The evolution of ecosystem processes: growth rate and elemental stoichiometry of a key herbivore in temperate and arctic habitats

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Abstract

Understanding the reciprocal interactions between the evolved characteristics of species and the environment in which each species is embedded is a major priority for evolutionary ecology. Here we use the perspective of ecological stoichiometry to test the hypothesis that natural selection on body growth rate affects consumer body stoichiometry. As body elemental composition (nitrogen, phosphorus) of consumers influences nutrient cycling and trophic dynamics in food webs, such differences should also affect biogeochemical processes and trophic dynamics. Consistent with the growth rate hypothesis, body growth rate and phosphorus content of individuals of the *Daphnia pulex* species complex were lower in Wisconsin compared to Alaska, where the brevity of the growing season places a premium on growth rate. Consistent with stoichiometric theory, we also show that, relative to animals sampled in Wisconsin, animals sampled in Alaska were poor recyclers of P and suffered greater declines in growth when fed low-quality, P-deficient food. These results highlight the importance of evolutionary context in establishing the reciprocal relationships between single species and ecosystem processes such as trophic dynamics and consumer-driven nutrient recycling.

Introduction

Evolutionary biologists are increasingly interested in understanding the functional consequences of genetic change at cellular, organismal, and ecological levels of organization (Thompson, 1994; Holt, 1995; Schlichting, 1998; Coen, 1999; Laland *et al*., 1999). Simultaneously, ecologists appear to have an expanding interest in the reciprocal relationships between the evolved traits of species and characteristics of the ecosystems in which those species have evolved and persist (Schulze & Mooney, 1994; Jones & Lawton, 1995). It has been proposed that ecological stoichiometry may provide a useful framework for integrating major ideas of organismal evolution with their ecosystem ramifications (Reiners, 1986; Elser *et al*., 1996). Ecological stoichiometry is the study of the balance of energy and multiple chemical elements in ecological interactions (Reiners, 1986; Sterner, 1995; Elser *et al*., 1996; Hessen, 1997). This approach considers the ecological consequences of the relative energetic and material requirements of individual organisms seeking to maximize their reproductive success. Thus, by focusing on the level of biological organization where natural selection operates, ecological stoichiometry may be more successful in integrating evolutionary and ecosystem thinking than previous approaches. Along these lines, Elser *et al*. (1996) have suggested that the cellular and biochemical machinery required for divergent life history strategies sets the stoichiometric requirements of individual organisms. Thus, stoichiometric analysis may be extended to various levels of biological integration (individuals, cells, biomolecules), a more generalized approach that might be called 'biological stoichiometry'. In this way we might better understand the proximate mechanisms by which evolutionary forces impinge on the flow of energy and matter in ecosystems and by which the relative

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availability of energy and multiple chemical elements in the environment might constrain evolutionary change. However, there have been few attempts thus far to evaluate the validity of these far-reaching connections.

Recent findings in pelagic ecology indicate that the relative requirements of consumers for major elements differ widely among species. Thus, planktonic taxa appear to provide an appropriate model system to test these ideas. In particular, crustacean zooplankton exhibit 4–5-fold variation in body P content, from <0.5% of body dry weight to more than 2.5% (Hessen & Lyche, 1991; Main *et al*., 1997). These differences have important ecological consequences, including direct mineral P limitation of growth of P-rich herbivores such as *Daphnia* (Urabe *et al*., 1997), alterations in rates and ratios of recycling of limiting nutrients by consumers (Sterner *et al*., 1992; Elser & Urabe, 1999), and stoichiometric constraints at the producer–herbivore interface that affect food-web dynamics (Andersen, 1997; Elser *et al*., 1998). Thus, consumer elemental composition is a key parameter influencing trophic dynamics and nutrient cycling in pelagic ecosystems. But why do animal taxa differ so much in their elemental composition? As mentioned above, it has been argued (Elser *et al*., 1996) that much of this proximate (elemental) variation in invertebrates is driven by the evolution of contrasting life history strategies that result in interspecific differences in somatic growth rate (the 'growth rate hypothesis' (GRH); see Fig. 1). Differences in growth rate result from differential cellular allocation to structures necessary for somatic growth, in particular, RNA-rich ribosomes. Thus, because RNA is an especially P-rich molecule and can comprise a significant proportion of biomass, natural selection favouring rapid growth rate should generate increased organism P content relative to conditions in which accelerated body growth rate is not favoured by natural selection. As body elemental composition is directly connected to trophic interactions and internal nutrient recycling, the evolution of body growth rate, a key integrating parameter of overall life history strategy (Arendt, 1997), may have unappreciated consequences for food-web dynamics and biogeochemical processing.

