NUTRIENT ENRICHMENT OF A DETRITUS-BASED STREAM ECOSYSTEM: EFFECTS ON INVERTEBRATE COMMUNITY STRUCTURE AND FUNCTION

by

WYATT FIRMIN CROSS

(Under the Direction of J. Bruce Wallace)

ABSTRACT

Nutrient enrichment of aquatic ecosystems is occurring globally as a result of anthropogenic changes to the availability and mobility of elements such as nitrogen (N) and phosphorus (P). Currently, very little is known about the effects of nutrient enrichment on detritus-based ecosystems despite the prevalence and global significance of detritus in most energetic budgets. This study was aimed at quantifying effects of an experimental nitrogen and phosphorus enrichment on invertebrate consumers and basal resources in a detritus-based headwater stream. Efforts were focused on examining nutrient-induced changes in a) the quantity and quality of detrital food resources and b) invertebrate growth rates, secondary production, food web dynamics, and stoichiometry. Enrichment had a large positive effect on the quality of basal resources (i.e., leaf litter, fine benthic organic matter, and epilithon) though increased N and P content. In contrast, the quantity of basal resources declined overall due to a significant reduction in benthic leaf litter. Both primary and secondary invertebrate consumers exhibited a positive response to enrichment through increased biomass and secondary production. However, there were large differences in response among individual taxa, which was related to an interaction between two dominant life history characteristics – larval lifespan and feeding behavior. Long-lived taxa (>1 year) showed little or no response to enrichment, whereas many short-lived taxa (<1 year) showed large positive responses to enrichment. Individual growth rates of chironomid larvae increased during enrichment, while those of *Tallaperla* spp. were not affected. Enrichment had no effect on the diets of consumers, but there were large effects on total organic matter flows to all functional feeding groups. Patterns of consumer stoichiometry indicated distinctly lower relative P content of detritus-based consumers in comparison to previously published data from plant- or algal-based communities. In addition, consumer P content was elevated in the treatment stream during enrichment, demonstrating that some insect consumers are much less homeostatic than other well-studied crustaceans. These data suggest potential evolutionary adaptations of detritus-based consumers to nutrient poor food resources. This dissertation should provide a basis for predicting how landscape-scale nutrient enrichment may affect community structure, production, and material cycling in forested headwater streams.

INDEX WORDS: Stream, Detritus, Invertebrate, Secondary production, Growth, Trophic basis of production, Ecological stoichiometry, Food web, Leaf litter, Coweeta

NUTRIENT ENRICHMENT OF A DETRITUS-BASED STREAM ECOSYSTEM: EFFECTS ON INVERTEBRATE COMMUNITY STRUCTURE AND FUNCTION

by

Wyatt Firmin Cross

B.A. The University of Colorado, 1994

M.S., The University of Alabama, 1999

A Dissertation Submitted to the Graduate Faculty of the University of Georgia in Partial Fulfillment of the Requirements for the Degree

DOCTOR OF PHILOSOPHY

ATHENS, GEORGIA

2004

NUTRIENT ENRICHMENT OF A DETRITUS-BASED STREAM ECOSYSTEM: EFFECTS ON INVERTEBRATE COMMUNITY STRUCTURE AND FUNCTION

by

WYATT FIRMIN CROSS

Major Professor:	J. Bruce Wallace
Committee:	Amy D. Rosemond Darold Batzer Mark D. Hunter Judy L. Meyer

Electronic Version Approved:

Maureen Grasso
Dean of the Graduate School
The University of Georgia
August 2004

ACKNOWLEDGEMENTS

I am deeply indebted to the huge number of people who helped throughout this dissertation. Most importantly, I thank my major professor (and colleague), J. Bruce Wallace, for his support, insight, and friendship throughout my stay. I also thank the other members of my committee, Amy D. Rosemond, Judy L. Meyer, Darold Batzer, and Mark D. Hunter for their unique approaches and input which greatly improved the study. Amy Rosemond deserves special thanks for putting so much thought and time into the study design, interpretation, and writing – and for being someone I could always go to for advice. I am also grateful to my family for periodically 'grounding' me, and for continual encouragement, interest, and faith that one day, I would finish – my mother Pat, my brother Jake, and my sister-in-law Courtney. I also owe a huge thanks to my girlfriend, Zab, for her help with many aspects of the study, and her paramount and unconditional support during the ups and downs. I thank the many (>20) laboratory assistants – most notably Lizzy Melda, Catherine Evans, Andrew Paloucci, and Gavin Veitch – who spent literally thousands of hours removing bugs from mud, or weighing dirt (glorious stuff). I also thank Boggs Benstead, Heidi Wilcox, Sue Eggert, and Sally Entrekin for their insight and help at various stages of the study. Many graduate students in both Ecology and Entomology were great to me during my stay, and will undoubtedly remain long-term friends. Loads of thanks also goes out to Janice Sand for making the Institute of Ecology what it is. Finally, thanks to Lake Oglethorpe for good times fishin' with good brobies and peebers.

TABLE OF CONTENTS

Page

CHAPTER 1

INTRODUCTION

General context

Concentrations of biologically available nutrients, such as nitrogen (N) and phosphorus (P), are increasing in aquatic and terrestrial ecosystems worldwide (e.g., Vitousek et al. 1997, Carpenter et al. 1998, Galloway et al. 2003). Humans have contributed substantially to this trend through activities such as fossil fuel combustion, fertilizer production and application, mining, and suburban/urbanization. The net effect has been large-scale mobilization of nutrients and changes in local and global biogeochemical cycles (e.g., Caraco 1993, Vitousek et al. 1997, Caraco and Cole 1999). Such changes can have drastic effects on species and ecosystems because the rates of many biochemical processes are often limited by the supply of N and P. A current challenge for ecologists is to understand the long-term consequences of increased nutrient availability on population and ecosystem dynamics, and to develop predictive conceptual and empirically based models to better forecast future trends. To this end, experimental studies will be critical for predicting ecological change associated with nutrient enrichment, providing theoretical insight, and for developing effective management strategies to minimize potentially deleterious effects (e.g., Carpenter et al. 1995). This dissertation was part of a long-term ecosystem-scale experiment aimed at understanding the effects of nutrient enrichment on the structure and function of detritus-based stream ecosystems.

While much is known concerning the effects of nutrients on living plant- or algal-based ecosystems (e.g., Tilman 1987, 1996, Vitousek and Howarth 1991, Peterson et al. 1993), heretofore little attention has been given to ecosystems based on detritus. This is surprising when one considers the prevalence and energetic importance of detritus in most food webs and ecosystems (e.g., Odum and de la Cruz 1963). Indeed, the vast majority of global primary production goes unconsumed by herbivores, and ultimately fuels detritus-based food webs (e.g., O'Neill and Reichle 1980). Research on nutrient effects in these systems is critical because the response of detritus-based communities and ecosystem processes may be quite different than those documented in plant- or algal-based systems.

A fundamental difference between detrital and plant-based systems is the composition of the predominant basal resource. In detrital food webs, the resource base is largely heterotrophic, and detritus and associated microbes (i.e., bacteria and fungi) occupy a similar trophic position as living plants or algae. This has important implications because nutrient enrichment can enhance the productivity of these detritus-associated microbes and accelerate the loss or mineralization of carbon at the base of the food web (e.g., Gulis and Suberkropp 2003). In contrast, nutrient enrichment of plant-based systems generally leads to a net carbon gain among producers at the base of the food web (e.g., Tilman 1996, Slavik et al. 2004)

Freshwater ecosystems

Freshwater ecosystems are among the most threatened by nutrient enrichment (Carpenter et al. 1998). This is due, in part, to their low-lying position in the landscape, acting as recipients and vectors for nutrient loading, processing, and transport. Additionally, freshwater systems are subject to considerable human development of the riparian zone leading to significant nutrient loading from point- and non point-sources (e.g., Caraco and Cole 1999). In general, our understanding of nutrient enrichment effects on freshwater ecosystems comes from theoretical and empirical work conducted in algal-based systems. In these systems, studies have shown that enrichment can lead to increased algal biomass or productivity (e.g., Schindler et al. 1978, Hart and Robinson 1990, Peterson et al. 1993, Mazumder and Edmonson 2002), changes in algal nutrient content (e.g., Stelzer and Lamberti 2001), and shifts in algal community structure or species diversity (e.g., Barnese and Schelske 1994). Such nutrient-induced alterations to the base of these food webs may 'cascade up' (Hunter and Price 1992) to primary and secondary consumers, having potentially significant effects on life history traits, community structure, and secondary production (e.g., Peterson et al. 1993, Mazumder and Edmonson 2002, Slavik et al. 2004). In some cases, enrichment can indirectly modify the strength or importance of top-down consumer effects (e.g., Rosemond et al. 1993, Peterson et al. 1993, Forkner and Hunter 2000).

In detritus-based stream ecosystems, terrestrially-derived leaf litter provides the dominant energetic source for microbial and metazoan productivity, and in-stream algal production is generally minimal (e.g., Webster et al. 1997, Wallace et al. 1999). Thus, nutrient enrichment effects are manifested through changes to detrital resources. Considerable research has examined the effects of nutrient enrichment on the decomposition (e.g., Elwood et al. 1981, Meyer and Johnson 1983, Chadwick and Huryn 2003, Gulis and Suberkropp 2003) and quality of detritus (Howarth and Fisher 1976, Cross et al. 2003, Stelzer et al. 2003), as well as biomass and production of detritus-associated microbes (Suberkropp 1995, Gulis and Suberkropp 2003, Rosemond et al. 2002, Ramirez et al. 2003). In general, nutrient effects on detrital quality are positive (via increased microbial biomass and nutrient content) and effects on quantity are negative (via accelerated decomposition). Few studies, however, have examined how these basal changes affect the long-term dynamics of higher consumers (but see Rosemond et al.2002, Ramirez and Pringle, in review); herein lies the primary goal of this dissertation. A conceptual working hypothesis which provided the theoretical backdrop for this project is depicted in Figure

1.1. In low-nutrient detritus-based streams (Figure 1.1A), benthic leaf litter is expected to decline slowly each year providing sufficient carbon throughout the year to support consumer growth and production. However, the quality of this detritus is relatively low. In nutrient-enriched streams (Figure 1.1B), rapid decomposition and mineralization of organic matter may lead to periods of time during the year when consumers are limited by carbon availability. The quality of this enriched organic matter, however, is expected to be high. To date, few studies have been conducted for sufficient duration to test this working hypothesis. This dissertation represents one of the first attempts to quantify in detail the response of detritivorous and predatory consumers to a long-term experimental enrichment (i.e., nitrogen and phosphorus) of a detritus-based ecosystem. The approach taken was to examine effects of nutrient enrichment at multiple hierarchical scales ranging from elemental patterns to ecosystem processes.

Experimental design

A paired-catchment design was used to examine long-term effects of nutrient enrichment. Streams draining two adjacent catchments at Coweeta Hydrologic Laboratory, North Carolina, were chosen for study because of similar physical, chemical, and biological characteristics (see Table 3.1). Research was conducted in both streams before and during 2 years of experimental enrichment. The mechanics of the experimental set-up are described in detail in Chapters 3, 4, and 5.

This ecosystem-level experiment was not replicated, employing 1 reference stream and 1 treated stream. This approach has been lauded for its realism and scale (e.g., Carpenter et al. 1995, Schindler 1998, Oksanen 2001), as well as criticized for its lack of strict replication or inference potential (Hurlbert 1984, 2004). The limitations of our design are recognized, but we felt that the scale of manipulation was essential for predicting realistic effects. Such catchment-level manipulations have indeed been critical for advancing our understanding of ecosystem dynamics and effects of perturbations (e.g., Likens et al. 1969, Wallace et al. 1997, Schindler 1998, Pace et al. 2004). Throughout the study, time-series data were examined statistically using randomized intervention analysis (Carpenter et al. 1989, Stewart-Oaten 2003, Murtaugh 2003). This method compares differences between the reference and treated systems before and after an experimental manipulation (i.e., in this case, nutrient enrichment). Generally, this statistic tests the null hypothesis of no change in the treated system relative to the reference system after the manipulation.

Dissertation

The goal of Chapter 2 is to describe the elemental composition of consumers and basal resources in the enriched stream and the reference stream. This chapter is a preliminary attempt to test some of the basic theoretical tenets of ecological stoichiometry. In doing so, it sets the stage for later chapters by reporting changes in the quality of organic matter and describing trends in nutrient content of invertebrates. Basic descriptive work on elemental composition of food webs (particularly non-algal) will be critical for determining the robustness of stoichiometric theory.

Chapter 3 examines in detail the response of 2 dominant detritivores (i.e., non-Tanypodinae chironomids and *Tallaperla* spp.) to nutrient enrichment. The goal was to contrast growth and secondary production of 2 taxa with highly disparate larval lifespan and feeding behavior. Such detailed growth studies were not logistically possible for all dominant primary consumers, so it was my hope that these taxa would represent opposite ends of the 'slow-fast' life history continuum. The differential response of these consumers provides insight into the interaction between lifespan and feeding behavior in determining the response of invertebrates to enrichment.

Chapter 4 examines the effects of enrichment on the structure and secondary production of the entire invertebrate community, as well as the storage dynamics of coarse and fine particulate organic matter. In addition relationships between predator and prey production are examined. Results are placed in a broader context by comparison with multiple years (21) of data collected from headwater streams at Coweeta. This chapter will be among the first to quantify long-term effects of nutrient enrichment on community secondary production in a detritus-based ecosystem.

The final study chapter (5) explores the effects of nutrient enrichment on food web dynamics. This study was designed to better understand mechanisms responsible for community-level changes in secondary production (Chapter 4). I quantified the trophic basis of production for all dominant consumers in both streams before and during enrichment. Organic matter flow food webs were also constructed to determine whether the magnitude or direction (i.e., major taxa involved) of flows was affected by enrichment. Such an approach can aid in determining whether nutrient-induced changes in secondary production were due to actual shifts in the diets of consumers or changes in the quality or basal resources.

Literature Cited

Barnese, L. E. and C. L. Schelske. 1994. Effects of nitrogen, phosphorus, and carbon enrichment on planktonic and periphytic algae in a softwater, oligotrophic lake in Florida, USA. Hydrobiologia 277:159-170.

Caraco, N. F. 1993. Disturbance of the phosphorus cycle: a case of indirect effects of human activity. Trends in Ecology and Evolution 8:51-54.

Caraco, N. F. and J. J. Cole. 1999. Human impact on nitrate export: an analysis using major world rivers. Ambio 28:167-170.

Carpenter, S. R., N. F. Caraco, D. L. Correll, R. W. Howarth, A. N. Sharpley, and V. H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. Ecological Applications 8:559-568.

Carpenter, S. R., S. W. Chisholm, C. J. Krebs, D. W. Schindler, and R. F. Wright. 1995. Ecosystem experiments. Science 269:324-327.

Carpenter S. R., T. M. Frost, D. Heisey, and T. K. Kratz. 1989. Randomized intervention analysis and the interpretation of whole-ecosystem experiments. Ecology 70:1142-1152.

Chadwick, M. A. and A. D. Huryn. 2003. Effect of a whole-catchment N addition on stream detritus processing. Journal of the North American Benthological Society 22:194-206.

Cross, W. F., J. P. Benstead, A. D. Rosemond , and J. B. Wallace. 2003. Consumer-resource stoichiometry in detritus-based streams. Ecology Letters 6:721-732.

Elwood, J. W., J. D. Newbold, A. F. Trimble, and R. W. Stark. 1981. The limiting role of phosphorus in a woodland stream ecosystem – effects of P-enrichment on leaf decomposition and primary producers. Ecology 62:146-158.

Forkner, R. E. and M. D. Hunter. 2000. What goes up must come down? Nutrient addition and predation pressure on oak herbivores. Ecology 81:1588-1600.

Galloway, J. N., J. D. Aber, J. W. Erisman, S. P. Seitzinger, R. W. Howarth, E. B. Cowling, and B. J. Cosby. 2003. The nitrogen cascade. BioScience 53:341-356.

Gulis, V. and K. Suberkropp. 2003. Leaf litter decomposition and microbial activity in nutrient-enriched and unaltered reaches of a headwater stream. Freshwater Biology 48:123-134.

Hart, D. D., and C. T. Robinson. 1990. Resource limitation in a stream community: phosphorus enrichment effects on periphyton and grazers. Ecology 71:1494-1502.

Howarth, R. W. and S. G. Fisher. 1976. Carbon, nitrogen, and phosphorus dynamics during leaf decay in nutrient-enriched stream micro-ecosystems. Freshwater Biology 6:221-228.

Hunter, M. D. and P. W. Price. 1992. Playing chutes and ladders: heterogeneity and the relative roles of bottom-up and top-down forces in natural communities. Ecology 73:724-732.

Hurlbert, S. H. 1984. Pseudoreplication and the design of ecological field experiments. Ecological Monographs 54:187-211.

Hurlbert, S. H. 2004. On misinterpretations of pseudoreplication and related matters: a reply to Oksanen. Oikos 104:591-597.

Likens, G. E., F. H. Bormann, N. M. Johnson, D. W. Fisher, and R. S. Pierce. 1969. Effects of forest cutting and herbicide treatment on nutrient budgets in the Hubbard Brook watershed-ecosystem. Ecological Monographs 40:23-47.

Mazumder, A. and J. A. Edmonson. 2002. Impact of fertilization and stocking on trophic interactions and growth of juvenile sockeye salmon (*Oncorhynchus nerku*). Canadian Journal of Fisheries and Aquatic Sciences 59:1361-1373.

Meyer, J. L. and C. Johnson. 1983. The influence of elevated nitrate concentration on rate of leaf decomposition in a stream. Freshwater Biology 13: 177-183.

Murtaugh, P. A. 2003. On rejection rates of paired intervention analysis: reply. Ecology 84:2799-2802.

Odum, E. P and A. A. de la Cruz. 1963. Detritus as a major component of ecosystems. AIBS Bulletin 13:39-40.

Oksanen, L. 2001. Logic of experiments in ecology: is pseudoreplication a pseudoissue? Oikos 94:27-38.

O'Neill, R. V. and D. A. Reichle. 1980. Dimensions of ecosystem theory. *In:* R. H. Waring, (ed) Forests: Fresh Perspectives From Ecosystem Analysis. Oregon State University Press, Corvallis, Oregon.

Peterson, B. J., L. Deegan, J. Helfrich, J. E. Hobbie, M. Hullar, B. Moller, T. E. Ford, A. Hershey, A. Hiltner, G. Kipphut, M. A. Lock, D. M. Fiebig, V. McKinley, M. C. Miller, J. R. Vestal, R. Ventullo, and G. Volk. 1993. Biological responses of a tundra river to fertilization. Ecology 74:653-672.

Ramirez, A. and C. M. Pringle. In review. Response of detrital consumers to phosphorus levels in lowland tropical streams.

Ramirez, A., C. M. Pringle, and L. Molina. 2003. Effects of stream phosphorus levels on microbial respiration. Freshwater Biology 48:88-97.

Rosemond, A. D., P. J. Mullholland, and J. W. Elwood. 1993. Top-down and bottom-up control of stream periphyton: effects of nutrients and herbivores. Ecology 74:1264-1280.

Rosemond, A. D., C. M. Pringle, A. Ramirez, M. J. Paul, and J. L. Meyer. 2002. Landscape variation in phosphorus concentration and effects on detritus-based tropical streams. Limnology and Oceanography 47:278-289.

Schindler, D. W. 1998. Replication versus realism: the need for ecosystem-scale experiments. Ecosystems 1:323-334.

Schindler, D. W., E. J. Fee, and T. Rusczynski. 1978. Phosphorus input and its consequences for phytoplankton standing crop and production in the experimental lakes area and in similar lakes. Journal Fisheries Research Board of Canada 35:190-196.

Slavik, K., B. J. Peterson, L. A. Deegan, W. B. Bowden, A. E. Hershey, and J. E. Hobbie. 2004. Long-term responses of the Kuparuk river ecosystem to phosphorus enrichment. Ecology 85:939-954.

Stelzer, R. S., J. Heffernan, and G. E. Likens. 2003. The influence of dissolved nutrients and particulate organic matter quality on microbial respiration and biomass in a forest stream. Freshwater Biology 48:1925-1937.

Stelzer, R. S. and G. A. Lamberti. 2001. Effects of N:P ratio and total nutrient concentration on stream periphyton community structure, biomass, and elemental composition. Limnology and Oceanography 46:356-367.

Stewart-Oaten, A. 2003. On rejection rates of paired intervention analysis: comment. Ecology 84:2795-2799.

Suberkropp, K. 1995. The influence of nutrients on fungal growth, productivity, and sporulation during leaf breakdown in streams. Canadian Journal of Botany 73:s1361-s1369.

Tilman, D. 1987. Secondary succession and the pattern of plant dominance along experimental nitrogen gradients. Ecological Monographs 57:189-214.

Tilman, D. 1996. Biodiversity: population versus ecosystem stability. Ecology 77:350-363.

Vitousek, P. M., J. D. Aber, R. W. Howarth, G. E. Likens, P. A. Matson, D. W. Schindler, W. H. Schlesinger, and D. G. Tilman. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecological Applications 7:737-750.

Vitousek, P. M. and R. W. Howarth. 1991. Nitrogen limitation on land and in the sea: how can in occur? Biogeochemistry 13:87-115.

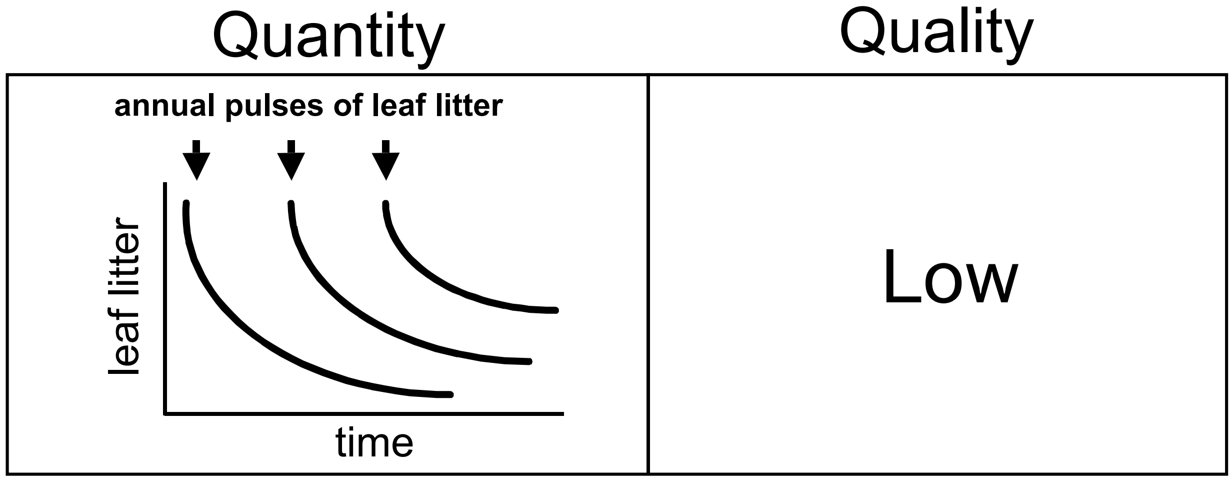
Wallace J. B., S. L. Eggert, J. L. Meyer and J. R. Webster. 1997. Multiple trophic levels of a forest stream linked to terrestrial litter inputs. Science 277:102-104.

Wallace J. B., S. L. Eggert, J. L. Meyer and J. R. Webster. 1999. Effects of resource limitation on a detrital-based ecosystem. Ecological Monographs 69:409-442.

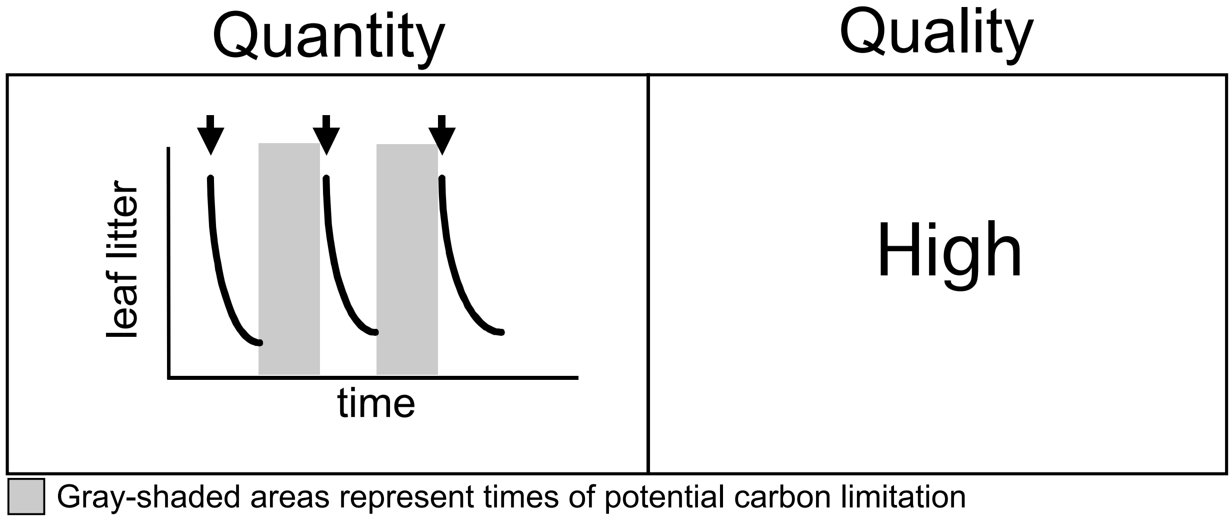
Webster, J. R., J. L. Meyer, J. B. Wallace, and E. F. Benfield. 1997. Organic matter dynamics in Hugh White Creek, Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:74-78.

Figure 1.1. Conceptual diagram of leaf litter quantity and quality in detritus-based streams at low (A) and high (B) nutrient concentrations. Black arrows represent pulsed annual inputs of allochthonous terrestrial leaf litter (i.e., figure is depicting 3 years). In streams with low nutrient concentrations (A), leaf litter is expected to decline each year at a relatively slow rate, and its quality is expected to be low because of low microbial biomass and low nutrient content. In streams with high nutrient concentrations (B), leaf litter is expected to decline rapidly each year with periods of time (gray-shaded areas) in which consumers may be limited by carbon availability. However, the quality of this leaf liter is expected to be high because of increased microbial biomass and nutrient content.

A. Low nutrient concentrations



B. High nutrient concentrations



CHAPTER 2

CONSUMER-RESOURCE STOICHIOIMETRY IN DETRITUS-BASED STREAMS[1]

---

[1] Cross, W. F., J. P. Benstead, A. D. Rosemond, and J. B. Wallace. 2003. *Ecology Letters* 6:721-732. Reprinted here with permission of publisher.

Abstract

Stoichiometric relationships between consumers and resources in detritus-based ecosystems have received little attention, despite the importance of detritus in most food webs. We analyzed carbon (C), nitrogen (N), and phosphorus (P) content of invertebrate consumers and basal food resources in two forested headwater streams (one reference, one nutrient-enriched). We found large elemental imbalances between consumers and food resources compared to living plant-based systems, particularly in regard to P content, which were reduced with enrichment. Enrichment significantly increased nutrient content of food resources (consistent with uptake of N and P by detritus-associated microbes). P content of some invertebrates also increased in the enriched vs reference stream, suggesting deviation from strict homeostasis. Nutrient content varied significantly among invertebrate functional feeding groups, orders, and to some extent, size classes. Future application of stoichiometric theory to detritus-based systems should consider the potential for relatively large consumer-resource elemental imbalances and P storage by insect consumers.

Introduction

Ecological stoichiometry is a conceptual framework that considers the relative balance of key elements in trophic interactions (Reiners 1986, Elser *et al.* 1996, Elser & Urabe 1999, Elser *et al.* 2000c, Sterner & Elser 2002). Within this framework, food items consumed are essentially packages of elements that may or may not be in balance with a consumer's elemental requirements. Limiting nutrients (e.g., N or P) are retained at higher efficiencies by the consumer, while others are consumed in excess and are egested or excreted. Data collected from a wide variety of invertebrates suggest that a consumer's chemical composition (i.e., carbon:nitrogen:phosphorus [C:N:P] ratio) is relatively homeostatic regardless of the chemical composition of its food (e.g., Hessen & Lyche 1991, Elser *et al.* 2000a, Sterner & Elser 2002).

According to stoichiometric theory, and considerable empirical support, the nutrient content and relative growth rate of a given consumer ultimately determine its nutritional requirements (e.g., Sterner & Elser 2002). Consumers (or specific life stages) with high body N or P content and high growth rates require food that is high in N or P, respectively, to maintain optimal growth. These nutrient-rich consumers are also most susceptible to reductions in growth or fitness if food resources are low in N or P relative to body demand (e.g., Sterner *et al.* 1993). Alternatively, consumers (or life stages) with low body N or P or low relative growth rates have lower requirements for these elements, and are less likely to suffer from reduced food quality (e.g., Schulz & Sterner 1999).

Several key factors are known to contribute to intra- and interspecific variation in consumer nutrient content. For example, a number of studies have linked variation in organism P content to differences in specific growth rate (e.g., Main *et al.* 1997, Elser *et al.* 2000c). Rapid growth rates during early stages of development or among species with fast turnover rates (i.e., r-selected species) have been correlated with high levels of P-rich ribosomal RNA and associated high body P demand (Elser *et al.* 2000c). In addition, variation in body nutrient content can arise from differences among organisms (or life stages) in the relative allocation of structural biomolecules such as chitin (high N:P ratio) or bone (high P) (Elser *et al.* 1996). Thus, to some degree, differences in ontogeny, life history strategy, and relative allocation of structural biomolecules may all contribute to the nutritional requirements of a given consumer.

In essence, stoichiometric theory implies that different food types do not have inherent 'qualities' *per se*; food quality is relative based on the nutritional requirements of individual consumers. For this reason, attention should be focused on the relative imbalances between consumer C:N:P and that of their food, instead of relying solely on measures of food quantity or nutrient content. Such knowledge of elemental imbalance may allow predictions about which taxa or life stages are most limited by nutrients (e.g., Urabe & Watanabe 1992), which taxa should respond positively (in terms of growth and secondary production) to increased nutrient content of food, and which taxa should dominate high- versus low-nutrient environments (e.g., Elser *et al.* 1988).

Important questions remain, however, concerning the importance of other evolutionary or ecological characteristics in determining consumer nutrient content (Frost *et al.* 2003). For example, little is known about relationships between phylogenetic position and nutrient content (but see Fagan *et al.* 2002, Vanni *et al.* 2002), or whether organisms adapt, via natural selection, to low or high-nutrient food resources. Moreover, basic information is still lacking about how consumer trophic level or feeding mode (i.e., functional feeding group, sensu Cummins 1973) may affect body nutrient content. However, some evidence suggests that nutrients tend to concentrate among consumers at higher trophic levels (e.g., Fagan *et al*. 2002, Sterner & Elser 2002).

Most empirical support for stoichiometric theory has come from algal-based food webs in lentic environments (Sterner & Elser 2002). In these systems, a stoichiometric perspective has led to considerable progress in understanding trophic dynamics, nutrient cycling, and competitive interactions (see Sterner & Elser 2002 and references therein). However, we still lack basic information about consumer-resource stoichiometry in other types of ecosystems, such as those based on detritus (but see Higashi *et al*. 1992). Study of detritus-based food webs is crucial because they represent the dominant pathway of energy flow in most ecosystems (i.e., >80 % of plant biomass ultimately ends up in detritus-based food webs; O'Neill & Reichle 1980, Wetzel & Ward 1992). Moreover, the nutritional quality of detritus is likely to be extremely low relative to the demands of detritivores (e.g., Enriquez *et al*. 1993), leading to potentially large elemental imbalances and growth constraints. In addition, there has been little research on benthic communities that are typically dominated by insect taxa (but see Frost & Elser 2002, Frost *et al*. 2002 a, b, Stelzer & Lamberti 2002, Frost *et al*. 2003).

Our primary objective in this paper was to test some of the basic tenets of ecological stoichiometry in a detritus-based ecosystem by contrasting elemental composition of larval insects and basal resources in two adjacent headwater streams of different trophic status (a nutrient poor vs. an experimentally nutrient-enriched stream). We determined the effects of nutrient enrichment on elemental composition of basal resources, and then examined how this enrichment affected the elemental composition of higher trophic levels and consumer-resource elemental imbalances. We also compared elemental composition of invertebrates among major taxonomic (orders), functional (functional feeding groups [FFG]), and life history (length of larval lifespan, body size) categories to extend our general understanding of consumer-resource stoichiometry to these nutrient-poor detritus-based food webs.

Methods

Study Site

This study was conducted in two adjacent headwater streams at the Coweeta Hydrologic Laboratory in Macon Co., North Carolina, USA. Coweeta is a large (1626 ha) heavily forested basin located in the Blue Ridge physiographic province of the southern Appalachian Mountains (see Swank & Crossley 1988). A network of small 1^{st}-3^{rd} order streams drains the Coweeta basin. Vegetation is dominated by mixed hardwoods (primarily maple, poplar, and oak) and a dense understory of *Rhododendron*, which shades the streams for most of the year. The two streams used in this study drain the relatively small catchments (C) 53 and 54 (hereafter, C53 and C54). In general, C53 and C54 have similar physical and chemical characteristics (i.e., catchment area, elevation, slope, discharge, temperature, pH, conductivity; Lugthart & Wallace 1992) but differ considerably in their concentrations of inorganic N and P as a result of an experimental nutrient enrichment. Nutrients (NH_4NO_3, K_2HPO_4, and KH_2PO_4) were dripped into C54 via a solar-powered metered-dose pump roughly every 10 m along the entire length of the stream for 2 years (July 2000 – August 2002). Before enrichment, stream water in both catchments was extremely low in N and P (ca. 29 μg (NH_4 + NO_3)-N/L, 7 μg/L SRP, biweekly samples, September 1999 – June 2000). After enrichment, nutrient levels in the enriched stream, C54, averaged 383 μg (NH_4 + NO_3)-N/L and 46 μg/L SRP (A.D. Rosemond, *unpublished data*). Invertebrate community structure was similar in both streams, and has been extensively described elsewhere (e.g., Lugthart & Wallace 1992). Headwater streams at Coweeta are extremely heterotrophic, and detritus, mostly in the form of leaf litter from the surrounding catchment, provides >90% of the energy base for microbial and invertebrate production (e.g., Wallace *et al.* 1997b). In-stream primary production is minimal (ca. 4-8 g C m^{-2} y^{-1}), and generally constitutes <1% of the total carbon entering these streams (Wallace *et al.* 1997a).

