Functional Ecology 2006
20, 514–521

Indirect effects of food web diversity and productivity on bacterial community function and composition

J. A. KRUMINS,† Z. T. LONG,* C. F. STEINER‡ and P. J. MORIN

Department of Ecology, Evolution and Natural Resources, 14 College Farm Road, Cook College, Rutgers University, New Brunswick, NJ 08901, USA

Summary

1. Previous evidence suggests that bacterially mediated decomposition of complex organic substrates increases with greater food web diversity. We attempted to identify changes in bacterial community composition and function associated with increased decomposition in more diverse food webs.

2. We used aquatic microcosms where we manipulated productivity with different initial nutrient concentrations. We created a diversity gradient by establishing communities of eukaryotes with zero (bacteria alone), one, two or four microbe species (protists and rotifers) in each of four trophic levels: producers, herbivores, bacterivores and predators. The initial bacterial community was standardized across all treatments. To determine effects of productivity and diversity on the bacterial community, we measured: decomposition, abundance, diversity of colony morphotypes (a measure of composition) and community level physiological profiles (CLPP) (a functional profile based on carbon substrate utilization).

3. Decomposition increased with greater eukaryotic species richness and was not influenced by productivity. Bacterial abundance remained constant with increasing eukaryotic species richness at low productivity, but significantly declined at high productivity. Eukaryotic species richness together with productivity influenced the composition of the bacterial community. However, the CLPP was strongly influenced by productivity and not species richness.

4. Food web diversity and productivity interact to influence bacterial community composition and function. In more diverse food webs, bacterial activity (decomposition) increased despite lower population abundance.

Key-words: bacteria, decomposition

Functional Ecology (2006) **20**, 514–521
doi: 10.1111/j.1365-2435.2006.01117.x

Introduction

The decomposition of organic matter by bacteria is a fundamental process in carbon cycling with potential impacts on community properties including patterns of species diversity (Jiang & Morin 2005), trophic structure (Hairston & Hairston 1993), nutrient flux (Harte & Kinzig 1993; Loreau 1994) and stability through increased mineralization of nutrients (Barsdate, Prentki & Fenchel 1974; DeAngelis 1992) and reduced fluctuations in nutrient availability (DeAngelis 1992). However, the connection between eukaryotic diversity and bacterial functioning remains poorly understood. This connection is important because bacteria play a central role in many ecosystem processes. Given that global declines in eukaryotic species diversity have occurred (Sala *et al*. 2000), it is important to explore the indirect effects of declining diversity on bacterial communities. Consumer effects on prey and productivity have been demonstrated in plant–herbivore models (De Mazancourt, Loreau & Abbadie 1998), and in experiments using microbial communities (Naeem & Li 1997), insect–plant interactions (Belovsky & Slade 2000) and ungulates grazing on the Serengeti grasslands (McNaughton 1976). The relationship between grazing by heterotrophic protozoa and bacterially mediated decomposition in aquatic

†Author to whom correspondence should be addressed. E-mail: jennadam@eden.rutgers.edu
*Present address: Institute of Marine Sciences University of North Carolina, Chapel Hill Morehead City, NC 28557, USA.
‡Present address: W. K. Kellogg Biological Station, 3700 E. Gull Lake Drive, Michigan State University, Hickory Corners, MI 49060, USA.

systems appears to be broadly similar to the positive effects of grazers on primary production in some terrestrial systems. Decomposition of organic matter (Fenchel & Harrison 1976; Sherr, Sherr & Berman 1982; Tso & Taghon 1999) and total community respiration (Coleman *et al*. 1978) both increase when protozoa graze on bacteria. The mechanisms causing this increase remain controversial. Grazing can liberate nutrients through bacterial cell lysis and excretion (Barsdate *et al*. 1974; Fenchel & Harrison 1976). Preferential grazing on larger bacteria can also change the composition of bacterial communities (Simek *et al*. 1995; Langenheder & Jurgens 2001). These results suggest different possible mechanisms underlying links between bacterial metabolic activity and consumer pressure. Much less is known about how the diversity of all trophic levels within a multitrophic food web can alter bacterial communities and decomposition (see McGrady-Steed, Harris & Morin 1997).

Numerous studies suggest that species diversity can influence various ecosystem properties including primary production, temporal stability and nutrient uptake (Tilman & Downing 1994; Hector *et al*. 1999; Tilman *et al*. 2001). The majority of this research has focused on primary producers and has not examined the effects of consumers (Duffy *et al*. 2001), or the effects of diversity distributed across an entire food web (Petchey *et al*. 2002). Within the small sample of studies that manipulated diversity over multiple trophic levels (Naeem *et al*. 1994; McGrady-Steed *et al*. 1997; Downing & Leibold 2002) and explored diversity effects on decomposition, only McGrady-Steed *et al*. (1997) found that an increase in decomposition accompanied increasing eukaryotic species richness. However, they did not identify proximal mechanisms for the response. Because bacterial diversity was not manipulated directly, positive indirect effects of eukaryotic species diversity on bacterially mediated processes seem plausible.