Testing the set of hypotheses just described requires comparing the somatic growth rates, elemental composition, trophic responses, and nutrient recycling characteristics of organisms that have evolved under contrasting selection for body growth rate. Although the hypothesized connection between P content and growth rate has been supported in cross-species comparisons (Main *et al*., 1997; Dobberfuhl, 1999), a more direct test of the GRH and its corollaries would be to compare closely related taxa in selective environments favouring different growth rates. In our study we contrasted populations of two species from the cosmopolitan *Daphnia pulex* species complex (Lynch, 1983; Colbourne & Hebert, 1996) in Wisconsin and in arctic Alaska. We reasoned that the extremely short growing season of arctic habitats imposes strong natural selection on growth rate and development time, as the animals must achieve sufficient body size to reproduce and produce resting eggs (ephippia) before the pond is solidly frozen. Indeed, other aspects of life history in this group appear to have responded to the strong contrast in selection regime. For example, Holarctic members of the *D. pulex* species complex are obligately parthenogenetic whereas temperate members are generally facultative parthenogens

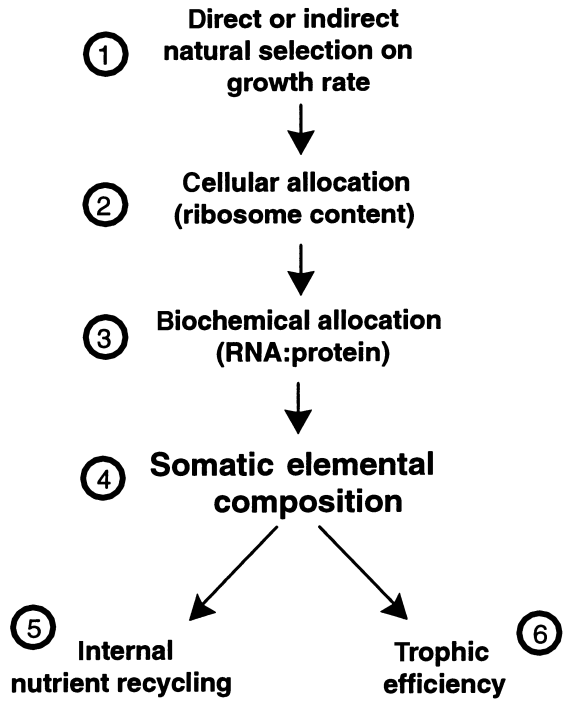

Fig. 1 Conceptual diagram illustrating the mechanistic connections linking direct or indirect natural selection on growth rate with ecosystem processes via effects on somatic elemental composition. More detailed information regarding this set of hypotheses has been presented elsewhere (Elser *et al*., 1996). In extreme environments (such as high latitudes, high altitudes or disturbed habitats), natural selection should favour rapid body growth and development (part 1 of the diagram). To grow rapidly, animals must raise their protein synthesis rate and to do so generally increase their allocation to ribosomes in the cell (part 2). As RNA is a major biochemical constituent of ribosomes, the biochemical consequence is increased RNA: protein ratios in body tissues, primarily owing to increases in rRNA (part 3). As RNA is one of the most P-rich molecules in cells and comprises a significant proportion of biomass in rapidly growing organisms, animals with elevated RNA levels should also be rich in P (part 4). As animals with P-rich tissues must retain a considerable amount of P from their food, rapidly growing animals should have reduced rates of P recycling relative to more slowly growing animals (part 5). Additionally, P-rich animals require a higher quality diet (in terms of mineral P content) to grow maximally. Thus, the trophic efficiency of rapidly growing animals (i.e. their efficiency of conversion of ingested food to new biomass) should decline more strongly when animals are challenged with poor quality food of high C : P ratio (part 6).

(Ward *et al*., 1994; Colbourne *et al*., 1998). Based on the GRH, we made the following predictions. Relative to temperate *Daphnia* sampled in Wisconsin, individuals of Alaskan populations should:

1 have higher body growth rates (given identical diet and temperature);

2 have higher body phosphorus content (% P by dry weight) and lower body N : P ratio, reflecting increased allocation to P-rich RNA for high growth rate;

3 retain a greater fraction of P from their diets and thus recycle N and P at high N : P ratio; and

4 suffer stronger reductions in growth when fed poor quality, high C : P food.

Our data were consistent with all but one of these expectations.