Basal Resources

Leaf litter – Submerged leaf litter was collected from C53 and C54 on a monthly basis from June 1999 to August 2002 for analysis of C, N, and P content. Five leaves were collected at 5 random points along each stream on each date (i.e., 25 total leaves/date) without regard to taxon. Monthly estimates of leaf C:N:P, therefore, represent a mixture of leaf species roughly in proportion to their natural abundance each month. Litter samples were transported to the laboratory, dried, and homogenized. Samples were subsequently weighed on a microbalance to the nearest μg. For C and N analysis, samples were weighed in tin capsules and analyzed with a Carlo Erba NA 1500 CHN analyzer. For P analysis, samples were weighed into acid-washed and pre-ashed ceramic crucibles, ashed at 500°C, acid digested, and analyzed spectrophotometrically (ascorbic acid method; APHA 1998). Ground pine needles (U.S. National Institute of Standards and Technology, 1575a) and poplar leaves (Analytical Chemistry Laboratory, University of Georgia) were used as external standards for P and N analyses (recovery 99% for P, 102% for N). All data are presented as either %C, N, and P of ash-free dry mass or as molar ratios.

Epilithon- Unglazed ceramic tiles (190 x 190 mm) were placed in C53 and C54 on 2 April 2002 to allow natural colonization of epilithon. We chose this pre-leaf-out period because it encompasses the time of peak chlorophyll *a* standing crop in Coweeta streams (typically ~10 mg/m^2; J. Greenwood, University of Georgia, *personal communication*). After 6 weeks, tiles were removed and immediately frozen. When tiles were thawed, epilithon was removed, filtered onto a glass fiber filter (pore size 45μm), dried at 50°C for >72h, and analyzed for C, N, and P as for other organic material. Freezing and thawing of tiles can potentially lyse algal cells, causing nutrient-rich cell contents to be lost upon filtration. Nonetheless, relative differences between streams in epiltihon nutrient content remain valid.

Fine particulate organic matter (FPOM)- Deposited inorganic and organic material (upper 5-10 cm) was collected haphazardly by hand from the entire length of C53 and C54 on 2 April 2002. Fine particulate organic matter retained on metal sieves (pore size <4.75 mm and >125 μm) was dried, homogenized, and analyzed for total C, N, and P as above.

Invertebrates

Invertebrates were collected from C53 and C54 between 14 March and 2 April 2002. Organic and inorganic substratum was collected from all dominant stream habitats (i.e., leaf packs, cobble, depositional areas, and bedrock outcrops) along the entire length of each stream. Substratum was rinsed onto stacked sieves with pore sizes ranging from 125 μm to 4.75 mm. Large invertebrates were removed live from material retained on sieves >1 mm. Chironomids and early instars of other taxa (all >1 mm in length) were removed live under a dissecting microscope from material retained on smaller sieves (125 – 250 μm mesh size). Invertebrates were frozen within 4 h of collection. Upon thawing, invertebrates were identified, measured in length to the nearest mm, their gut contents carefully removed (except chironomids and first instars of a few other taxa), refrozen at –80°C, lyophilized, homogenized, weighed, and analyzed for total C, N, and P as above. Analysis of C, N, and P content was generally conducted on composite samples of a given taxon consisting of 3-100 individuals of similar-size (within 1 mm size classes). Otherwise, if enough material was present, chemical analysis was performed on individual larvae. The majority of samples contained enough material for C, N, and P analysis; however, when sample weight was limited, we restricted our analysis to %P. Biomass of individuals was obtained using previously established length-weight regressions for Coweeta stream taxa (Benke *et al.* 1999). Functional feeding groups (FFG) were designated according to Merritt and Cummins (1996) and knowledge of the local fauna based on gut-content analyses (J. B. Wallace, *unpublished data*). Functional feeding groups, which are based primarily on mouth-part morphology and behavioral characteristics (Cummins 1973), included scrapers (scrape biofilm from hard surfaces), shredders (consume primarily leaf material), collector-gatherers (gather fine organic particles), collector-filterers (filter fine particles), and predators (consume other animals).

Statistical analyses

Leaf litter- We analyzed the time series of leaf litter nutrient content using randomized intervention analysis (RIA, Carpenter *et al.* 1989). RIA uses paired, before-and-after time-series data from a manipulated and a reference system to detect changes caused by the manipulation. In this study, RIA was used to test the null hypothesis that no change in leaf litter nutrient content occurred in the treatment stream relative to the reference stream following the initiation of nutrient enrichment.

Invertebrates- Overall differences between streams in invertebrate %C, %N, %P, C:P, N:P, and C:N were assessed with Kolmogorov-Smirnov goodness-of-fit tests. To test the null hypothesis of no difference in invertebrate nutrient content between streams (i.e., chemical homeostasis), samples of similarly sized taxa (within 2 mm) from both streams were compared with paired t-tests. Differences among functional feeding groups and orders were analyzed with two-way analysis of variance; significant ANOVAs were followed by Tukey's HSD multiple comparisons. Relationships between body size (mg AFDM) or length of larval lifespan (days) and invertebrate nutrient content were examined with linear regression. All such regressions were performed on the entire data set, within functional feeding groups, and within orders. Data were either log(x+1) or arcsin-square root transformed to meet assumptions of normality and homoscedasticity. No adjustments were made to a for protection against experimentwise error (e.g., Bonferroni) because of relatively low statistical power, and the potential for increased type II error (Perneger 1998).

Results

Basal resources

Nutrient enrichment of C54 resulted in significantly higher nutrient content of leaf litter, including increases in %P, and consequent decreases in N:P and C:P ratios relative to leaf litter in the control stream ($P < 0.00001$ for all three tests, RIA; Fig. 2.1, Table 2.1). Post-enrichment values of P content were on average 59% higher in C54 compared to C53 (range 14-111%). There was also a small, but significant ($P = 0.04$, RIA), increase in %C in the enriched stream (data not shown). No change in %N or C:N ratio of leaf litter was observed (see Table 2.1 for C:N; %N data not shown).

FPOM contained slightly more P and N, and less C in the enriched stream than in the reference stream (P: 0.1% vs. 0.07%; N: 0.9% vs. 0.8%; C: 23% vs. 25%). These minor differences were reflected in FPOM elemental ratios (Table 2.1).

Epilithon in the enriched stream contained ~4X more P and ~5.5X more N than in the reference stream (P: 0.08% vs. 0.02%; N: 12.1% vs. 2.2%). Enrichment also increased %C in the enriched stream relative to the reference (27.7% vs. 16.5%), presumably because of differences in standing crop, and a higher proportion of organic (including microbes) vs. inorganic matter on tiles in the enriched stream. Epilithon C:P, N:P, and C:N ratios differed correspondingly (Table 2.1).

Invertebrates

We analyzed the nutrient content of 40 invertebrate taxa from C53 and C54, ranging from 1–36 mm in length and 0.002 – 86 mg AFDM in weight. Larval lifespan of these taxa ranges from < 2 weeks to > 3 years (see Wallace *et al.* 1999). Invertebrates exhibited considerable variability in %P, %N, and %C (Fig. 2.2). Overall, the coefficient of variation in %P (CV = 52%) was much higher than %N (CV = 13%) or %C (CV = 8%). Coefficients of variation for %P, %N, and %C were, on average, 30-45% lower in the enriched stream (C54) than in the reference stream (C53). High variability of %P was reflected in invertebrate C:P, N:P, and C:N ratios, where overall variability was much higher in C:P (CV = 60%) and N:P (CV = 51%) ratios than in the C:N (CV = 15%) ratio (Fig. 2.2). Coefficients of variation for C:P, N:P, and C:N were also 30-40% lower in the enriched stream (C54) than in the reference stream (C53). Overall, there was no difference in %P, %C, %N, C:P, N:P, and C:N ratios of invertebrates between C53 and C54 (Kolmogorov-Smirnov tests, $P > 0.05$).

Paired comparisons of similar-sized taxa between C53 and C54 revealed patterns that indicate some invertebrate taxa may not be strictly homeostatic with regard to body nutrient content. Invertebrates in the enriched stream had significantly higher %P ($P = 0.03$) and lower

C:P ($P = 0.01$) and N:P ($P = 0.02$) ratios than in the reference stream (Fig. 2.3). No difference was detected in %C, %N, or C:N ratios among paired taxa between streams ($P > 0.05$).

Significant differences in %P, %N, and %C were found among major insect orders when data from both streams were considered together (two-way ANOVA, order: $P < 0.0001$, Fig. 2.4). Trichoptera and Ephemeroptera were generally lower in %P and %N than Diptera, Odonata, and Plecoptera (Fig 2.4). Significant differences also existed in C:P, N:P, and C:N among the major insect orders (two-way ANOVA, order: $P < 0.001$, Fig. 2.4). The only significant difference found in invertebrate nutrient content between streams was among Trichoptera, in which C:P ratios were lower in the enriched stream (C54) than in the reference stream (C53) (two-way ANOVA, stream: $P = 0.03$, stream x order interaction: $P = 0.002$, Fig. 2.4).

Percent P, %N, and %C varied significantly among invertebrate FFGs when considering all data together (two-way ANOVA, FFG: $P < 0.0001$, Fig. 2.5). Again, most of the variation was associated with P content. On average, predators contained more P and N than the other FFGs. There were no significant effects of enrichment on %P, %N, and %C for any FFG (two-way ANOVA, stream and stream x FFG interaction non-significant: $P > 0.05$). Carbon:P and C:N ratios also differed significantly among FFGs (two-way ANOVA, FFG: $P < 0.001$, Fig. 2.5). Carbon:P ratios of shredders were higher than those of collector-gatherers and predators. Carbon:N ratios were higher among shredders and collector-filterers than predators. No difference in N:P ratios was apparent among FFGs. Additionally, there were no overall differences in C:P, N:P, and C:N ratios between C53 and C54 (two-way ANOVA, stream and interaction non-significant: $P > 0.05$).

Invertebrate body size (mg AFDM) did not explain any significant variation in invertebrate nutrient content when all data were considered together (linear regression, all P values > 0.05), and when invertebrate orders were examined separately for each stream. However, when body size-nutrient content relationships were examined by FFG irrespective of stream, some significant patterns emerged. Collector-gatherer C:P ($r^2 = 0.58$, $P < 0.01$), N:P ($r^2 = 0.48$, $P < 0.01$), and C:N ($r^2 = 0.21$, $P = 0.1$) ratios all increased with body size (mg AFDM); %P decreased with body size ($r^2 = 0.25$, $P = 0.04$). In addition, collector-filterers exhibited an increase in N:P ratio ($r^2 = 0.27$, $P = 0.08$) and a concomitant decrease in %P ($r^2 = 0.24$, $P = 0.05$) with body size. No such relationships existed for scrapers, shredders, or predators.

No significant relationships were found between larval lifespan (d) and any measure of body nutrient content.

Discussion

Basal food resources in streams at Coweeta had very low nutrient content (Table 2.1). Leaf litter, which forms the basis for most heterotrophic production (Wallace *et al.* 1997b), was extremely low in N and P. Carbon:P and N:P ratios were among the highest reported for vascular plant detritus (Enriquez *et al.* 1993). These values are also considerably higher than those of living terrestrial foliage (Table 2.1), suggesting nutrient loss via absorption by trees before leaf abscission, the dominance of structural C compounds in leaf litter, and leaching of soluble N and P upon contact with stream water. Increases in P of leaf litter after enrichment coincided with significant increases in bacterial and fungal production on leaves (K. Suberkropp, *unpublished data*), suggesting increased microbial biomass associated with leaf litter as the primary cause of changes in nutrient content.

Nitrogen and P content of epilithon was much higher than that of leaf litter, but still low relative to many previously reported values for streams (Table 2.1, e.g., Kahlert 1998: C:P range (99-603), N:P range (10-49)). Fine particulate organic matter (<4 mm, FPOM), which is also directly consumed by many stream invertebrates, generally had a higher nutrient content than large leaf particles (Table 2.1; Sinsabaugh & Linkins 1990); this was likely a result of increased surface:volume ratios leading to a higher proportion of nutrient-rich microbes (i.e., bacteria) on smaller particles, and the presence of microbial and animal exudates within stream FPOM (Findlay *et al.* 2002).

Carbon:P and N:P ratios of stream consumers were considerably higher than those of invertebrate consumers in littoral benthic, planktonic, and terrestrial environments (Table 2.1). C:N ratios of consumers did not differ among these environments (Table 2.1). Differences in P content were unexpected, as invertebrate nutrient content has been thus far reported as relatively constant across a wide range of systems with very different basal resource elemental composition (Table 2.1, e.g., Elser *et al.* 2000a, Frost *et al.* 2003). Such low relative P content may be characteristic of detritus-based systems. Nutritional constraints in these systems may be severe enough to cause evolutionary adaptation to low nutrient food resources via lowered body nutrient content and hence lowered requirements for P (e.g., Elser *et al.* 2000b, Fagan *et al.* 2002). In addition, some prominent detritivores at Coweeta (e.g., *Tipula* spp., Diptera) harbor gut flora or fauna that potentially aid in the assimilation of low nutrient leaf litter (e.g., Klug & Kotarski 1980), an adaptation also seen among wood-eating termites (Higashi *et al.* 1992).

To estimate simple consumer-resource elemental imbalances, we calculated arithmetic differences between the C:P and C:N of consumers and their food resources (Table 2.1). This provides a comparison of the relative stoichiometric constraints among trophic groups in detritus-based streams versus systems based on living plants or algae. It is evident that stream consumers dependent on leaf litter (i.e., shredders) in C53 are far out of balance with their food resources (C:P 4360, C:N 66, Table 2.1). In comparison, the elemental imbalances between terrestrial herbivores and living plants (C:P 852, C:N 30) are ca. 5X and 2X lower than those of stream shredders, in terms of P and N, respectively. Thus, limits to growth and production of leaf-eating consumers are potentially more severe in detritus-based food webs than in those based on living plant tissue. Imbalances of other stream consumers (i.e., collectors, scrapers) were also relatively high in comparison to lake zooplankton, but fell within the range of some lake benthic invertebrates (Table 2.1). Stream predators were generally the least out of balance with their food, based on consumption of N and P-rich prey. For all stream trophic groups, nutrient enrichment reduced consumer-resource imbalances, potentially alleviating nutrient limitation for some taxa (C54, Table 2.1).

A central tenet of ecological stoichiometry is that consumers maintain elemental homeostasis within a relatively small range (Elser *et al.* 1996, Sterner & Elser 2002), regardless of the elemental composition of their food. Here we have shown that this may not be strictly true for some invertebrates at Coweeta that exhibited up to 4-fold differences in C:P and N:P ratios between C53 and C54. This deviation from strict homeostasis may have been due to either increased P storage in insect tissue or hemolymph (Woods *et al.* 2002), or increased rRNA allocation associated with higher growth rates (e.g., Schade *et al.* 2003). Woods *et al.* (2002) demonstrated that a larval lepidopteran (*Manduca sexta*) was capable of storing excess P as a-glycerophosphate in hemolymph. The authors suggested that P storage might act as a potential buffer against short-term decreases in P availability. Interestingly, the taxa in our study that consistently showed the largest differences in P content between C53 and C54 were Trichoptera, which share a close evolutionary history with Lepidoptera (Wheeler *et al.* 2001), and may be similarly capable of P storage. An alternative explanation is that higher P content of some consumers in the enriched stream was due to higher growth rates and greater cellular allocation to rRNA (e.g., Schade *et al.* 2003). Frost and Elser (2002) showed that mayfly larvae (*Ephemerella* sp.) fed P-enriched epilithon harbored significantly more P in their body tissue than those that were fed low-P periphyton. In this case, growth rates were higher on P-rich epilithon, suggesting a growth-rRNA effect on mayfly P content. A thorough test of these alternative hypotheses for organisms that deviate from strict homeostasis will require careful estimates of daily growth rate, rRNA content, and C:N:P content of consumers and basal resources.

Our analyses relating body size and larval lifespan to nutrient content were made in an effort to test the 'growth rate hypothesis' (i.e., rapid growth associated with P-rich rRNA, Elser *et al.* 1996, Main *et al.* 1997). Although we did not find that body size was consistently related to nutrient content, we did find some patterns that support previous stoichiometric theory. For example, N and P content decreased with body size among collector-gatherers and collector-filterers. These results suggest that, for some taxa, later instars contain less N and P, have decreased growth rates (as shown for Chironomidae, Huryn 1990, W. F. Cross, *unpublished data*), and may have lower physiological requirements for N and P than early instars. Although no relationship was found between larval lifespan (a proxy for turnover rate) and P content, patterns may be difficult to detect among organisms that vary in size or life-stage. For example, relationships may be obscured when comparing early instars of long-lived taxa (i.e., high P for that taxon due to high growth rates at small size, low P relative to other taxa due to long-lived life history), and late instars of short-lived taxa (i.e., low P for that taxon, high relative P).

We found significant differences among stream invertebrate functional and taxonomic groups. Nitrogen and P content were generally highest among predators, which is consistent with the results of Fagan *et al.* (2002), who found that terrestrial invertebrate predators contained higher amounts of N than invertebrate herbivores. Multiple valid hypotheses exist for this trophic difference, including the effect of higher nutrient content in the diet of predators (Fagan *et al.* 2002), but proper understanding will require further analysis. Among orders, Diptera consistently harbored the highest amount of P, which may be related to a disproportionate number of fast-growing taxa (i.e., chironomids) within this order (Huryn 1990). We found no apparent relationship between nutrient content and invertebrate phylogenetic position (i.e., ancient Ephemeroptera vs. recently derived Diptera) (e.g., Fagan *et al.* 2002).

A large disparity exists between the temporal scale of leaf-litter sampling in this study (monthly for 3 years), and that of other food web components (once). This disparity raises important questions about seasonal differences in consumer-resource elemental imbalance. For example, temporal changes in resource elemental composition could potentially alleviate nutrient imbalances for some invertebrates during critical times of the year (i.e., during growth spurts). Although our long-term leaf litter data showed no obvious seasonal trends, this does not preclude the potential for seasonal variability in FPOM, epilithon, or, to a lesser extent, invertebrates. Future studies geared towards understanding temporal variability of consumer-resource C:N:P content will be important in recognizing the potential for seasonal differences in stoichiometric constraints.

We sampled most food web components from both streams after the experimental enrichment had begun in one of them. As a consequence, it is not certain that stoichiometric differences between streams were actually due to the enrichment. However, both of the study streams were similarly low in inorganic N and P before the enrichment. In addition, large changes in leaf-litter nutrient content coincided directly and significantly with experimental enrichment (Fig. 2.1). Therefore, it is highly likely that between-stream elemental differences in epilithon, FPOM, and invertebrates were caused by the enrichment.

This study is one of the first to describe stoichiometric relationships among consumers and basal resources in a detritus-based ecosystem. We have shown that larval invertebrates in these systems harbor low relative amounts of P in their body tissue compared to invertebrates in other food webs based on living plant tissue. We have also shown that some invertebrate taxa do not exhibit strict elemental homeostasis, and may have the potential to avoid nutrient limitation via storage of P. Indeed, insects may cope with physiological nutrient constraints in a different manner (e.g., store nutrients) than some well-studied crustaceans (i.e., copepods, cladocerans). Nutrient enrichment of detritus-based ecosystems may alleviate nutrient limitation of some invertebrate taxa, and potentially lead to long-term alteration of community structure, secondary production, and food web dynamics. More research in detritus-based systems will be necessary to determine whether stoichiometric relationships in these systems are fundamentally different from those in systems based on living plants.

Acknowledgements

We are grateful to Keller Suberkropp for collection of leaf litter, and Molly Neely-Burnam, Nathan Taylor, and Roger Hilten for laboratory assistance. We also thank Judy Meyer, Elizabeth Reese, and three anonymous reviewers for helpful advice that improved the manuscript. Research funding was provided by the National Science Foundation (DEB –

9806610; A.D. Rosemond, J.B. Wallace, K. Suberkropp, and P.J. Mulholland), and a Sigma Xi Grant in Aid of Research (W. Cross).

Literature Cited

American Public Health Association. (1998). Standard methods for the examination of water and wastewater, 20th edition, American Public Health Association, Washington, DC, USA.

Benke A.C., Huryn A.D., Smock L.A. & Wallace J.B. (1999). Length-mass relationships for freshwater macroinvertebrates in North America with particular reference to the southeastern United States. *J. N. Am. Benthol. Soc.*, 18, 308-343.

Carpenter S.R., Frost T.M., Heisey D. & Kratz T.K. (1989). Randomized intervention analysis and the interpretation of whole-ecosystem experiments. *Ecology*, 70, 1142-1152.

Cummins K.W. (1973). Trophic relations of aquatic insects. *Ann. Rev. Entomol.*, 18, 183-206.

Elser J.J. & Urabe J. (1999). The stoichiometry of consumer-driven nutrient recycling: theory, observations, and consequences. *Ecology*, 80, 735-751.

Elser J.J., Dobberfuhl D.R., MacKay N.A. & Schampel J.H. (1996). Organism size, life history, and N:P stoichiometry. *BioScience*, 46, 674-684.

Elser J.J., Elser M.M., Mackay N.A. & Carpenter S.R. (1988). Zooplankton-mediated transitions between N-limited and P- limited algal growth. *Limnol. Oceanogr.,* 33, 1-14.

Elser J.J., Fagan W.F., Denno R.F., Dobberfuhl D.R., Folarin A., Huberty A., Interlandi S., Kilham S.S., McCauley E., Schulz K.L., Siemann E.H. & Sterner R.W. (2000a). Nutritional constraints in terrestrial and freshwater food webs. *Nature*, 408, 578-580.

Elser J.J., O'Brien J.O., Dobberfuhl D.R. & Dowling T.E. (2000b). The evolution of ecosystem processes: growth rate and elemental stoichiometry of a key herbivore in temperate and arctic habitats. *J. Evol. Biol.*, 13, 845-853.

Elser J.J., Sterner R.W., Gorokhova E., Fagan W.F., Markow T.A., Cotner J.B., Harrison J.F., Hobbie S.E., Odell G.M. & Weider L.J. (2000c). Biological stoichiometry from genes to ecosystems. *Ecol. Lett.*, 3, 540-550.

Enriquez S., Duarte C.M. & Sandjensen K. (1993). Patterns in decomposition rates among photosynthetic organisms - the importance of detritus C-N-P content. *Oecologia*, 94, 457-471.

Fagan W.F., Siemann E., Mitter C., Denno R.F., Huberty A.F., Woods H.A. & Elser J.J. (2002). Nitrogen in insects: implications for trophic complexity and species diversification. *Am. Nat.*, 160, 784-802.

Findlay S., Tank J., Dye S., Valett H.M., Mulholland P.J., McDowell W.H., Johnson S.L., Hamilton S.K., Edmonds J., Dodds W.K., Bowden W.B. (2002). A cross-system comparison of bacterial and fungal biomass in detritus pools of headwater streams. *Microb. Ecol.*, 43, 55-66.

Frost P.C. & Elser J.J. (2002). Growth responses of littoral mayflies to the phosphorus content of their food. *Ecol. Lett.*, 5, 232-240.

Frost P.C., Elser J.J. & Turner M.A. (2002a). Effects of caddisfly grazers on the elemental composition of epilithon in a boreal lake. *J. N. Am. Benthol. Soc.*, 21, 54-63.

Frost P.C., Stelzer R. S., Lamberti G.A. & Elser J. J. (2002b). Ecological stoichiometry of trophic interactions in the benthos: understanding the role of C:N:P ratios on littoral and lotic habitats. *J. N. Am. Benthol. Soc.,* 21, 515-528.

Frost P.C., Tank S.E., Turner M.A. & Elser J.J. (2003). Elemental composition of littoral invertebrates from oligotrophic and eutrophic Canadian lakes *J. N. Am. Benthol. Soc.*, 22, 51-62.

Hessen D.O. & Lyche A. (1991). Interspecific and intraspecific variations in zooplankton element composition. *Archiv. Hydrobiol.*, 121, 343-353.

Higashi M., Abe T. & Burns T.P. (1992). Carbon-nitrogen balance and termite ecology. *Proc R. Soc. Lond.* B, 249, 303-308.

Huryn A.D. (1990). Growth and voltinism of lotic midge larvae: patterns across an Appalachian mountain basin. *Limnol. Oceanogr.*, 35, 339-351.

Kahlert M. (1998). C:N:P ratios of freshwater benthic algae. *Arch. Hydrobiol. Spec. Issues Advanc. Limnol.*, 51, 105-114.

Klug M.J. & Kotarski S. (1980). Bacteria associated with the gut tract of larval stages of the aquatic cranefly Tipula abdominalis (Diptera: Tipulidae). *App. Env. Microb.*, 40, 408-416.

Lugthart G.J. & Wallace J.B. (1992). Effects of disturbance on benthic functional structure and production in mountain streams. *J. N. Am. Benthol. Soc.*, 11, 138-164.

Main T.M., Dobberfuhl D.R. & Elser J.J. (1997). N:P stoichiometry and ontogeny of crustacean zooplankton: a test of the growth rate hypothesis. *Limnol. Oceanogr.*, 42, 1474-1478.

Merritt R.W. & Cummins K.W. (1996). *An Introduction to the Aquatic Insects of North America*. 3rd edn. Kendall/Hunt Publishing Co., USA.

O'Neill R.V. & Reichle D.A. (1980). Dimensions of ecosystem theory. In: *Forests: Fresh Perspectives from Ecosystem Analysis* (ed. Waring R.H.). Oregon State University Press, Corvallis, Oregon, pp. 11-26.

Perneger T.V. (1998). What's wrong with Bonferroni adjustments. *Brit. Med. J.*, 316, 1236-1238

Reiners W.A. (1986). Complementary models for ecosystems. *Am. Nat.*, 127, 59-73.

Schade J.D., Kyle M., Hobbie S.E., Fagan W.F. & Elser J.J. (2003). Stoichiometric tracking of soil nutrients by a desert insect herbivore. *Ecol. Lett.*, 6, 96-101.

Schulz K.L. & Sterner R.W. (1999). Phytoplankton phosphorus limitation and food quality for *Bosmina*. *Limnol. Oceanogr.*, 44, 1549-1556.

Sinsabaugh R.L. & Linkins A.E. (1990). Enzymic and chemical analysis of particulate organic matter from a boreal river. *Freshwat. Biol.*, 23, 301-309.

Stelzer R.S. & Lamberti G.A. (2002). Ecological stoichiometry in running waters: periphyton chemical composition and snail growth. *Ecology*, 83, 1039-1051.

Sterner R.W., Hagemeier D.D. & Smith W.L. (1993). Phytoplankton nutrient limitation and food quality for *Daphnia*. *Limnol. Oceanogr.*, 38, 857-871.

Sterner R.W. & Elser J.J. (2002). *Ecological Stoichiometry*. Princeton University Press, Princeton, USA.

Swank W.T & Crossley D.A. (1988). *Forest hydrology and ecology at Coweeta.* Springer-Verlag, New York, USA.

Urabe J. & Watanabe Y. (1992). Possibility of N-limitation or P-limitation for planktonic cladocerans - an experimental test. *Limnol. Oceanogr.*, 37, 244-251.

Vanni M.J., Flecker A.S., Hood J.M. & Headworth J.L. (2002). Stoichiometry of nutrient recycling by vertebrates in a tropical stream: linking species identity and ecosystem processes. *Ecol. Lett.*, 5, 285-293.

Wallace J.B., Cuffney T.F., Eggert S.L. & Whiles M.R. (1997a). Stream organic matter inputs, storage, and export for Satellite Branch at Coweeta Hydrologic Laboratory, North

Carolina, USA. *J. N. Am. Benthol. Soc.*, 16, 67-74.

Wallace J.B., Eggert S.L., Meyer J.L. & Webster J.R. (1997b). Multiple trophic levels of a forest stream linked to terrestrial litter inputs. *Science*, 277, 102-104.

Wallace J.B., Eggert S.L., Meyer J.L. & Webster J.R. (1999). Effects of resource limitation on a detrital-based ecosystem. *Ecol. Monogr.*, 69, 409-442.

Wetzel R.G. & Ward A.K. (1992). Primary production. In: *The Rivers Handbook, Volume 1* (ed Calow P. & Petts G.E.). Blackwell Scientific, Oxford, UK, pp. 354-369.

Wheeler W.C., Whiting M, Wheeler Q.D. & Carpenter J.M. (2001). The phylogeny of the extant hexapod orders. *Cladistics*, 2, 113-169.

Woods H.A., Perkins M.C., Elser J.J. & Harrison J.F. (2002). Absorption and storage of phosphorus by larval *Manduca sexta*. *J. Ins. Phys.*, 48, 555-564.

Table 2.1. Comparisons of C:P, N:P, and C:N among invertebrate trophic groups and food resources from lake, stream, and terrestrial habitats[1]. Values in bold are from this study. C53 (reference), C54 (enriched). All ratios are molar.

	Trophic Group						**Food Resource**		**Elemental imbalance[2]**	
	C53			C54[3]			C53	C54[3]	C53	C54
	mean	median	range	mean	median	range	mean	mean		
	stream shredders						***leaf detritus***			
C:P	**498**	**493**	**(136–877)**	**252**	**221**	**(123-610)**	**4858**	**3063**	**4360**	**2565**
C:N	**6.7**	**6.4**	**(5.4-8.9)**	**6.4**	**6.3**	**(5.0-7.7)**	**73**	**82**	**66**	**75**
N:P	**73**	**76**	**(17-125)**	**39**	**30**	**(19-97)**	**67**	**39**	**--**	**--**
	stream collectors						***FPOM***			
C:P	**277**	**208**	**(93-574)**	**227**	**219**	**(80-358)**	**1015**	**673**	**738**	**396**
C:N	**6.4**	**6.0**	**(5.2-9.0)**	**6.0**	**5.8**	**(5.3-7.2)**	**34**	**29**	**28**	**23**
N:P	**43**	**38**	**(14-78)**	**37**	**37**	**(14-59)**	**28**	**23**	**--**	**--**
	stream scraper-herbivores						***stream epilithon***			
C:P	**369**	**--**	**--**	**287**	**304**	**(155-371)**	**1741**	**845**	**1372**	**476**
C:N	**6.2**	**--**	**--**	**5.8**	**5.4**	**(5.1-7.1)**	**8.7**	**4.6**	**2.5**	**-1.6**
N:P	**59**	**--**	**--**	**51**	**56**	**(22-68)**	**201**	**318**	**--**	**--**

Table 2.1. (cont.)

	Trophic Group						**Food Resource**		**Elemental imbalance**[2]	
	C53			C54[3]			C53	C54[3]	C53	C54
	mean	median	range	mean	median	range	mean	mean		
	stream predators						***stream prey***			
C:P	**223**	**215**	**(102-351)**	**227**	**215**	**(78-430)**	**324**	**236**	**101**	**13**
C:N	**5.1**	**5.2**	**(4.9-5.4)**	**5.6**	**5.5**	**(5.0-6.8)**	**6.1**	**5.9**	**1.0**	**0.8**
N:P	**43**	**42**	**(20-65)**	**40**	**37**	**(15-75)**	**52**	**40**	**--**	**--**
	terrestrial herbivores						*terrestrial plants*			
C:P				116			968		852	
C:N				6.5			36		30	
N:P				26			28		--	
	lake zooplankton						*lake phytoplankton*			
C:P				124			307		183	
C:N				6.3			10		3.9	
N:P				22			30		--	

Table 2.1. (cont.)

	Trophic Group						**Food Resource**		**Elemental imbalance**[2]	
	C53			C54[3]			C53	C54[3]	C53	C54
	mean	median	range	mean	median	range	mean	mean		
				lake benthic invertebrates			*lake benthic algae*			
C:P				148			(98-1496)		(-50-1348)	
C:N				5.5						
N:P				27						

[1] Terrestrial herbivores and plants, lake phytoplankton and zooplankton from Elser *et al*. 2000a; Lake benthic algae and benthic invertebrates from Frost & Elser 2002 and Frost *et al*. 2003.

[2] Elemental imbalance is calculated as the arithmetic difference between a consumer and its food resource.

[3] Post-enrichment values.

Figure 2.1. Changes in (a) %P, (b) N:P, and (c) C:P of leaf litter in the reference stream, (C53, open symbols) and the enriched stream (C54, closed symbols) from June 1999 to August 2002. Arrow indicates start of nutrient enrichment of C54. All ratios are molar. Data are means (*N* = 5, except first date where *N* = 3).