Indirect effects of eukaryotic diversity on the bacterial processes that drive decomposition could arise from two potentially interdependent mechanisms. First, the more diverse metabolic processes associated with greater eukaryotic species richness might enhance nutrient cycling and promote greater metabolic activity by bacteria (Barsdate *et al*. 1974; DeAngelis 1992). Second, a greater diversity of eukaryotic grazers may consume a wider variety of bacterial taxa, causing a change in bacterial community composition and functioning (Langenheder & Jurgens 2001). Here we test these hypotheses by describing effects of eukaryotic species diversity on decomposition and bacterial community composition and functional profiles. Microcosms are ideal for testing these hypotheses as they allow easy manipulation of diversity and provide a tractable model system to follow complex food web patterns over multiple generations (Gause 1934; Fenchel & Harrison 1976; McNaughton 1988; McGrady-Steed *et al*. 1997; Naeem & Li 1997; Petchey *et al*. 2002).

Methods

MICROCOSM ASSEMBLY

Aquatic microcosms were established with two levels of nutrient concentrations to manipulate productivity: 0·7 g l^{-1} of protist pellet (a nutrient source for protist cultivation from Carolina Biological Supply, Burlington, NC) and 1·67 g l^{-1} soil, or 0·07 g l^{-1} protist pellet and 0·167 g l^{-1} soil. Soil came from an organic garden on the Rutgers University campus and was used to supplement the media with trace nutrients and minerals. High-productivity and low-productivity treatments had total phosphorus concentrations of 145·8 µg l^{-1} and 25·4 µg l^{-1} respectively. These concentrations span the mesotrophic to hypereutrophic range (Wetzel 2001). Past research using similar manipulations of nutrient concentrations shows that they produce increases in the abundance and biomass of organisms that are consistent with increased productivity (Kaunzinger & Morin 1998; Jiang & Morin 2005; Steiner *et al*. 2005).

On day 10 of the experiment, one sterile, dried and preweighed wheat seed was added to each microcosm to provide the target substrate for decomposition. Gradients of diversity were created by establishing zero (bacteria control), one, two or four eukaryotic species in each of four trophic levels: primary producers (unicellular algae), algal consumers (protozoa and rotifers), bacterivores (protozoa) and top predators (protozoa). The zero eukaryotic diversity treatment was intended to create a control containing only bacteria (although contamination by one unidentified algal species and one small ciliate affected all treatments). The standardized bacteria-only control was established by inoculating microcosms with three species of bacteria (*Serratia marcescens*, *Bacillus cereus* and *Bacillus subtillus*) consumed by all bacterivores in our study plus an inoculum pooled from cultures of all eukaryotic species used in this experiment. Within each diversity treatment level, we established four different compositional combinations (Table 1) to avoid confounding eukaryotic species composition with diversity. Compositional combinations served as replicates for the diversity treatments, and species were randomly drawn from a pool of candidate species at each trophic level. Every treatment and compositional combination was replicated twice, except for the functional profile analysis (see below), which was replicated three times. Starting during the second week of the experiment and continuing weekly thereafter, 10% of the media (by volume) was replaced with fresh and sterile media. Microcosms were incubated at 22 °C with 12:12 h light : dark, and the experiment was maintained for 40 days.

SAMPLING

To evaluate the effects of eukaryotic diversity and productivity on bacterial community structure and

Table 1. Compositional combinations of diversity treatments

Trophic level	Organism	Diversity treatment level*		
		1	2	4
Producers	*Ankistrodesmus*		c	a,c,d
	Chlorella	c	a	a,b
	Chrysopsis		b,c	d
	Clamydomonas	b		c
	Cyclotella		a	a,b,c
	Euglena gracilis	d	b,d	a,b,c,d
	Unknown diatom sp.	a	d	
Herbivores	*Colpidium striatum*	d		b
	Frontonia	b	a	d
	Gastropus		d	
	Lepadella		b	a,c
	Monostyla		c	a,b
	Paramecium aurelia		a	b,c
	Paramecium caudatum	c		a,c,d
	Paramecium tetraurelia		b,d	
	Rotaria		c	b,c,d
	Stylonychia	a		a,d
Bacterivores	*Chilomonas*		b,c	c
	Coleps	d	a	c
	Colpoda cucullus	b	d	b,c
	Colpoda inflata		a	d
	Gastrotrich sp.			a,b
	Loxocephalus		c,d	
	Spirostomum ambiguum			a,d
	Tetrahymena pyriformis			b,c
	Tetrahymena thermophila	c		a,d
	Tilline		b	
	Unknown protist sp.	a		a,b,d
Top predators	*Actinosphaerium*		b	a,b,d
	Amoeba sp.		b	a,c
	Blepharisma americanum	b		d
	Didinium			b,d
	Dileptus	d	b	b,d
	Euplotes	c	c	b
	Oxytricha		a,c	
	Stentor coeruleus		d	a,b
	Tetrahymena vorax	a	d	a,c

*Compositional combinations a, b, c and d are shown to illustrate which species from each trophic level are represented in the different diversity treatments. Four compositional combinations were used for each diversity treatment level.

function, the following variables were measured: decomposition (as percentage dry weight lost of a wheat seed), bacterial abundance and community structure based on colony morphotypes on R2A agar (Difco Laboratories, Inc., Detroit, MI, a standard oligotrophic medium frequently used to culture aquatic environmental bacteria; Franklin *et al*. 2001; Garland *et al*. 2001; Muller *et al*. 2002), and community level physiological profiles (CLPP) (Garland & Mills 1991). The CLPP method serves as a relative measure of the functional diversity of a bacterial community using Ecolog (Biolog, Hayward, CA) microtitre plates containing an array of 31 different carbon substrates and a water control well. The response of the community to the carbon substrates is determined spectrophotometrically and provides a profile of that bacterial community's metabolic diversity.