Materials and methods

Study species

To test the set of predictions emerging from the GRH, we sampled populations from the *D. pulex* species complex during summer 1997 and 1998 in the vicinity of the University of Notre Dame Environmental Research Center (Wisconsin) and the Toolik Lake field station (Alaska) (Table 1). The taxonomic status of the *D. pulex* species complex is somewhat problematic as uncertainty exists about whether phylogenetic lineages within this group are sufficiently distinct to constitute separate species (Lehman *et al*., 1995). Our analysis is guided by the recent phylogenetic analysis by Colbourne *et al*. (1998) of arctic and temperate members of the *D. pulex* species complex. Using sequence diversity of a 498 nt sequence in a rapidly evolving mitochondrial gene (NADH dehydrogenase subunit 5; ND5), Colbourne *et al.* (1998) identified three major clades within the complex. One of these main clades includes panarctic *D. pulex*, *D. middendorfiana*, *D. melanica*, and three lineages of *D. pulicaria* (Eastern, Western and Polar). According to distribution maps and habitat and morphological criteria developed by these workers, several members of this species complex are present in Wisconsin. However, Alaskan *Daphnia* north of the Brooks Range have never been characterized for ND5. We chose populations most likely to correspond with *D. pulicaria* (probably the Western lineage; sampled in Wisconsin) and Alaskan populations that we thought most likely to be panarctic *D. pulex*. These lineages have probably experienced strongly divergent selection on growth rate and development time. Although *D. pulicaria* occur in stratified lakes with a temperate growing season that lasts several months, panarctic *D. pulex* inhabit small ponds where the growing season rarely exceeds 3–4 weeks. Thus, our study assesses the consequences of the evolutionary divergence of *D. pulicaria* and panarctic *D. pulex* during the time that these taxa have come to occupy strongly contrasting habitats. The consequences of interest are potential changes in growth rate, body C : N : P stoichiometry, and nutrient cycling and trophic dynamic characteristics of these taxa in ecosystems where they are important components of the food web.

To confirm the taxonomic status of sampled populations and to evaluate the relative extent of evolutionary divergence, selected samples were characterized for variation in the same segment of the ND5 gene characterized by Colbourne *et al*. (1998). We were especially interested in confirming the taxonomic status of the populations from Alaska. DNA was extracted from frozen samples of *Daphnia* using the Qiagen Qiaamp tissue kit following the manufacturer's instructions. A large fragment of ND5 (896 bp) was amplified with the primers ND5a and ND5b (Colbourne *et al*., 1998) using the following parameters: 35–40 cycles of denaturation at 94 °C for 1 min; primer annealing at 48 °C for 1 min; and extension at 72 °C for 2 min. Resulting amplification products were cleaned by centrifugation through filter units as indicated by the supplier (Millipore Corp., Bedford, MA, USA), and 495 bp of sequence determined

Table 1 Sampling sites and dates for studies of Alaska and Wisconsin populations of the *Daphnia pulex* species complex. Populations sampled in 1997 were used for determinations of growth rate, body elemental composition, and N and P release rates. Populations sampled in 1998 were used for assessment of trophic response to stoichiometric food quality. Animals from sites indicated by * were analysed for ND5 sequences as described in the text. More precise information about localities can be obtained by contacting the authors.

D. pulex populations in vicinity of Toolik Lake field station, Alaska (latitude: 68°38′N)	*D. pulicaria* populations in vicinity of University of Notre Dame Environmental Research Center, Wisconsin/Michigan (latitude: 46°13′N)
Walden Pond (NE-2) (4 July 1997, 19 June 1998*)	Tender Bog (2 June 1997, 18 May 1998)
Airstrip Pond (5 July 1997, 20 June 1998*)	Tenderfoot Lake (3 June 1997, 15 May 1998*)
Old Camp Road East Pond (7 July 1997, 22 June 1998*)	Paul Lake (3 June 1997)
PB Pond (9 July 1997)	Big Musky Lake (10 June 1997, 27 May 1998*)
Green Cabin Pond (11 July 1997)	Plum Lake (14 June 1997)
GPS-22 Pond (11 July 1997, 30 June 1998*)	West Long Lake (16 June 1997, 22 May 1998)
Berm Pond (13 July 1997, 30 June 1998*)	Raspberry Lake (31 May 1998)
Fog Pond #29 (25 June 1998*)	
Fog Pond #30 (25 June 1998*)	

with primer ND5b using an ABI 377 automated sequencer. Sequences are available upon request to T.E.D. Sequences were aligned by eye using the homologous sequence of *D. pulex* (GenBank #5835848) as a frame of reference. Taxonomic placement was determined using PAUP4.0b2a (Swofford, 1998). Sequences from our unknown samples were added to the 79 analysed by Colbourne *et al*. (1998) and relative similarity evaluated by the neighbour-joining option of PAUP using Jukes-Cantor distances. Relative strength of nodes was determined by completion of 1000 bootstrap replicates in PAUP.