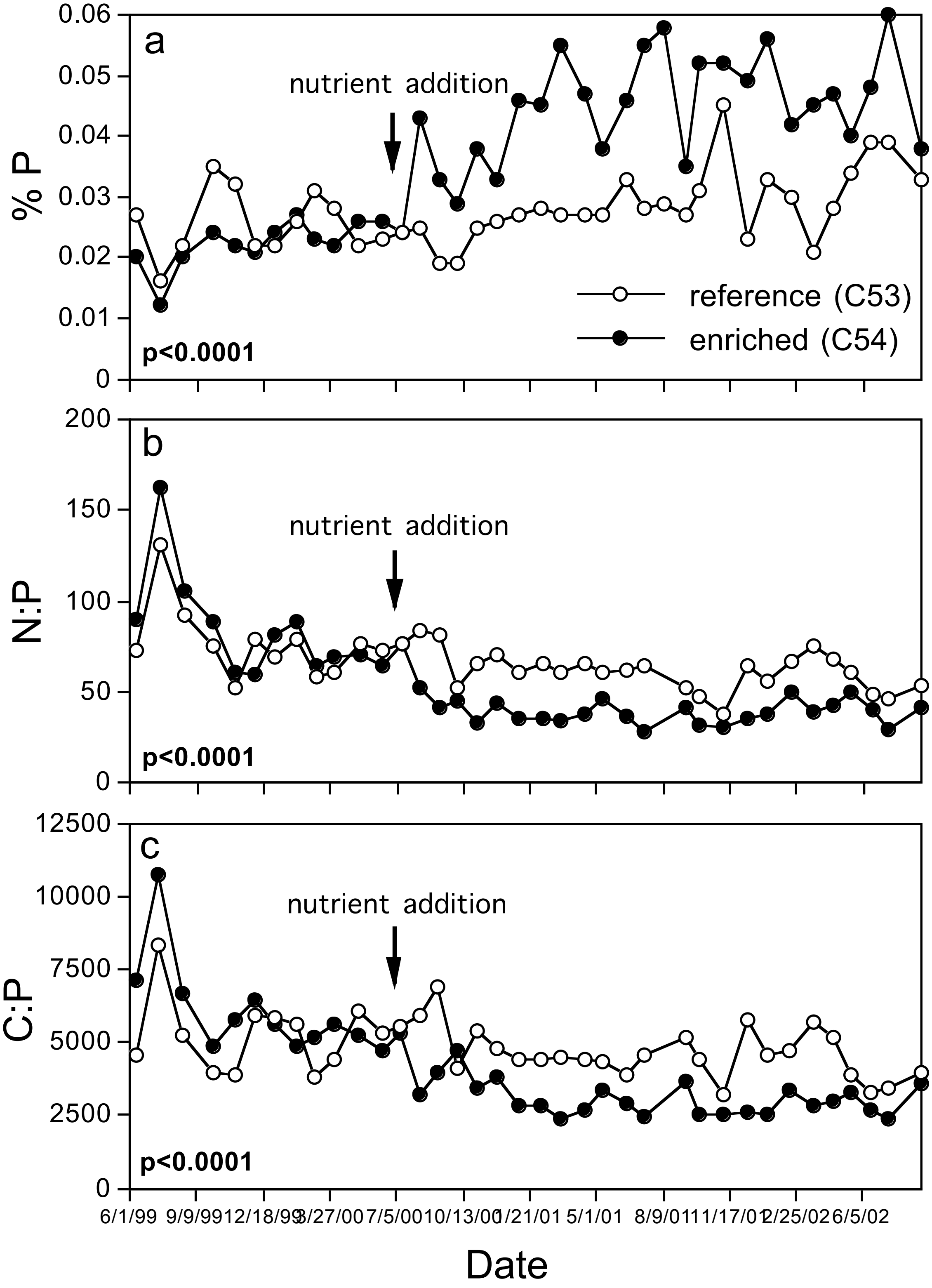

a
b
c
% P
N:P
C:P
nutrient addition
reference (C53)
enriched (C54)
p<0.0001
0.06
0.05
0.04
0.03
0.02
0.01
0
200
150
100
50
12500
10000
7500
5000
2500
6/1/99
9/9/99
12/18/99
3/27/00
7/5/00
10/13/00
1/21/01
5/1/01
8/9/01
11/17/01
2/25/02
6/5/02
Date

Figure 2.2. Frequency histograms of invertebrate body % P, % C, % N, C:P, N:P, and C:N. Histograms include all data from C53 and C54 together; no significant difference was found between streams for any measure of nutrient content when all data were considered together (Kolmogorov-Smirnov tests, $P > 0.05$). All ratios are molar.

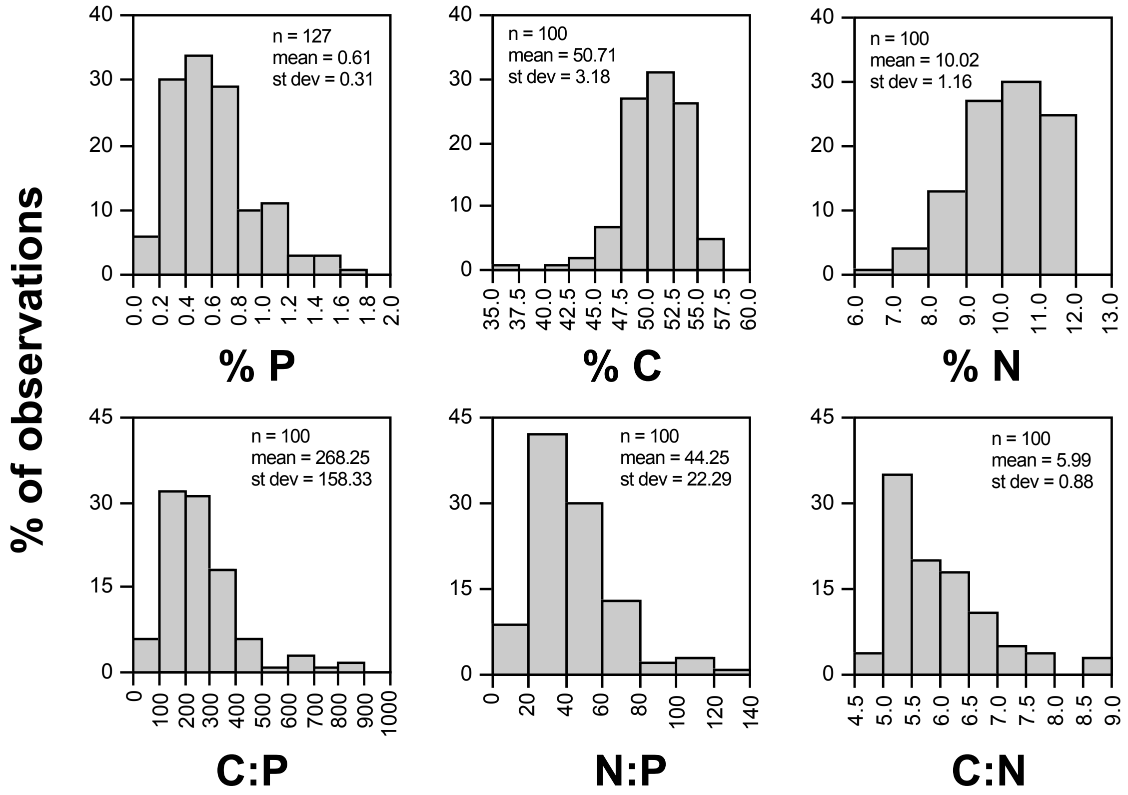
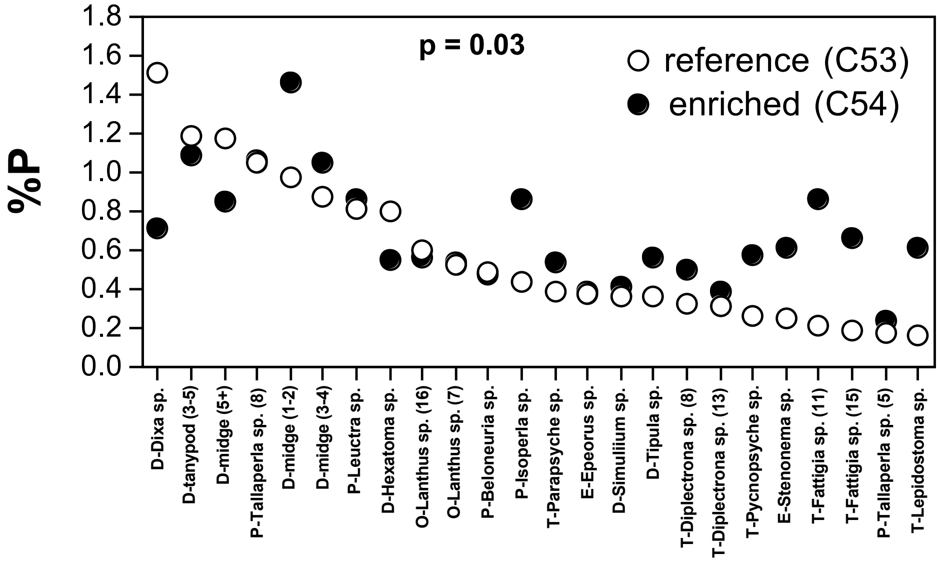
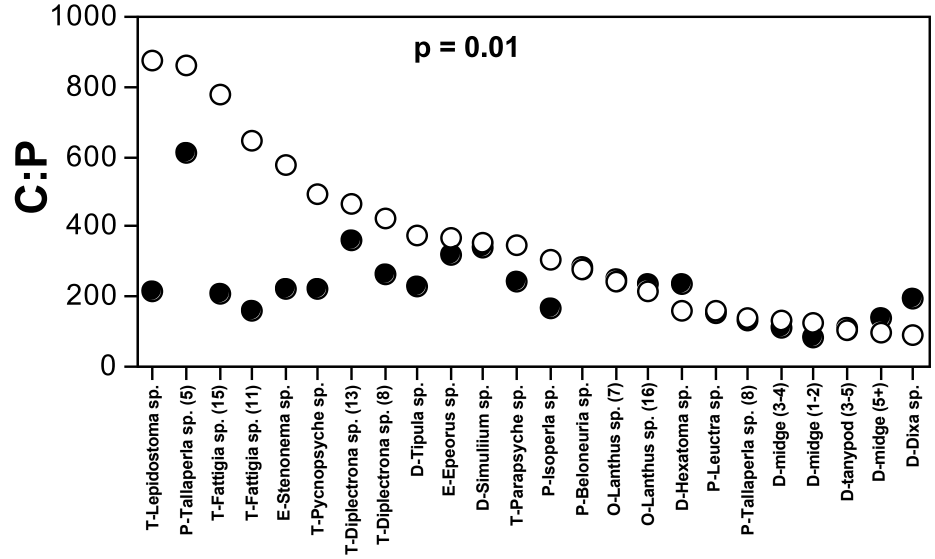

% of observations
n = 127
mean = 0.61
st dev = 0.31
0.0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
1.6
1.8
2.0
% P
n = 100
mean = 50.71
st dev = 3.18
35.0
37.5
40.0
42.5
45.0
47.5
50.0
52.5
55.0
57.5
60.0
% C
n = 100
mean = 10.02
st dev = 1.16
6.0
7.0
8.0
9.0
10.0
11.0
12.0
13.0
% N
n = 100
mean = 268.25
st dev = 158.33
0
100
200
300
400
500
600
700
800
900
1000
C:P
n = 100
mean = 44.25
st dev = 22.29
0
20
40
60
80
100
120
140
N:P
n = 100
mean = 5.99
st dev = 0.88
4.5
5.0
5.5
6.0
6.5
7.0
7.5
8.0
8.5
9.0
C:N
0
10
20
30
40
15
45

Figure 2.3. Percent P, C:P, and N:P ratios of paired invertebrates of similar size from C53 (reference) and C54 (enriched). Capital letters before the genus indicate insect order. D = Diptera; E = Ephemeroptera; O = Odonata; P = Plecoptera; T = Trichoptera. Where more than one size class of a given taxon is presented, numbers in parentheses indicate length of larvae in mm. P values are shown for paired t-tests between streams. All ratios are molar.


%P
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
p = 0.03
○ reference (C53)
● enriched (C54)
D-Dixa sp.
D-tanypod (3-5)
D-midge (5+)
P-Tallaperla sp. (8)
D-midge (1-2)
D-midge (3-4)
P-Leuctra sp.
D-Hexatoma sp.
O-Lanthus sp. (16)
O-Lanthus sp. (7)
P-Beloneuria sp.
P-Isoperla sp.
T-Parapsyche sp.
E-Epeorus sp.
D-Simulium sp.
D-Tipula sp.
T-Diplectrona sp. (8)
T-Diplectrona sp. (13)
T-Pycnopsyche sp.
E-Stenonema sp.
T-Fattigia sp. (11)
T-Fattigia sp. (15)
P-Tallaperla sp. (5)
T-Lepidostoma sp.


C:P
1000
800
600
400
200
0
p = 0.01
T-Lepidostoma sp.
P-Tallaperla sp. (5)
T-Fattigia sp. (15)
T-Fattigia sp. (11)
E-Stenonema sp.
T-Pycnopsyche sp.
T-Diplectrona sp. (13)
T-Diplectrona sp. (8)
D-Tipula sp.
E-Epeorus sp.
D-Simulium sp.
T-Parapsyche sp.
P-Isoperla sp.
P-Beloneuria sp.
O-Lanthus sp. (7)
O-Lanthus sp. (16)
D-Hexatoma sp.
P-Leuctra sp.
P-Tallaperla sp. (8)
D-midge (3-4)
D-midge (1-2)
D-tanypod (3-5)
D-midge (5+)
D-Dixa sp.

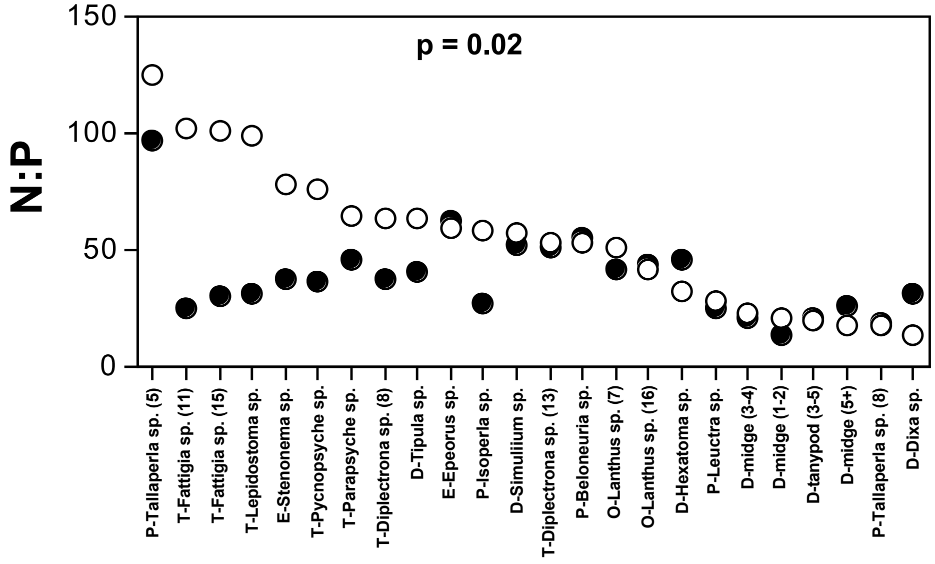

N:P
150
100
50
0
p = 0.02
P-Tallaperla sp. (5)
T-Fattigia sp. (11)
T-Fattigia sp. (15)
T-Lepidostoma sp.
E-Stenonema sp.
T-Pycnopsyche sp.
T-Parapsyche sp.
T-Diplectrona sp. (8)
D-Tipula sp.
E-Epeorus sp.
P-Isoperla sp.
D-Simulium sp.
T-Diplectrona sp. (13)
P-Beloneuria sp.
O-Lanthus sp. (7)
O-Lanthus sp. (16)
D-Hexatoma sp.
P-Leuctra sp.
D-midge (3-4)
D-midge (1-2)
D-tanypod (3-5)
D-midge (5+)
P-Tallaperla sp. (8)
D-Dixa sp.

Figure 2.4. Mean %P, %N, %C, C:P, N:P, and C:N ratio (+1 SD) of invertebrates from 5 dominant insect orders in the reference stream (C53, white bars) and the enriched stream (C54, grey bars). DIPT = Diptera; ODON = Odonata; PLEC = Plecoptera; TRIC = Trichoptera; EPHE = Ephemeroptera. Results of 2 way ANOVA: ORDER factor significant for all variables: %P****, %N****, %C****, C:P****, N:P***, C:N****, STREAM factor significant for C:P*, ORDER x STREAM interaction significant for C:P** and N:P**. Different capital letters above bars indicate significant differences among orders (Tukey's HSD) using data from both streams (i.e., STREAM factor not significant). Different lower-case letters above bars (as for C:P) indicate significant differences (Tukey's HSD) considering the reference and enriched streams separately (i.e., STREAM factor significant). *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, ****$p < 0.0001$. For %P in C53 & C54, DIPT (N = 17, 23), ODON (N = 5, 6), PLEC (N = 9, 14), TRIC (N = 12, 23), EPHE (N = 6, 10). For other graphs in C53 & C54, DIPT (N = 11, 17), ODON (N = 5, 6), PLEC (N = 8, 12), TRIC (N = 8, 20), EPHE (N = 4, 7). All ratios are molar.

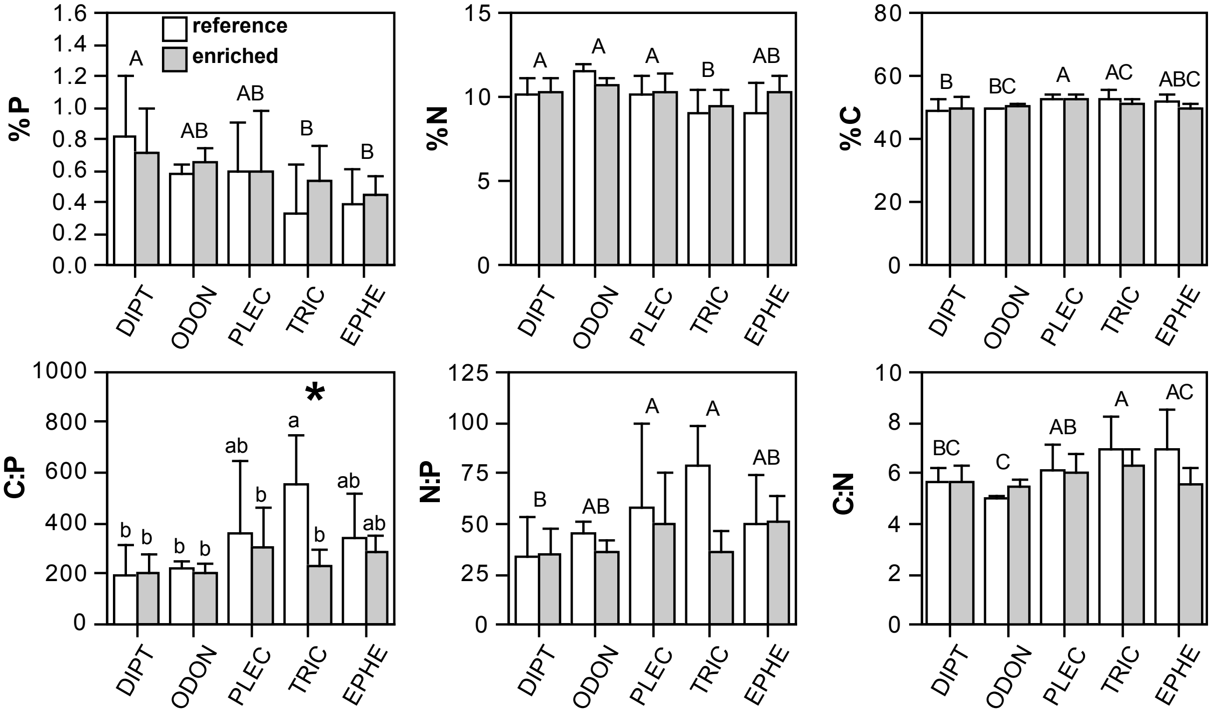
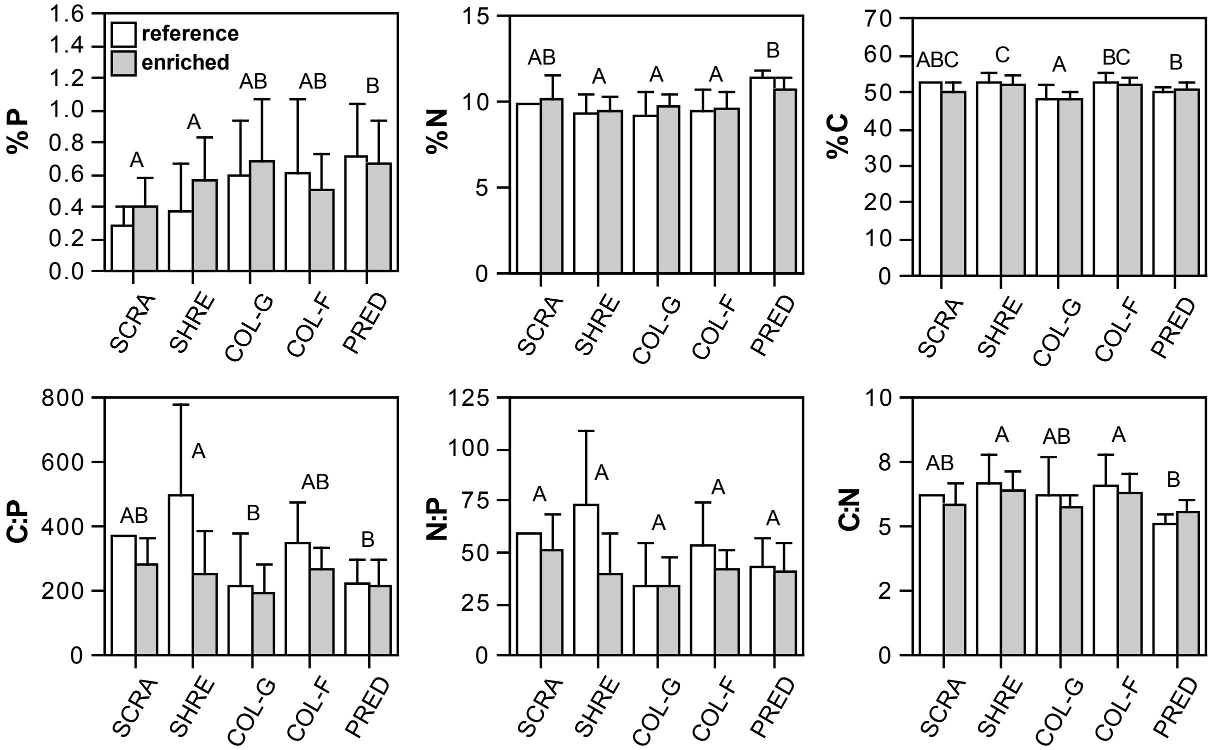

%P
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
reference
enriched
A
AB
AB
B
B
%N
15
10
5
0
A
A
A
B
AB
%C
80
60
40
20
0
B
BC
A
AC
ABC
DIPT
ODON
PLEC
TRIC
EPHE
C:P
1000
800
600
400
200
0
*
b
b
b
b
ab
b
a
b
ab
ab
N:P
125
100
75
50
25
0
B
AB
A
A
AB
C:N
10
8
6
4
2
0
BC
C
AB
A
AC
DIPT
ODON
PLEC
TRIC
EPHE

Figure 2.5. Mean %P, %N, %C, C:P, N:P, and C:N ratio (+1 SD) of invertebrates from 4 functional feeding groups in the reference stream (C53, white bars) and the enriched stream (C54, gray bars). SCRA = scraper; SHRE = shredder; COL-G = collector-gatherer; COL-F = collector-filterer; PRED = predator. Results of 2 way ANOVA: FFG factor significant for all variables except N:P: %P**, %N****, %C****, C:P***, C:N****, STREAM factor not significant for any variables, FFG x STREAM interaction not significant for any variables. Different capital letters above bars indicate significant differences (Tukey's HSD) based on results from both streams combined. *$p < 0.05$, **$p < 0.001$, ***$p < 0.001$, ****$p < 0.0001$. For %P in C53 & C54, SCR (*N* = 2, 10), SHRE (*N* = 13, 20), COL-G (*N* = 9, 9), COL-F (*N* = 9, 8), PRED (*N* = 16, 29). For other graphs in C53 & C54, SCR (*N* = 1, 5), SHRE (*N* = 11, 16), COL-G (*N* = 7, 7), COL-F (*N* = 6, 6), PRED (*N* = 11, 28). All ratios are molar.


reference
enriched
% P
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
A
A
AB
AB
B
% N
15
10
5
0
AB
A
A
A
B
% C
70
60
50
40
30
20
10
0
ABC
C
A
BC
B
C:P
800
600
400
200
0
AB
A
B
AB
B
N:P
125
100
75
50
25
0
A
A
A
A
A
C:N
10
8
5
2
0
AB
A
AB
A
B
SCRA
SHRE
COL-G
COL-F
PRED

CHAPTER 3

EFFECTS OF LONG-TERM NUTRIENT ENRICHMENT ON GROWTH AND PRODUCTION OF TWO STREAM DETRITIVORES[1]

---

[1] Cross, W. F., B. R. Johnson, J. B. Wallace, and A. D. Rosemond. To be submitted to *Limnology and Oceanography*.

Abstract

The effects of nutrient enrichment (i.e., nitrogen and phosphorus) on the dynamics of primary consumers in detritus-based aquatic ecosystems is poorly understood. Nutrient enrichment may simultaneously increase the nutritional quality of detritus for consumers while accelerating its loss via increased metabolism of heterotrophic microbes such as bacteria and fungi. In this study we examined in detail the response of growth and secondary production of two dominant stream detritivores (non-Tanypodinae chironomids and *Tallaperla* spp.) to a 2-year large-scale experimental enrichment of a small headwater stream. We purposely contrasted the response of 2 detritivores at opposite ends of the 'slow-fast' life-history continuum and with distinct feeding behavior. Nutrient enrichment had large positive effects (~50% increase) on individual growth rates of chironomids, but no detectable effects on growth rates of *Tallaperla* spp.. On a per square meter basis, enrichment had large positive effects secondary production (~183% increase), and production/biomass ratios of chironomid larvae, but these effects were largely habitat-specific. In contrast, production of *Tallaperla* spp. per square meter was unaffected by enrichment. When expressed on a per gram organic matter basis (i.e., per gram leaf litter or fine benthic organic material), enrichment had a large positive effect on secondary production of chironomids and *Tallaperla* spp.. Together, these results suggest nutrient-induced changes to organic matter quality positively affected both taxa; however, for *Tallaperla* spp., positive effects on resource quality were offset by negative effects on resource (i.e., leaf litter) quantity. We explored the generality of these results by examining the response of many other stream taxa to enrichment (Chapter 4) in relation to larval lifespan and feeding behavior. For leaf-eating taxa (i.e., shredders), life history and feeding behavior strongly interact to determine the response of these taxa to enrichment. Differential response of consumers to nutrient

perturbation underscores the importance of species-specific characteristics such as larval lifespan, feeding behavior, and elemental composition in determining the population and community-level responses of detritivores to enrichment.

Introduction

Nutrient enrichment of aquatic ecosystems is occurring worldwide as a result of human-induced changes to global nitrogen and phosphorus cycling (e.g., Caraco 1993, Bennett et al. 2001, Galloway et al. 2003). Fertilizer production and application, fossil fuel combustion, and suburban/urbanization have all contributed to increased mobilization of nitrogen (N) and phosphorus (P) and elevated concentrations of these elements in streams, lakes, and coastal marine environments (e.g., Caraco 1993, Vitousek et al. 1997, Carpenter et al. 1998, Howarth et al. 2002). Such changes in nutrient availability can have strong effects on population dynamics, community structure, and ecosystem processes because many functional attributes of aquatic systems (e.g., respiration, decomposition, primary and secondary production) are limited by the supply of N and P (e.g., Carpenter et al. 1998). Our general understanding of nutrient effects on aquatic ecosystems is limited, however, because the majority of studies have been conducted in algal- or plant-based systems, where effects of enrichment are often manifested in increased primary production and eutrophication (e.g., Hart and Robinson 1990, Peterson et al. 1993, Carpenter et al. 1998, Howarth et al. 2002).

In most aquatic ecosystems, detritus is a dominant basal resource and provides the energetic basis for diverse and productive detritus-based food webs (e.g., Wetzel 1995). However, despite the prevalence of these detrital pathways, our understanding of nutrient enrichment effects in detritus-based systems lags far behind that of plant- or algal-based systems.

Responses of detritus-based food webs to enrichment may be fundamentally different than their living plant-based counterparts (e.g., Polis and Strong 1996, Rosemond et al. 2001). Stimulation of microbial production at the base of the food web may provide an enriched resource for primary consumers in the short-term, but may ultimately have negative effects on these consumers by increasing the rate at which detritus is metabolized or lost from the system (e.g., Gulis and Suberkropp 2003).

In the eastern US, and many other forested regions of the world, detritus-based headwater streams dominate the total length of stream networks (e.g., Wallace 1988, Meyer and Wallace 2001). In these streams, productivity is driven by pulsed inputs of allochthonous leaf litter, and in-stream autotrophic production can be extremely low (e.g., Webster et al. 1997). Recent studies have shown that elevated levels of inorganic N and P in detritus-based streams can lead to increased biomass and productivity of microbes (i.e., bacteria and fungi) associated with detritus (e.g., Rosemond et al. 2002, Gulis and Suberkropp 2003, Ramirez et al. 2003, Stelzer et al. 2003), and consequent increased rates of organic matter decomposition (e.g., Elwood et al. 1981, Robinson and Gessner 2000, Grattan and Suberkropp 2001, Chadwick and Huryn 2003, Gulis and Suberkropp 2003). Thus, both the quality (i.e., through increased biomass of nutrient-rich microbes) and quantity (i.e., through increased rates of decomposition) of detritus can be affected by nutrient availability. However, we know very little about how nutrient-mediated changes in detritus affect the growth and productivity of primary consumers (but see Ward and Cummins 1979, Rosemond et al. 2001, 2002). This is an important line of research because primary consumers limit the flow of energy and materials to higher trophic levels, and often play prominent roles in the retention, processing, and export of organic matter and nutrients in forested streams (Wallace et al. 1982, Cuffney et al. 1990, Pringle et al. 1999, Crowl et al. 2001, Cross et al. in press).

Assemblages of primary consumers typically exhibit a broad range of life-history characteristics, with potentially large differences among coexisting taxa in terms of growth rates, lifespan duration, and feeding behavior (e.g., Huryn and Wallace 2000). These fundamental characteristics are likely to have considerable influence on the response of individual taxa to perturbations such as nutrient enrichment. For example, lifespan duration may influence the rate of population response because short-lived taxa may undergo more generations during a given perturbation than long-lived taxa. Additionally, feeding mode or behavior may affect the response of consumers if food resources are differentially affected by a given perturbation. Understanding emergent community-level responses to perturbations may require careful consideration of taxa that occupy disparate 'ends' of life-history continua and feeding behavior.

In this study, we experimentally enriched a detritus-based headwater stream with inorganic N and P for 2 years. Our objective was to examine effects of nutrient enrichment on the abundance, biomass, growth rates, and secondary production of two detritivorous taxa that exhibit very different life history strategies (i.e., fast growth and short life span vs. slow-growth and long life span) and feeding behavior (i.e., leaf litter vs. fine benthic organic detritus). We compared the response of these taxa in the treatment stream to an adjacent reference stream to isolate the effects of enrichment from natural inter-annual variability.

We predicted that increased food quality as a result of nutrient enrichment (Cross et al. 2003, Gulis and Suberkropp 2003) would positively influence growth rates and assimilation of both short and long-lived taxa. We tested this hypothesis by comparing the growth rates and production per gram organic matter (i.e., per gram leaf litter or fine benthic organic matter, a proxy for assimilation) of both taxa in the reference and treatment streams. In addition, we predicted that changes in organic matter quantity as a result of enrichment (i.e., decline in leaf litter) would negatively affect long-lived taxa that depend on continuous availability of leaf litter. This hypothesis was tested by examining consumer production on a per square meter basis. Because we examined 2 taxa that differ in both lifespan duration and feeding behavior, our ability to examine these characteristics independently is confounded. Therefore, we examined the response of multiple taxa to enrichment from a concurrent study (Cross Chapter 4) in relation to larval lifespan and feeding mode. Our analysis provided a unique opportunity to explore the multiple influences of larval lifespan, food quantity, and food quality on response of detritivores to a long-term nutrient enrichment.

Study organisms

Non-Tanypodinae chironomids (Diptera) were chosen as representative *r*-selected taxa (i.e., rapid growth, high reproductive rates). Chironomids are among the most abundant macroinvertebrates in most freshwater ecosystems (Armitage et al. 1995) and generally exhibit rapid growth rates (e.g., Huryn 1990, Hauer and Benke 1991, Johnson et al. 2003), short larval life spans (e.g., Huryn 1990), and multivoltine life histories (i.e., complete multiple generations per year, Huryn 1990). Most non-tanypod chironomids are functionally classified as collector-gatherers (Merritt and Cummins 1996), and their diet in southern Appalachian streams is dominated by amorphous detritus associated with fine benthic organic matter (FBOM) (e.g., Hall et al. 2000, Rosi-Marshall and Wallace 2002). More than 25 genera of non-Tanypodinae have been collected from the study streams (Wallace et al. 1991), but 7 dominant genera represent > 80% of their abundance (see Huryn 1990).

Tallaperla spp. (Plecoptera: Peltoperlidae) larvae were chosen as representative *K*-selected taxa (i.e., slow growth, low reproductive rates). These stoneflies exhibit slow growth rates (e.g., O'Hop et al. 1984, Johnson et al. 2003), have relatively long larval life spans (≈ 540 days), and comprise a significant proportion of the abundance and biomass of the study stream communities (Lugthart and Wallace 1992, Wallace et al. 1999, this study). *Tallaperla* spp. are functionally classified as leaf-eating shredders (Merritt and Cummins 1995), although early instars may predominantly feed on amorphous detritus (Cross, personal observation). Up to four species of *Tallaperla* spp. potentially co-occur in Coweeta headwater streams: *T. maria, T. anna, T. cornelia, and T. elisa* (Huryn and Wallace 1987, Stewart and Stark 1993). These species are indistinguishable as larvae but share the same semivoltine life cycle. Previous work has shown that growth rates of *Tallaperla* spp. are sensitive to changes in resource (i.e., leaf) quantity (Johnson et al. 2003).

Methods

Study site and experimental enrichment

This study was conducted in two adjacent headwater streams at the Coweeta Hydrologic Laboratory, Macon County, North Carolina, USA. Coweeta is a large (2185-ha) heavily forested basin located in the Blue Ridge physiographic province of the southern Appalachian Mountains (see Swank and Crossley 1988). Forest vegetation is dominated by mixed hardwoods (primarily oak, maple, and poplar) and a dense understory of *Rhododendron maximum* which shades the streams throughout the year. Headwater streams at Coweeta are extremely heterotrophic, and allochthonous inputs of detritus provide >90% of the energy base for microbial and invertebrate production (Wallace et al. 1997b, Hall et al. 2000). In-stream primary production is low (ca. 4 –

8 g C $m^{-2} y^{-1}$) and constitutes < 1% of the total carbon entering these streams (Wallace et al. 1997a).

The two streams used for this study drain catchments (C) 53 and 54. These streams have very similar physical and chemical characteristics (Table 3.1), but differ (since July 2000) in their concentrations of dissolved N and P as a result of our experimental nutrient enrichment of C54 (Table 3.1).

Starting in July 2000, nitrogen (NH_4NO_3) and phosphorus (K_2HPO_4 and KH_2PO_4) were dripped continuously into C54 to increase concentrations of dissolved inorganic N and P to ≈ 6-15 X background levels (Table 3.1). Nutrient solution was added along 140 m of C54 with a solar powered metered-dose pump (LMI, Acton, Massachusetts, USA) connected to a streamwater-fed plastic pipe laid along the streambed. The plastic pipe was fitted with garden irrigation valves every 10 m to evenly distribute nutrients along the length of the stream. Stream water nutrient concentrations were held relatively constant across a range of discharge by connecting the pump to a discharge data logger (Isco Inc., Lincoln, NE, USA) located at the base of the stream; the pump was engaged every time a known volume of water (generally 50-100 liters) passed through the downstream weir. Concentrations of (NO_3 + NO_2)-N, NH_4-N, and soluble reactive phosphorus (SRP) were measured biweekly at the weir of each study stream (APHA 1998). During enrichment, nutrient concentrations were also measured at 5 locations along the length of C54 to confirm that nutrients were evenly distributed along the stream. Nutrient concentrations in the enriched stream (Table 3.1) were well within the range of natural concentrations in streams in the region (Scott et al. 2002), and thus provided a realistic assessment of moderate enrichment effects. Water temperature (°C) was monitored every 30 min throughout the study in both streams (K. Suberkropp, University of Alabama, unpublished data) with Optic StowAway temperature probes (Onset Computer Corp., Pocasset, MA, USA).

