits than the elemental composition alone (Hall et al. 2004). Taxa may differ in their P acquisition, assimilation and retention efficiencies, their ability to graze selectively or in their resource allocation. In fact, even within one species, microevolutionary adaptation to LP food has been shown to involve changes in sexual investment of a plankton consumer without demonstrable changes in body elemental composition (Declercq et al. 2015). In communities without predators such as ours, we expect that the capacity to exploit limiting resources should be a better predictor of relative performance than P-related growth rate (Iwabuchi & Urabe 2012a,b). Nevertheless, our Price-based analyses confirm that an increase in seston C:P has resulted in a net increase in zooplankton C:P via a shift towards a dominance of C-rich taxa, a conclusion that we would not have easily reached from the consideration of the responses of individual taxa alone.

In our multifactorial experiment, we observed a strong interaction effect of light and P supply on phytoplankton C:P (see also Dickman et al. 2006). This response of phytoplankton C:P was strongly in accordance with the light–nutrient hypothesis (Sternner et al. 1997; Elser et al. 2003; Hall et al. 2004; Urabe et al. 2010), which predicts high phytoplankton C to nutrient ratios when light supply is high relative to nutrients. At low P, zooplankton biomass failed to respond to an increase in phytoplankton biomass caused by increased light availability. It is unlikely that this lack of response is due to shifts in the functional composition of the phytoplankton community because all treatments in the experiment were dominated by edible phytoplankton (Geller & Müller 1981). More likely it reflects a stoichiometry-driven reduction in herbivore resource use efficiency (Urabe & Sternner 1996; Urabe

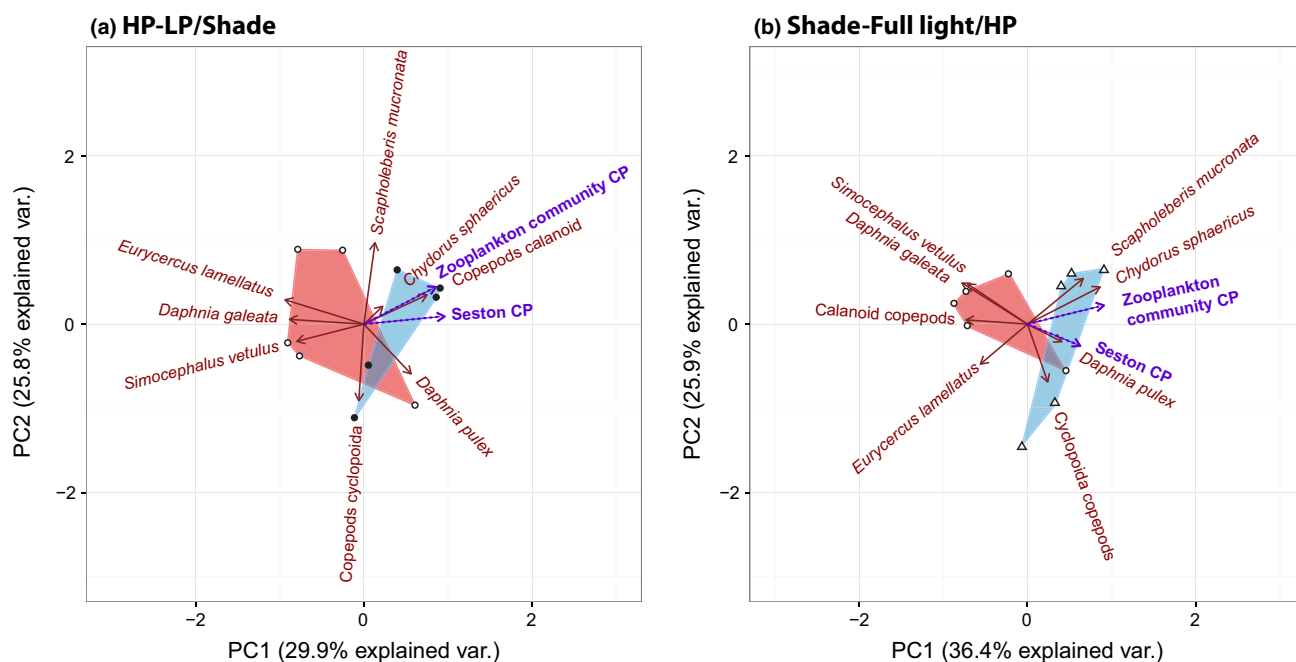


Figure 6 PCA plots of the zooplankton community composition along experimental treatments of (a) HP to LP under shaded conditions and (b) Shaded to Full light under HP conditions. Seston C:P and zooplankton community C:P ratio are plotted in blue (dashed arrow) as neutral variables. Open symbols indicate HP tanks and closed symbols LP tanks, circles represent shaded tanks, whereas triangles represent full light tanks.

et al. 2010). It is noteworthy that in higher ranges of producer C:P values (C:P > 270), strong increases in seston C:P (up to 677; due to increased light at low P supply or reduced P availability at full day light) resulted in no further increase in zooplankton C:P. For these treatment combinations, dbRDA indicated no significant changes in zooplankton community composition. These observations altogether indicate that seston C:P had surpassed a threshold above which zooplankton communities were unable to respond via phenotypic plasticity or species sorting.