In the following we discuss these results in terms of 'Alaska *Daphnia*' vs. 'Wisconsin *Daphnia*'. This nomenclature is primarily for clarity in presentation as we recognize that we do not have a sufficiently extensive sample to generalize about *Daphnia* in broad regions. We also recognize that the contrasts that we document may not be entirely the result of contrasting selection pressures under arctic vs. temperate conditions *per se*. For example, similar differences within the temperate zone might be observed in comparing lake–dwelling *D. pulicaria* with pond–dwelling *D. pulex*. However, such differences would likely be derived from the same general mechanisms argued for here; that is, a contrast in selection pressure on growth rate driven by habitat ephemerality. In our study we sought the strongest selection contrast we could examine within this species complex and thus chose to compare the characteristics of a taxon inhabiting large water bodies with a long-growing season with those of a taxon inhabiting small water bodies with an extremely abbreviated growing season. Finally, as discussed further below, our ability to distinguish between adaptation and artefacts of phylogenetic history in interpreting any observed differences is limited because our study is modest in scope and confined to a single pair of closely related species.

Organismal parameters

Growth potential

To measure the growth rate potential of animals collected from different ponds and lakes, animals were collected from study sites and isolated into narrow size classes by screening or hand-separation. For each population sampled, 5–10 individuals were placed in each of 150-mL clear plastic bottles for incubation at three temperature treatments (10, 15 and 20 °C); 5–6 replicate bottles were used for each temperature. Additional animals were set aside for determination of initial body mass based on body length using length–weight regressions. Each bottle was filled with a standardized food preparation consisting of dried, nutrient-sufficient (cellular C : N : P was 183 : 29 : 1 by atoms), *Scenedesmus acutus* raised under chemostat conditions (Sterner *et al*., 1993) the previous spring. Previous studies showed that drying the algae does not substantially alter its elemental and biochemical composition and that dried algae is of similar quality for *Daphnia* growth as fresh algae (Dobberfuhl & Elser, 1999). Dried algae were re-suspended in filtered lake water to achieve a concentration of 4 mg dry weight L^{-1}, sufficient to saturate grazer feeding. Food was replaced daily and bottles were gently mixed frequently throughout the incubation period. After 3 days of incubation in water baths maintained at the appropriate temperatures, animals remaining in bottles were collected, rates of survivorship noted, and individual body lengths measured. Growth rates were determined as μ (day^{-1}) = ln(final body mass/initial body mass)/3. Data were subjected to analysis of covariance (ANCOVA) to assess the statistical significance of sampling site (Alaska vs. Wisconsin) and temperature while correcting for differences in body size of animals used in different experiments. To avoid pseudoreplication, data for separate replicate bottles for each temperature were used to calculate a mean growth rate for each temperature and for each lake population sampled. These independent values for different lake populations were then used in the ANCOVA.

Elemental composition

For each population sampled, animals from the same group as used in the growth experiment were hand-isolated and immediately frozen until return from the field laboratory. Animals were then dried at 60 °C, ground with a pestle, and separated into weighed subsamples for C and N analysis using a Europa Scientific ANCA-SL isotope ratio mass spectrometer. Two to three analytical replicates for each population were analysed. Samples for P analysis were prepared in the same manner as for N, after which the P content of weighed subsamples was determined by wet chemical methods (APHA, 1992). Two to three analytical replicates for each population were analysed. For each population, somatic N : P ratio was calculated as the ratio of somatic % N to somatic % P for that population and then converted to atomic values. N content, P content, and N : P ratio did not vary with body mass; therefore, mean values were compared directly using *t*-tests. P content and N : P ratio were analysed using one-tailed *t*-tests as we had the *a priori* expectation that arctic *Daphnia* would have higher P content and lower N : P ratio than temperate *Daphnia*. As we did not have *a priori* expectations regarding the direction of differences in body % N, means for % N were compared using a two-tailed *t*-test. Again, mean values of elemental composition for a given lake population were computed and each population mean was then used as a single observation in the *t*-tests.