Growth rates

In situ daily growth rates (mg mg^{-1} d^{-1}) of chironomids and *Tallaperla* spp. were quantified on a seasonal basis in both streams following methods developed by Huryn and Wallace (1986) and Huryn (1990). Chironomid growth rates were measured between November 1999 and July 2002 (n = 10 seasons; 3 before treatment, 7 during treatment), and *Tallaperla* spp. growth rates were measured between November 1999 and November 2001 (n = 8 seasons; 3 before treatment, 5 during treatment).

During each season, larvae were obtained from leaf litter and FBOM collected in nearby undisturbed Coweeta headwater streams. Organic matter was rinsed with stream water through a series of nested metal sieves and examined under a dissecting microscope (chironomids) or by eye (*Tallaperla* spp.) for removal of larvae. Body length of larvae was measured to the nearest 0.01 mm under a dissecting microscope fitted with an ocular micrometer, and larvae were separated into 1-mm size classes irrespective of species identity. Size classes used for each growth incubation were chosen based on the natural size distribution of larvae at the time of collection. In general, seasonal incubations consisted of 3 distinct size classes of chironomids and 2-4 size classes of *Tallaperla* spp. in each stream.

Groups of larvae within a given size class were placed into partially submerged triangular growth chambers constructed of Plexiglas® and Nitex® mesh. (base: 20 cm x 20 cm x 14 cm, sides: 16 cm height, described in Huryn and Wallace 1986). The mesh size of chambers (63 μm) was small enough to prevent immigration or emigration of stream invertebrates, but sufficiently large to allow entry of FBOM. Chambers were placed directly in the stream with the triangular point facing upstream, their tops extending above the water surface, and anchored to the stream bottom with rocks placed along external Plexiglas® flanges. The shape of growth chambers greatly reduced the amount of drag and debris-clogging of the nitex mesh. Larval densities in chambers (Appendix A) fell within the range of naturally occurring densities of chironomids, and were slightly higher than natural densities of *Tallaperla* spp. in the study streams (Lugthart et al. 1990, Wallace et al. 1999, this study). On occasion, two widely disparate size classes of *Tallaperla* spp. were placed in the same growth chamber to increase the number of growth estimates during a given season. Because these size classes were easily distinguishable, growth rates could be estimated separately.

At the beginning of growth incubations, 8-10 leaves collected from either stream were rinsed to remove invertebrates and placed in each of the chambers. Leaves placed in the chambers represented a mixture of species readily available during the time of incubation. The most commonly used leaf types were maple (*Acer* spp.), beech (*Fagus granifolia*, Ehrh.), oak (*Quercus* spp.), rhododendron, yellow poplar (*Liriodendron tulipifera* L.), and dogwood (*Cornus florida* L.). Efforts were made to keep leaf species and leaf condition relatively constant among chambers and between streams.

Growth incubations lasted ≈ 1-2 weeks for chironomids and ≈ 2 months for *Tallaperla* spp. (see Appendix A). Leaves were replaced half way through *Tallaperla* spp. incubations to prevent food limitation of larvae. At the end of growth incubations chamber contents were preserved in Kahle's solution, larvae were removed, and final body length of larvae was measured as described above. Initial and final lengths were converted to biomass estimates (ash-free dry mass, AFDM) using previously established length-mass regressions for these taxa

(Benke et al. 1999). Size-specific daily growth rates (g, mg mg^{-1} d^{-1}) of larvae were calculated as: $g = (\ln M_f - \ln M_i)/t$, where M_f is the mean final AFDM of surviving larvae, M_i is the mean intial AFDM of larvae introduced into chambers, and t is the duration of the incubation in days. Linear equations were derived from relationships between ln(initial length[mm]) and daily growth rate. Growth estimates were not used in these relationships if fewer than 2 individuals survived the incubation. This assemblage-based method of estimating growth rates assumes that changes in the average weight of similarly-sized mixtures of species accurately reflects size-specific growth rates of the entire taxonomic group (Huryn and Wallace 1986).

Food was always provided in excess during growth incubations to control for food quantity; thus, between-stream differences in growth were viewed as a test for effects of food quality.

Benthic sampling and secondary production

Quantitative benthic sampling was conducted monthly in each stream for 4 yrs between September 1998 and August 2002. Each month, samples were taken in two distinct stream habitats: mixed substrate (i.e., cobble, pebble, gravel, sand, silt) and bedrock outcrops. The mixed substrate habitat dominates the total area of both streams (C53: 73%, C54: 65%), with bedrock comprising the remainder (C53: 27%, C54: 35%). Mixed substrate samples were collected at 4 predetermined random locations in each stream with a core sampler (400 cm^2). Cores were firmly placed in the streambed, and all material was removed to a depth of 15 cm by hand or cup and placed in a large plastic jar. Bedrock samples (15 cm x 15 cm) were taken at 3 randomly determined locations in each stream by brushing and scraping moss and associated particles from a known area (15 cm x 15 cm) with a dish scrubber and knife into a plastic bag pressed flush against the bedrock.

Benthic samples were brought to the laboratory, refrigerated, and processed within 24 hours. Samples were rinsed onto nested metal sieves (pore sizes 1 mm and 250 μm), and material retained on each sieve was elutriated to separate organic from inorganic material. Organic material was then preserved separately for each size fraction (> 1mm and < 1 > 250 μm) with formalin solution (6-8%). All invertebrates were removed from the > 1 mm fraction by hand picking under a dissecting microscope at 15x magnification. Organic material in the smaller size fraction (<1 mm>250 μm) was subsampled (1/8 to 1/64 of whole samples) using a sample splitter (Waters 1969), and animals were removed from subsamples with a dissecting microscope at 15x magnification. The amount of coarse and fine particulate organic matter in each sample was also quantified, and those data are presented elsewhere (Cross Chapter 4).

All Non-Tanypodinae chironomids and *Tallaperla* spp. were counted and their body length measured to the nearest mm using a graduated microscope stage. Biomass (mg AFDM) of individual larvae was determined with length-mass relationships as described above, and total biomass per square meter was calculated for each sample. Secondary production was estimated for each sampling interval using the community-level instantaneous growth method (Huryn and Wallace 1986, Benke 1993). Production (mg AFDM m^{-2} $interval^{-1}$) for each sampling interval (P_{int}) was calculated as: $P_{int} = \sum_{i = 1 \text{ to } n} ([B_{t+1} + B_t]/2) \times g_i \times d$, where n = 1 mm size classes, B_{t+1} = mean larval biomass at sampling interval t+1, B_t = mean larval biomass at sampling interval t, g_i = size-specific instantaneous growth rate, and d = number of days in the interval. Annual production was calculated as the sum of all P_{int} values for a given year. Size-specific growth rates were obtained from empirically-derived linear growth equations described above. Larval abundance (no. m^{-2}), biomass (mg AFDM m^{-2}), and interval or annual secondary production (mg AFDM m^{-2} interval[d^{-1}] or mg AFDM m^{-2} y^{-1}) were estimated separately for each stream habitat. Habitat-weighted values for the entire stream were calculated according to the relative proportion of each habitat in each stream (e.g., Huryn and Wallace 1987). Production/biomass (P/B) ratios were calculated for each sampling interval (P/B_{int}) and for each year of study. Annual P/B ratios can be used as a relative assessment of biomass turnover rates (Benke 1993, Benke 1998).

To examine the influence of changes in food *quality* on production of chironomids and *Tallaperla* spp., annual production during the study was additionally expressed on a per gram leaf litter (for *Tallaperla* spp.) or FBOM (for chironomids) basis (organic matter data from Cross Chapter 4).

Statistical analyses

A t-test was used to test for temperature differences between streams. We used stepwise multiple regression analysis to build parsimonious models for predicting growth rates or mortality from mean daily stream temperature, number of degree days, density of larvae in chambers, and initial length of larvae (mm) ($\alpha \leq 0.1$ to enter model). Analysis of covariance (ANCOVA) was used to test for differences in growth rates between streams before and during enrichment; initial length was used as the covariate because early instars tend to grow faster than late instars for both taxonomic groups examined (e.g., Huryn 1990, Johnson et al. 2003).

Time series of chironomid and *Tallaperla* spp. abundance, biomass, and secondary production were analyzed with randomized intervention analysis (RIA, Carpenter et al. 1989, also see Murtaugh 2003, Stewart-Oaten 2003). RIA uses paired, before-and-after time series data from a manipulated and a reference system to detect changes caused by the manipulation.

In this study, RIA was used to test the null hypothesis that no change in abundance, biomass, or secondary production occurred in the treatment stream relative to the reference stream following the initiation of nutrient enrichment.

Differences in the proportion of larvae distributed among size classes between mixed substrate and bedrock outcrop habitats were tested with chi-square tests. We examined relationships between P/B_{int} in reference and enriched streams before and during treatment. Deviations from the 1:1 line before and during treatment were examined with Wilcoxon signed-ranks tests. All data were appropriately transformed (log[x+1] or arcsin-square root) when necessary to meet assumptions of normality and homoscedasticity.

Results

Temperature

Throughout the study, temperature ranged from 1 – 19°C and did not differ between study streams ($P > 0.05$, K. Suberkropp, unpublished data).

Growth rates

Numbers of chironomid larvae introduced into individual growth chambers ranged from 13 to 94 (mean: 47). Average mortality of chironomids was high at 69% (range: 37% - 100%) (Appendix A), and daily mortality rates averaged 6.1%/d (range: 2.5 – 11.8%/d). Rates of mortality were not influenced by larval density, larval initial length, or the number of degree days of growth incubations (regression analysis: r^2 values < 0.1, P values > 0.05). However, mean daily stream temperature (°C) during incubations had an overall significant positive effect on mortality rates ($r^2 = 0.18$, $P = 0.002$). When streams were analyzed separately, the effect of temperature was only significant for C54 ($r^2 = 0.41$, $P < 0.001$), and did not differ between pre- and post-enrichment periods ($P > 0.05$).

Growth rates of chironomid larvae in the reference and treatment stream were relatively high (mean: 0.07 mg mg^{-1} d^{-1}) and ranged from 0.015 to 0.153 mg mg^{-1} d^{-1} (Figure 3.1A). Mean daily temperature (°C), number of degree days, and larval density did not affect chironomid growth rates (r^2 values < 0.10, $P > 0.1$), and were therefore dropped from the model. Overall, growth rate was inversely related to initial length in both streams ($r^2 = 0.48$, $P < 0.0001$) and this relationship was best described by the equation: $g = -0.065 * \ln(\text{initial length[mm]}) + 0.122$. Prior to the nutrient enrichment, no difference was found between growth rates in the reference stream and the treatment stream (ANCOVA, $P > 0.05$, Figure 3.1A). These data were thus combined to yield a single 'non-enriched' growth equation: $g = -0.054 * \ln(\text{initial length[mm]}) + 0.095$ ($r^2 = 0.49$, $P < 0.0001$, Figure 3.1A: dotted line).

Nutrient enrichment of C54 had a highly significant positive effect on chironomid growth rates (ANCOVA, $P < 0.0001$, Figure 3.1A). On average, chironomids grew 53% faster in the treatment stream during enrichment than in the reference stream and treatment before enrichment. This relationship was best described by the linear equation: $g = -0.083 * \ln(\text{initial length[mm]}) + 0.143$ ($r^2 = 0.78$, $P < 0.0001$, Figure 3.1: solid line). Differences between enriched and non-enriched conditions were most pronounced among small larvae (Figure 3.1A).

Numbers of *Tallaperla* spp. introduced into growth chambers ranged from 5 to 40 (mean: 25). Average mortality of *Tallaperla* spp. was slightly lower than chironomids at 42% (range: 6 – 100%), and daily mortality rates averaged 0.7%/d (range: 0.1 – 1.4%/d). Rates of mortality were not significantly affected by initial length or density of larvae, mean daily temperature (°C), or degree days during incubation (r^2 values < 0.10, $P > 0.05$).

Growth rates of *Tallaperla* spp. were an order of magnitude lower than chironomids (overall average: 0.006 mg mg^{-1} d^{-1}) and ranged from 0 to 0.015 mg mg^{-1} d^{-1} (Figure 3.1B). There was no significant effect of larval density, mean daily temperature (°C), or number of degree days on *Tallaperla* spp. growth rates (r^2 values < 0.10, $P > 0.1$). A large proportion of the variation in growth rates was explained by initial length of larvae ($r^2 = 0.50$, $P < 0.0001$, Figure 3.1B), and this negative relationship was best described by the linear equation: $g = -0.007 * \ln(\text{initial length[mm]}) + 0.018$.

Growth rates of *Tallaperla* spp. did not differ significantly among the reference stream, the treatment stream before enrichment, and the treatment stream during enrichment (ANCOVA, $P > 0.05$, Figure 3.1B).

Assemblage responses to nutrient enrichment

Nutrient enrichment had a large positive effect on habitat-weighted chironomid abundance, biomass, and interval secondary production per square meter (RIA, P values all < 0.0001, Figure 3.2, Table 3.2). The greatest response was seen in chironomid production (183% increase in comparison to pretreatment period), followed by biomass (86% increase) and abundance (70% increase). These increases are conservative estimates because concomitant decreases were seen in abundance, biomass, and production in the reference during treatment. A closer examination of individual habitats revealed that positive effects of enrichment only occurred in the mixed substrate habitat and not on bedrock outcrops (Table 3.2). Annual habitat-weighted chironomid production in the reference stream (C53) was similar before (mean of yrs 1 and 2: 1098 mg AFDM m^{-2} y^{-1}) and during (mean of yrs 3 and 4: 1050 mg AFDM m^{-2} y^{-1}) enrichment of C54 (Table 3.3). Annual habitat-weighted production in the treatment stream

(C54) increased considerably from an average of 968 mg m^{-2} y^{-1} before enrichment to 2531 mg AFDM m^{-2} y^{-1} during enrichment (Table 3.3).

We expressed annual production of chironomids in the mixed substrate habitat on a per gram organic matter (FBOM) basis (Table 3.4). During the 2 pretreatment years, chironomid production per gram FBOM was considerably lower (48% on average) in the treatment stream than in the reference stream (years 1 & 2, Table 3.4). During the 2 years of nutrient enrichment, production per gram FBOM increased in the enriched stream to levels comparable to that of the reference stream (years 3 & 4, Table 3.4). The quantity of FBOM was relatively stable in both streams before and during enrichment (Cross Chapter 4).

Nutrient enrichment did not significantly affect habitat-weighted *Tallaperla* spp. biomass or secondary production per square meter (RIA, P values > 0.05, Figure 3.3, Table 3.2). Habitat-weighted abundance increased 60% in the treatment stream (RIA, P = 0.01, Figure 3.3), while abundances in the reference stream decreased by 37% during enrichment (Table 3.2). This significant effect was likely driven by large recruitment events in the reference stream before (Sept-Dec 1998 and 1999), but not during, enrichment, and small increases in abundance in the treated stream during the enrichment (Figure 3.3). Abundance, but not biomass or production, of *Tallaperla* spp. increased in the mixed substrate habitat during enrichment (RIA, P = 0.01, Table 3.2). On bedrock outcrops, abundance, biomass, and production appeared to decrease in the treated stream during enrichment (RIA, P values < 0.05, Table 3.2). However, the significance of this RIA resulted from large increases in the reference stream during enrichment as opposed to increases in the enriched stream. Throughout the study, annual habitat-weighted production was consistently higher (≈ 75%) in the reference stream (mean: 308 mg AFDM m^{-2} y^{-1}) than in the enriched stream (175 mg AFDM m^{-2} y^{-1}), and nutrient enrichment had no effect on this difference (Table 3.3).

Annual production of *Tallaperla* spp. per gram leaf litter in the mixed substrate habitat was lower (55% on average) in the treatment stream than in the reference stream during the 2 years of non-enriched conditions (years 1 & 2, Table 3.4). In contrast, production of *Tallaperla* spp. per gram leaf litter increased during nutrient enrichment relative to the reference stream and was 47% greater during the second year of enrichment (year 4, Table 3.4).

Annual P/B ratios of chironomids ranged from 7.5 to 19.5 in the mixed substrate habitat, and 16.2 to 27.4 in the bedrock outcrops. These P/B ratios suggest average community biomass turnover rates between 49 and 13 days, with the fastest turnover rates occurring on bedrock outcrops (Table 3.3). In general, the highest annual P/B values were found in C54 during enrichment (Table 3.3). Higher P/B ratios on bedrock outcrops versus mixed substrate habitat was due to slightly higher proportions of smaller instars on bedrock. Plots of habitat-weighted P/B values during each sampling interval (P/B_{int}) in the reference versus the treatment stream demonstrated that before enrichment, values were spread evenly around the 1:1 ratio line (Figure 3.4A, Wilcoxon signed ranks test, arithmetic differences between C53 and C54 not significantly different than 0, $P > 0.05$). In contrast, during nutrient enrichment, 81% of P/B_{int} values fell above the 1:1 line (Figure 3.4A, $P < 0.05$), indicating consistently higher biomass turnover rates of chironomids in the enriched stream.

Annual P/B ratios of *Tallaperla* spp. were considerably lower than chironomids, ranging from 1.6 to 2.7 in the mixed substrate habitat, and 2.4 to 3.2 on bedrock outcrops (Table 3.3). These annual P/B values correspond to biomass turnover rates of 228 – 114 days. Because larval life span (or cohort production interval) of *Tallaperla* spp. in Coweeta streams is ≈ 540 days, this suggests at least two overlapping cohorts of *Tallaperla* spp. are present in these streams (e.g., O'Hop et al. 1984). Nutrient enrichment had no apparent effect on *Tallaperla* spp. P/B_{int} values (Figure 3.4B, Wilcoxon signed ranks tests, P values > 0.05).

Discussion

Although a number of studies have demonstrated a causal link between nutrient enrichment and increased microbial activity, detrital quality, and decomposition rates, this study is among the first to examine the long-term consequences of these basal changes on growth and production of detritivorous macroconsumers (but see Rosemond et al. 2002, Ramirez and Pringle, in review). We observed a marked difference between the response of chironomids and *Tallaperla* spp. to enrichment, and this difference provides valuable insight into the interaction between larval lifespan and feeding behavior in determining consumer response to nutrient enrichment.

Larval development time, or cohort production interval (CPI, Benke 1979), is an important life history characteristic that is central to determining population turnover rate and contributing to estimates of secondary production (e.g., Benke 1993). Differences in CPI among taxa largely determine the extent to which species can exploit disturbed habitats, or respond to environmental perturbations (e.g., Wallace 1990). Larval development times for non-Tanypodinae chironomids at Coweeta range from 46 – 365 days (Huryn 1990), indicating that some of these taxa are capable of completing up to 8 generations per year. Such fast development times likely enabled chironomids to increase their production rapidly in response to nutrient enrichment.

Nutrient-induced changes in food quality is the most probable mechanism responsible for increased production of chironomids. Fine benthic organic matter (the dominant food resource for chironomids) is extremely abundant in headwater streams at Coweeta (ca. 1000 g m^{-2}), and its quantity is relatively stable throughout the year (Cross Chapter 4). Nutrient enrichment caused increased N and P content of FBOM in the treatment stream (Cross et al. 2003), which generally translates to increased food quality for chironomids (e.g., Ward and Cummins 1979, Vos et al. 2000). In contrast, enrichment had little effect on FBOM quantity (Cross Chapter 4). Results from our growth incubations indicated that changes in food quality led to increased growth rates of chironomids. Additionally, chironomid production per gram FBOM increased in the treatment stream relative to the reference stream during enrichment (Table 3.4), further supporting the notion that changes were driven by food quality.

Enrichment also had a positive effect on the quality of leaf litter, the dominant food resource of *Tallaperla* spp.. Concurrent research demonstrated that leaf nutrient content (Cross et al. 2003, Gulis and Suberkropp), as well as fungal and bacterial biomass (K. Suberkropp, University of Alabama, unpublished data) increased significantly in the treatment stream during enrichment. At first glance, our results conflict with regard to the effects of increased food quality on *Tallaperla* spp.. We did not detect an effect of enrichment on individual growth rates (i.e., no quality effect), but we found that secondary production per gram leaf litter increased in the treatment stream during enrichment (i.e., positive quality effect). One possible explanation for this disparity is that growth incubations were not conducted for long enough time periods to detect a response. Each growth trial lasted 2 months, which only represents ca. 11% of the larval lifespan of *Tallaperla* spp.. During incubations, growth was sufficiently low that differences caused by enrichment may have been swamped by measurement error or variability among individuals. In contrast, 2 week growth incubations of chironomids represented ca. 50% of the lifespan of many chironomid taxa (Huryn 1990).

Alternatively, it is possible that growth rates of *Tallaperla* spp. are physiologically 'fixed', and effects of enrichment are only manifested as increased survivorship or increased carrying capacity per gram resource. Between-taxon differences in growth response may have been due to fundamental differences in elemental requirements (sensu Elser et al. 1996). Chironomids at Coweeta grow rapidly, and, in accordance with stoichiometric theory, have relatively high amounts of body P (1.1%) and low C:P ratios (113) (Elser et al. 1996, Cross et al. 2003). In contrast, *Tallaperla* spp. have slow growth rates and, on average, lower body P (0.5%) and higher C:P ratios (419) (Cross et al. 2003). Theory predicts that invertebrates containing high amounts of body P (or N) require P-(or N) rich food resources for optimal growth (Elser et al. 1996). Those that contain low amounts of these elements are less constrained by low-nutrient resources, and may not respond to increased food nutrient content. Thus, the absence of an enrichment effect on growth rates of *Tallaperla* spp. may have been due to a low physiological requirement for P and a lack of pronounced P limitation.

Despite evidence for potentially positive effects of food quality on *Tallaperla* spp., our results suggest that production per square meter was ultimately limited by food quantity. Larval development times for *Tallaperla* spp. are much longer than chironomids (480 – 550 days, O'Hop et al. 1984), and these semivoltine taxa complete less than one generation per year. Thus, *Tallaperla* spp. require consistent availability of leaf litter throughout the year. Nutrient enrichment led to major reductions in benthic leaf litter via increased leaf decomposition rates (Gulis and Suberkropp 2003, Greenwood 2004, Cross Chapter 4). In fact, during summer months of the enrichment (particularly year 2), leaf litter was virtually absent from the treatment stream (K. Suberkropp, unpublished data, Cross Chapter 4). Nonetheless, total secondary production per square meter was unaffected by enrichment. Therefore, overall, any positive effects of nutrient enrichment on food quality were offset by the negative effects of decreased food quantity. In this case, a long larval development time may be considered a constraint for *Tallaperla* spp. in terms of their ability to take full advantage of increased food quality.

Were differences in the response of chironomids and *Tallaperla* spp. to enrichment primarily caused by lifespan duration or feeding behavior? Can these life-history characteristics be viewed separately in terms of their effect on invertebrate response? To get at these questions we first examined the range of larval lifespans among invertebrate taxa in Coweeta headwater streams (Figure 3.5A). Larval lifespans ranged from ca. 28 – 1140 days. Next, we identified those taxa that increased in production more than 100% in the treatment stream during enrichment relative to the pretreatment period (data from Cross Chapter 4); these taxa are represented as dark circles on Figure 3.5A. It is clear that positive effects of enrichment were limited to taxa with lifespans less than or equal to 1 year. Thus, it appears that larval lifespan is indeed an important determinant of invertebrate response to enrichment. Next, we explored the response of long- versus short-lived shredders (Figure 3.5B). Here, our analysis revealed a minimal response among long-lived shredders, and a consistently large response among short-lived shredders. Interestingly, one of the main contributors to increased production in the treatment stream (Cross Chapter 4) was *Pycnopsyche* spp., a shredder with a relatively short lifespan (275 d). The dominant species of *Pycnopsyche* enters a non-feeding final instar in the spring, prior to the time leaf litter was limiting in the treatment stream. Thus, both the duration and timing of larval development may be important determinants of shredder response to enrichment. For gatherer taxa, 1 long-lived taxon did not respond to enrichment, and there was considerably variability in response among short-lived taxa (3.5C). This pattern suggests that larval lifespan may be less important in determining the response of gatherer taxa to enrichment because the quantity of their food base is unaffected by enrichment.

General growth and production comparisons

Daily growth rates of chironomids were high, but not outside the range of those of previous studies conducted at Coweeta (Huryn and Wallace 1986, Huryn 1990, Johnson et al. 2003). Growth rates were also comparable to those found for chironomids in other field-based (e.g., Gresens 1997) and laboratory studies (e.g., Mackey 1977, Vos et al. 2000), although growth is highly variable in these studies as a result of differences in taxonomic composition, thermal regime, and food quality. We found that nutrient enrichment had a large positive effect on chironomid growth (ca. 50% increase). However, the highest growth rates measured in this study (0.153 d^{-1}) were still considerably lower than values reported from warmer streams in the southeastern U.S. (Stites and Benke 1989, Hauer and Benke 1991), the southwestern U.S. (Fisher and Gray 1983), and the tropics (Jackson and Sweeney 1995, Rosemond et al. 2001, Ramirez and Pringle, in review).

Annual secondary production of chironomids under non-enriched conditions (mean: 1038 mg AFDM m^{-2} y^{-1}) was slightly lower than previous studies of undisturbed headwater streams at Coweeta (Lugthart et al. 1990: 2254 mg AFDM m^{-2} y^{-1}; Wallace et al. 1999: 1379 mg AFDM m^{-2} y^{-1}), but did not fall outside the 95% confidence interval (CI) of these estimates (1009 – 2319 mg AFDM m^{-2} y^{-1}). However, this comparison is conservative because a) growth equations used to estimate production in prior studies (Huryn 1990) generally predict higher production than equations used in this study, and b) production was abnormally high (3636 mg AFDM m^{-2} y^{-1}) in

C53 during 1984-1985 (Lugthart et al. 1990) during a year of low precipitation. During nutrient enrichment, annual production increased by ca. 160% compared to pre-enrichment values. The highest annual production value (2963 mg AFDM m^{-2} y^{-1}) was outside the 95% CI of previous studies, but was still lower than the highest reported value at Coweeta (3636 mg AFDM m^{-2} y^{-1} Lugthart et al. 1990). Drought conditions, such as those experienced in the mid-80s (Lugthart et al. 1990), and during this study, can positively influence invertebrate production by increasing organic matter retention (see Wallace et al. 1991, Wallace et al. 1999). However, in our study discharge was consistently low during pre- and post treatment periods (data not shown), suggesting that increased production of chironomids during enrichment was due factors other than increased retention, such as aforementioned changes in food quality.

As expected, growth rates and secondary production of *Tallaperla* spp. were significantly lower than chironomids (Tank 1996, Johnson et al. 2003). Estimated production values (mean: 242 mg AFDM m^{-2} y^{-1}) were similar, but slightly lower than those reported for other streams at Coweeta (mean: 358 mg AFDM m^{-2} y^{-1}, 95% CI: 236 – 480 mg AFDM m^{-2} y^{-1}, O'Hop et al. 1984, Wallace et al. 1999).

Interestingly, temperature did not influence growth rates of chironomids or *Tallaperla* spp. in this study. Others have found that temperature is an important determinant of chironomid growth rates (e.g., Mackey 1977, Huryn and Wallace 1986, Huryn 1990, Gresens 1997), as well as growth rates of many other benthic invertebrate taxa (e.g., Vanote and Sweeney 1980, Huryn and Wallace 2000). It is possible that the range of incubation temperatures used in this study (6.7 – 17.1°C) was not wide enough to detect a significant effect. Indeed, previous studies at Coweeta have measured growth rates at temperatures as low as 2°C (Huryn and Wallace 1986).

Long-term implications

In detritus-based ecosystems, enrichment is capable of stimulating microbial respiration (e.g., Ramirez et al. 2003, Stelzer et al. 2003), invertebrate production (e.g., this study, Cross Chapter 4), and leaf litter decomposition (e.g., Elwood et al. 1981, Gulis and Suberkropp 2003). Over a sufficient time period, these factors may cause a net loss of carbon from the system (A. D. Rosemond, unpublished data, Greenwood 2004). This carbon loss has the potential to cause major shifts in community structure by favoring species with short life spans that do not require continuous availability of coarse particulate organic matter. Our study suggests that community change in response to enrichment will largely depend on life history characteristics of the initial species composition. Such functional changes in the detritivore community, if persistent, will have major consequences for the storage, processing, and export of organic matter from headwater streams (e.g.,Wallace and Webster 1996).

Nutrient enrichment of aquatic ecosystems continues to threaten the long-term sustainability of aquatic communities and the ecosystem services they provide. Predicting the consequences of enrichment and developing effective management strategies will require considerable research in ecosystems that are based on detritus. Detritus-based food webs truly dominate the earth's biosphere (e.g., Odum and de la Cruz 1963), and increasing knowledge of their response to enrichment is a high priority.

Acknowledgements

We thank Heidi Wilcox and Sally Entrekin for field assistance and patience during long grueling hours of measuring live chironomids. We also thank Jonathan Benstead, Elizabeth

Reese, and Sue Eggert for insightful comments on an earlier draft. Research funding was provided by NSF (DEB – 9806610).

Literature Cited

Armitage, P, P. S. Cranston, and L. C. V. Pinder. 1995. The Chironomidae: Biology and Ecology of Non-Biting Midges. Chapman and Hall.

American Public Health Association. 1998. Standard Methods for the Examination of Water and Wastewater, 20th edn. American Public Health Association.

Benke, A. C. 1979. A modification of the Hynes method for estimating secondary production with particular significance for multivoltine populations. Limnology and Oceanography 24:168-171.

Benke, A. C. 1993. Concepts and patterns of invertebrate production in running waters. Verh. Internat. Verein. Limnol. 25:1-24.

Benke, A. C. 1998. Production dynamics of riverine chironomids: extremely high biomass turnover rates of primary consumers. Ecology 79:899-910.

Benke, A.C., A. D. Huryn, L. A. Smock, and J. B. Wallace. 1999. Length-mass relationships for freshwater macroinvertebrates in North America with particular reference to the southeastern United States. Journal of the North American Benthological Society 18: 308–343.

Bennett, E. M., S. R. Carpenter, and N. F. Caraco. 2001. Human impact on erodable phosphorus and eutrophication: a global perspective. BioScience 51:227-234.

Caraco, N. F. 1993. Disturbance of the phosphorus cycle: a case of indirect effects of human activity. Trends in Ecology and Evolution 8:51-54.

Carpenter, S. R., T. M. Frost, D. Heisey, and T. K. Kratz. 1989. Randomized intervention analysis and the interpretation of whole-ecosystem experiments. Ecology 70:1142-1152.

Carpenter, S. R., N. F. Caraco, D. L. Correll, R. W. Howarth, A. N. Sharpley, and V. H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. Ecological Applications 8:559-568.

Chadwick, M. A. and A. D. Huryn. 2003. Effect of a whole-catchment N addition on stream detritus processing. Journal of the North American Benthological Society 22:194-206.

Cross, W. F., A. D. Rosemond, J. P. Benstead, S. L. Eggert, and J. B. Wallace. in press. Differential effects of consumers on C, N, and P dynamics: insights from long-term research. Food Webs. deRuiter and Moore (eds.). Elsevier.

Cross, W. F., J. P. Benstead, A. D. Rosemond , and J. B. Wallace. 2003. Consumer-resource stoichiometry in detritus-based streams. Ecology Letters 6:721-732.

Crowl, T. A., W. H. McDowell, A. P. Covich, and S. L. Johnson. 2001. Freshwater shrimp effects on detrital processing and nutrients in a tropical headwater stream. Ecology 82:775-783.

Cuffney, T. F., J. B. Wallace, and G. J. Lugthart. 1990. Experimental evidence quantifying the role of benthic invertebrates in organic matter dynamics of headwater streams. Freshwater Biology 23:281-299.

Elser, J. J., D. R. Dobberfuhl, N. A. MacKay, and J. H. Schampel. 1996. Organism size, life history, and N:P stoichiometry. BioScience 46:674-684.

Elwood, J. W., J. D. Newbold, A. F. Trimble, and R. W. Stark. 1981. The limiting role of phosphorus in a woodland stream ecosystem: effects of P enrichment on leaf decomposition and primary producers. Ecology 62:146-158.

Fisher, S. G. and L. J. Gray. 1983. Secondary production and organic matter processing by collector macroinvertebrates in a desert stream. Ecology 64:1217-1224.

Galloway, J. N., J. D. Aber, J. W. Erisman, S. P. Seitzinger, R. W. Howarth, E. B. Cowling, and B. J. Cosby. 2003. The nitrogen cascade. BioScience 53:341-356.

Grattan, R. M., and K. Suberkropp. 2001. Effects of nutrient enrichment on yellow poplar leaf decomposition and fungal activity in streams. Journal of the North American Benthological Society 20:33-43.

Greenwood, J. 2004. The response of heterotrophic and autotrophic resources to a long-term nutrient enrichment in an Appalachian headwater stream ecosystem. Dissertation. University of Georgia, Athens, GA, USA.

Gresens, S. E. 1997. Interactive effects of diet and thermal regime on growth of the midge *Pseudochironomus richardsoni* Malloch. Freshwater Biology 38:365-373.

Gulis, V. and K. Suberkropp. 2003. Leaf litter decomposition and microbial activity in nutrient-enriched and unaltered reaches of a headwater stream. Freshwater Biology 48:123-134.

Hall, R. O. J., J. B. Wallace, and S. L. Eggert. 2000. Organic matter flow in stream food webs with reduced detrital resource base. Ecology 81:3445-3463.

Hart, D. D., and C. T. Robinson. 1990. Resource limitation in a stream community: phosphorus enrichment effects on periphyton and grazers. Ecology 71:1494-1502.

Hauer, F. R. and A. C. Benke. 1991. Growth of snag-dwelling chironomids in a blackwater river – the influence of temperature and discharge. Journal of the North American Benthological Society 10:154-164.

Howarth, R. W., A. Sharpley, and D. Walker. 2002. Sources of nutrient pollution to coastal waters in the United States: implications for achieving coastal water quality goals.