Our Price-based analysis suggests qualitative differences in the way zooplankton community stoichiometry responded to different factors. In shade the increase in community C:P observed in response to a decreased P availability could only be explained by changes in the relative abundance of species (CDE_n). In contrast, the community C:P increase observed in response to an augmentation of light intensity at high P levels was not only realised through compositional shifts but also via phenotypic elemental plasticity (CDE_p). The absence of plasticity in the first case may reflect a stronger tendency of zooplankton organisms to remain homeostatic than in the second case. Invertebrates are known to maintain elemental homeostasis in the face of nutrient limitation, for example, by increasing P uptake (e.g. compensatory or selective grazing), P assimilation (e.g. through increased gut passage time or production of digestive enzymes) and P retention. These strategies are energetically costly, resulting in enhanced respiration and therefore loss of carbon in the form of CO_2 , which may also contribute to maintaining the C:P balance. In contrast, the elemental plasticity observed with varying light intensity at high P levels may reflect a tolerance to a higher body C content when P is saturating, for example,

under the form of lipid storage (Becker & Boersma 2005). To summarise, our results show that different mechanisms (P limitation or excess of light) may result in similar increases in seston C:P to which zooplankton C:P responds. Although the zooplankton C:P responds to both environmental factors were of comparable magnitude, the Price-based analysis revealed differences in the mechanisms underlying these responses (see also Supplement 3).

There has been a long tradition of research into the relationship between biodiversity and ecosystem functioning (Schmid *et al.* 2001). Although the potential importance of stoichiometric plasticity is increasingly recognised as a determinant of ecosystem functioning (Hall *et al.* 2007; Dickman *et al.* 2008; Rowland *et al.* 2015), it remains unclear to what extent this plasticity is determined by species composition and diversity. Hall *et al.* (2007) broadened the bottom-up perspective of the light–nutrient hypothesis by emphasising the potential importance of grazer-induced changes in the taxonomic composition of producer communities. Similarly, Liess & Kahlert (2009) and Mette *et al.* (2011) studied the stoichiometric response of producer communities to light and nutrient gradients in the presence and absence of grazers and reported associations between phytoplankton community composition and seston C:P. In these studies, however, the relative importance of compositional changes and of intraspecific stoichiometry plasticity was not disentangled. Recently, a few studies have addressed the relationship between species diversity and stoichiometry at the base of the food web. Striebel *et al.* (2009a) suggested that spectral niche complementarity associated with high species diversity may enhance phytoplankton C:P via an increase in resource use efficiency. With laboratory and field experiments, Striebel *et al.* (2009b) demonstrated a

positive association between phytoplankton diversity and algal C:P ratios. Plum *et al.* (2015) demonstrated that a richness-induced enhancement of phytoplankton C:P resulted in a decrease in food quality large enough to negatively affect herbivore growth. In these studies, diversity is treated as a factor independent of resource supply, whereas, in reality, diversity itself often responds to gradients in relative resource supply. Due to such covariation, it may prove difficult to disentangle the effects of diversity change from other mechanisms that affect community stoichiometry along such gradients. Along a resource supply gradient, new species may establish, whereas others may disappear. Some species may become relatively more abundant, whereas others become scarce. Provided that species differ in body elemental composition, each of these changes will have its effects on community stoichiometry, in addition to the effects of phenotypic plasticity in elemental composition. The strength of our Price-based approach lies in its ability to reveal the independent impact of species gains, species losses, compositional change and phenotypic plasticity along gradients of relative resource supply.

Although initial communities were composed of zooplankton mixtures from multiple sources, species sorting was likely constrained. For this reason, we may regard the observed community elemental plasticity as conservative. It is expected that stronger shifts in community elemental plasticity would be observed in systems more connected to a regional species pool as the latter would allow the colonisation and establishment of species with a broader range of elemental phenotypes (cf. 'boundary species', Danger *et al.* 2008). Through our Price-based approach such a hypothesis may be tested, effectively linking community stoichiometry to metacommunity dynamics.

The Price equation has already been successfully applied in studies addressing the role of biodiversity in ecosystem functioning (e.g. Winfree *et al.* 2015). We extend its use to the analysis of community-level stoichiometry, and propose a statistical framework for the analysis of its partitions in replicated community experiments. Our approach has the limitation that the sum of the proportional components is not equal to the total; ratios may only be evaluated and tested in isolation. Still, the approach can yield fundamental insights into the mechanisms that shape community stoichiometry in a reproducible way, as is illustrated by our experimental results.

CONCLUSIONS

Gradients in seston C:P were shown to lead to C:P shifts in the consumer community larger than expectations based on observed levels of elemental plasticity within individual species. Therefore, studies of single-species' consumer plasticity in response to food stoichiometry are likely to underestimate the potential of whole-community plasticity in natural systems. Our results illustrate that species sorting is a major factor in governing community-level stoichiometry shifts in response to food quality. As such, species sorting may dampen negative effects of stoichiometric mismatches on important ecosystem functions such as grazing and nutrient cycling by consumers (Danger *et al.* 2008).

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STATEMENT OF AUTHORSHIP

ST and SAJD conceived the study. ST, SAJD, MV and DBW designed and ST, MV and DS carried out the experiment. ST analysed the data with help from MV, LG and SADJ where discussions with DBW, DS and ED helped with interpreting the results. ST and SAJD wrote the manuscript with important input from all other authors (MV, DS, DBW, ED and LG).

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