Ecological parameters

Nutrient recycling

Measurements of N and P release rates on standard food followed the approach of Dobberfuhl & Elser (1999). Rates of N release were determined by measuring the

accumulation of NH_4-N over 24 h in bottles containing zooplankton relative to bottles without zooplankton. Animals from the same group collected and isolated for growth rate and elemental composition measurements were placed in sets of 150-mL PVC bottles (5–6 replicates per set, 20–30 individuals per bottle) filled with the standardized food preparation used in the growth rate studies. An additional set of bottles received food but no animals as a control to determine potential changes in NH_4-N owing to release or immobilization of N by the dried algae or by bacteria introduced with the animals. One set of bottles was immediately harvested and its contents filtered through a glass-fibre filter (GF/F) for NH_4 analysis of the filtrate using conventional wet chemistry techniques (Kamphake *et al*., 1967). The remaining bottles were incubated for 24 h at 15 °C after which the contents were filtered and analysed for NH_4-N. N release rate was calculated on a mass-specific basis for each sampling site as: $([NH_4]_{Daphnia} - [NH_4]_{control})$/ *Daphnia* biomass concentration added/24 h. Measurements of P release were made simultaneously with NH_4-N release by measuring concentrations of soluble reactive phosphorus (APHA, 1992) in experimental bottles. Nutrient release rates did not vary with body mass and so data were compared directly using *t*-tests, again using mean values of nutrient release rates for each independent population sampled. As we did not have *a priori* expectations regarding the predicted direction of differences in mass-specific N release rate, these data were analysed using a two-tailed test. P release data were compared using a one-tailed test as we had an *a priori* expectation that P release by arctic animals would be lower relative to temperate animals.

Response of body growth rate to stoichiometric food quality

Animals were isolated from fresh field collections and for each population sampled two sets of bottles (3–5 bottles per set) were prepared by adding 10–15 individuals to each bottle. One set of bottles contained high-quality food (nutrient-sufficient *Scenedesmus* (C : N : P of 183 : 29 : 1), as used in the previous experiments) whereas the other set contained low-quality food (P-limited *Scenedesmus* (C : N : P of 823 : 70 : 1), grown at reduced dilution rates but otherwise similar conditions as the high-quality food (Sterner *et al*., 1993). In both treatments, food was offered at a concentration of 4 mg dry weight L^{-1} and food was replaced daily. As before, animals were incubated for 3 days at 15 °C after which growth rates were calculated from changes in body length relative to animals measured at the start of the experiment. Food quality response did not vary with body mass and so data were compared directly using a one-tailed *t*-test as we had the *a priori* expectation that, relative to temperate animals, growth of arctic animals would decrease more strongly on a poor quality diet. Once again, to avoid pseudoreplication, mean values of growth response to food quality were computed for each population and then used in the *t*-test to compare sampling regions.

Results and discussion

DNA samples of *Daphnia* from seven Alaska and two Wisconsin sites were characterized by obtaining sequence from 495 bp of ND5. Sequences were similar within geographical locations, with average divergence among Alaskan and Wisconsin samples of 2.7 and 2.0%, respectively. Divergence estimates between regions were higher, averaging 7.0%. Neighbour-joining analysis of these samples with those of Colbourne *et al*. (1998) clustered Alaskan *Daphnia* with 'panarctic *D. pulex*' whereas those from Wisconsin were most similar to 'Western *D. pulicaria*'. Bootstrap support was high in both instances, with the node defining each of these two groups found in 97% of the 1000 replicates. Given this level of support, we feel confident that our samples from Alaska and Wisconsin represent panarctic *D. pulex* and Western *D. pulicaria*, respectively. Using a universal calibration for arthropods, Colbourne *et al*. (1998) dated the divergence of the groups containing panarctic *D. pulex* and Western *D. pulicaria* to approximately 2.2 million years ago. Although the method of calibration leaves this date open to conjecture, estimates of sequence divergence obtained from this gene indicate a relatively recent divergence of these taxa.

Our data are consistent with all but one prediction of the GRH. When grown on the same diet at the same temperatures, *Daphnia* from the arctic ponds grew ~1.3 times more rapidly than those from the temperate lakes in Wisconsin (Fig. 2A). Increases in growth rate and decreases in development time from south to north have frequently been observed (e.g. in ant lions, Arnett & Gotelli, 1999). Although the growth rate difference matches our *a priori* hypothesis that accelerated animal growth rate in the Arctic would be adaptive, interpretations should be made with caution owing to the modest nature of our study. As we focused on a single pair of species, we cannot necessarily distinguish between an interpretation that differences in growth rate represent an adaptation to local conditions (i.e. arctic vs. temperate environments) from an interpretation that the difference reflects a happenstance outcome in which a fast growth rate form came to inhabit the Alaska habitats owing to an accident of phylogenetic history. However, our results do suggest that a more thorough study considering additional pairs of closely related arctic and temperate species from distinct phylogenetic lineages would be fruitful.