Estuaries 25:656-676.

Huryn, A. D. 1990. Growth and voltinism of lotic midge larvae: patterns across an Appalachian mountain basin. Limnology and Oceanography 35:339-351.

Huryn, A. D., and J. B. Wallace. 1986. A method for obtaining in situ growth rates of larval Chironomidae (Diptera) and its application to studies of secondary production. Limnology and Oceanography 31:216-222.

Huryn, A. D., and J. B. Wallace. 1987. Local geomorphology as a determinant of macrofaunal production in a mountain stream. Ecology 68:1932-1942.

Huryn, A. D., and J. B. Wallace. 2000. Life history and production of stream insects. Annual Review of Entomology 45:83-110.

Jackson, J. K, and B. W. Sweeney. 1995. Egg and larval development times for 35 species of tropical stream insects from Costa Rica. Journal of the North American Benthological Society 14:115-130.

Johnson, B. R., W. F. Cross, and J. B. Wallace. 2003. Long-term resource limitation reduces insect detritivores growth in a headwater stream. Journal of the North American Benthological Society 22:565-574.

Lugthart, G. J., and J. B. Wallace. 1992. Effects of disturbance on benthic functional structure and production in mountain streams. Journal of the North American Benthological Society 11: 138-164.

Lugthart, G. J., J. B. Wallace, and A. D. Huryn. 1990. Secondary production of chironomid communities in insecticide-treated and untreated headwater streams. Freshwater Biology 24:417-427.

Mackey, A. P. 1977. Growth and development of larval Chironomidae. Oikos 28:270-275.

Merritt, R.W. and K. W. Cummins. 1996. An Introduction to the Aquatic Insects of North America. 3rd edn., Kendall/Hunt Publishing Co., USA.

Meyer, J. L. and J. B. Wallace. 2001. Lost linkages and lotic ecology: rediscovering small streams. In: Ecology: Achievement and Challenge, M. C. Press, N. J. Huntly, and S. Levin (eds.), Blackwell Science Press.

Murtaugh, P. A. 2003. On rejection rates of paired intervention analysis: reply. Ecology 84:2799-2802.

Odum, E. P and A. A. de la Cruz. 1963. Detritus as a major component of ecosystems. AIBS Bulletin 13:39-40.

O'Hop, J., J. B. Wallace, and J. D. Haefner. 1984. Production of a stream shredder, *Peltoperla maria* (Plecoptera: Peltoperlidae) in disturbed and undisturbed hardwood catchments. Freshwater Biology 14:13-21.

Peterson, B. J., L. Deegan, J. Helfrich, J. E. Hobbie, M. Hullar, B. Moller, T. E. Ford, A. Hershey, A. Hiltner, G. Kipphut, M. A. Lock, D. M. Fiebig, V. McKinley, M. C. Miller, J. R. Vestal, R. Ventullo, and G. Volk. 1993. Biological responses of a tundra river to fertilization. Ecology 74:653-672.

Polis, G. A., and D. R. Strong. 1996. Food web complexity and community dynamics. American Naturalist 147:813-846.

Pringle, C. M., N. Hemphill, W. H. McDowell, A. Bednarek, and J. G. March. 1999. Linking species and ecosystems: different biotic assemblages cause interstream differences in organic matter. Ecology 80:1860-1872.

Ramirez, A. and C. M. Pringle. In review. Response of detrital consumers to phosphorus levels in lowland tropical streams.

Ramirez, A., C. M. Pringle, and L. Molina. 2003. Effects of stream phosphorus levels on microbial respiration. Freshwater Biology 48:88-97.

Robinson, C. T., and M. O. Gessner. 2000. Nutrient addition accelerates leaf breakdown in an alpine springbrook. Oecologia 122:258-263.

Rosemond, A. D., C. M. Pringle, A. Ramirez, and M. J. Paul. 2001. A test of top-down and bottom-up control in a detritus-based food web. Ecology 82:2279-2293.

Rosemond, A. D., C. M. Pringle, A. Ramirez, M. J. Paul, and J. L. Meyer. 2002. Landscape variation in phosphorus concentration and effects on detritus-based tropical streams. Limnology and Oceanography 47:278-289.

Rosi-Marshall, E. J. and J. B. Wallace. 2002. Invertebrate food webs along a stream resource gradient. Freshwater Biology 47:129-141.

Scott, M. C., G. S. Helfman, M. E. McTammany, E. F. Benfield, and P. V. Bolstad. 2002. Multiscale influences on physical and chemical stream conditions across blue ridge landscapes. Journal of the American Water Resources Association 38:1379-1392.

Stelzer, R. S., J. Heffernan, and G. E. Likens. 2003. The influence of dissolved nutrients and particulate organic matter quality on microbial respiration and biomass in a forest stream. Freshwater Biology 48:1925-1937.

Stewart, K.W., and B.P. Stark. 2002. Nymphs of North American Stonefly Genera (Plecoptera). 2nd edition. The Caddis Press, Columbus, Ohio, USA.

Stewart-Oaten, A. 2003. On rejection rates of paired intervention analysis: comment. Ecology 84:2795-2799.

Stites, D. L., and A. C. Benke. 1989. Rapid growth-rates of chironomids in 3 habitats of a subtropical blackwater river and their implications for P-B ratios. Limnology and

Oceanography 34:1278-1289.

Swank, W.T., and D.A. Crossley. 1988. Forest Hydrology and Ecology at Coweeta. Ecological Studies Series Volume 66, Springer-Verlag, New York.

Tank, J. L. 1996. Microbial activity on wood in streams: exploring abiotic and biotic factors affecting the structure and function of wood biofilms. Dissertation. Virginia Polytechnic Institute and State University.

Vannote, R. L., and B. W. Sweeney. 1980. Geographic analysis of thermal equilibria: a conceptual model for evaluating the effect of natural and modified thermal regimes on aquatic insect communities. American Naturalist 115:667-695.

Vitousek, P. M., J. D. Aber, R. W. Howarth, G. E. Likens, P. A. Matson, D. W. Schindler, W. H. Schlesinger, and D. G. Tilman. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecological Applications 7:737-750.

Vos, J. H., M. A. G. Oojevaar, J. F. Postma, and W. Admiraal. 2000. Interaction between food availability and food quality during growth of early instar chironomid larvae. Journal of the North American Benthological Society 19:158-168.

Wallace, J. B. 1988. Aquatic invertebrate research. In: Ecological Studies, Vol. 66: Forest Hydrology and Ecology at Coweeta, W. T. Swank and D. A. Crossley, Jr. (eds.), Springer-Verlag.

Wallace, J. B. 1990. Recovery of lotic macroinvertebrate communities from disturbance. Environmental Management 14:605-620.

Wallace, J. B., J. R. Webster, and T. F. Cuffney. 1982. Stream detritus dynamics: regulation by invertebrate consumers. Oecologia 53:197-200.

Wallace, J. B., T. F. Cuffney, S. L. Eggert, and M. R. Whiles. 1997. Stream organic matter inputs, storage, and export for Satellite Branch at Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:67-74.

Wallace, J. B., S. L. Eggert, J. L. Meyer, and J. R. Webster. 1997. Multiple trophic levels of a forest stream linked to terrestrial litter inputs. Science 277:102-104.

Wallace, J. B., A. D. Huryn, and G. J. Lugthart. 1991. Colonization of a headwater stream during three years of seasonal insecticidal applications. Hydrobiologia 211:65-76.

Wallace, J. B., S. L. Eggert, J. L. Meyer, and J. R. Webster. 1999. Effects of resource limitation on a detrital-based ecosystem. Ecological Monographs 69:409-442.

Wallace, J. B., and J. R. Webster. 1996. The role of macroinvertebrates in stream ecosystem function. Annual Review of Entomology 41:115-139.

Ward, G. M. and K. W. Cummins. 1979. Effects of food quality on growth of a stream detritivores: *Paratendipes albimanus* (Meigen) (Diptera: Chironomidae). Ecology 60:57-64.

Waters, T. F. 1969. Subsampler for dividing large samples of stream invertebrate drift. Limnology and Oceangraphy 14:813.

Webster, J. R., J. L. Meyer, J. B. Wallace, and E. F. Benfield. 1997. Organic matter dynamics in Hugh White Creek, Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:74-78.

Wetzel, R. G. 1995. Death, detritus, and energy flow in aquatic ecosystems. Freshwater Biology 33:83-89.

Table 3.1. Physical and chemical characteristics of headwater streams draining catchments 53 (reference) and 54 (enriched) at the Coweeta Hydrologic Laboratory. Elevations were measured at the gauging weirs.

Variable	**C53 (reference)**	**C54 (enriched)**
Catchment		
Area (ha)	5.2	5.5
Elevation (m asl)	829.0	841.0
Channel		
Gradient (cm m^{-1})	27.0	33.0
Length (m)	145.0	282.0
Bankfull Area (m^2)	327.0	443.0
Discharge (l s^{-1})		
Average*	1.2	1.5
Maximum*	47.2	35.5
Average this study¶	0.3	0.5
Maximum this study¶	3.8	4.8
Substrate composition (%)		
Mixed substrates	73.0	65.0
Bedrock outcrop	27.0	35.0
Temperature (°C)¶		
Annual average	12.0	12.0
Minimum	2.6	4.8
Maximum	18.6	16.7
Water chemistry (range)		
pH	6.6 (6.2-7)	6.9 (6.6-7.9)
(NO_3-N + NO_2)-N (μg l^{-1})		
pretreatment: 1999 – 2000	15.4 (9.4-25.8)	18.8 (4-39.5)
treatment: 2000 – 2002	16.9 (bd-151)	308.9 (11-1711)
NH_4-N (μg l^{-1})		
pretreatment: 1999 – 2000	9.4 (bd-30)	9.9 (bd-25)
treatment: 2000 – 2002	10.4 (bd-76)	105.5 (6-566)
SRP‡ (μg l^{-1})		
pretreatment: 1999 – 2000	7.6 (bd-20)	8.8 (bd-22)
treatment: 2000 – 2002	3.7 (bd-17)	51.2 (bd-268)

*C53 values from 12 years (1984-1996), C54 values from 8 years (1985-1992).

‡Soluble reactive phosphorus. ¶September 1998 – August 2002. bd = below detection limit.

Table 3.2. Mean monthly abundance, biomass, and interval secondary production of non-tanypod Chironomidae and *Tallaperla* spp. in the reference stream (C53) and enriched stream (C54) before (pretreatment: September 1998 – June 2000) and during (treatment: July 2000 – August 2002) nutrient enrichment of C54.

Taxon/habitat	Abundance (no m^{-2})					Biomass (mg m^{-2})					Production (mg m^{-2} interval^{-1})				
	reference			enriched		reference			enriched		reference			enriched	
Chironomidae															
Mixed Substrate	mean	±SE		mean	±SE	mean	±SE		mean	±SE	mean	±SE		mean	±SE
pretreatment	44979	(5894)	***	24470	(3859)	115.5	(14.4)	***	89.8	(19.2)	123.9	(14.8)	***	86.6	(12.4)
treatment	32899	(2412)		49330	(4627)	90.6	(6.7)		187.7	(23.7)	118.4	(6.8)		302.5	(24.8)
Bedrock Outcrops															
pretreatment	8832	(1085)	ns	16092	(2764)	19.3	(3.8)	ns	38.8	(7.3)	29.0	(3.5)	ns	64.2	(11.5)
treatment	9628	(2002)		13098	(1804)	25.1	(6.3)		33.8	(6.7)	36.8	(6.9)		75.1	(10.1)
Habitat-Weighted															
pretreatment	35219	(4328)	***	21538	(3024)	89.5	(10.4)	***	72.0	(13.2)	98.3	(10.7)	***	78.8	(9.4)
treatment	26616	(1994)		36649	(3197)	72.9	(5.6)		133.9	(16.8)	96.4	(5.4)		222.9	(18.2)
***Tallaperla* spp.**															
Mixed Substrate															
pretreatment	1193	(327)	**	276	(58)	105.7	(19.3)	ns	78.3	(25.5)	26.2	(4.7)	ns	12.2	(2.4)
treatment	627	(82)		598	(129)	164.8	(22.6)		138.6	(31.5)	35.5	(4.4)		25.5	(4.1)
Bedrock Outcrops															
pretreatment	294	(71)	**	315	(82)	53.6	(16.9)	*	44.0	(13.1)	14.3	(3.1)	*	10.2	(2.2)
treatment	511	(65)		211	(54)	97.0	(17.1)		33.5	(8.6)	24.4	(3.4)		9.3	(1.8)
Habitat-Weighted															
pretreatment	950	(240)	*	289	(52)	91.7	(14.9)	ns	66.3	(17.2)	23.0	(3.7)	ns	11.5	(1.8)
treatment	595	(64)		462	(90)	146.5	(18.6)		101.8	(20.5)	32.5	(3.6)		19.8	(2.7)

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$, Randomized intervention analysis. ns = not significant, $P > 0.05$. This analysis compares before- and after-treatment time series data from C53 and C54.

Table 3.3. Annual secondary production (mg m^{-2} y^{-1}) and P/B ratios for chironomids and *Tallaperl*a spp. during each year of the study in the reference (C53) and enriched (C54) streams. Year 1 = September 1998 – August 1999, year 2 = September 1999 – August 2000, year 3 = September 2000 – August 2001, year 4 = September 2001 – August 2002. Years 1 and 2 are before the experimental nutrient enrichment. Also see Figures 2 and 3.

	year 1		year 2		year 3		year 4	
Chironomidae	P	P/B	P	P/B	P	P/B	P	P/B
Mixed substrate								
reference	1005.4	9.2	1704.1	12.7	1260.9	15.2	1279.0	13.1
enriched	802.6	10.0	1456.8	7.5	3968.0	19.5	2817.4	15.7
Bedrock outcrop								
reference	292.9	16.2	321.6	17.9	332.8	17.2	581.8	16.7
enriched	479.7	16.2	854.0	19.9	1097.2	25.0	764.4	27.4
Habitat-Weighted								
reference	838.2	9.9	1356.8	13.2	1007.9	15.3	1090.7	13.5
enriched	692.8	11.1	1243.0	8.8	2963.2	20.0	2098.8	16.6
***Tallaperla* spp.**								
Mixed substrate								
reference	217.5	2.4	333.9	2.6	442.1	2.7	359.5	2.1
enriched	81.0	1.6	222.2	2.1	334.8	2.0	227.3	2.0
Bedrock outcrop								
reference	142.7	3.2	159.7	2.9	275.2	2.9	333.2	3.0
enriched	90.4	2.4	134.8	2.7	90.6	2.6	80.0	2.5
Habitat-Weighted								
reference	197.3	2.5	286.9	2.7	397.0	2.8	352.4	2.2
enriched	84.3	1.8	191.6	2.2	249.3	2.0	175.7	2.1

Table 3.4. Annual secondary production of non-Tanypodinae chironomids and *Tallaperla* spp. expressed per gram fine benthic organic matter (FBOM) or leaf litter throughout the study in the reference and treatment streams. A description of organic matter methods and results can be found in Chapter 4. Year 1 = September 1998 – August 1999, year 2 = September 1999 – August 2000, year 3 = September 2000 – August 2001, year 4 = September 2001 – August 2002.

	chironomid production mg mg^{-1} FBOM m^{-2} y^{-1}			*Tallaperla* spp. production mg mg^{-1} leaf litter m^{-2} y^{-1}		
	reference	treatment	% higher or lower in enriched stream	reference	enriched	% higher or lower in enriched stream
year 1	0.00096	0.00055	(-) 42%	0.00148	0.00046	(-) 69%
year 2	0.00204	0.00068	(-) 67%	0.00125	0.00074	(-) 41%
year 3	0.00164	0.00191	(+) 16%	0.00160	0.00147	(-) 8%
year 4	0.00156	0.00140	(-) 10%	0.00154	0.00225	(+) 47%

Figure 3.1. Size-specific daily growth rates (d^{-1}) of non-Tanypodinae chironomids (A) and *Tallaperla* spp. (B) in the reference stream (C53, white circles), the treatment stream before enrichment (C54, gray circles), and the treatment stream during the experimental enrichment (C54, black circles). Lines represent significant linear regressions (see text for equations). For chironomids (A), the dashed line represents data from the reference stream and the treatment stream before enrichment (white and grey circles), and the solid line represents data from the treatment stream during enrichment (black circles). For *Tallaperla* spp. (B), the solid line represents data from all time periods.

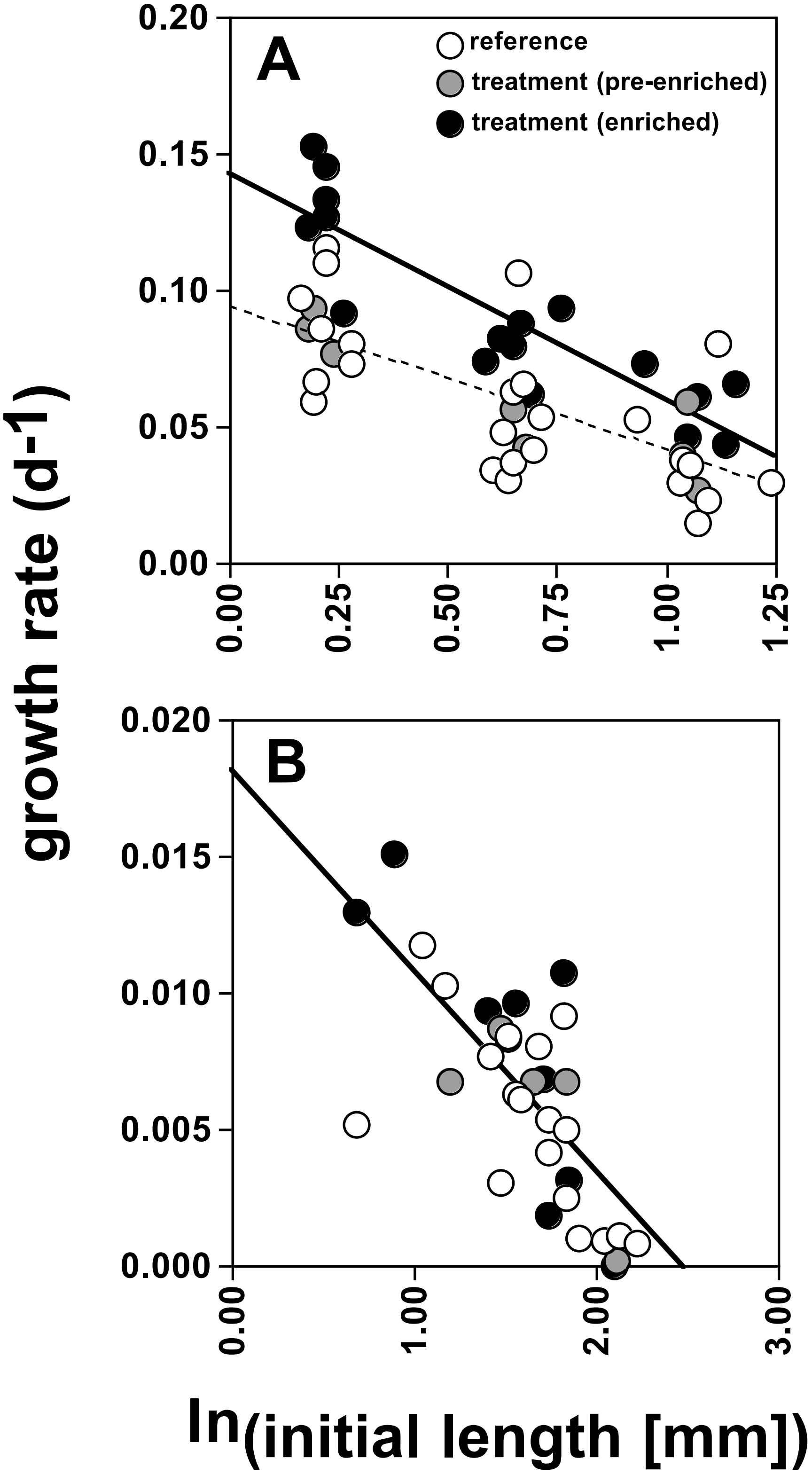

A
○ reference
● treatment (pre-enriched)
● treatment (enriched)
0.20
0.15
0.10
0.05
0.00
0.00
0.25
0.50
0.75
1.00
1.25
B
0.020
0.015
0.010
0.005
0.000
0.00
1.00
2.00
3.00
growth rate (d-1)
ln(initial length [mm])

Figure 3.2. Mean monthly abundance and biomass, and interval secondary production, of non-Tanypodinae chironomids in the reference stream (C53, white circles) and the treatment stream (C54, black circles) from September 1998 to August 2002. The arrow indicates the initiation of nutrient enrichment of C54. Differences between streams were tested with randomized intervention analysis.

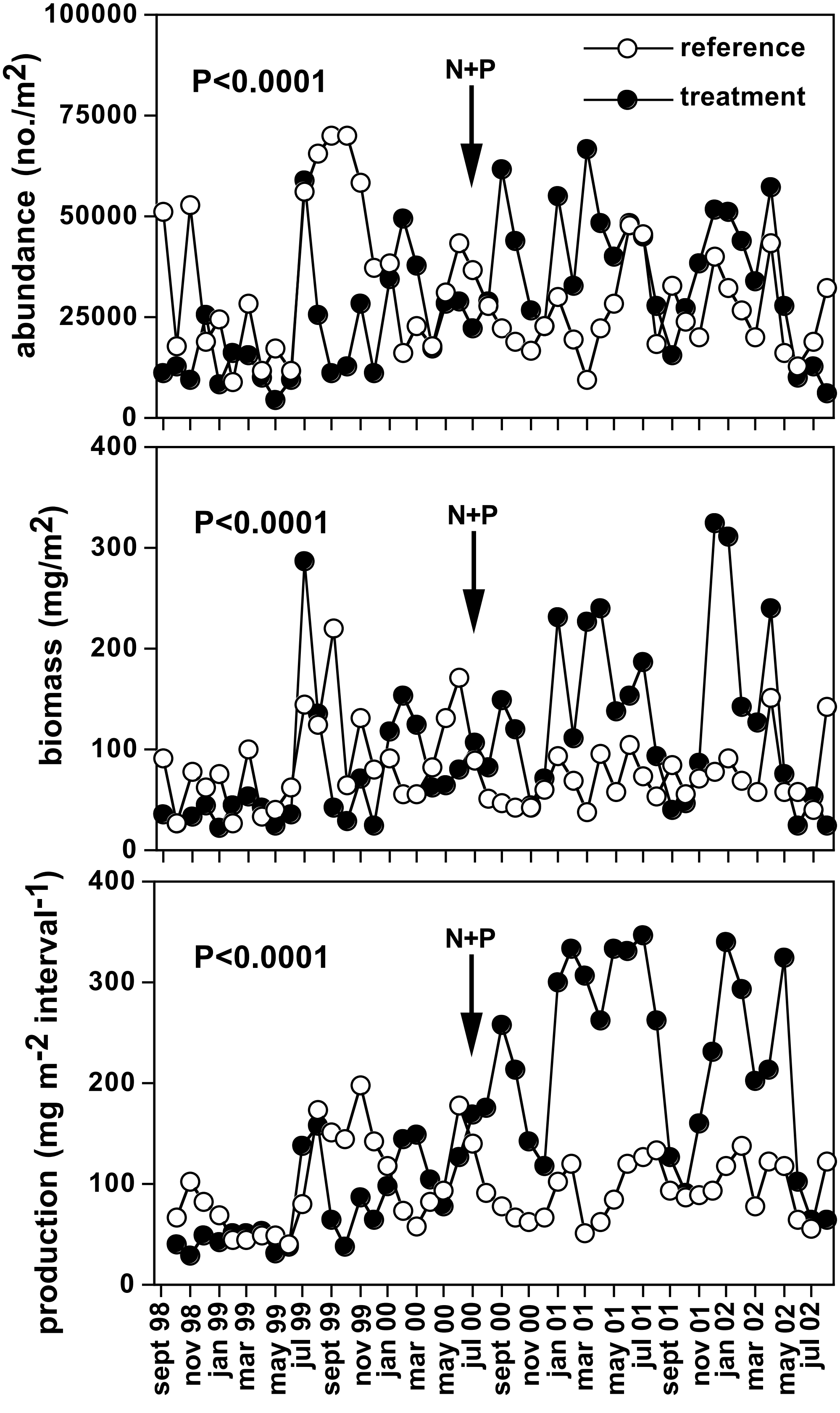

abundance (no./m^2)
100000
75000
50000
25000
0
P<0.0001
N+P
reference
treatment
biomass (mg/m^2)
400
300
200
100
0
P<0.0001
N+P
production (mg m^{-2} interval^{-1})
400
300
200
100
0
P<0.0001
N+P
sept 98
nov 98
jan 99
mar 99
may 99
jul 99
sept 99
nov 99
jan 00
mar 00
may 00
jul 00
sept 00
nov 00
jan 01
mar 01
may 01
jul 01
sept 01
nov 01
jan 02
mar 02
may 02
jul 02

Figure 3.3. Mean monthly abundance and biomass, and interval secondary production, of non-*Tallaperla* spp. in the reference stream (C53, white circles) and the treatment stream (C54, black circles). The arrow indicates the initiation of nutrient enrichment of C54. Differences between streams were tested with randomized intervention analysis.

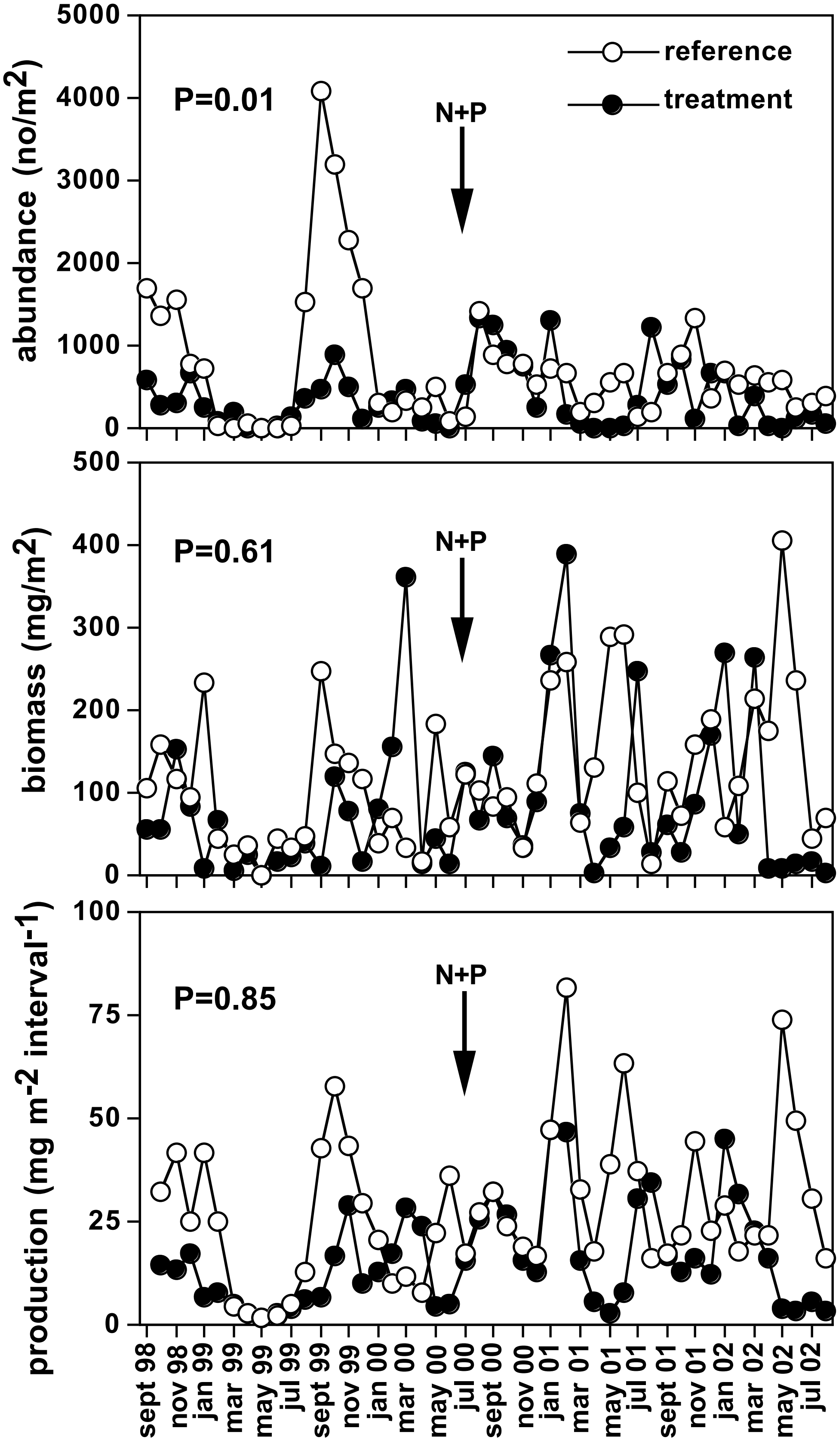

abundance (no/m^2)
0
1000
2000
3000
4000
5000
P=0.01
N+P
reference
treatment
biomass (mg/m^2)
0
100
200
300
400
500
P=0.61
N+P
production (mg m^{-2} interval^{-1})
0
25
50
75
100
P=0.85
N+P
sept 98
nov 98
jan 99
mar 99
may 99
jul 99
sept 99
nov 99
jan 00
mar 00
may 00
jul 00
sept 00
nov 00
jan 01
mar 01
may 01
jul 01
sept 01
nov 01
jan 02
mar 02
may 02
jul 02

Figure 3.4. Relationship between interval P/B (P/B_{int}) in the reference stream (C53) and P/B_{int} in the treatment stream (C54) before (pretreatment, white circles) and during enrichment of C54 (enrichment, black circles) for non-Tanypodinae chironomids (A) and *Tallaperla* spp. (B). Differences from the 1:1 line during pretreatment and treatment periods tested with Wilcoxon signed-ranks tests. ns = not significant ($P > 0.05$).

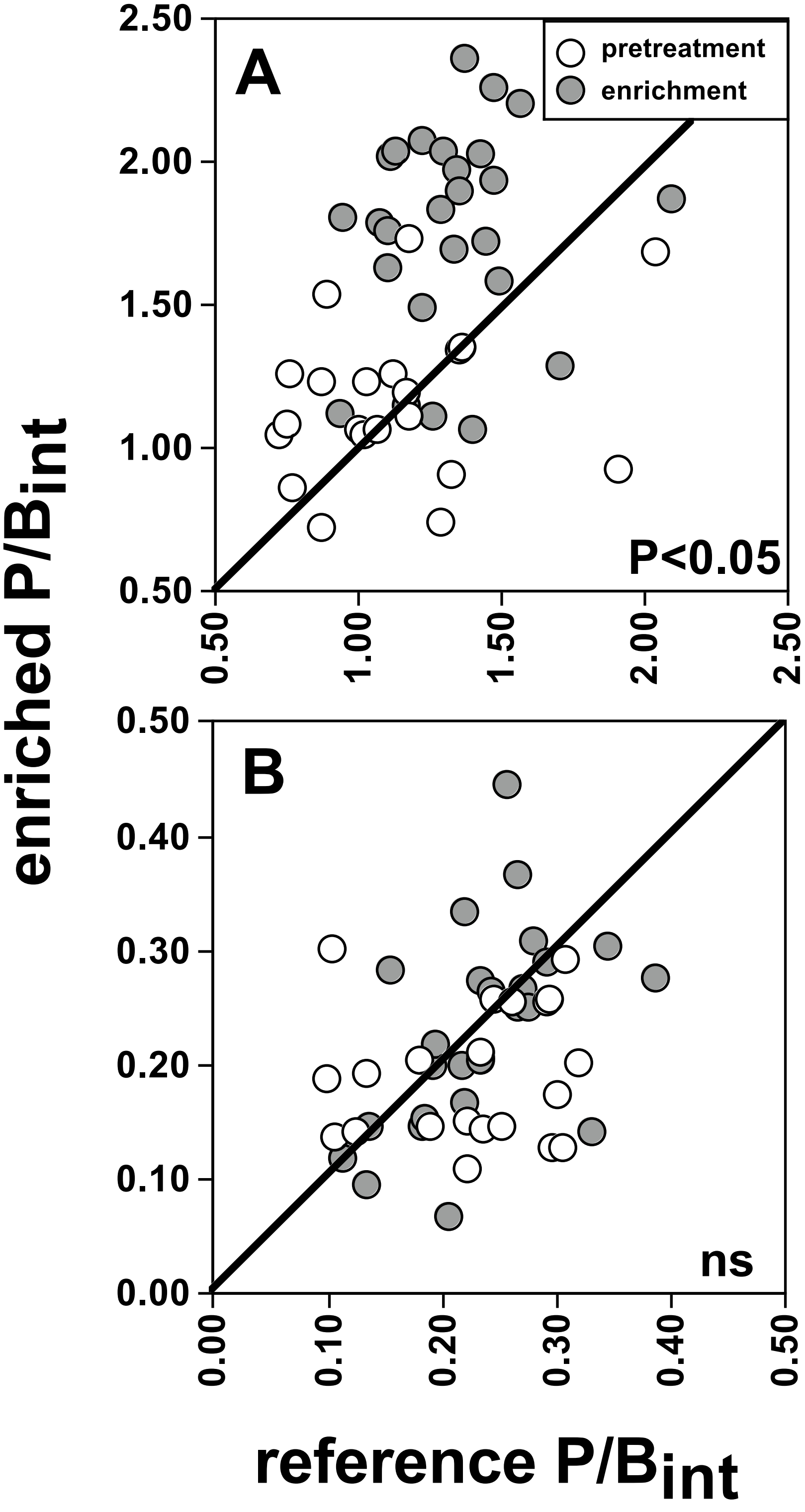
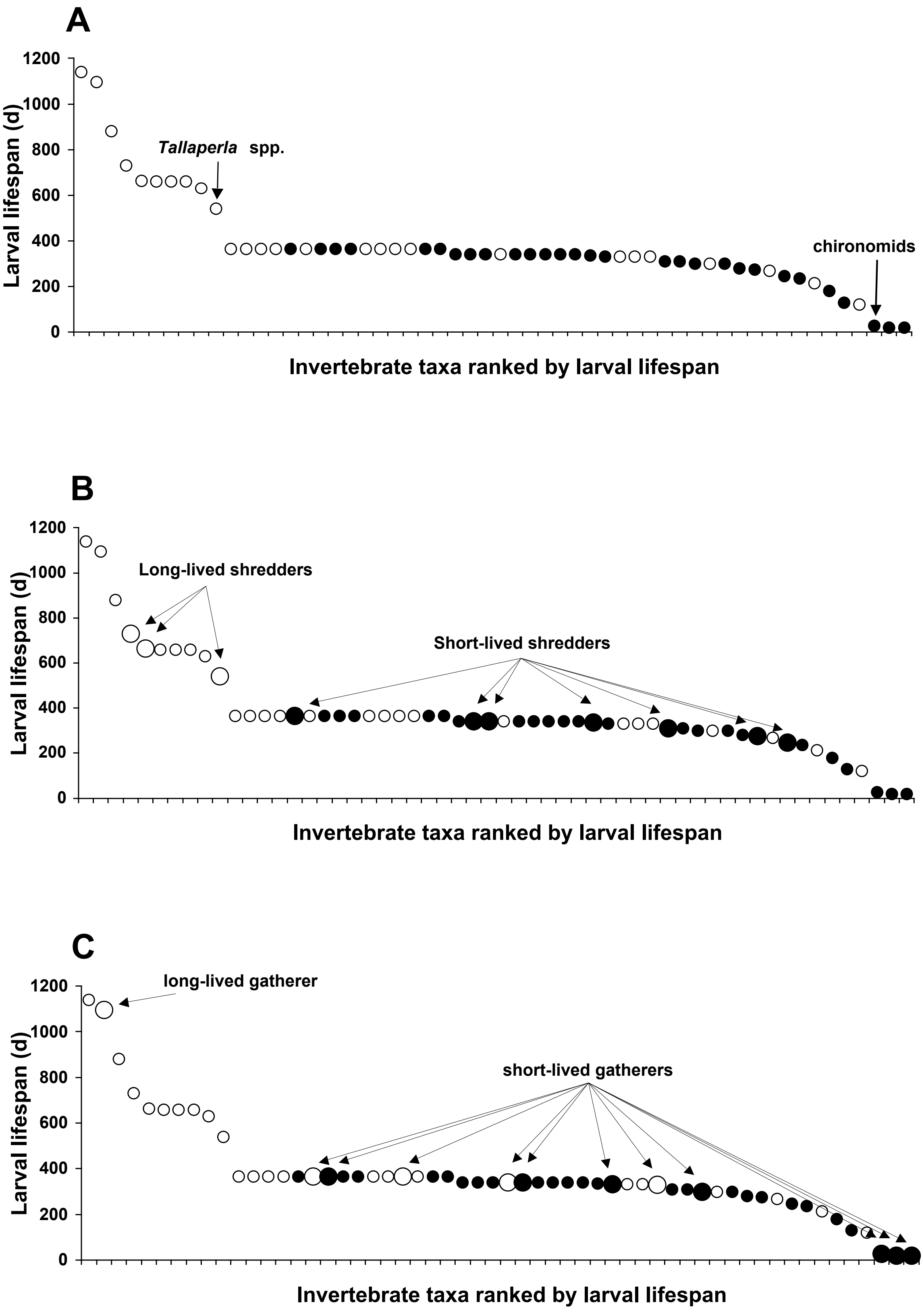

A
B
○ pretreatment
● enrichment
P<0.05
ns
2.50
2.00
1.50
1.00
0.50
0.50
1.00
1.50
2.00
2.50
0.50
0.40
0.30
0.20
0.10
0.00
0.00
0.10
0.20
0.30
0.40
0.50
enriched P/B_{int}
reference P/B_{int}

Figure 3.5. Coweeta invertebrate taxa ranked by larval lifespan. A. This panel shows the larval lifespan of chironomids and *Tallaperla* spp. in relation to other consumers. Dark circles indicate taxa that increased in production 100% or more in the treatment stream during enrichment relative to the pretreatment period. B. This panel points out the position of long-lived versus short-lived shredders. C. This panel points out the position of long-lived versus short-lived gatherers.