To monitor food web diversity, rotifers and protists were sampled every 3–4 days up to day 22 and then every 3–6 days up to the final date of the experiment. Microcosms were first gently mixed and between 900 and 1500 µl medium were removed and examined with a dissecting microscope. Rare taxa were enumerated by counting the entire sample volume while abundant taxa were counted in smaller subsamples of known volume. Algae and small microflagellates were enumerated using a haemocytometer and a compound microscope.

Decomposition and bacterial enumeration

Wheat seeds were removed on day 40, dried for 48 h at 70 °C and weighed to determine mass loss, which was used to calculate percent decomposition. On day 20, bacterial abundance was measured using dilution plating on R2A agar. Microcosm suspensions were diluted tenfold, and plates were inoculated and incubated at room temperature for 48 h prior to counting.

Colony morphotype analysis

Colony morphotype analysis was conducted in a second experiment of similar design. Morphotypes were identified using a colony counter or dissecting microscope. Each morphotype was characterized by size, colour, margin, edge and elevation. The experiment was established as outlined above, but diversity treatments were limited to high (at least three species at each trophic level) and low levels (one species at each trophic level), and did not include compositional replicates. Up to 14 colony types were distinguishable on R2A agar and the relative abundance of the 10 present at the end of the experiment were estimated to characterize bacterial community composition. We acknowledge the difficulty in considering all bacterial taxa with culture-based methodology, and we emphasize that this approach provides only a relative comparison of the cultivable bacteria over our treatments (Ovreas & Torsvik 1998; Hughes *et al*. 2001; Ward 2002). However, prior studies using culture-based methods have effectively captured relative differences among experiment treatments (Garland *et al*. 2001; Muller *et al*. 2002). Hence, plate counts can be used as effective proxy measures of relative bacterial diversity and abundance.

Bacterial functional profile

CLPP were sampled on day 20 by diluting samples tenfold and inoculating each sample into Ecolog (Biolog Inc.) microtitre plates. Plates were incubated for 48 h in the dark at room temperature. Biolog plates contain a redox dye in each well that indicates metabolism of the substrate by bacteria. The response to the carbon substrates was determined using a spectrophotometer and absorbance at 590 nm; all substrate wells were blanked against the water control well. Owing to order

of magnitude differences in the abundance of bacteria between high- and low-productivity treatments, absorbance data acquired in the CLPP were normalized against the well with the maximum absorbance for each productivity treatment.

DATA ANALYSIS

Eukaryotic species richness is presented as 'average realized species richness' rather than as the initial richness in each treatment because not all introduced species persisted throughout the course of the experiment (Table 1). This value was calculated as the mean of eukaryotic richness over the course of the experiment for each replicate. An analysis of covariance (ANCOVA) explored the relationship between average realized species richness and decomposition and bacterial abundance. Productivity was treated as a fixed effect and average realized species richness as a continuous covariate in testing the hypothesis that bacterially mediated decomposition and abundance increased with increasing food web diversity at both productivity levels.

Principal components analysis (PCA) summarized the bacterial community profiles from both the colony morphotype analysis and the CLPP. The abundance of each colony morphotype was logarithmically transformed prior to analysis. The 10 colony morphotypes were treated as separate variables. To test the hypothesis that eukaryotic species richness and productivity altered the metabolic functional profile of the bacterial community, normalized absorbance measures of the CLPP were transformed into binary measures (1 = substrate used and 0 = substrate not used) and relationships between the diversity treatments and productivity summarized using PCA. All PCA analyses were followed by a multivariate analysis of variance (MANOVA). Productivity and diversity treatments were fixed factors while the first two component scores were the dependent variables. We conducted all statistical analyses using SAS version 9·1 (SAS Institute, Cary, NC).

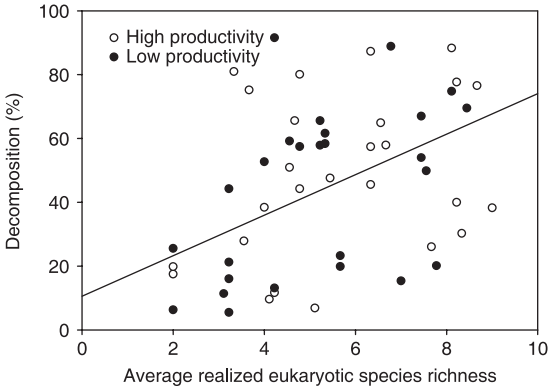

Fig. 1. The relationship between average realized species richness of protists and rotifers and percent decomposition of sterile dried wheat seeds. Productivity level is indicated by fill, and the trend line of all data is displayed for ease of interpreting data (ANCOVA species richness effect: $F = 8{\cdot}44$, $P < 0{\cdot}01$, $n = 52$) .