How did differences in body growth rate translate into differences in elemental composition? Although Alaska and Wisconsin animals did not differ in N content (Fig. 2B), as predicted by the GRH panarctic *D. pulex* had significantly higher body P content than Western *D. pulicaria* samples in Wisconsin (~1.1 times higher;

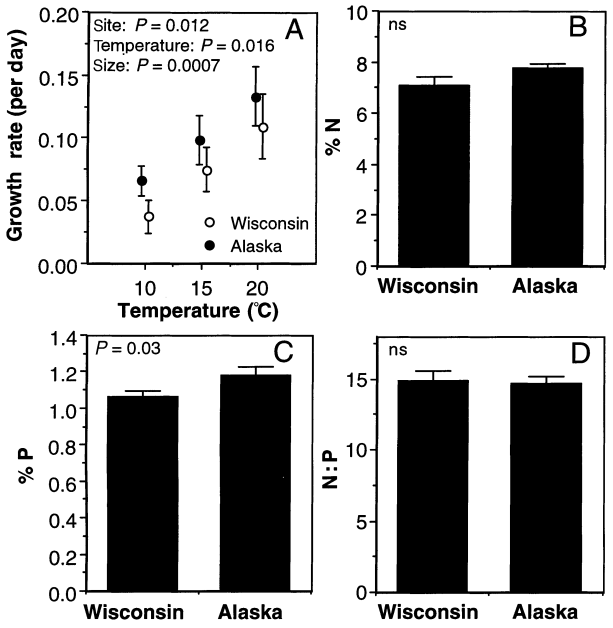

Fig. 2 Somatic growth and elemental composition of study *Daphnia* sampled in Alaska and Wisconsin. (A) Body growth rates at three temperatures for animals from the two study regions. Error bars here and in the following indicate ±1 standard error based on variation among ponds and lakes at a given sampling site. Six independent populations in Alaska and six in Wisconsin were sampled. ANCOVA (with initial body size as the covariate) indicated a significance of sampling site (Alaska vs. Wisconsin, $P = 0.012$), temperature ($P = 0.016$), and initial body size ($P = 0.007$). (B) Somatic nitrogen content (percentage of body dry weight contributed by N). *t*-test: $t = 1.79$, $P = 0.06$ (two-tailed test), $n = 6$ Alaska populations, $n = 6$ Wisconsin populations. (C) Somatic phosphorus content (percentage of body dry weight contributed by P). *t*-test: $t = 2.06$, $P = 0.03$ (one-tailed test), $n = 7$ Alaska populations, $n = 6$ Wisconsin populations. (D) Somatic N : P ratio (by atoms). *t*-test: $t = 0.26$, $P = 0.40$ (one-tailed test), $n = 6$ Alaska populations, $n = 6$ Wisconsin populations.

Fig. 2C). This is consistent with increased cellular allocation to P-rich rRNA associated with increased growth rate (Elser *et al*., 1996; Dobberfuhl, 1999). However, there was no difference in body N : P ratio between Alaska and Wisconsin populations (Fig. 2D). This lack of significance may have been the result of several factors, including the low overall power of the study design owing to limited sample size, the slight increase in % N in the arctic populations (thus compensating for elevated % P), as well as increased variance in N : P associated with taking the mean of two variables.

How were these divergences in growth rate and body phosphorus composition reflected in trophic ecology and nutrient cycling aspects of *Daphnia* collected in the two regions? The ecological consequences were pronounced and consistent with stoichiometric theory, which predicts that P-rich animals should be less efficient recyclers of P and more sensitive to poor quality, high C : P food. Although no differences in N recycling were observed (Fig. 3A), Alaska *Daphnia* released little P whereas Wisconsin animals recycled P rather efficiently (Fig. 3B). Indeed, in five of seven experiments in Alaska we detected no P release whatsoever but measured substantial P release in all six nutrient recycling experiments in Wisconsin. Consistent with the close scavenging of P from the artificial food that we observed in the nutrient release experiments, Alaska animals also had more strongly reduced growth rates when raised on high C : P food (Fig. 3C). Note that these differences represent an underestimation of the food quality effect. We measured growth rates using length–weight regressions but it has been shown that animals grown on poor quality, P-limited food have reduced mass per unit length relative to animals grown on high-quality food (Sterner *et al*., 1993). Thus, consistent with stoichiometric theory and as anticipated by Arendt (1997), the high growth rate lifestyle of panarctic *D. pulex* exacts a more significant growth penalty when dietary conditions deteriorate. The magnitude of differences in P recycling rates and trophic response to food quality between Alaska and Wisconsin *Daphnia* (Fig. 3B,C) are striking in the light of relatively subtle difference in body phosphorus content that we observed (Fig. 2C). This suggests that the ecophysiological challenge of maintaining an elevated growth rate of a P-rich biomass may be even more difficult than previously imagined. Indeed, reduced growth on poor quality food may represent an ecological cost constraining the evolution of high growth capacity (Sterner & Schulz, 1998).