A
1200
1000
800
600
400
200
0
Larval lifespan (d)
Tallaperla spp.
chironomids
Invertebrate taxa ranked by larval lifespan
B
1200
1000
800
600
400
200
0
Larval lifespan (d)
Long-lived shredders
Short-lived shredders
Invertebrate taxa ranked by larval lifespan
C
1200
1000
800
600
400
200
0
Larval lifespan (d)
long-lived gatherer
short-lived gatherers
Invertebrate taxa ranked by larval lifespan

CHAPTER 4

CONSUMER RESPONSE TO LONG-TERM NUTRIENT ENRICHMENT OF A DETRITUS-BASED STREAM ECOSYSTEM[1]

---

[1] Cross, W. F., J. B. Wallace, A. D. Rosemond, and S. L. Eggert. To be submitted to *Ecology*.

Abstract

Increased availability and mobilization of nutrients, such as nitrogen and phosphorus, is having large effects on the structure, function, and diversity of aquatic and terrestrial ecosystems globally. Much of our understanding of these effects, however, comes from systems or food webs based on living plants or algae. In contrast, little is known about the effects of enrichment in systems based on non-living detritus, despite the prevalence and energetic importance of detritus in most ecosystems. We experimentally enriched a detritus-based headwater stream for 2 years to examine the effects of nutrient-induced changes at the base of the food web on higher metazoan (predominantly invertebrate) consumers. Our paired-catchment design was aimed at quantifying organic matter and invertebrate dynamics in the enriched stream and an adjacent reference stream for 2 years prior to enrichment and 2 years during enrichment. Enrichment had a strong negative effect on standing crop of leaf litter, but no apparent effect on fine benthic organic matter. Abundance, biomass and secondary production of many invertebrate taxa increased in response to enrichment, and taxon-specific differences were related to length of larval lifespan – a prominent life history characteristic. Production of invertebrate predators was also positively influenced by enrichment, and closely tracked the increased production of invertebrate prey. Response of invertebrates was largely habitat-specific with little effect of enrichment on food webs inhabiting bedrock outcrops. Invertebrate secondary production during the enrichment was the highest ever reported for streams at Coweeta, and was much higher than predicted production based on 27 years of background data from these streams. Despite reductions in leaf litter quantity caused by enrichment, production was extremely high underscoring the potential importance of nutrient-induced changes in food quality in driving consumer response.

Introduction

Humans are altering global carbon (C), nitrogen (N), and phosphorus (P) budgets. Activities such as fossil fuel combustion, agriculture, mining, and urban/suburbanization have led to changes in the dynamics of these elements (e.g., Galloway et al. 2003), and large-scale mobilization of nutrients such as N and P (e.g., Vitousek et al. 1997, Carpenter et al. 1998, Bennett et al. 2001, Paul and Meyer 2001). Increased nutrient availability in terrestrial and aquatic ecosystems may cause long-term changes in ecosystem structure and function because these elements often limit the rate of many ecological processes (e.g., Vitousek et al. 1997). While much is known about how nutrients function in plant- or algal-based ecosystems (e.g., Schindler et al. 1978, Tilman 1987, Tamm 1991, Vitousek and Howarth 1991, Peterson et al. 1993, Carpenter et al. 1998, Harvey et al. 1998), few studies have addressed the effects of enrichment in ecosystems based on detritus (i.e., non-living organic matter) (but see Chen and Wise 1999, Scheu and Schaefer 1999, Pearson and Connolly 2000, Rosemond et al. 2001, 2002, Ramirez et al. 2003).

Detritus-based ecosystems may respond to nutrient enrichment in a fundamentally different way than ecosystems based on living-plants. This is largely due to differences in composition of the dominant basal resource. In detrital systems, detritus and associated heterotrophic microbes (i.e., bacteria and fungi) occupy a similar trophic position as living plants or algae. This is of paramount importance because nutrient enrichment can stimulate the metabolism of these heterotrophic microbes (e.g., Pace and Funke 1991), as opposed to plants or algae, and this may result in a net loss of carbon at the base of the food web. Evidence from soils, lakes, streams, and coastal marine systems indicates that nutrients may indeed stimulate the biomass and metabolism of heterotrophic microbes (e.g., Pace and Funke 1991, Gallardo and

Schlesinger 1994, Pomeroy et al. 1995, Joergensen and Scheu 1999, Gulis and Suberkropp 2003). In addition, both correlational and experimental studies of forested streams and soils have linked increased microbial activity to higher decomposition or mineralization rates and carbon depletion (e.g., Elwood et al. 1981, Suberkropp and Chauvet 1995, Scheu and Schaefer 1998, Gulis and Suberkropp 2003, but see Aerts et al. 2003).

The degree to which nutrient-induced changes at the base of detrital food webs affects the dynamics of higher consumers has received very little attention (e.g., Scheu and Schaefer 1998, Pearson and Connolly 2000, Rosemond et al. 2001, 2002). Consumers that rely on detritus and associated microbes as a dominant food source may be positively or negatively affected by nutrient enrichment. In the short term, stimulation of microbial activity can lead to higher nutrient content of detritus, and increased food quality for consumers (e.g., Cross et al. 2003, Stelzer et al. 2003). Indeed, growth rates of detritivores have been shown to increase with detrital nutrient content (e.g., Iversen 1974, Tenore 1977, Ward and Cummins 1979, Vos et al. 2000). However, in the long term, accelerated decomposition rates may lead to decreased food quantity which could negatively affect long-lived consumers that depend on the availability of detritus throughout their entire life cycles.

Although all natural systems include important detrital pathways (e.g., Odum and de al Cruz 1963, Wetzel 1995, Moore et al., in press), detritus represents the dominant carbon source in forested headwater streams in the eastern U.S., and many other regions of the world (e.g., Wallace et al. 1997b). In these streams, productivity and metabolism are driven by inputs of allochthonous leaf detritus from the terrestrial catchment (e.g., Fisher and Likens 1973, Wallace et al. 1997b, 1999), and in-stream autotrophic production is generally very low (e.g., Fisher and Likens 1973, Webster et al. 1997). A number of studies have demonstrated strong positive links between the quantity of detritus and stream consumer abundance or productivity (e.g., Minshall 1967, Richardson 1991, Wallace et al. 1997, 1999). However, the quality of detritus is also likely to be important in determining limits to consumer production; this is particularly true in detritus-based systems where large elemental imbalances exist between consumers and food resources (Cross et al. 2003). Although many studies have demonstrated significant effects of nutrient enrichment on stream detritus quality (e.g., Howarth and Fisher 1976, Elwood et al. 1981, Cross et al. 2003, Stelzer et al. 2003), microbial activity (e.g., Ramirez et al. 2003, Stelzer et al. 2003) and decomposition rates (e.g, Elwood et al. 1981, Meyer and Johnson 1983, Rosemond et al. 2002, Gulis and Suberkropp 2003), our current knowledge of enrichment effects on productivity of higher consumers is limited by a lack of long-term experimental manipulations at the whole ecosystem scale. Such large-scale experiments are critical for assessing the response of ecosystems to environmental change and human-induced perturbations (e.g., Carpenter et al. 1995, Schindler 1998).

In this study, our main objective was to examine the effects of an ecosystem-scale experimental nutrient enrichment on community structure and productivity of consumers in detritus-based headwater streams. We examined the dynamics of invertebrates and benthic organic matter in the experimental stream and an adjacent reference stream for 2 years prior to enrichment and 2 years during enrichment. Concurrent studies in these streams demonstrated a strong positive effect of nutrients on the quality of detritus via increased detritus-bound microbial biomass and production (Gulis and Suberkropp 2003, K. Suberkropp, University of Alabama, unpublished data) and increased detrital nutrient content (Cross et al. 2003, Gulis and Suberkropp 2003). We predicted that these microbially-mediated changes would positively influence total production of invertebrate detritivores and predators. However, we hypothesized that the quantity of detritus (particularly leaf litter) would be negatively affected by enrichment, and that production of some leaf-eating taxa (i.e, shredders) would suffer from this quantitative decline in their resource base. Finally, we predicted that differences among taxa in response to changes in resource quantity and quality would be dependent upon larval lifespan, a dominant life history characteristic.

Study sites

This study was conducted in two adjacent headwater streams at the Coweeta Hydrologic Laboratory, Macon County, North Carolina, USA. Coweeta is a large (2185 ha) heavily forested basin located in the Blue Ridge physiographic province of the southern Appalachian Mountains (see Swank and Crossley 1988). Forest vegetation is dominated by mixed hardwoods (primarily oak, maple, and poplar) and a dense understory of *Rhododendron maximum* which shades the streams throughout the year. Headwater streams at Coweeta are extremely heterotrophic, and allochtonous inputs of detritus provide >90% of the energy base for microbial and invertebrate production (Wallace et al. 1997b, Hall et al. 2000). In-stream primary production is very low (ca. 4 – 8 g C m^{-2} y^{-1}) and constitutes < 1% of the total carbon entering these streams (Wallace et al. 1997a). Both streams are fishless, but contain vertebrate larval salamanders.

The two streams used for this study are first order, and drain the small catchments (C) 53 and 54 (hereafter C53 and C54). These streams have similar physical and chemical characteristics (Table 4.1), but differ (since July 2000) in their concentrations of dissolved N and P as a result of our experimental nutrient enrichment of C54 (Table 4.1). Natural concentrations of inorganic N and P in these streams are very low (Table 4.1). The dominant stream substrate is a heterogeneous mixture of cobble, pebble, gravel, sand, and silt (hereafter 'mixed substrate')

(Table 4.1). This habitat is disrupted periodically by moss-covered bedrock outcrops (hereafter 'bedrock outcrops') (Table 4.1). The bedrock outcrops comprise a smaller proportion of the benthos than the mixed substrate habitat (Table 4.1). Both streams have rough topography and abundant woody debris, which facilitate high retention and processing of organic matter (Wallace et al. 1995).

Discharge was monitored continuously during the study with FW-1 stage and ISCO 3230 Bubble Flow Meter (ISCO Inc., Lincoln, Nebraska, USA) recorders attached to 30 cm H-flumes at the base of each catchment. Water temperature (°C) was monitored every 30 minutes in both streams with Optic StowAway temperature probes (Onset Computer Corp., Pocasset, MA, USA K. Suberkropp, University of Alabama, unpublished data).

Precipitation during the study was considerably lower than the long term average of 182 cm y^{-1} (based on 64-y record from Coweeta Hydrologic Laboratory). When expressed as a percentage of the long-term average, year 1 (September 1998 – August 1999) was 83%, year 2 (September 1999 – August 2000) was 66%, year 3 (September 2000 – August 2001) was 74%, and year 4 (September 2001 – August 2002) was 78%.

Methods

Experimental Enrichment

Our study consisted of a ~2 year pretreatment period (September 1998 – June 2000) and a ~2 year experimental enrichment (July 2000 – August 2002) of C54. Starting in July 2000, nitrogen (NH_4NO_3) and phosphorus (K_2HPO_4 and KH_2PO_4) were dripped continuously into the treatment stream to increase concentrations of dissolved inorganic N and P to ≈ 6-15 X background levels (Table 4.1). Nutrient solution was added along 140 m of the treatment stream with a solar powered metered-dose pump (LMI, Acton, Massachusetts, USA) connected to a streamwater-fed plastic pipe laid along the streambed. The plastic pipe was fitted with garden irrigation valves roughly every 10 m to evenly distribute nutrients along the length of the stream. Stream water nutrient concentrations were held relatively constant across a range of discharge by connecting the pump to an Isco data logger (Isco Inc., Lincoln, NE, USA) located at the base of the stream; the pump was engaged every time a known volume of water (generally 50-100 liters) passed through the downstream weir. Concentrations of (NO_3 + NO_2)-N, NH_4-N, and soluble reactive phosphorus (SRP) were measured biweekly at the flume of each study stream (APHA 1998). During the experimental enrichment, nutrient concentrations were also measured at 5 locations along the length of C54 to confirm that nutrients were evenly distributed along the stream. Nutrient concentrations in the treatment stream during enrichment (Table 4.1) were well within the range of natural concentrations of streams in the region (Scott et al. 2002), and thus provided a realistic assessment of moderate enrichment effects.

This ecosystem-level experiment was not replicated, employing a paired-catchment design (e.g., Likens et al. 1969) with one treatment stream and one reference stream. This approach has been lauded for its realism and scale (e.g., Carpenter et al. 1995, Schindler 1998, Oksanen 2001), as well as criticized for its lack of strict replication (Hurlbert 1984, 2004). The limitations of our design are recognized, but we felt that the scale of manipulation was essential for predicting realistic effects.

Benthic sampling

We quantified invertebrate secondary production, as well as abundance and biomass, because production estimates capture population-level dynamics of growth, mortality, and fecundity (Benke 1993), as well as provide a measurement of energetic flow through consumers. Quantitative benthic sampling was conducted monthly in each stream for 4 years between September 1998 and August 2002. Each month, samples were taken from mixed substrate and bedrock outcrop habitats. Mixed substrate samples were collected at 4 random locations in each stream with a stove pipe core sampler (400 cm^2). Cores were firmly placed in the streambed, and all material was removed to a depth of 15 cm by hand or cup and placed in a large plastic jar. Following sample removal, five water depths were taken inside the core to estimate the total volume of material remaining in the water. This water was then stirred, subsampled with a plastic cup, and brought to the laboratory to estimate the amount of fine particulate organic matter remaining in the sampler (OMW). Bedrock samples were taken at 3 randomly determined locations in each stream by brushing and scraping moss and associated particles from a known area (15 cm x 15 cm) with a dish scrubber and knife into a plastic bag and hess net (250 μm mesh size) pressed flush against the bedrock.

Benthic samples were brought to the laboratory, refrigerated, and processed within 24 hours. Samples were rinsed with tap water onto nested metal sieves (pore sizes 1 mm and 250 μm), and material retained on each sieve was elutriated to separate organic from inorganic material. All organic material, including invertebrates and salamanders, was then preserved separately for each size fraction (> 1mm and < 1 > 250 μm) in formalin (6-8%). Animals were removed from the > 1 mm fraction by hand picking under a dissecting microscope at 15x magnification. Organic material in the smaller size fraction (<1 mm>250 μm) was subsampled (1/8 to 1/64 of whole samples) using a sample splitter (Waters 1969), and animals were removed from subsamples with a dissecting microscope at 15x magnification.

The amount of coarse (>1 mm, CBOM), fine (<1 mm>250 μm, FBOM), and very-fine (<250 μm, VFBOM) benthic organic matter was quantified in each sample (also see Lugthart and Wallace 1992). CBOM was separated into leaf, wood, seed, moss, and miscellaneous categories, then dried (50°C), weighed to the nearest 0.01 g, ashed (at 500°C), and reweighed to quantify ash-free dry mass (AFDM). FBOM and VFBOM were quantified during sample processing by placing material that was retained (FBOM) or had passed through (VFBOM) the 250 μm sieve into a graduated bucket with a known volume of water. This material was stirred, sub-sampled (< 1%) with a graduated syringe, and filtered onto pre-ashed, pre-weighed glass fiber filters (Gelman A/E). Filters were dried, weighed to the nearest 0.01 mg, ashed, and reweighed to quantify AFDM. OMW was subsampled with a syringe, filtered, dried, weighed, ashed, and re-weighed as for FBOM. In this study, total FBOM refers to the summation of FBOM, VFBOM, and OMW (i.e., all organic particles <1 mm).

Invertebrates and larval salamanders were counted and identified. All insects, except Chironomidae, were identified to the genus or species level. Larval chironomids were identified as either Tanypodinae or non-Tanypodinae. Most non-insect taxa were identified to the ordinal level or higher. Taxa were assigned to functional feeding groups according to Merritt and Cummins (1996) and our knowledge of the local fauna. In this study, all functional feeding group designations (i.e., shredder, collector-gatherer [=gatherers], scraper, collector-filterer [=filterers], and predator) follow Wallace et al. (1999). The body length of each individual was measured to the nearest mm with a dissecting microscope at 12X magnification and a graduated stage. Total snout-vent length and carapace length were measured on salamanders and crayfish, respectively. For most taxa, biomass (AFDM) was obtained using previously established length-weight regressions for invertebrates at Coweeta, or, for a few taxa, nearby North Carolina streams (Benke et al. 1999). For Copepoda, Hydracarina, Nematoda, and Cladocera, biomass was obtained from mean mass of >50 individuals in subsamples of representative size classes (J. B. Wallace, unpublished data).

Secondary production

Annual production was estimated for most taxa using the size-frequency method (Hamilton 1969) corrected for the cohort production interval (CPI, Benke 1979). Invertebrate CPIs were the same as those used by Wallace et al. (1999), and were estimated from length-frequency histograms constructed from monthly benthic samples taken in the study streams (Lugthart and Wallace 1992) or another stream at Coweeta (Huryn and Wallace 1987a, b). Larval development times for salamanders were obtained from Lugthart (1991).

Annual production of non-Tanypodinae chironomids and *Tallaperla* spp. (Plecoptera: Peltoperlidae) was estimated using the community-level instantaneous growth method (Huryn and Wallace 1986, Huryn 1990). Size-specific growth rates were measured *in situ* in both streams throughout the study and applied to monthly biomass values (Cross Chapter 3). Production of Copepoda was estimated using an empirically-derived production/biomass (P/B) value of 18 (O'Doherty 1985). For the remaining taxa, the product of standing stock biomass and annual P/B ratio was used to estimate production. P/B values of 5 and 10 were used for several taxa considered to by univoltine and bivoltine, respectively (Waters 1977, Lugthart and Wallace 1992). With the exception of oligochaetes, these remaining taxa comprised a small proportion of total community biomass. A conservative P/B value of 5 was used for oligochaetes (but see Brinkhurst and Cook 1980).

Annual production was estimated separately for each taxon in the mixed substrate and bedrock outcrop habitats. Habitat-weighted production values for the entire stream were calculated according to the relative proportion of each habitat in each stream (e.g., Huryn and Wallace 1987a, Lugthart and Wallace 1992, Table 4.1).

Statistical analyses

We used randomized intervention analysis (RIA, Carpenter et al. 1989, also see Stewart-Oaten 2003, Murtaugh 2003) to test for effects of nutrient enrichment on benthic organic matter and abundance and biomass of invertebrate taxa and functional feeding groups. RIA uses paired before- and after-treatment time-series data from a manipulated and a reference system to detect changes caused by the manipulation. In this study, RIA was used to test the null hypothesis of no change in the variable of interest in the treatment stream relative to the reference stream following initiation of nutrient enrichment. To increase the statistical power of RIAs, we included data from a previous unmanipulated year (October 1984 – September 1985) in both streams (see Lugthart and Wallace 1992). These previous data are presented throughout the study as the first year of pretreatment (Pre-1). Data were log (x+1) transformed to meet assumptions of normality and homoscedasticity.

Relationships between organic matter standing crop and invertebrate production were analyzed with linear regression analysis. Regression was also used to analyze relationships between primary consumer or total invertebrate production and predator production. For these relationships the y-intercept was set equal to zero (i.e., no prey production = no predator production). Although predators are included in both axes of the relationship between total and predator production, this analysis is justified based on the prevalence of intraguild predation in these streams (Wallace et al. 1999, Hall et al. 2000, W. F. Cross, personal observation).

Long-term relationships between organic matter standing crop and invertebrate production were also examined using data from the present study, as well as previously published estimates from the study streams (Lughtart and Wallace 1992, Wallace et al. 1999) and another headwater stream at Coweeta (C55, Wallace et al. 1999). These long-term data include 7 years of leaf litter reduction in C55 (Wallace et al. 1999).

We calculated percent difference in production for each taxon between pre- and post-enrichment periods in the enriched stream. To test for significant differences between the response of short lived (<365 days) and long-lived (>365 days) taxa we used a non-parametric Wilcoxon test.

Results

Nutrient enrichment

The experimental enrichment successfully elevated concentrations of NO_3-N, NO_2-N, NH_4-N, and soluble reactive phosphorus (SRP) to 6-15 X background levels in the treatment stream (Table 4.1). During enrichment, the molar N:P ratio of streamwater in the treated stream was 19.7. This ratio was considerably higher than that of the stock nutrient solution added (11.4), suggesting preferential uptake of P in the enriched stream.

Benthic organic matter

Nutrient enrichment had a significant negative effect on standing crop of leaf litter detritus (RIA, P = 0.002, Figure 4.1, Table 4.2) in the mixed substrate habitat. Although both streams attained similar maxima during annual leaf fall, benthic leaf litter disappeared more rapidly in the treatment stream during the two years of enrichment (Figure 4.1). Between-stream differences in leaf litter standing crop were most pronounced during the final year of the study in which the treatment stream contained on average only 43% of leaf litter in the reference stream (101 g AFDM m^{-2} vs. 234 g AFDM m^{-2}). Although nutrient enrichment appeared to have a significant positive influence on total FBOM standing crop (RIA, $P = 0.001$, Figure 4.1, Table 4.1), closer examination of the data revealed that changes in the treatment stream occurred more than a year prior to the enrichment (Figure 4.1). Total CBOM standing crop in the mixed substrate habitat was not affected by nutrient enrichment (RIA, P values > 0.05, Figure 4.1, Table 4.2).

Standing crop of benthic organic matter on bedrock outcrop habitat was not influenced by nutrient enrichment (RIA, P values > 0.05, Table 4.2). All categories of bedrock organic matter (ie., total FBOM, total CBOM, and moss) were higher before enrichment than during enrichment in both streams (Table 4.2).

Invertebrates

Nutrient enrichment had a significant positive effect on total invertebrate abundance and biomass in the mixed substrate habitat (RIA, P values <0.000001, Figure 4.2). During enrichment, abundance and biomass in the treatment stream were 104% and 71% higher than average pretreatment values, respectively. In contrast, invertebrate abundance and biomass in the reference stream decreased during the 2 yrs of enrichment in comparison to pretreatment values (abundance: -42%; biomass: -15%, Figure 4.2). Total invertebrate secondary production in the mixed substrate habitat was similar in both streams during the 3 years of pretreatment

(Figure 4.2). During enrichment, production in the treatment stream was ca. 117% higher than pretreatment values and ca. 135% higher than production in the reference stream (Figure 4.2). Total secondary production in the reference stream was slightly lower (-19%) during the treatment in comparison to pretreatment years.

In sharp contrast to mixed substrate habitat, nutrient enrichment had no significant effect on total invertebrate abundance and biomass on bedrock outcrops (RIA, P values > 0.05, Figure 4.2). Excepting the first year of pretreatment data, total abundance was similar between streams before and after enrichment (Figure 4.2). Total biomass was consistently higher in the treatment stream than in the reference stream before (25%) and during (43%) treatment, with the largest difference (71%) occurring during the first year of enrichment (Figure 4.2). Annual secondary production was also consistently higher in the treatment stream than in the reference stream before (23%) and during (65%) enrichment, with the largest difference (102%) occurring during the first year of enrichment (Figure 4.2).

Functional feeding groups

In mixed substrate habitat, abundance and biomass of all functional feeding groups except scrapers were positively influenced by nutrient enrichment (RIA, all P values < 0.05, Figure 4.3, Table 4.3). In the treatment stream, percent increase in abundance between pre- and post-enrichment periods was greatest for filterers (170%), followed by shredders (143%), gatherers (111%), and predators (79%) (Table 4.3); percent increase in biomass was greatest for filterers (140%), and followed by gatherers (98%), predators (56%), and shredders (50%). Annual secondary production of all functional feeding groups except scrapers increased during the enrichment period (Table 4.5). Filterers exhibited the largest increase in production (192%), followed by shredders (178%), gatherers (108%), and predators (60%). For many functional feeding groups, concomitant decreases in abundance and biomass occurred in the reference stream during the period of nutrient enrichment (Figure 4.3, Table 4.5).

Although invertebrates on bedrock outcrops did not respond to enrichment as a group (Table 4.4), some functional feeding groups showed significant changes following nutrient enrichment (Figure 4.4, Tables 4.4 and 4.5). Shredder abundance and biomass in the treatment stream decreased on bedrock following nutrient enrichment (abundance: -51%; biomass: -60%, Table 4.4). Although predator abundance on bedrock significantly changed following enrichment (RIA, P = 0.004), pre- and post-treatment differences only occurred in the reference stream (-49%, Figure 4.4). Biomass of gatherers increased (82%) in the treatment stream with nutrient enrichment (Figure 4.4, Table 4.4). Secondary production of scrapers and gatherers in the treatment stream increased by 74% and 51% respectively on bedrock outcrops following enrichment (Table 4.5). Shredder production on bedrock outcrops decreased (-50%), while no change was observed for filterers or predators (Table 4.5).

Taxon-specific responses

Twenty five out of 65 taxa (38%) in the mixed substrate habitat exhibited a significant change in abundance, biomass, or both in the treatment stream relative to the reference stream (Table 4.6). These taxa comprised ca. 75% of total secondary production of these streams. All changes in the mixed substrate of the treatment stream were positive, except for the crayfish, *Cambarus bartoni*; however, this result should be viewed with caution because crayfish numbers were extremely low throughout the study (Appendix B). Individual shredder taxa that showed increased abundance or biomass with enrichment included plecopterans (*Leuctra* spp.), trichopterans (*Lepidostoma* spp., *Pycnopsyche* spp.), and dipterans (*Molophilus* sp.) (Table 4.6). Both *Lepidostoma* spp. and *Pycnopsyche* spp. increased in biomass, but not abundance, suggesting either increased body size of individuals or increased survivorship. Gatherer taxa that exhibited a significant increase in abundance or biomass included ephemeropterans (*Serratella* sp., *Stenonema* sp), plecopterans (*Amphinemur*a sp.), dipterans (non-tanypod chironomids, *Leptotarsus* sp.), Copepoda, Nematoda, and Oligochaeta (Table 4.6). Filterer taxa that increased with enrichment included trichopterans (*Diplectrona modesta*), dipterans (Simuliidae), and molluscs (Sphaeridae).

Nutrient enrichment also led to increased abundance or biomass of 9 out of 19 predator taxa in the mixed substrate habitat (Table 4.6). These secondary consumers included odonates (*Cordulegaster* sp.), plecopterans (*Sweltsa* sp., *Beloneuria* sp., *Isoperla* spp.), dipterans (Ceratopogonidae, *Dicranota* spp., Tanypodinae, Empididae), and aquatic mites (Acari). There was no change in abundance or biomass of larval salamanders following enrichment, although there was evidence that individual growth rates of salamanders increased in the treatment stream during the first year of enrichment (Johnson 2002). In the mixed substrate habitat, 22 of 25 taxa (88%) that responded to enrichment have larval development times less than, or equal to, 1 year.

Of the 10 taxa that contributed most to total invertebrate biomass on mixed substrate habitat, the largest response was seen for the shredder, *Pycnopsyche* sp. (Figure 4.5). Smaller increases were seen among the gatherers Ceratopogonidae and Chironomidae (Figure 4.5).

On bedrock outcrops, 10 out of 35 taxa, representing ca. 30% of total bedrock production, exhibited significant changes in abundance or biomass following nutrient enrichment (Table 4.6). Three of these taxa, of which all were shredders (*Leuctra* spp, *Tallaperla* spp, *Tipula* spp.), actually declined on bedrock outcrops in the treatment stream following enrichment (Table 4.6,

Appendix B). Significant change in biomass of *Pycnopsyche* spp., was due to a decrease in the reference stream as opposed to an increase in the treatment stream (Appendix B). In contrast, 3 gatherer taxa (*Serratella* sp., *Amphinemura* sp., Nematoda), 1 filterer (Simuliidae), and 3 predators (*Pseudogoera singularis*, *Dicranota* spp., Acari) on bedrock outcrops increased in abundance or biomass in the treatment stream following enrichment (Table 4.6). All 7 taxa which responded positively to nutrient enrichment on bedrock outcrops have larval development times less than, or equal to, 1 year.

Of the 10 dominant taxa in terms of biomass on bedrock outcrops, the most notable positive response was seen for the mayfly gatherer, *Serratella* sp. (Figure 4.5).

Taxon-specific percent increase in production in the treatment stream was related to length of larval lifespan (Figure 4.6). Although there was no significant linear relationship between these variables, the response of taxa with lifespans longer than 1 year was significantly lower than that of taxa with lifespans less than, or equal to, 1 year ($P<0.001$, Wilcoxon test).

Predator-prey relationships

Annual habitat-weighted invertebrate production data from both study streams ($n = 10$) were combined to examine relationships between predator and prey production. Both primary consumer production and total invertebrate production explained a significant amount (61% and 27%, respectively, P values < 0.001) of variation in predator production (Figure 4.7). Slopes of these relationships (0.27 and 0.37) were close to known bioenergetic efficiencies (production/ingestion ≈ 0.35) of invertebrate predators (Slansky and Scriber 1982, Wallace et al. 1999).

Organic matter – secondary production relationships

In the mixed substrate habitat, relationships between annual mean organic matter standing crop (i.e., leaf litter and total benthic organic matter) and annual secondary production or shredder production were not significant when considering all 10 stream-years of data from this study ($r^2 < 0.1$, all P values > 0.05). Moreover, when values from the treatment stream during enrichment were excluded from the analysis, relationships were still non-significant ($r^2 < 0.1$, all P values > 0.05).

We examined these data in the context of long-term data from headwater streams at Coweeta (n = 29 stream years). For relationships between annual mean leaf litter standing crop and total invertebrate production or shredder production, all non-enriched years of the present study (C53: 1998 – 2002, C54: 1998 – 2000, n = 6) were in close agreement with previous years (Figure 4.8a and b). In sharp contrast, total secondary production during the two years of enrichment was 100% (yr 1) and 282% (yr 2) higher than predicted production based on the long-term relationship (Figure 4.8a). In addition, shredder production during the 2 years of enrichment was even further from predicted values at 125% and 493% of predicted production (Figure 4.8b). These two years of enrichment comprised the largest residuals of these relationships, were identified as outliers, and were thus not included in regression equations (Figure 4.8). Excluding enriched years, leaf litter standing crop explained 64% and 70% of the variation in total and shredder secondary production, respectively (Figure 4.8a and b, P < 0.0001). Total benthic organic matter also explained a large amount (54%) of the variation in total secondary production (Figure 4.8c, P < 0.0001). Although the two years of enrichment were among the largest residuals, they were not identified as outliers, and were included in the regression. However, the two years of enrichment were 90% and 118% higher than the final year of pretreatment in the treated stream which had similarly high levels of total benthic organic matter (Figure 4.8c).

Discussion

To our knowledge, this study is the first to examine the effects of long-term nutrient enrichment on whole-community macroconsumer production in a detritus-based ecosystem. We have shown that enrichment can lead to increased production of both primary and secondary invertebrate consumers despite major reductions in the quantity of leaf litter, a dominant allochthonous food resource. As predicted by theory, release of bottom-up nutrient limitation in this donor-controlled food web led to increased biomass of consumers at both intermediate and top trophic positions (sensu Polis and Strong 1996).