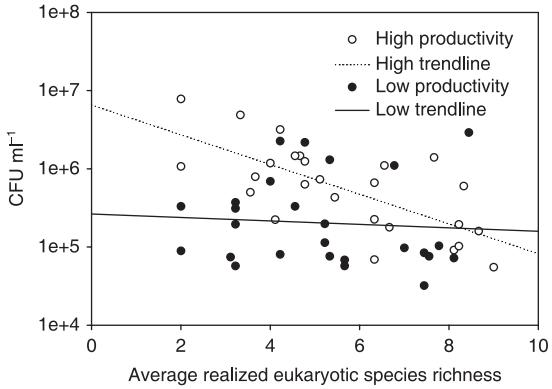

Fig. 2. The relationship between average realized species richness of protists and rotifers and bacterial abundance measured by dilution plating. Productivity level is indicated by fill, and trend lines are displayed for each productivity level for ease of interpreting data (ANCOVA species richness effect: $F = 7{\cdot}55$, $P < 0{\cdot}01$, $n = 26$; productivity effect: $F = 4{\cdot}17$, $P < 0{\cdot}05$, $n = 26$; interaction: $F = 10{\cdot}18$, $P < 0{\cdot}001$).

Results

Decomposition increased with increasing realized eukaryotic species richness (Fig. 1). ANCOVA showed that average realized species richness positively influenced decomposition ($F_{1,51} = 8{\cdot}44$, $P < 0{\cdot}01$), but productivity ($F_{1,51} = 0{\cdot}61$, $P = 0{\cdot}43$) had no detectable effect. Eukaryotic species diversity ($F_{1,51} = 7{\cdot}55$, $P < 0{\cdot}01$) and productivity ($F_{1,51} = 4{\cdot}17$, $P < 0{\cdot}05$) interacted to affect bacterial abundance (productivity × diversity, $F_{1,51} = 10{\cdot}18$, $P < 0{\cdot}001$, Fig. 2). At low productivity, bacterial abundance showed little change with increasing eukaryotic diversity, while at high productivity, bacterial abundance decreased with increasing eukaryotic diversity (Fig. 2).

Eukaryotic species diversity significantly altered bacterial community composition in the second experiment (Fig. 3 and Table 2, Wilk's lambda $F_{6,32} = 21{\cdot}28$, $P < 0{\cdot}0001$, a diversity effect). The high-diversity treatments in both high- and low-productivity treatments produced a different distribution of colony morphotypes than the low-diversity treatment or the control. The composition of the bacterial communities in the low-diversity treatments and the bacterial control were more similar in the same productivity level (Fig. 3, Wilk's lambda $F_{3,16} = 22{\cdot}12$, $P < 0{\cdot}0001$, a productivity effect) than the same diversity level.

Eukaryotic diversity treatments did not alter the functional profile of the bacterial community measured by the CLPP analyses. However, productivity significantly changed the functional profile of the bacterial communities (Fig. 4, Wilks' lambda $F_{3,68} = 53{\cdot}9$ and $P < 0{\cdot}0001$). Approximately half of the carbon substrates utilized in the microtitre plates were significantly correlated with variation described by principal component (PC) axis 1 (Table 3).

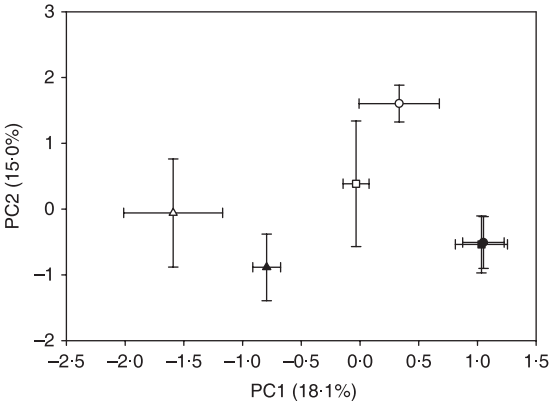

Fig. 3. The separation of bacterial community composition (summarized by the PCA of bacterial colony morphotypes) across high productivity (open symbols), low productivity (closed symbols) and the bacteria-only control (circle), low-diversity treatment (square), high-diversity treatment (triangle). For each diversity treatment symbol $n = 4$ and error bars indicate standard deviation.

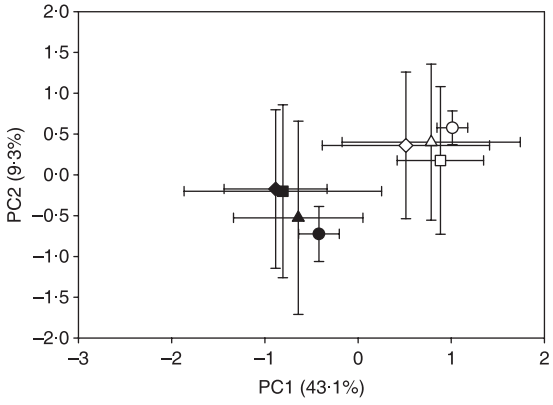

Fig. 4. The separation of bacterial community functional profiles (summarized by the PCA of bacterial carbon substrate utilization) across high productivity (open symbols), low productivity (closed symbols) and the bacteria-only control (circle), diversity treatments: level 1 (square), level 2 (triangle), level 4 (diamond). For each diversity treatment symbol $n = 3$ for the bacterial control and $n = 12$ for the diversity treatments. Error bars indicate standard deviation.