Our data may shed some light on puzzling aspects of the cluster of life history features associated with the *D. pulex* species complex and other taxa whose ranges extend across environmental extremes. The syndrome of special features associated with this species complex includes small genome size (Beaton & Hebert, 1988), a tendency for polyploidy (Ward *et al*., 1994), unexpected reductions in DNA content under polyploidy (Beaton & Hebert, 1988), and reproductive strategies that include obligate, rather than cyclical, parthenogenesis (Beaton & Hebert, 1988). All of these features change at high latitudes in this group, with increased polyploidy, decreased DNA content per genome, and a shift towards obligate parthenogenesis. To this we add our evidence of accelerated somatic growth rate (Fig. 2A). This complex of features can perhaps be better understood if we consider strong natural selection for rapid growth and development operating within stoichiometric constraints set by the quality of food available for conversion into P-intensive cellular machinery. As we have already argued, rapid growth rate requires enhanced capabilities for protein synthesis and thus elevated rRNA levels are often seen in rapidly growing organisms (Sutcliffe, 1970; Elser *et al*., 1996; Dobberfuhl, 1999). In addition to increased growth rates, extreme environments (high latitudes and altitudes, disturbed habitats, islands) also appear to favour polyploidy (Suomalainen, 1962), perhaps so that animals can overcome transcription

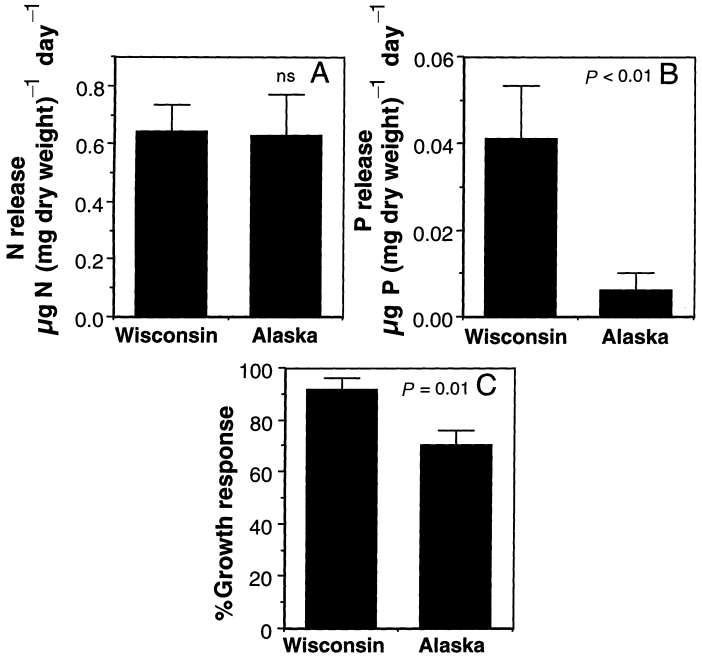

Fig. 3 Consequences for ecosystem processes (nutrient recycling, trophic efficiency) of differences in growth rate and P composition for *Daphnia* sampled in Alaska and Wisconsin. (A) Rate of N release per unit *Daphnia* biomass for temperate and arctic populations. Error bars here and in the following indicate ±1 standard error based on variation among ponds and lakes at a given sampling site. *t*-test: $t = 0.30$, $P = 0.76$ (two-tailed test), $n = 7$ Alaska populations, $n = 6$ Wisconsin populations. (B) Rate of P release per unit *Daphnia* biomass for temperate and arctic populations. *t*-test: $t = 2.91$, $P = 0.007$ (one-tailed test), $n = 7$ Alaska populations, $n = 6$ Wisconsin populations. Estimates of particle elimination rates indicated that arctic and temperate animals did not differ in rates of ingestion of P. Thus, the reduced P recycling rate for arctic animals also indicates a reduction in the percentage of ingested P recycled by the animals. (C) Response of body growth rate to poor food quality for temperate and arctic populations. Data are expressed as a percentage calculated as: 100 × (mean growth of animals fed low-quality food/mean growth of animals fed high-quality food). *t*-test: $t = 2.85$, $P = 0.01$ (one-tailed test), $n = 5$ Alaska populations, $n = 5$ Wisconsin populations. Estimates of particle elimination rates indicated that animals from the two regions did not differ in feeding rates. Thus, the larger reduction in growth rate for Alaskan animals when fed high C : P food indicates an actual reduction in trophic transfer efficiency.