Nutrient enrichment led to significant increases in microbial biomass and production (Gulis and Suberkropp 2003, K. Suberkropp, University of Alabama, unpublished data, Greenwood 2004), detrital nutrient content (Cross et al. 2003, Gulis and Suberkropp 2003), and leaf decomposition rates (Gulis and Suberkropp 2003, Greenwood 2004). For invertebrate consumers, the positive effects of enrichment on detrital quality (i.e., increased microbial activity and nutrient content) appeared to outweigh the negative effects of enrichment on carbon loss (i.e., increased decomposition and respiration). Despite significant reductions in leaf litter at the base of the food web (ca. 45% lower in the enriched stream), invertebrate production in the dominant stream habitat was ca. 135% higher in the treatment stream than in the reference stream. Moreover, production during the two years of enrichment were statistical outliers when compared to long-term production data from Coweeta (Figure 4.8). For invertebrates that consume leaf litter (i.e., shredders), these results suggest that either a) assimilation efficiencies and/or survivorship were much higher on nutrient-rich leaf litter, or b) shredders switched diets to include other high quality food items such as wood biofilm (e.g., Tank and Webster 1998, Eggert 2003). The former explanation is the most plausible based on gut content analyses (Cross Chapter 5) in which no major dietary changes among shredders were observed. This finding, however, does not discount the importance of leaf litter quantity to consumer production in the treatment stream. Even under enriched conditions, during the final 4 months of the study shredder production appeared to be limited by low standing crop of leaf litter; six out of 8 dominant shredder taxa had lower biomass in the enriched stream than the reference stream during these months. Therefore, despite overall elevated production of shredders during enrichment, accelerated decomposition rates may lead to resource ‘crunches’ (sensu Wiens 1977) during spring and summer months in which consumer production may be limited by the availability of leaf litter.

Increased production of other detritivorous taxa, such as gatherers and filterers, was likely due to higher nutrient content of FBOM (Cross et al. 2003) during enrichment. Although FBOM quantity was similar among the final 3 years of the study, invertebrate production during the 2 enriched years was ca. 100% higher than the year preceding enrichment, suggesting a significant effect of FBOM quality. Indeed, individual growth rates of a dominant gatherer, non-tanypod chironomids, were significantly higher in the treatment steam than the reference stream during the enrichment (Cross Chapter 3). Other studies have also demonstrated a positive influence of detrital nutrient content on detritivore growth or physiological condition (e.g., Iversen 1974, Tenore 1977, Pearson and Connolly 2000, Vos et al. 2000). FBOM-feeding taxa are potentially less vulnerable than shredders to nutrient-induced carbon loss because FBOM is much more abundant in Coweeta streams than leaf litter (Table 4.2). Moreover, FBOM storage in stream sediments is sufficiently high that quantitative reductions of FBOM, even in the absence of leaf litter inputs, has been shown to take many years (Wallace et al. 1999, J. B. Wallace, unpublished).

Nutrient enrichment also had a positive effect on production of invertebrate predators, whose trophic position is generally multiple links removed from the ultimate source of enrichment; however, the magnitude of this effect was not as great as that observed for detritivores. This pattern is consistent with McQueen et al.'s (1986) bottom up:top down model, which predicts that bottom-up control should be strong at the base of the food web and weaker at the top. Others have found support for this model in lake ecosystems (e.g., McQueen et al. 1989, Brett and Goldman 1997). Predator production also appeared to closely track prey production over the course of the study in both streams (Figure 4.7), and the slopes of these relationships were similar to gross production efficiencies reported for invertebrate predators (production/ingestion ~ 0.35, Slansky and Scriber 1982). Others have observed increased abundance and biomass of soil invertebrate predators in response to an enhanced resource base (Chen and Wise 1999, Scheu and Schaefer 1998). In addition, Wallace et al. (1999) demonstrated that experimental reduction of allochthonous detritus (i.e., leaf litter) drastically reduced production of stream invertebrate predators and growth rates of vertebrate salamanders (Johnson 2002) in a nearby Coweeta stream. Together, these studies highlight the prevalence of resource limitation among consumers at all trophic positions in detritus-based food webs (sensu Polis and Strong 1996). Interestingly, the contribution of predators to total secondary production declined during the final year of study in the treatment stream (Figure 4.7, final year was well below predicted line). This observation suggests a potential saturation of predator functional response (Holling 1959), which is supported by reduced estimates of the proportion of available prey consumed by predators during the final year (Cross Chapter 5).

The response of invertebrates to nutrient enrichment was strongly habitat-dependent. In contrast to mixed substrate habitat, enrichment had a relatively small effect on bedrock outcrop invertebrate production. Moreover, abundance and biomass of few individual taxa associated with bedrock increased during enrichment. One potential explanation for this difference between habitats is that space, as opposed to organic matter quality or quantity, limits invertebrate production on bedrock outcrops. Many filtering taxa such as hydropsychid caddisflies or black flies are known to compete for space on stable substrata (e.g., Hemphill 1991, Hiltner and Hershey 1992). Such competition may be particularly strong when drifting food abundance is low, as in streams at Coweeta. Interestingly, Hall et al. (2000) showed that cannabalism among filter-feeding caddisflies was common during periods of larval recruitment, and they suggested that space-limitation may have accentuated such intraguild predation. Scrapers, which maintain very low densities in Coweeta headwater streams, also showed no significant response to enrichment despite increased periphyton biomass (Greenwood 2004) and nutrient content (Cross et al. 2003). Close examination of the data revealed that mean scraper abundance and biomass was actually higher in the treatment stream during periods of high periphyton availability (March - May), but this effect was far too variable to be detected as significant, and a similar pattern was seen during the first non-enriched year of the study (Table 4.5). Interestingly, 3 out of 5 shredder taxa on bedrock outcrops actually decreased with enrichment. Generally, shredders occupying bedrock habitat, such as *Lepidostoma* spp. or *Tallaperla* spp., are dominated by early instars. We can only speculate that this effect was due to the increased movement of early instars away from bedrock because of higher nutritional quality of organic matter in the mixed substrate habitat.

There are multiple factors to consider in understanding mechanisms leading to increased production of invertebrates in response to enrichment. Positive effects on production could be driven by changes in individual growth rates, survivorship, fecundity, or a combination of these factors (Benke 1984, 1993, Huryn and Wallace 2000). Invertebrate growth rates at Coweeta are difficult to assess from benthic samples because a large number of taxa have overlapping generations or asynchronous development. However, visual examination of size-frequency histograms (data not shown), as well as positive results from field-based growth studies (Cross Chapter 3) indicate that increased production was potentially due to both increased survivorship and higher individual growth rates. Although fecundity was not directly quantified in this study, larger terminal size of many taxa in the enriched stream likely led to increased egg production per female (e.g., Vannote and Sweeney 1980). Further detailed observation of life history and physiology of individual taxa is necessary to tease out the relative importance of different mechanisms leading to increased production.

We found that length of larval lifespan, a dominant life history trait, was related to the magnitude of invertebrate response to enrichment in the treatment stream. In general, there was extreme variability among taxa with lifespans less than one year, but a consistently weak response of longer-lived taxa (Figure 4.6). This pattern generally supports the idea that species with rapid turnover rates are more likely to respond to enrichment because they can complete multiple generations and increase rates of fecundity. Longer lived taxa are faced with multiple constraints on production. For example, if many generations are necessary to elicit an increase in production, response of some taxa may take much longer than the timeframe of this study.

However, species with different life history strategies may have very different elemental composition or metabolism which can lead to very distinct nutritional requirements (e.g., Elser et al. 1996). For example, long-lived, slow growing taxa may not be nutrient limited because of evolutionarily-fixed slow rates of metabolism or low nutrient requirements. Interestingly, we found no effect of enrichment on individual growth rates of a long-lived shredder, *Tallaperla* spp. (Cross Chapter 3), which suggests a longer time of enrichment may have little effect on this species. *Tallaperla* spp. also had a low amount of body P, suggesting relatively low requirements for this element (e.g., Elser et al. 2000, Cross et al. 2003). For some long-lived taxa – particularly those that consume large leaf particles – carbon availability may have primacy in limiting secondary production. Further research dealing with life history evolution and nutritional requirements will be fruitful for understanding mechanisms behind community change or response to elevated nutrient concentrations.

During the two years of nutrient enrichment we saw greatly increased invertebrate production. However, questions remain concerning the long-term sustainability of this boost in secondary production. Multiple lines of evidence suggest that continued enrichment would lead to decreased secondary production via carbon limitation. Benthic samples (this study), leaf litter surveys (K. Suberkropp, University of Alabama, unpublished data), and litter bag decomposition studies (Gulis and Suberkropp 2003, Greenwood 2004) all demonstrated elevated breakdown and metabolism of carbon during the enrichment. Invertebrates undoubtedly contributed these losses via increased feeding activity. In addition, biweekly samples of organic matter export (A. D. Rosemond, unpublished data) indicated a significant increase in FBOM export from the treatment stream during enrichment. Even though FBOM standing crop in benthic samples did not decrease during the study, reductions may take much longer than the time span of this study.

This is especially true considering the large inputs of leaf litter which occur each autumn. We expect that continued nutrient enrichment would ultimately lead to an alternate, albeit lower, equilibrium state of total organic matter standing crop. Invertebrate production would likely decrease to levels comparable to the reference stream considering the new combination of lower organic matter quantity and higher organic matter quality. Interestingly, despite greater overall production during the 2nd year of enrichment, ca. 50% of invertebrate species had lower production during the final year of enrichment compared to the first year of enrichment. This result coincides with lower organic matter standing crop during the second year.

In summary, our study was the first to show that nutrient enrichment can stimulate whole-community production through detrital-microbial pathways. We demonstrated strong bottom-up effects of nutrient enrichment in a detritus-based stream ecosystem which propagated from microbes to top invertebrate predators. Although net carbon loss occurred in the treatment stream (Gulis and Suberkrop 2003, Greenwood 2004, A. D. Rosemond, and K. Suberkropp, unpublished data), total secondary production during the 2 years of enrichment was still much higher than that predicted based on long-term relationships. However, anecdotal evidence suggested that this loss may have lessened the effects of enrichment on some leaf-eating consumers. In combination with other studies examining resource limitation of detritus-based systems, our study reinforces the current view that overall productivity of both primary and secondary consumers is resource limited in these donor-controlled food webs.

Acknowledgements

We thank the many hardworking undergraduates who removed invertebrates from benthic samples - without which this work would not have been possible. In particular, Catherine Evans, Lizzy Melda, Andrew Paloucci, and Gavin Veitch, dedicated many hours and maintained remarkably good attitudes. In addition, Lizzy Melda provided valuable laboratory assistance. We also thank Jennifer Greenwood, Sue Eggert, and Elizabeth Reese for comments on an earlier version. Funding was provided by NSF (DEB – 9806610) to Amy D. Rosemond, J. Bruce Wallace, Keller Suberkropp, and Pat J. Mulholland.

Literature Cited

Aerts, R., H. De Caluwe, and B. Beltman. 2003. Plant community mediated vs. nutritional controls on litter decomposition rates in grasslands. Ecology 84:3198-3208.

American Public Health Association. 1998. Standard methods for the examination of water and wastewater, 20th edition. American Public Health Association, Washington, DC, USA.

Arsuffi, T. L. and K. Suberkropp. 1985. Selective feeding by stream caddisfly (Trichoptera) detritivores on leaves with fungal-colonized patches. Oikos 45:50-58.

Benke, A. C. 1979. A modification of the Hynes method for estimating secondary production with particular significance for multivoltine populations. Limnology and Oceanography 24:168-171.

Benke, A. C. 1984. Secondary production of aquatic insects. *In*: Resh, V. H. and D. M. Rosenberg (eds). The Ecology of Aquatic Insects. Praeger, pp. 289-322.

Benke, A. C. 1993. Concepts and patterns of invertebrate production in running waters. Verh. Internat. Verein. Limnol. 25:1-24.

Benke A. C., A. D. Huryn, L. A. Smock and J. B. Wallace. 1999. Length-mass relationships for freshwater macroinvertebrates in North America with particular reference to the southeastern United States. Journal of the North American Benthological Society

18:308-343.

Bennett, E. M., S. R. Carpenter, and N. F. Caraco. 2001. Human impact on erodable phosphorus and eutrophication: a global perspective. BioScience 51:227-234.

Brett, M. T. and C. R. Goldman. 1997. Consumer versus resource control in freshwater pelagic food webs. Science 275:384-386.

Brinkhurst, R. O. and D. G. Cook, editors. 1980. Aquatic Oligochaete Biology. Plenum Press, New York, USA.

Caraco, N. F. 1993. Disturbance of the phosphorus cycle: a case of indirect effects of human activity. Trends in Ecology and Evolution 8:51-54.

Carpenter, S. R., S. W. Chisholm, C. J. Krebs, D. W. Schindler, and R. F. Wright. 1995. Ecosystem experiments. Science 269:324-327.

Carpenter S. R., T. M. Frost, D. Heisey, and T. K. Kratz. 1989. Randomized intervention analysis and the interpretation of whole-ecosystem experiments. Ecology 70:1142-1152.

Carpenter, S. R., N. F. Caraco, D. L. Correll, R. W. Howarth, A. N. Sharpley, and V. H. Smith. 1998. Nonpoint pollution of surface waters with phosphorus and nitrogen. Ecological Applications 8:559-568.

Chen, B. and D. H. Wise. 1999. Bottom-up limitation of predaceous arthropods in a detritus-based terrestrial food web. Ecology 80:761-772.

Cross, W. F., J. P. Benstead, A. D. Rosemond , and J. B. Wallace. 2003. Consumer-resource stoichiometry in detritus-based streams. Ecology Letters 6:721-732.

Eggert, S. L. 2003. Resource use by detritivorous macroinvertebrates in southern Appalachian headwater streams. Dissertation, University of Georgia, Athens, GA, USA.

Elser J. J., D. R. Dobberfuhl, N. A. MacKay and J. H. Schampel. 1996. Organism size, life history, and N:P stoichiometry. BioScience 46:674-684.

Elser J. J., R. W. Sterner, E. Gorokhova, W. F. Fagan, T. A. Markow, J. B. Cotner, J. F. Harrison, S. E. Hobbie, G. M. Odell and L. J. Weider. 2000. Biological stoichiometry from genes to ecosystems. Ecology Letters 3:540-550.

Elwood, J. W., J. D. Newbold, A. F. Trimble, and R. W. Stark. 1981. The limiting role of phosphorus in a woodland stream ecosystem – effects of P-enrichment on leaf decomposition and primary producers. Ecology 62:146-158.

Fisher, S. G. and G. E. Likens. 1973. Energy flow in Bear Brook, New Hampshire: integrative approach to stream ecosystem metabolism. Ecological Monographs 43:421-439.

Gallardo, A. and W. H. Schlesinger. 1994. Factors limiting microbial biomass in the mineral soil and forest floor of a warm-temperate forest. Soil Biology and Biochemistry 26:1409-1415.

Galloway, J. N., J. D. Aber, J. W. Erisman, S. P. Seitzinger, R. W. Howarth, E. B. Cowling, and B. J. Cosby. 2003. The nitrogen cascade. BioScience 53:341-356.

Greenwood, J. 2004. The response of heterotrophic and autotrophic resources to a long-term nutrient enrichment in an Appalachian headwater stream ecosystem. Dissertation. University of Georgia, Athens, GA, USA.

Gulis, V. and K. Suberkropp. 2003. Leaf litter decomposition and microbial activity in nutrient-enriched and unaltered reaches of a headwater stream. Freshwater Biology 48:123-134.

Hall, R. O. Jr., J. B. Wallace, and S. L. Eggert. 2000. Organic matter flow in stream food webs with reduced detrital resource base. Ecology 81:3445-3463.

Hamilton, A. L. 1969. On estimating annual production. Limnology and Oceanography 14: 771-782.

Harvey, C. J., B. J. Peterson, W. B. Bowden, A. E. Hershey, M. C. Miller, L. A. Deegan, and J. C. Finlay. Biological responses to fertilization of Oksrukuyik Creek, a tundra stream. Journal of the North American Benthological Society 17:190-209.

Hemphill, N. 1991. Disturbance and variation in competition between 2 stream insects. Ecology 72:864-872.

Hiltner, A. L. and A. E. Hershey. 1992. Black fly (Diptera, Simuliidae) response to phosphorus enrichment of an arctic tundra stream. Hydrobiologia 240:259-265.

Holling, C. S. 1959. The components of predation as revealed by a study of small mammal predation of the European pine sawfly. Canadian Entomologist 91: 293-320.

Howarth, R. W. and S. G. Fisher. 1976. Carbon, nitrogen, and phosphorus dynamics during leaf decay in nutrient-enriched stream micro-ecosystems. Freshwater Biology 6:221-228.

Hurlbert, S. H. 1984. Pseudoreplication and the design of ecological field experiments. Ecological Monographs 54:187-211.

Hurlbert, S. H. 2004. On misinterpretations of pseudoreplication and related matters: a reply to Oksanen. Oikos 104:591-597.

Huryn, A. D. 1990. Growth and voltinism of lotic midge larvae: patterns across an Appalachian mountain basin. Limnology and Oceanography 35:339-351.

Huryn, A. D., and J. B. Wallace. 1986. A method for obtaining in situ growth rates of larval Chironomidae (Diptera) and its application to studies of secondary production. Limnology and Oceanography 31:216-222.

Huryn, A. D., and J. B. Wallace. 1987a. Local geomorphology as a determinant of macrofaunal production in a mountain stream. Ecology 68:1932-1942.

Huryn, A. D., and J. B. Wallace. 1987b. The exopterygote insect community of a mountain stream in North Carolina, USA – life histories, production, and functional structure. Aquatic Insects 9:229-251.

Huryn, A. D., and J. B. Wallace. 2000. Life history and production of stream insects. Annual Review of Entomology 45:83-110.

Iversen, T. M. 1974. Ingestion and growth in *Sericostoma personatum* (Trichoptera) in relation to the nitrogen content of ingested leaves. Oikos 25:278-282.

Joergensen, R. G. and S. Scheu. 1999. Response of soil microorganisms to the addition of carbon, nitrogen, and phosphorus in a forest Rendzina. Soil Biology and Biochemistry 31:859-866.

Johnson , B. R. 2002. Effects of resource manipulation on selected primary and secondary consumers in two detritus-based southern Appalachian streams. Dissertation. University of Georgia.

Likens, G. E., F. H. Bormann, N. M. Johnson, D. W. Fisher, and R. S. Pierce. 1969. Effects of forest cutting and herbicide treatment on nutrient budgets in the Hubbard Brook watershed-ecosystem. Ecological Monographs 40:23-47.

Lugthart, G. J. 1991. Macrofaunal community structure and production in disturbed and undisturbed headwater streams. Dissertation. University of Georgia, Athens, GA, USA.

Lugthart G. J. and J. B. Wallace. 1992. Effects of disturbance on benthic functional structure and production in mountain streams. Journal of the North American Benthological Society 11: 138-164.

McQueen, D. J., M. R. S. Johannes, J. R. Post, T. J. Stewart, and D. R. S. Lean. 1989. Bottom-up and top-down impacts on freshwater pelagic community structure. Ecological Monographs 59:289-309.

McQueen, D. J., J. R. Post, and E. L. Mills. 1986. Trophic relationships in freshwater pelagic ecosystems. Canadian Journal of Fisheries and Aquatic Sciences 43:1571-1581.

Meyer, J. L. and C. Johnson. 1983. The influence of elevated nitrate concentration on rate of leaf decomposition in a stream. Freshwater Biology 13: 177-183.

Minshall, G. W. 1967. Role of allochthonous detritus in trophic structure of a woodland springbrook community. Ecology 48:139-149.

Moore, J. C., E. L. Berlow, D. C. Coleman, P. C. de Ruiter, Q. Dong, A. Hastings, N. C. Johnson, K. S. McCann, K. Melville, P. J. Morin, K. Nadelhoffer, A. D. Rosemond, D. M. Post, J. L. Sabo, K. M. Scow, M. J. Vanni, and D. H. Wall. In press. Detritus, trophic dynamics and biodiversity. Ecology Letters.

Murtaugh, P. A. 2003. On rejection rates of paired intervention analysis: reply. Ecology 84:2799-2802.

O'Doherty, E. C. 1985. Stream dwelling copepods: their life history and ecological significance. Limnology and Oceanography 30:554-564.

Odum, E. P and A. A. de la Cruz. 1963. Detritus as a major component of ecosystems. AIBS Bulletin 13:39-40.

Oksanen, L. 2001. Logic of experiments in ecology: is pseudoreplication a pseudoissue? Oikos 94:27-38.

Pace, M. L. and E. Funke. 1991. Regulation of planktonic microbial communities by nutrients and herbivores. Ecology 73:904-914.

Paul, M. J. and J. L. Meyer. 2001. Streams in the urban landscape. Annual Review of Ecology and Systematics 32:333-365

Pearson, R. G. and N. M. Connolly. 2000. Nutrient enhancement, food quality and community dynamics in a tropical rainforest stream. Freshwater Biology 43:31-42.

Peterson, B. J., L. Deegan, J. Helfrich, J. E. Hobbie, M. Hullar, B. Moller, T. E. Ford, A. Hershey, A. Hiltner, G. Kipphut, M. A. Lock, D. M. Fiebig, V. McKinley, M. C. Miller, J. R. Vestal, R. Ventullo, and G. Volk. 1993. Biological responses of a tundra river to fertilization. Ecology 74:653-672.

Polis, G. A. and D. R. Strong. 1996. Food web complexity and community dynamics. American Naturalist 147:813-846.

Pomeroy, L. R., J. E. Sheldon, W. M. Sheldon, Jr., and F. Peters. 1995. Limits to growth and respiration of bacterioplankton in the Gulf of Mexico. Mar. Ecol. Prog. Ser. 117:259-269.

Ramirez, A., C. M. Pringle, and L. Molina. 2003. Effects of stream phosphorus levels on microbial respiration. Freshwater Biology 48:88-97.

Richardson, J. S. 1991. Seasonal food limitation of detritivores in a montane stream – an experimental test. Ecology 72:873-887.

Rosemond, A. D., C. M. Pringle, A. Ramirez, and M. J. Paul. 2001. A test of top-down and bottom-up control in a detritus-based food web. Ecology 82:2279-2293.

Rosemond, A. D., C. M. Pringle, A. Ramirez, M. J. Paul, and J. L. Meyer. 2002. Landscape variation in phosphorus concentration and effects on detritus-based tropical streams. Limnology and Oceanography 47:278-289.

Schindler, D. W. 1998. Replication versus realism: the need for ecosystem-scale experiments. Ecosystems 1:323-334.

Schindler, D. W., E. J. Fee, and T. Rusczynski. 1978. Phosphorus input and its consequences for phytoplankton standing crop and production in the experimental lakes area and in similar lakes. Journal Fisheries Research Board of Canada 35:190-196.

Scheu, S. and M. Schaefer. 1998. Bottom-up control of the soil macrofauna community in a beechwood on limestone: manipulation of food resources. Ecology 79:1573-1585.

Scott, M. C., G. S. Helfman, M. E. McTammany, E. F. Benfield, and P. V. Bolstad. 2002. Multiscale influences on physical and chemical stream conditions across blue ridge landscapes. Journal of the American Water Resources Association 38:1379-1392.

Slansky, F. and J. M. Scriber. 1982. Selected bibliography and summary of quantitative food utilization by immature insects. Bulletin of the Entomological Society of America 28:43-55.

Slavik, K., B. J. Peterson, L. A. Deegan, W. B. Bowden, A. E. Hershey, and J. E. Hobbie. 2004. Long-term responses of the Kuparuk river ecosystem to phosphorus enrichment. Ecology 85:939-954.

Stelzer, R. S., J. Heffernan, and G. E. Likens. 2003. The influence of dissolved nutrients and particulate organic matter quality on microbial respiration and biomass in a forest stream. Freshwater Biology 48:1925-1937.

Stelzer, R. S. and G. A. Lamberti. 2001. Effects of N:P ratio and total nutrient concentration on stream periphyton community structure, biomass, and elemental composition. Limnology and Oceanography 46:356-367.

Stewart-Oaten, A. 2003. On rejection rates of paired intervention analysis: comment. Ecology 84:2795-2799.

Suberkropp, K. and E. Chauvet. 1995. Regulation of leaf breakdown by fungi in streams: influences of water chemistry. Ecology 76:1433-1445.

Swank W.T and D. A. Crossley. 1988. Forest Hydrology and Ecology at Coweeta. Springer-

Verlag, New York, USA.

Tamm, C. O. 1991. Nitrogen in terrestrial ecosystems. Springer-Verlag. Berlin, Germany.

Tank, J. L. and J. R. Webster. 1998. Interaction of substrate and nutrient availability on wood biofilm processes in streams. Ecology 79:2168-2179.

Tenore, K. R. 1977. Growth of *Capitella capatata* cultured on various levels of detritus from different sources. Limnology and Oceangraphy 22:936-941.

Tilman, D. 1987. Secondary succession and the pattern of plant dominance along experimental nitrogen gradients. Ecological Monographs 57:189-214.

Vannote, R. L. and B. W. Sweeney. 1980. Geographic analysis of thermal equilibria – a conceptual model for evaluating the effect of natural and modified thermal regimes on aquatic insect communities. American Naturalist 115:667-695.

Vitousek, P. M., J. D. Aber, R. W. Howarth, G. E. Likens, P. A. Matson, D. W. Schindler, W. H. Schlesinger, and D. G. Tilman. 1997. Human alteration of the global nitrogen cycle: sources and consequences. Ecological Applications 7:737-750.

Vitousek, P. M. and R. W. Howarth. Nitrogen limitation on land and in the sea – how can it occur. Biogeochemistry 13:87-115.

Vos, J. H., M. A. G. Oojevaar, J. F. Postma, and W. Admiraal. 2000. Interaction between food availability and food quality during growth of early instar chironomid larvae. Journal of the North American Benthological Society 19:158-168.

Wallace J. B., T. F. Cuffney, S. L. Eggert and M. R. Whiles. 1997a. Stream organic matter inputs, storage, and export for Satellite Branch at Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:67-74.

Wallace J. B., S. L. Eggert, J. L. Meyer and J. R. Webster. 1997b. Multiple trophic levels of a forest stream linked to terrestrial litter inputs. Science 277:102-104.

Wallace J. B., S. L. Eggert, J. L. Meyer and J. R. Webster. 1999. Effects of resource limitation on a detrital-based ecosystem. Ecological Monographs 69:409-442.

Wallace, J. B., M. R. Whiles, S. L. Eggert, T. F. Cuffney, G. J. Lugthart, and K. Chung. 1995. Long-term dynamics of coarse particulate organic matter in three Appalachian Mountain streams. Journal of the North American Benthological Society 14:217-232.

Ward, G. M. and K. W. Cummins. 1979. Effects of food quality on growth of a stream detritivores: *Paratendipes albimanus* (Meigen) (Diptera: Chironomidae). Ecology 60:57-64.

Waters, T. F. 1969. Subsampler for dividing large samples of stream invertebrate drift. Limnology and Oceanography 14:813-815.

Waters, T. F. 1977. Secondary production of inland waters. Advances in Ecological Research 10:91-164.

Webster, J. R., J. L. Meyer, J. B. Wallace, and E. F. Benfield. 1997. Organic matter dynamics in Hugh White Creek, Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:74-78.

Wetzel, R. G. 1995. Death, detritus, and energy flow in aquatic ecosystems. Freshwater Biology 33:83-89.

Whiles, M. R. and J. B. Wallace. 1992. First-year recovery of a headwater stream following a 3-year insecticide-induced disturbance. Freshwater Biology 28:81-91.

Whiles, M. R. and J. B. Wallace. 1995. Macroinvertebrate production in a headwater stream during recovery from anthropogenic disturbance and hydrologic extremes. Canadian Journal of Fisheries and Aquatic Sciences 52:2402-2422.

Wiens, J. A. 1977. On competition and variable environments. American Scientist 65: 590-597.

Table 4.1. Physical and chemical characteristics of headwater streams draining the reference (C53) and treated (C54) catchments at Coweeta Hydrologic Laboratory. Elevations were measured at the gauging weirs.

Variable	**Reference (C53)**	**Treatment (C54)**
Catchment		
Area (ha)	5.2	5.5
Elevation (m asl)	829.0	841.0
Channel		
Gradient (cm m^{-1})	27.0	33.0
Length (m)	145.0	282.0
Bankfull Area (m^2)	327.0	443.0
Discharge (l s^{-1})		
Average*	1.2	1.5
Maximum*	47.2	35.5
Average this study¶	0.3	0.5
Maximum this study¶	3.8	4.8
Substrate composition (%)		
Mixed substrates	73.0	65.0
Bedrock outcrop	27.0	35.0
Temperature (°C)¶		
Annual average	12.0	12.0
Minimum	2.6	4.8
Maximum	18.6	16.7
Water chemistry (range)		
pH	6.6 (6.2-7)	6.9 (6.6-7.9)
$(NO_3\text{-N} + NO_2)\text{-N}$ ($\mu g\ l^{-1}$)		
pretreatment: 1999 – 2000	15.4 (9.4-25.8)	18.8 (4-39.5)
enrichment: 2000 – 2002	16.9 (bd-151)	308.9 (11-1711)
$NH_4\text{-N}$ ($\mu g\ l^{-1}$)		
pretreatment: 1999 – 2000	9.4 (bd-30)	9.9 (bd-25)
enrichment: 2000 – 2002	10.4 (bd-76)	105.5 (6-566)
SRP‡ ($\mu g\ l^{-1}$)		
pretreatment: 1999 – 2000	7.6 (bd-20)	8.8 (bd-22)
enrichment: 2000 – 2002	3.7 (bd-17)	51.2 (bd-268)

*C53 values from 12 years (1984-1996), C54 values from 8 years (1985-1992).

‡Soluble reactive phosphorus. ¶September 1998 – August 2002. bd = below detection limit. For nutrient data: reference stream - pretreatment n = 5, treated stream - pretreatment n = 12, reference stream - enrichment n = 33, treated stream - enrichment n = 44.

Table 4.2. Organic matter standing crops (g AFDM m^{-2}) for mixed substrate and bedrock outcrop habitats before[†], and after[‡], the initiation of nutrient enrichment in the reference stream (C53) and the treatment stream (C54).

Habitat	reference, before	reference, after	treatment, before	treatment, after	RIA probability	n
Mixed substrates						
Total FBOM	900.0	784.7	1327.4	2023.0	**(+) 0.001**	60
Total CBOM	1157.0	1236.6	1060.7	1330.0	0.213	60
Leaf detritus	204.2	244.7	244.3	168.5	**(-) 0.002**	60
Bedrock Outcrop						
Total FBOM	28.0	20.1	45.8	28.7	0.336	60
Total CBOM	51.6	30.6	45.2	17.6	0.346	60
Moss	9.1	3.5	15.2	4.5	0.157	60

[†] ‘before’ data include October 1984 – September 1985 (Lugthart and Wallace 1992) and September 1998 – June 2000 (this study); [‡] ’after’ initiation of enrichment includes data from July 2000 – August 2002. Total FBOM = all benthic organic matter <1 mm, Total CBOM = all benthic organic matter >1 mm. n = total number of months included in RIA. Bold values indicate significant RIAs. (+) and (-) indicate the direction of effect due to enrichment.

Table 4.3. Average abundance and biomass of invertebrate functional feeding groups in mixed substrate habitat of the reference stream (C53) and treatment stream (C43) before (October 1984 – September 1985 & September 1998 – June 2000) and after (July 2000 – August 2002) the initiation of nutrient enrichment.

Functional group	reference, before	reference, after	enriched, before	enriched, after	RIA probability[†]
Abundance (no. m^{-2})					
Scrapers	45	33	16	17	0.668
Shredders	3365	3311	1843	4476	**(+) <0.000001**
Gatherers	115304	69684	64931	137039	**(+) <0.000001**
Filterers	690	527	306	825	**(+) 0.016**
Total primary consumers	119350	73514	67069	142284	**(+) <0.000001**
Predators	16840	9492	7556	13525	**(+) <0.000001**
Biomass (mg AFDM m^{-2})					
Scrapers	7	4	14	1	0.477
Shredders	1203	916	1090	1635	**(+) 0.006**
Gatherers	457	338	357	706	**(+) <0.000001**
Filterers	46	29	47	113	**(+) <0.000001**
Total primary consumers	1707	1286	1501	2443	**(+) <0.000001**
Predators	1022	739	920	1433	**(+) <0.000001**

[†] Randomized intervention analysis (RIA) tests the null hypothesis of no change in benthic abundance and biomass for each functional feeding group between the reference and enriched stream. Bold values indicate significant RIAs. Total primary consumer numbers are slighty less than the sum of all primary consumer groups because a fraction of *Parapsyche cardis* and *Diplectrona* spp. (omnivorous filtering caddisflies) was placed into the predator category. (+) and (-) indicate the direction of effect due to enrichment. n = 60 for all RIAs.

Table 4.4. Average abundance and biomass of invertebrate functional feeding groups in bedrock outcrop habitat of the reference stream (C53) and treatment stream (C43) before (October 1984 – September 1985 and September 1998 – June 2000) and after (July 2000 – August 2002) the initiation of nutrient enrichment.

Functional group	reference, before	reference, after	treatment, before	treatment, after	RIA probability[†]
Abundance (no. m^{-2})					
Scrapers	175	233	200	262	0.273
Shredders	556	827	695	338	**(-) <0.000001**
Gatherers	18035	12941	16716	15960	0.126
Filterers	889	1057	693	816	0.623
Total primary consumers	19484	14839	18151	17211	0.232
Predators	3820	1952	2120	1863	**(+) 0.004**
Biomass (mg AFDM m^{-2})					
Scrapers	22	35	74	60	0.393
Shredders	135	145	113	45	**(-) <0.000001**
Gatherers	125	74	126	229	**(+) 0.003**
Filterers	151	128	190	167	0.459
Total primary consumers	399	347	454	456	0.954
Predators	128	87	154	129	0.287

[†] Randomized intervention analysis (RIA) tests the null hypothesis of no change in benthic abundance and biomass for each functional feeding group between the reference and enriched stream. Bold values indicate significant RIAs ($P < 0.05$). (+) and (-) indicate the direction of effect due to enrichment. Total primary consumer numbers are slighty less than the sum of all primary consumer groups because a fraction of *Parapsyche cardis* and *Diplectrona* spp. (omnivorous filtering caddisflies) was placed into the predator category. (+) and (-) indicate the direction of effect due to enrichment. n = 60 for all RIAs.