Discussion

Decomposition increased with increasing species richness in our food webs, a result that is consistent with previous findings in similar systems (Fenchel & Harrison 1976; McGrady-Steed *et al*. 1997). This result was not influenced by variation in productivity. The absence of a productivity effect was somewhat surprising since metabolic activity at all trophic levels should increase with nutrient availability. For example, Fenchel & Harrison (1976) found an increase in rate of decomposition in media enriched with NO_3^- and PO_4^{-3} presumably because bacteria were previously limited by those nutrients. Instead, we found that interactions between bacteria and the eukaryotes compensated for what would otherwise be a nutrient-limited environment. This conclusion is supported by an increase in wheat seed decomposition despite lower bacterial population size (Figs. 1 and 2). Thus, despite the fact that a higher diversity of consumers and producers apparently limited bacterial population sizes, those that remained were more metabolically active.

The patterns seen in our high-productivity treatment are analogous to the impacts of increased herbivore diversity on algal biomass observed in other studies (Naeem & Li 1997; Norberg 2000). Our finding suggests that effects of consumer diversity on the abundance of basal species are broadly similar regardless of whether those basal species are autotrophs or heterotrophs. Both producers and decomposers assimilate carbon and inorganic nutrients, but decomposers also mineralize organic matter into CO_2 and inorganic nitrogen and phosphorus making it available to producers and consumers in the food web. Although it was not directly measured in our study, increased material and nutrient flow associated with grazing may have accompanied a greater diversity of species across the entire food web. This is suggested by the decline in bacterial abundance concurrent with an increase in decomposition of the wheat seed (Figs. 1 and 2).

Theory suggests that a more diverse assemblage of species will more completely exploit a set of available resources (Tilman, Lehman & Thompson 1997). Likewise, we expect that a more diverse assemblage of eukaryotic consumers and osmotrophs might utilize more bacterial taxa and dissolved substrates. The resulting increase in material flow and diversity of metabolites may in turn have a positive effect on the metabolic activity of bacterial communities by potentially releasing them from nutrient limitation. Interactions between producers and decomposers (Harte & Kinzig 1993; Naeem, Hahn & Schuurman 2000; Loreau 2001) and autotrophs and heterotrophs (Naeem 2001) involve a reciprocal and obligatory exchange of material between these major functional groups. As decomposers are assumed to be carbon

Table 2. Correlations between bacterial colony morphotypes and principal component scores

	PC1 (18·1% variance)		PC2 (15·0% variance)	
Colony morphotype	*r*	*P*-value	*r*	*P*-value
Small clear	–0·43158	0·035	*	*
Butter	–0·78153	<0·0001	*	*
Yellow spreader	0·85279	<0·0001	0·42226	0·038
Tiny white	–0·68131	0·0002	*	*
Super tiny	*	*	*	*
Tiny yellow	*	*	–0·46526	0·022
Big white	0·42085	0·041	0·73576	<0·0001
Medium bright white	0·72438	<0·0001	*	*
Tiny orange	0·42732	0·037	*	*
Tiny pink	*	*	0·88045	<0·0001

*No significant correlation.

Table 3. Correlations between carbon substrates and principal component scores

	PC1 (43·1% variance)		PC2 (9·3% variance)	
Carbon substrate	*r*	*P*-value	*r*	*P*-value
Pyruvic acid methyl ester	0·76203	<0·0001	*	*
Tween 40	–0·35419	0·0015	*	*
Tween 80	*	*	*	*
α-Cyclodextrin	*	*	*	*
Glycogen	0·83275	<0·0001	0·49639	<0·0001
D-Cellobiose	0·76991	<0·0001	*	*
α-D-Lactose	*	*	*	*
B-Methyl-D-glucoside	0·85066	<0·0001	–0·25543	<0·05
D-Xylose	0·46258	<0·0001	*	*
I-Erythritol	*	*	*	*
D-Manitol	0·88405	<0·0001	*	*
N-Acetyl-D-glucosamine	0·91466	<0·0001	– 0·28187	0·0124
D-Glucosaminic acid	*	*	–0·23781	0·036
Glucose-1-phosphate	0·89144	<0·0001	*	*
D,L-α-Glycerol phosphate	0·60573	<0·0001	*	*
D-Galactonic acid-γ-lactone	0·80913	<0·0001	–0·32005	0·0043
D-Galacturonic acid	*	*	*	*
2-Hydroxybenzoic acid	*	*	*	*
4-Hydroxybenzoic acid	0·68383	<0·0001	*	*
γ-Hydroxybutyric acid	0·69685	<0·0001	0·3043	0·0068
Itaconic acid	*	*	*	*
α-Ketobutyric acid	*	*	*	*
D-Malic acid	0·45821	<0·001	0·40878	0·0002
L-Argenine	0·37272	0·0008	0·37018	0·0009
L-Asparagine	*	*	*	*
L-Phenylalanine	*	*	*	*
L-Serine	0·25506	0·02	*	*
L-Threonine	*	*	*	*
Glycyl- L-glutamic acid	*	*	*	*
Phenylethylamine	0·68228	<0·0001	0·4579	<0·0001
Putrescine	*	*	*	*

*No significant correlation.