limitations during accelerated mitosis. Accelerated growth via enhanced RNA levels and polyploidy involves investment in nucleic acids, one of the most P-rich classes of molecules in organisms (Elser *et al.*, 1996). However, an animal's ability to construct this P-intensive biochemical machinery at a rapid rate may be constrained by the stoichiometric quality of its food, especially food P content. Evidence for this is seen in our Alaska *Daphnia* samples (Fig. 3B,C). To overcome such P limitations, selection should favour P-sparing mechanisms, as the animal that develops fastest per unit P used will be favoured in competition. The known reduction in genomic DNA, especially repetitive, noncoding elements, in members of the *D. pulex* species complex (Bachmann & Rhinesmith, 1973) may be at least partially a manifestation of this response; however, other factors (such as reduction in genomic parasites under asexual reproduction) may also play a major role (Zeyl & Bell, 1996). Thus, reduction in overall genome size and in DNA content per genome in arctic *Daphnia* may be the consequence of evolution of a rapid life cycle within the stoichiometric constraint of available food. It is interesting to note that haploidy in diatoms has been proposed as a P-sparing adaptation increasing competitive ability under nutrient limitation (Lewis, 1985). Thus, the stoichiometric consequences of altered life history strategy may be a key but unappreciated aspect relevant to understanding the constraining factors operating on organism development rate (Arendt, 1997).

What functional consequences for nutrient cycling and trophic dynamics in lakes and ponds might we expect based on the differences in growth stoichiometry for Alaska and Wisconsin animals that we observed? To the extent that our sample populations are representative of general regions, one effect is that the P recycling regime driven by *Daphnia* should differ among regions. Provided that food C : P values were generally moderate, Wisconsin *Daphnia* would recycle a higher percentage of ingested P, thus sustaining further production by P-limited phytoplankton in the continuous cycle that is emphasized by conventional views of consumer-driven nutrient cycling (Lehman, 1980; Carpenter & Kitchell, 1984; Sterner, 1986). However, in a *Daphnia*-dominated food web in Northern Alaska, P appears to be on a 'one-way trip'.

P incorporated by algae that are then ingested by *Daphnia* would largely be retained by those animals until their death at the end of the brief growing season; rapidly growing Alaska *Daphnia* would largely be an internal 'sink' for P (Andersen, 1997). In oceanographic parlance (Dugdale & Goering, 1967), rapid growth rate lifestyles of dominant zooplankters in Alaskan lakes may result in pelagic ecosystems in which P-limited primary production is nearly 100% 'new' production (that is, entirely dependent on external sources). In contrast, in systems where grazers may have lower somatic P requirements, such as in the temperate Wisconsin lakes sampled, ecosystems would have a greater component of 'recycled' production in addition to the 'new' production supported by external P sources. As consumer trophic response also depends on body elemental composition owing to food quality effects, regional variation in consumer body stoichiometry should also affect food-web dynamics. In particular, stoichiometric modelling of algae–grazer interactions indicates that, relative to low-P animals, P-rich consumers such as those we sampled in arctic Alaska should have more erratic population dynamics and be more vulnerable to local deterministic extinction (Andersen, 1997).

Even though we cannot completely discount the role of phylogenetic history in production of the observed patterns, these results still emphasize the importance of extending consideration of the functional consequences of evolutionary processes to include biogeochemical processes and food–web interactions. Furthermore, the findings indicate that incorporating species into ecosystem frameworks requires consideration of the evolutionary context within which particular species are found. Much controversy has been associated with previous attempts to integrate evolutionary and ecosystem perspectives (Hagen, 1992). Some of this dispute may reflect misunderstandings by ecosystem ecologists about the level at which natural selection generally operates. Stoichiometric analysis avoids these difficulties by emphasizing the individual organism and its adaptive machinery in development of hypotheses in both evolutionary and ecosystem arenas (Fig. 1). Our findings also support the idea that biological stoichiometry is an appropriate framework for achieving a conceptual integration among biological subdisciplines and levels of organization (Reiners, 1986; Elser *et al*., 1996), a key priority for biology (Pickett *et al*., 1994).

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