limited (Hairston, Smith & Slobodkin 1960; Harte & Kinzig 1993; Naeem 2001), it is likely that any increase in carbon and nutrient flow associated with the distribution and diversity of eukaryotic species in a food web will facilitate bacterial metabolic activity. In this work, we hypothesized that a greater diversity of metabolites will result from more eukaryotic species in the food web, thus maximizing potential for metabolic activity by bacteria. We saw an increase in decomposition (and presumably bacterial metabolic activity) with increasing eukaryotic diversity. However, that change was not accompanied by a change in the metabolic profile (CLPP) of the bacterial assemblage (Fig. 3). Substrate use by bacterial communities did not vary with eukaryotic diversity, but we did observe differences associated with the productivity level of the microcosm. Bacterial communities in high- and low-productivity treatments were markedly different in the carbon sources they utilized (Table 3), but they did not differ overall in their aggregate activity affecting decomposition. It is interesting that the bacteria utilized a different suite of carbon substrates depending on their environmental productivity level. This may be due to the metabolic state of the bacterial assemblage (i.e. stationary *vs* growth phase) that is secondary to the metabolic benefits of a diverse food web that we propose here.

Consumer-mediated shifts in species composition may influence the whole community's ability to utilize complex carbon sources such as wheat seeds, which served as a convenient proxy for other kinds of allochthonous carbon degraded by aquatic microbial communities. In fact, we hypothesized that a greater diversity of grazers in the food web will utilize a wider variety of bacterial taxa and alter the composition of the bacterial community. In this case, if we assume that resources were plentiful in the high-productivity treatments, bacterial abundance was limited by a greater diversity of consumers that were perhaps capable of exploiting more bacterial taxa (Fig. 2). Similar patterns have been observed for relationships between an assemblage of primary producers and their consumers in other studies (Naeem & Li 1997). However, in this experiment, the grazers in high-diversity treatments not only consumed more bacteria, but also changed bacterial community composition (Fig. 3 and Table 2). This result, together with the decline in bacterial abundance, suggests that grazing selects for more productive bacteria in more diverse food webs.

Productivity and consumer diversity interact to indirectly increase bacterially mediated decomposition. This interaction is illustrated by the combined influence of nutrient limitation in low-productivity, low-diversity food webs and by more pressure on bacterial taxa when diversity is high. The complex interactions between productivity, eukaryotic diversity and bacterial communities may be explained by trophic level interactions. For instance, diversity in multitrophic communities yields increased stability (Steiner *et al*. 2005) and predictability (McGrady-Steed *et al*. 1997; Morin & McGrady-Steed 2003), and bacterially mediated decomposition stabilizes communities through regulation of nutrients in the environment (DeAngelis 1992). The combination of bottom-up effects from bacterially mediated decomposition and top-down effects from multitrophic diversity may work synergistically to create stable and productive communities. Research and theory have demonstrated that the interactions between functional groups can be complex (Harte & Kinzig 1993; Naeem *et al*. 2000; Loreau 2001).

We propose that the indirect effects of grazers on decomposition occurred through a combination of two influences: (1) media enrichment associated with heterotroph and autotroph activity leading to increased metabolic activity and (2) selection for a different and more productive bacterial community composition. The implications of this result to biodiversity and ecosystem function research are important. First, the consequences of increased species richness to ecosystem function are not limited to interactions within discrete trophic levels (Duffy *et al*. 2001; Petchey *et al*. 2002). Second, bacterial communities and the processes that they control must be studied in the context of complete

food webs to avoid missing key interactions with other functional groups (Cochran-Stafira & von Ende 1998). We propose that indirect consequences of altered biodiversity may be as important as direct effects of diversity on function.

Knowledge about the important causes and consequences of biodiversity is growing, but there is still a critical need for research that explores the consequences of changing diversity on multiple trophic levels (Hooper *et al*. 2005). The difficulty with incorporating bacteria into such multitrophic level studies involves very different scales of sampling (Hughes *et al*. 2001; Horner-Devine, Carney & Bohannan 2003) and different interpretations given to prokaryotic and eukaryotic diversity (Martiny *et al*. 2006). Our work did not directly manipulate bacterial diversity and functioning (see, e.g., Bell *et al*. 2005). However, we evaluated mechanisms that highlight the potentially important role of bacterial consumers in diversity studies. Such interactions are likely to be important in most food webs where bacteria and eukaryotes interact in potentially complex ways.

Acknowledgements

This work was partially funded by NASA's Graduate Student Researchers Program to J. A. Krumins, the Cook College NJAES and an NSF Microbial Biology Postdoctoral Fellowship to C. F. Steiner. We would like to thank Valdis Krumins, Aabir Banerji and Lin Jiang for valuable comments and discussion. Two anonymous reviewers made comments that greatly improved this manuscript.

References

Barsdate, R.J., Prentki, R.T. & Fenchel, T. (1974) Phosphorus cycle of model ecosystems: significance for decomposer food chains and effect of bacterial grazers. *Oikos* **25**, 239–251.

Bell, T., Newman, J.A., Silverman, B.W., Turner, S.L. & Lilley, A.K. (2005) The contribution of species richness and composition to bacterial services. *Nature* **436**, 1157–1160.

Belovsky, G.E. & Slade, J.B. (2000) Insect herbivory accelerates nutrient cycling and increases plant production. *Proceedings of the National Academy of Sciences USA* **97**, 14412–14417.

Cochran-Stafira, D.L. & von Ende, C.N. (1998) Integrating bacteria into food webs: studies with *Sarracenia purpurea inquilines*. *Ecology* **79**, 880–898.

Coleman, D.C., Anderson, R.V., Cole, C.V., Elliott, E.T., Woods, L. & Campion, M.K. (1978) Trophic interactions in soils as they affect energy and nutrient dynamics. IV. Flows of metabolic and biomass carbon. *Microbial Ecology* **4**, 373–380.

DeAngelis, D.L. (1992) *Dynamics of Nutrient Cycling and Food Webs*. Chapman and Hall, New York.

De Mazancourt, C., Loreau, M. & Abbadie, L. (1998) Grazing optimization and nutrient cycling: when do herbivores enhance plant production? *Ecology* **79**, 2242–2252.

Downing, A.L. & Leibold, M.A. (2002) Ecosystem consequences of species richness and composition in pond food webs. *Nature* **416**, 837–841.

Duffy, J.E., Macdonald, K.S., Rhode, J.M. & Parker, J.D. (2001) Grazer diversity, functional redundancy, and productivity in sea grass beds: an experimental test. *Ecology* **82**, 2417–2434.

Fenchel, T. & Harrison, P. (1976) The significance of bacterial grazing and mineral cycling for the decomposition of particulate detritus. *The Role of Terrestrial and Aquatic Organisms in the Decomposition Process* (ed. A. Macfadyn). Blackwell Scientific Publications, London.

Franklin, R.B., Garland, J.L., Bolster, C.H. & Mills, A.L. (2001) Impact of dilution on microbial community structure and functional potential: comparison of numerical simulations and batch culture experiments. *Applied and Environmental Microbiology* **67**, 702–712.

Garland, J.L. & Mills, A.L. (1991) Classification and characterization of heterotrophic microbial communities on the basis of patterns on community-level-sole-carbon-source utilization. *Applied and Environmental Microbiology* **57**, 2351–2359.

Garland, J.L., Cook, K.L., Adams, J.L. & Kerkhof, L. (2001) Culturability as an indicator of succession in microbial communities. *Microbial Ecology* **42**, 150–158.

Gause, G.F. (1934) *The Struggle for Existence*. Williams and Wilkins, Baltimore.

Hairston, N.G. Jr & Hairston, N.G. Sr (1993) Cause effect relationship in energy flow, trophic structure, and interspecific interactions. *American Naturalist* **142**, 379–411.

Hairston, N.G., Smith, F.E. & Slobodkin, L.B. (1960) Community structure, population control, and competition. *American Naturalist* **94**, 421–425.

Harte, J. and Kinzig, A.P. (1993) Mutualism and competition between plants and decomposers: implications for nutrient allocation in ecosystems. *American Naturalist* **141**, 829–846.

Hector, A., Schmid, B., Beierkuhnlein, C., Caldeira, M.C., Diemer, M., Dimitrakopoulos, P.G., Finn, J.A., Freitas, H., Giller, P.S., Good, J., Harris, R., Hogberg, P., Huss-Danell, K., Joshi, J., Jumpponen, A., Korner, C., Leadley, P.W., Loreau, M., Minns, A., Mulder, C.P.H., O'Donovan, G., Otway, S.J., Pereira, J.S., Prinz, A., Read, D.J., Scherer-Lorenzen, M., Schulze, E.D., Siamantziouras, A.S.D., Spehn, E.M., Terry, A.C., Troumbis, A.Y., Woodward, F.I., Yachi, S. & Lawton, J.H. (1999) Plant diversity and productivity experiments in European grasslands. *Science* **286**, 1123–1127.

Hooper, D.U., Chapin, S.F., Ewel, J.J., Hector, A., Inchausti, P., Lavorel, S., Lawton, J.H., Lodge, D.M., Loreau, M., Naeem, S., Schmid, B., Setala, H., Symstad, A.J., Vandermeer, J.H. & Wardle, D.A. (2005) Effects of biodiversity on ecosystem functioning: a consensus of current knowledge. *Ecological Monographs* **75**, 3–35.

Horner-Devine, M.C., Carney, K.M. & Bohannan, B.J.M. (2003) An ecological perspective on bacterial biodiversity. *Proceedings of the Royal Society of London B* **271**, 113–122.

Hughes, J.B., Hellmann, J.J., Ricketts, T.H. & Bohannan, B.J.M. (2001) Counting the uncountable: statistical approaches to estimating microbial diversity. *Applied and Environmental Microbiology* **67**, 4399–4406.

Jiang, L. & Morin, P.J. (2005) Predator diet breadth influences the relative importance of bottom-up and top-down control of prey biomass and diversity. *American Naturalist* **165**, 350–363.

Kaunzinger, C.M.K. & Morin, P.J. (1998) Productivity controls food chain properties in microbial communities. *Nature* **395**, 495–497.

Langenheder, S. & Jurgens, K. (2001) Regulation of bacterial biomass and community structure by metazoan and protozoan predation. *Limnology and Oceanography* **46**, 121–134.

Loreau, M. (1994) Material cycling and the stability of ecosystems. *American Naturalist* **143**, 508–513.

Loreau, M. (2001) Microbial diversity, producer–decomposer interactions and ecosystem processes: a theoretical model. *Proceedings of the Royal Society of London Series B* **268**, 303–309.

Martiny, J.B.H., Bohannan, B.J.M., Brown, J.H., Colwell, R.K., Fuhrman, J., Green, J., Horner-Devine, M.C., Kane, M., Krumins, J.A., Kuske, C., Morin, P.J., Naeem, S., Ovreas, L. & Reysenbach, A.-L. (2006) Microbial biogeography: putting microbes on the map. *Nature Reviews Microbiology* **4**, 102–112.

McGrady-Steed, J., Harris, P.M. & Morin, P.J. (1997) Biodiversity regulates ecosystem predictability. *Nature* **390**, 162–165.

McNaughton, S.J. (1976) Serengeti migratory wildebeest: facilitation of energy flow by grazing. *Science* **191**, 92–94.

McNaughton, S.J. (1988) Diversity and stability. *Nature* **333**, 204–205.

Morin, P.J. & McGrady-Steed, J. (2003) Biodiversity and ecosystem functioning in aquatic microbial systems: a new analysis of temporal variation and species richness. *Oikos* **104**, 458–466.

Muller, A.K., Westergaard, K., Christensen, S. & Sorensen, S.J. (2002) The diversity and function of soil microbial communities exposed to different disturbances. *Microbial Ecology* **44**, 49–58.

Naeem, S. (2001) Autotrophic–heterotrophic interactions and their impacts on bidiversity and ecosystem function. *The Functional Consequences of Biodiversity* (ed. D. Tilman), Vol. 33. Princeton University Press, Princeton, NJ.

Naeem, S. & Li, S. (1997) Biodiversity enhances ecosystem reliability. *Nature* **390**, 507–509.

Naeem, S., Thompson, L.J., Lawler, S.P., Lawton, J.H. & Woodfin, R.M. (1994) Declining biodiversity can alter the performance of ecosystems. *Nature* **368**, 734–737.

Naeem, S., Hahn, D.R. & Schuurman, G. (2000) Producer–decomposer co-dependency influences biodiversity effects. *Nature* **403**, 762–764.

Norberg, J. (2000) Resource-niche complementarity and autotrophic compensation determines ecosystem-level responses to increased cladoceran species richness. *Oecologia* **122**, 264–272.

Ovreas, L. & Torsvik, V. (1998) Microbial diversity and community structure in two different agricultural soil communities. *Microbial Ecology* **36**, 303–315.

Petchey, O.L., Morin, P.J., Hulot, F.D., Loreau, M., McGrady-Steed, J. & Naeem, S. (2002) Contributions of aquatic model systems to our understanding of biodiversity and ecosystem functioning. *Biodiversity and Ecosystem Functioning: Synthesis and Perspectives* (ed. P. Inchausti). Oxford University Press, Oxford.

Sala, O.E., Chapin, S.F., Armesto, J.J., Berlow, E., Bloomfield, J., Dirzo, R., Huber-Sanwald, E., Huenneke, L.F., Jackson, R.B., Kinzig, A.P., Leemans, R., Lodge, D.M., Mooney, H.A., Oesterheld, M., Poff, N.L., Sykes, M.T., Walker, B.F., Walker, M. & Wall, D.H. (2000) Global biodiversity scenarios for the year 2100. *Science* **287**, 1770–1774.

Sherr, B.F., Sherr, E.B. & Berman, T. (1982) Decomposition of organic detritus: a selective role for microflagellate Protozoa. *Limnology and Oceanography* **27**, 765–769.

Simek, K., Bobkova, J., Macek, M., Nedoma, J. & Pseener, R. (1995) Ciliate grazing on picoplankton in a eutrophic reservoir during the summer phyoplankton maximum: a study at the species and community level. *Limnology and Oceanography* **40**, 1077–1090.

Steiner, C.F., Long, Z.T., Krumins, J.A. & Morin, P.J. (2005) Temporal stability of aquatic food webs: partitioning the effect of species diversity, species composition and enrichment. *Ecology Letters* **8**, 819–828.

Tilman, D. & Downing, J.A. (1994) Biodiversity and stability in grasslands. *Nature* **367**, 363–365.

Tilman, D., Lehman, C. & Thompson, K.T. (1997) Plant diversity and ecosystem productivity: theoretical considerations. *Proceedings of the National Academy of Sciences USA* **94**, 1857–1861.

Tilman, D., Reich, P.B., Knops, J., Wedin, D., Mielke, T. & Lehman, C. (2001) Diversity and productivity in a long-term grassland experiment. *Science* **297**, 843–845.

Tso, S.F. & Taghon, G.L. (1999) Factors affecting predation by *Cyclidium* sp. and *Euplotes* sp. on PAH-degrading and nondegrading bacteria. *Microbial Ecology* **37**, 3–12.

Ward, B. (2002) How many species of prokaryotes are there? *Proceedings of the National Academy of Sciences USA* **99**, 10234–10236.

Wetzel, R.G. (2001) *Limnology: Lake and River Ecosystems*, 3rd edn. Academic Press, New York.

Received 23 November 2005; revised 25 February 2006; accepted 3 March 2006
Editor: Owen Petchey