Table 4.5. Annual invertebrate secondary production (mg AFDM m^{-2} y^{-1}) in the reference stream (C53) and treatment stream (C54) by functional feeding group in mixed substrate and bedrock outcrop habitats. RIA was not possible on these annual data because of limited data points before (n=3) and after (n=2) treatment.

Functional group	Stream	Pre – 1[†]	Pre – 2	Pre – 3	Enr – 1	Enr – 2
Mixed substrates						
Scrapers	reference	67	25	19	6	16
	treatment	162	2	9	24	1
Shredders	reference	4017	3198	3434	3327	4768
	treatment	4953	2636	3531	7802	12805
Gatherers	reference	7123	3572	3389	2248	2442
	treatment	4755	1695	3077	6775	6451
Filterers	reference	526	114	289	177	279
	treatment	113	170	420	666	706
Predators	reference	4655	2468	3635	3745	2839
	treatment	4318	1732	4598	6036	5297
Bedrock outcrops						
Scrapers	reference	44	50	175	131	131
	treatment	207	52	96	222	190
Shredders	reference	1137	407	376	438	1024
	treatment	916	395	395	163	410
Gatherers	reference	2390	704	629	743	1227
	treatment	2091	863	1698	2813	1879
Filterers	reference	658	960	1459	842	871
	treatment	1820	1207	972	968	1626
Predators	reference	741	257	203	184	386
	treatment	705	385	567	559	581

[†] Pre – 1, 2, and 3 = pretreatment October 1984 – September 1985, September 1998 – August 1999, and September 1999 – August 2000, respectively. Enr – 1 and 2 = nutrient enrichment September 2000 – August 2001, and September 2001 – August 2002. Data for 1984 – 1985 are from Lugthart and Wallace (1992); *Stenonema* sp. was moved from scrapers and placed in gatherers for 1984 – 1985.

Table 4.6. Results of randomized intervention analyses (RIA) for abundance and biomass of individual taxa in mixed substrate and bedrock outcrop habitats. RIA tests the null hypothesis of no change in abundance or biomass between the reference stream and treatment stream. Data (n = 60) include 34 months of pretreatment data (October 1984 – September 1985, September 1998 – June 2000) and 26 months of post-enrichment data (July 2000 – August 2002).

		Mixed Substrate		Bedrock Outcrops	
Functional group and taxon	Order	P value, abundance	P value, biomass	P value, abundance	P value, biomass
Scrapers					
Epeorus sp.	E	--	--	0.896	0.760
Baetis sp.	E	--	--	0.467	0.389
Hydroptila sp.	T	--	--	0.928	0.963
Neophylax sp.	T	--	--	0.053	0.127
Ectopria sp.	C	0.256	0.378	0.651	0.154
Elmidae	C	--	--	--	--
Shredders					
Leuctra spp.	P	**(+) <0.000001**	**(+) 0.003**	**(-) 0.006**	**(-) 0.005**
Tallaperla spp.	P	0.634	0.249	**(-) <0.000001**	**(-) <0.000001**
Lepidostoma spp.	T	0.931	**(+) 0.009**	0.057	0.393
Pycnopsyche spp.	T	0.214	**(+) 0.006**	0.995	**(+) 0.026**
Fattigia pele	T	0.250	0.305	--	--
Psilotreta sp.	T	0.237	0.348	--	--
Molophilus sp.	D	**(+) 0.029**	**(+) 0.004**	--	--
Tipula sp.	D	0.334	0.657	**(-) 0.002**	**(-) 0.002**
Lipsothrix sp.	D	0.334	0.293	--	--
Limonia sp.	D	0.507	0.345	--	--
Anchytarsus sp.	C	0.600	0.531	--	--
Cambarus bartoni	NI	**(-) 0.010**	0.060	--	--
Collector-gatherers					
Paraleptophlebia sp.	E	0.095	0.059	0.496	0.291
Serratella sp.	E	0.345	**(+) 0.008**	**(+) 0.028**	**(+) 0.007**

Table 4.7. (cont.)

		Mixed Substrate		Bedrock Outcrops	
Functional group and taxon	Order	P, abundance	P, biomass	P, abundance	P, biomass
Collector-gatherers (cont.)					
Stenonema sp.	E	**(+) 0.004**	**(+) <0.000001**	--	--
Amphinemura sp.	P	0.101	**(+) 0.001**	0.188	**(+) 0.014**
Soyedina sp.	P	0.845	0.542	--	--
Lype diversa	T	0.731	0.192	--	--
Chironomidae (non-Tanypodinae)	D	**(+) <0.000001**	**(+) <0.000001**	0.274	0.945
Leptotarus sp.	D	**(+) <0.000001**	**(+) 0.011**	--	--
Nymphomyiidae	D	--	--	0.510	0.287
Ormosia sp.	D	0.139	0.117	--	--
Sciaridae	D	0.806	0.394	--	--
Copepoda	NI	**(+) <0.000001**	**(+) <0.000001**	0.087	0.201
Nematoda	NI	**(+) <0.000001**	**(+) <0.000001**	**(+) 0.005**	**(+) 0.004**
Oligochaeta	NI	**(+) <0.000001**	**(+) 0.040**	0.717	0.909
Collector-filterers					
Diplectrona modesta	T	0.469	**(+) 0.001**	0.363	0.837
Diplectrona metaqui	T	--	--	--	--
Parapsyche cardis	T	--	--	0.952	0.764
Wormaldia spp. (summer cohort)	T	0.913	0.978	0.947	0.758
Wormaldia spp. (winter cohort)	T	0.816	0.060	0.424	0.452
Dolophilodes distinctus	T	--	--	--	--
Simuliidae	D	0.234	**(+) 0.024**	0.249	**(+) 0.002**
Dixa sp.	D	0.631	0.765	0.310	0.564
Sphaeridae	NI	**(+) <0.000001**	**(+) <0.000001**	--	--
Predators					
Cordulegaster sp.	O	**(+) 0.037**	**(+) 0.002**	--	--
Lanthus sp.	O	0.062	0.076	--	--
Sweltsa sp.	P	0.089	**(+) 0.027**	--	--
Beloneuria sp.	P	0.500	**(+) 0.036**	0.618	0.968

Table 4.7. (cont.)

Functional group and taxon	Order	Mixed Substrate P, abundance	Mixed Substrate P, biomass	Bedrock Outcrops P, abundance	Bedrock Outcrops P, biomass
Predators (cont.)					
Isoperla spp.	P	**(+) <0.000001**	**(+) 0.001**	0.164	0.125
Malerikus hastatus	P	--	--	--	--
Rhyacophila spp.	T	0.097	0.228	0.915	0.222
Pseudogoera singularis	T	0.962	0.100	**(+) 0.050**	**(+) 0.003**
Ceratopogonidae	D	**(+) <0.000001**	**(+) 0.001**	0.317	0.740
Hexatoma spp.	D	0.782	0.429	0.054	0.179
nr. *Pedicia* sp.	D	0.190	0.519	--	--
Pedicia sp.	D	0.100	0.095	--	--
Dicranota spp.	D	**(+) 0.029**	**(+) <0.000001**	**(+) 0.004**	**(+) <0.000001**
Glutops sp.	D	0.143	0.177	--	--
Tanypodinae	D	**(+) <0.000001**	**(+) <0.000001**	0.711	0.431
Empididae	D	0.186	**(+) 0.011**	0.658	0.495
Pilaria sp.	D	--	--	--	--
Pseudolimnolphila sp.	D	0.971	0.131	--	--
Rhabdomastix sp.	D	--	--	--	--
Dolichopodidae	D	0.494	0.023	--	--
Turbellaria	NI	0.606	0.912	0.110	0.226
Acari	NI	**(+) 0.003**	**(+) <0.000001**	**(+) 0.001**	**(+) 0.004**
Vertebrate predators					
Eurycea sp.	NI	0.353	0.353	--	--
Desmognathus spp.	NI	0.565	0.390	--	--
Total salamanders		0.195	0.136	--	--

Notes: All analyses were conducted on log (x+1) transformed data based on monthly means for mixed substrate and bedrock outcrop habitats (n = 60 for each habitat). Insect orders are as follows: E = Ephemeroptera, P = Plecoptera, T = Trichoptera, C = Coleoptera, O = Odonata, and NI = non-insect. Bold values indicate significant RIAs (i.e., $P < 0.05$). (+) and (-) indicate the direction of effect due to enrichment. -- indicates too few data points for RIA analysis.

Figure 4.1. Long-term time series of log (x+1) leaf litter, FBOM, and CBOM standing crop (g AFDM m^{-2}) in the reference stream (C53, white circles) and treatment stream (C54, black circles). The vertical dashed line refers to the initiation of nutrient enrichment in the treatment stream. Elapsed days –365 through 0 are between October 1984 and September 1985; days 0 through 1460 are between September 1998 and August 2002. P values are from randomized intervention analysis (RIA) on differences between streams before and after the manipulation. NS = non-significant ($P > 0.05$).

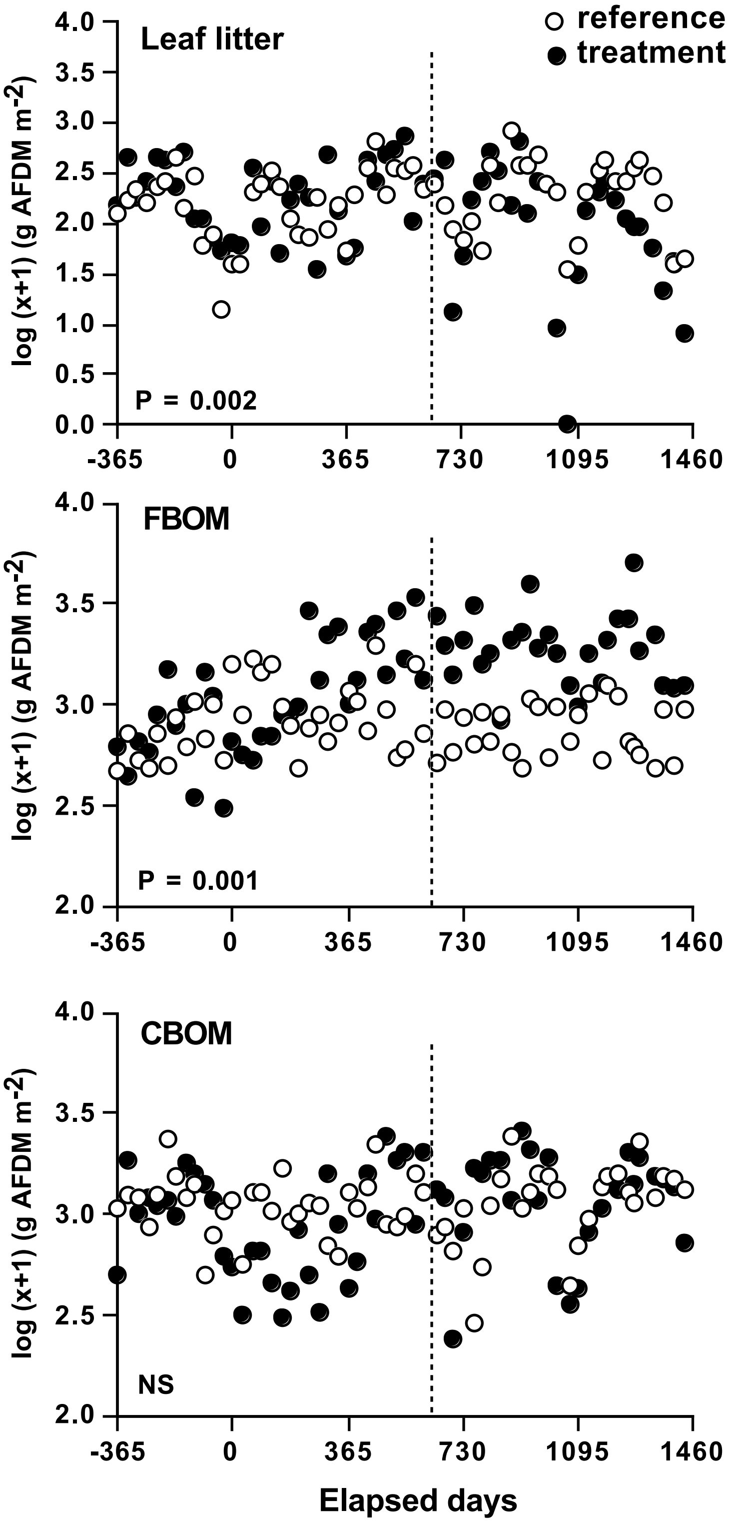

reference
treatment
Leaf litter
P = 0.002
FBOM
P = 0.001
CBOM
NS
log (x+1) (g AFDM m-2)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-365
0
365
730
1095
1460
Elapsed days

Figure 4.2. Annual mean abundance, biomass, and annual secondary production during the five years of study in the reference stream (white bars) and treatment stream (grey bars) in mixed substrate and bedrock outcrop habitats. The arrow refers to the initiation of nutrient enrichment in the treatment stream. Pre-1, 2, and 3 = October 1984 – September 1985, September 1998 – August 1999, and September 1999 – August 2000, respectively. Enr-1 and 2 = September 2000 – August 2001 and September 2001 – August 2002. Error bars for abundance and biomass are ±1 SE ($n = 12$). P values are from randomized intervention analysis (RIA) on differences between streams before and after the manipulation. NS = non-significant ($P > 0.05$). RIA was not possible on annual production data because of limited data points before ($n = 3$) and during ($n = 2$) enrichment.

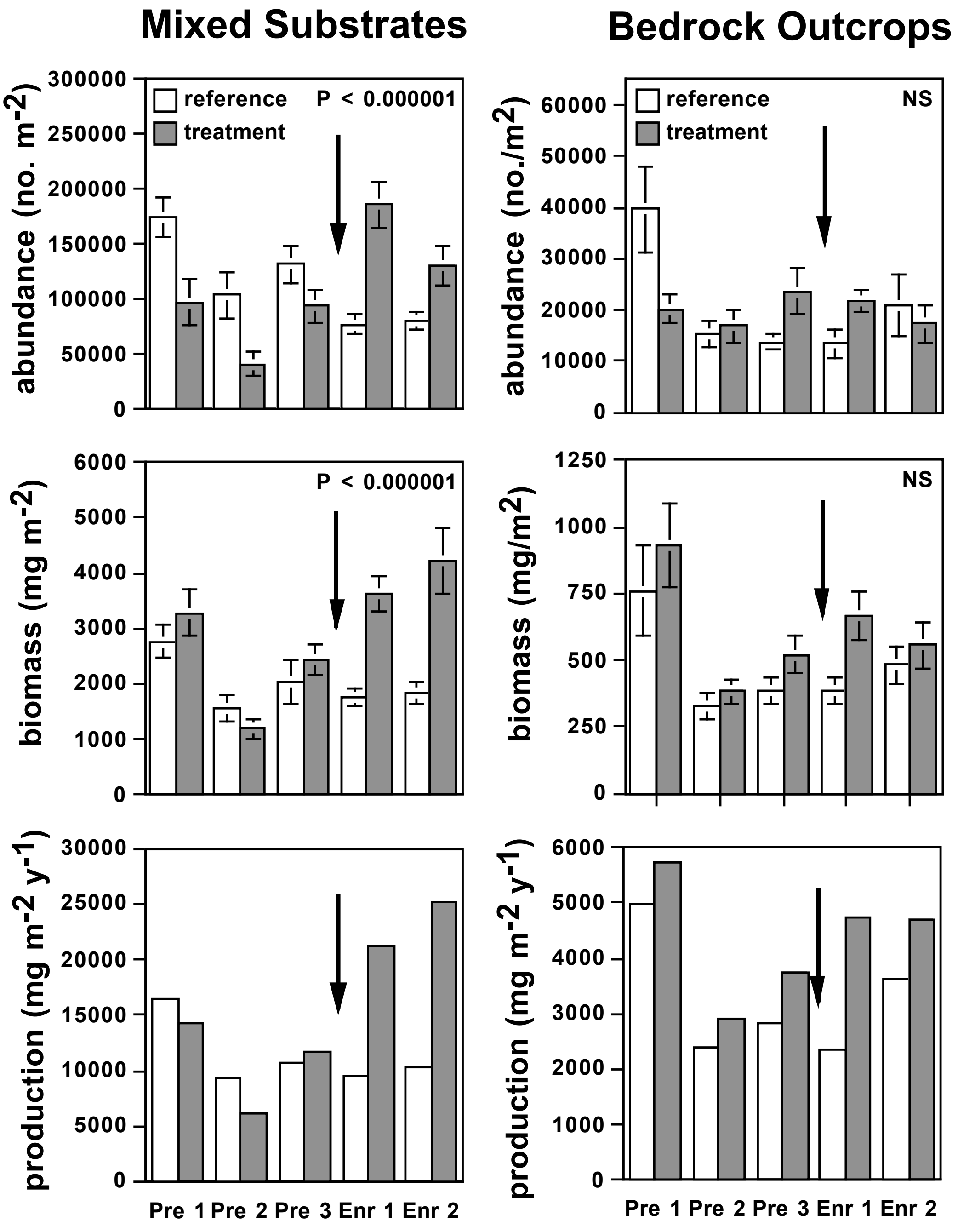
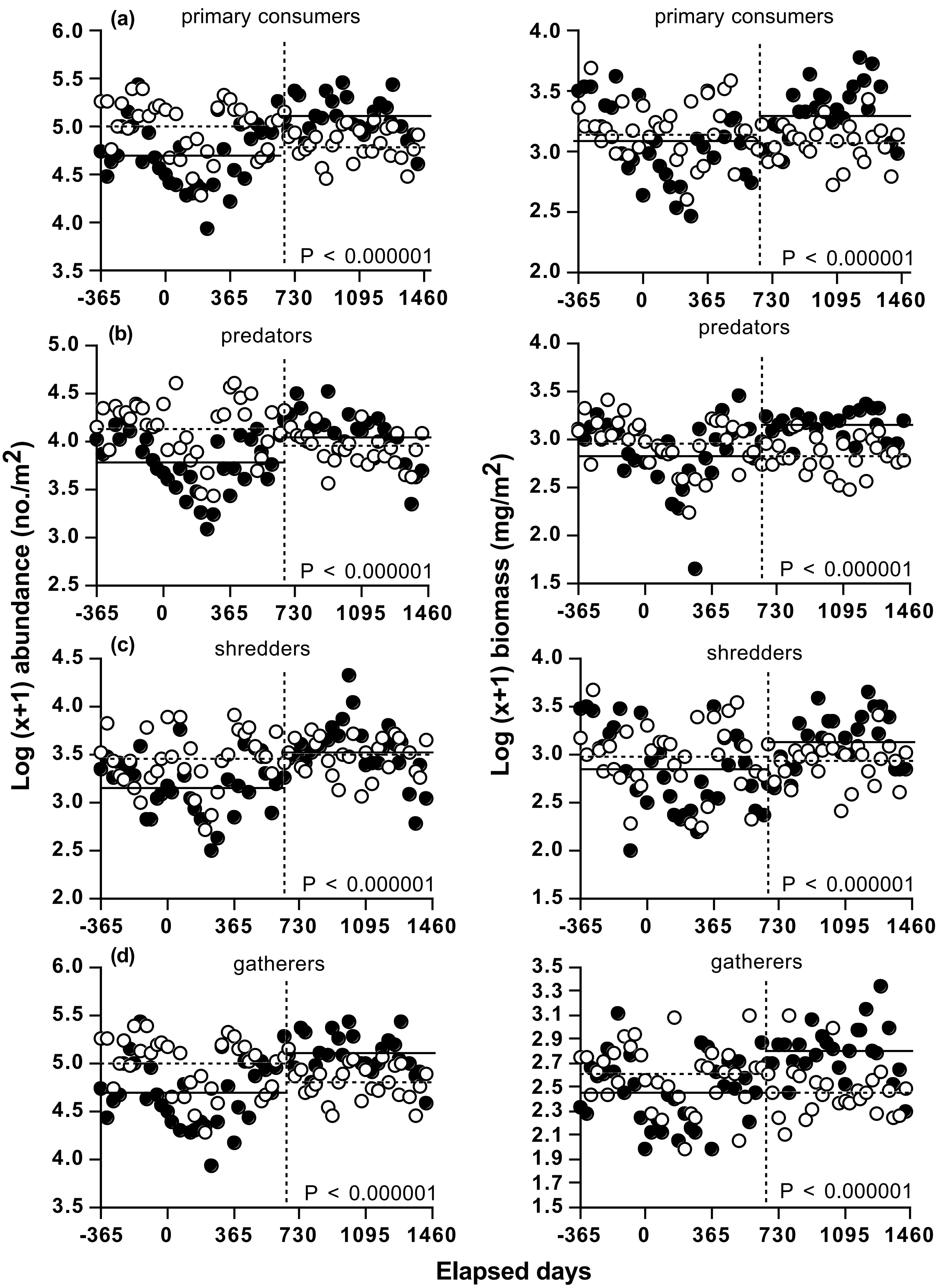

Mixed Substrates
Bedrock Outcrops
reference
treatment
P < 0.000001
NS
abundance (no. m-2)
abundance (no./m2)
300000
250000
200000
150000
100000
50000
0
60000
50000
40000
30000
20000
10000
0
P < 0.000001
NS
biomass (mg m-2)
biomass (mg/m2)
6000
5000
4000
3000
2000
1000
0
1250
1000
750
500
250
0
production (mg m-2 y-1)
30000
25000
20000
15000
10000
5000
0
6000
5000
4000
3000
2000
1000
0
Pre 1
Pre 2
Pre 3
Enr 1
Enr 2

Figure 4.3. Time series of log (x+1) abundance and biomass of (a) primary consumers, (b) predators, (c) shredders, (d) gatherers, (e) filterers, and (f) scrapers in the mixed substrate habitat in the reference stream (C53, white circles) and treatment stream (C54, black circles). The vertical dashed line refers to the initiation of nutrient enrichment in the treatment stream. Horizontal dashed lines represent mean values in the reference stream before and during enrichment. Horizontal solid lines represent mean values in the treatment stream before and during treatment. Elapsed days –365 through 0 are between October 1984 and September 1985; days 0 through 1460 are between September 1998 and August 2002. P values are from randomized intervention analysis (RIA) on differences between streams before and after the manipulation. NS = non-significant ($P > 0.05$).


(a)
primary consumers
6.0
5.5
5.0
4.5
4.0
3.5
P < 0.000001
primary consumers
4.0
3.5
3.0
2.5
2.0
P < 0.000001
(b)
predators
5.0
4.5
4.0
3.5
3.0
2.5
P < 0.000001
predators
4.0
3.5
3.0
2.5
2.0
1.5
P < 0.000001
(c)
shredders
4.5
4.0
3.5
3.0
2.5
2.0
P < 0.000001
shredders
4.0
3.5
3.0
2.5
2.0
1.5
P < 0.000001
(d)
gatherers
6.0
5.5
5.0
4.5
4.0
3.5
P < 0.000001
gatherers
3.5
3.3
3.1
2.9
2.7
2.5
2.3
2.1
1.9
1.7
1.5
P < 0.000001
-365
0
365
730
1095
1460
Log (x+1) abundance (no./m2)
Log (x+1) biomass (mg/m2)
Elapsed days

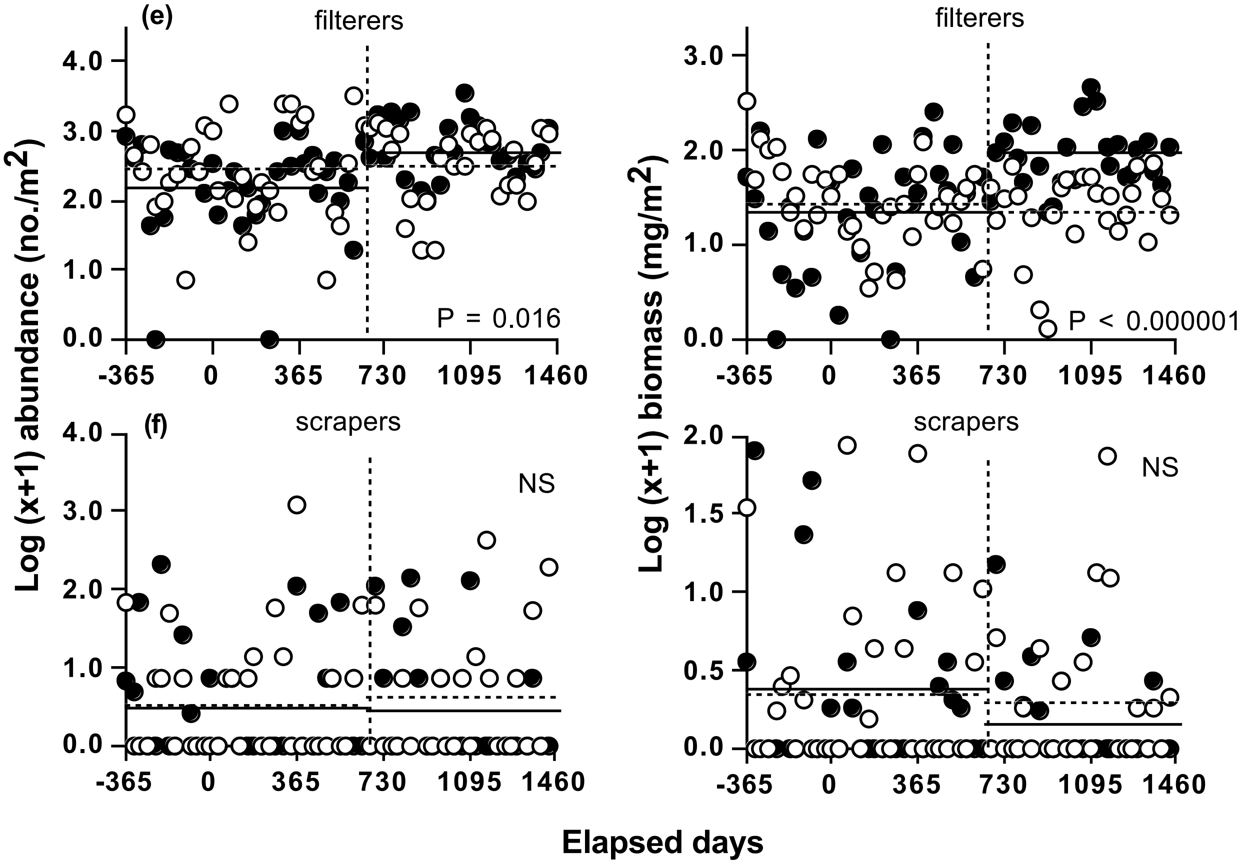
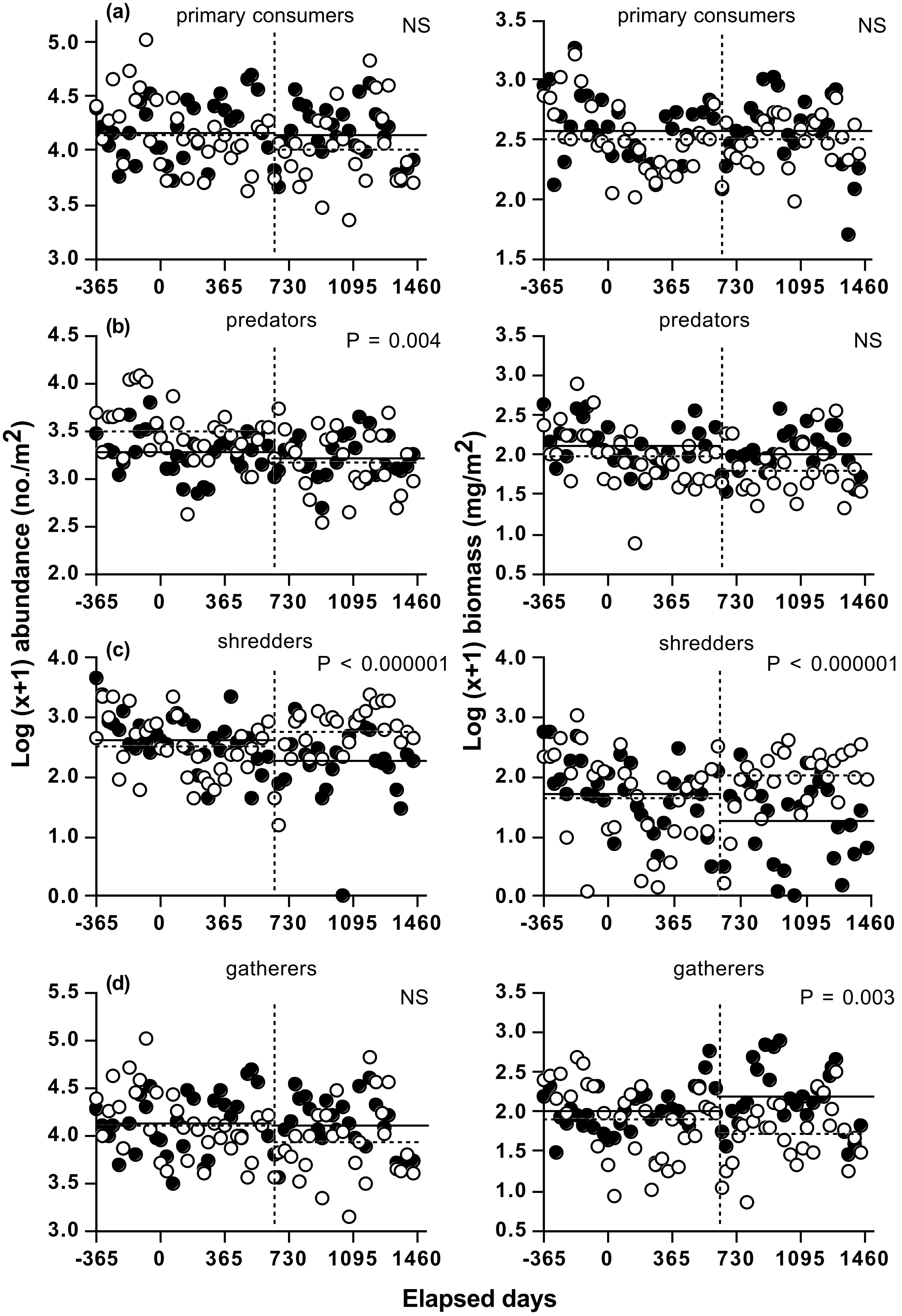


figure 3 (cont.)

Figure 4.4. Time series of log (x+1) abundance and biomass of (a) primary consumers, (b) predators, (c) shredders, (d) gatherers, (e) filterers, and (f) scrapers in the bedrock outcrop habitat in the reference stream (C53, white circles) and treatment stream (C54, black circles). The vertical dashed line refers to the initiation of nutrient enrichment in the treatment stream. Horizontal dashed lines represent mean values in the reference stream before and during enrichment. Horizontal solid lines represent mean values in the treatment stream before and during treatment. Elapsed days –365 through 0 are between October 1984 and September 1985; days 0 through 1460 are between September 1998 and August 2002. P values are from randomized intervention analysis (RIA) on differences between streams before and after the manipulation. NS = non-significant ($P > 0.05$).


(a)
primary consumers
NS
primary consumers
NS
(b)
predators
P = 0.004
predators
NS
(c)
shredders
P < 0.000001
shredders
P < 0.000001
(d)
gatherers
NS
gatherers
P = 0.003
Log (x+1) abundance (no./m²)
Log (x+1) biomass (mg/m²)
Elapsed days
-365
0
365
730
1095
1460
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0

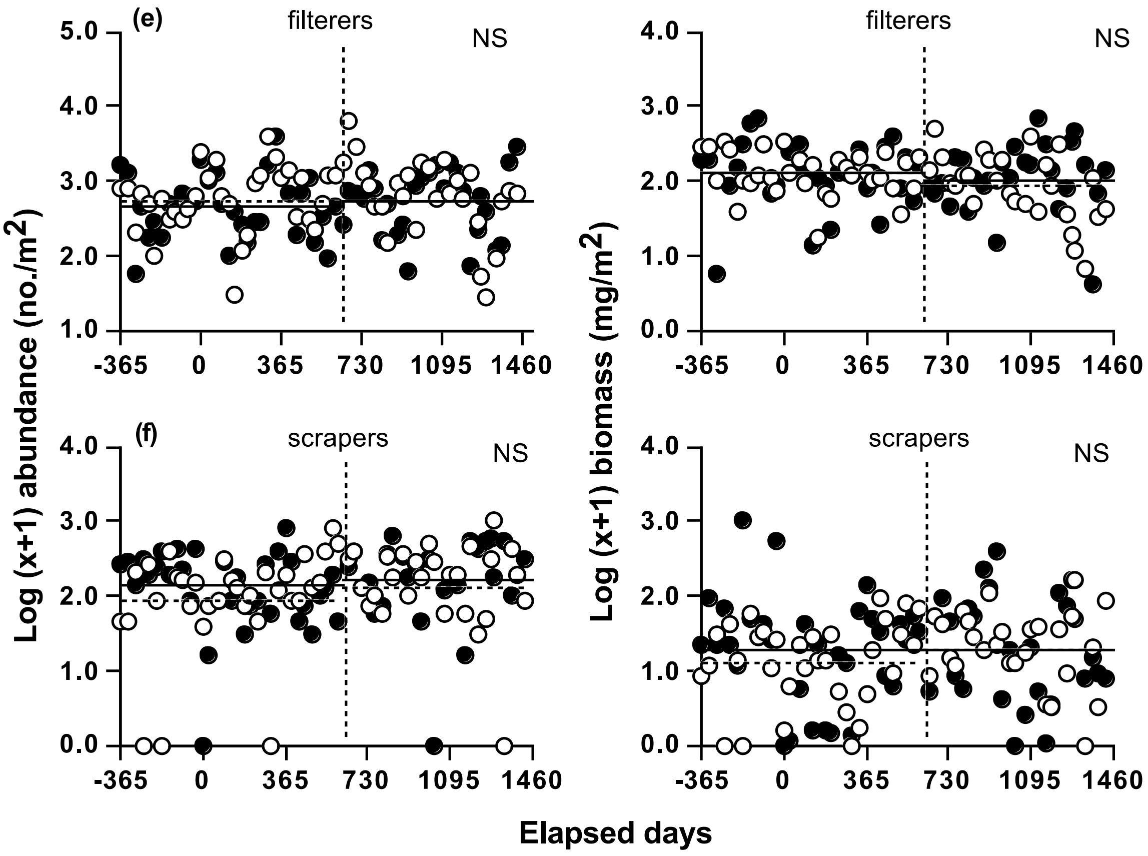
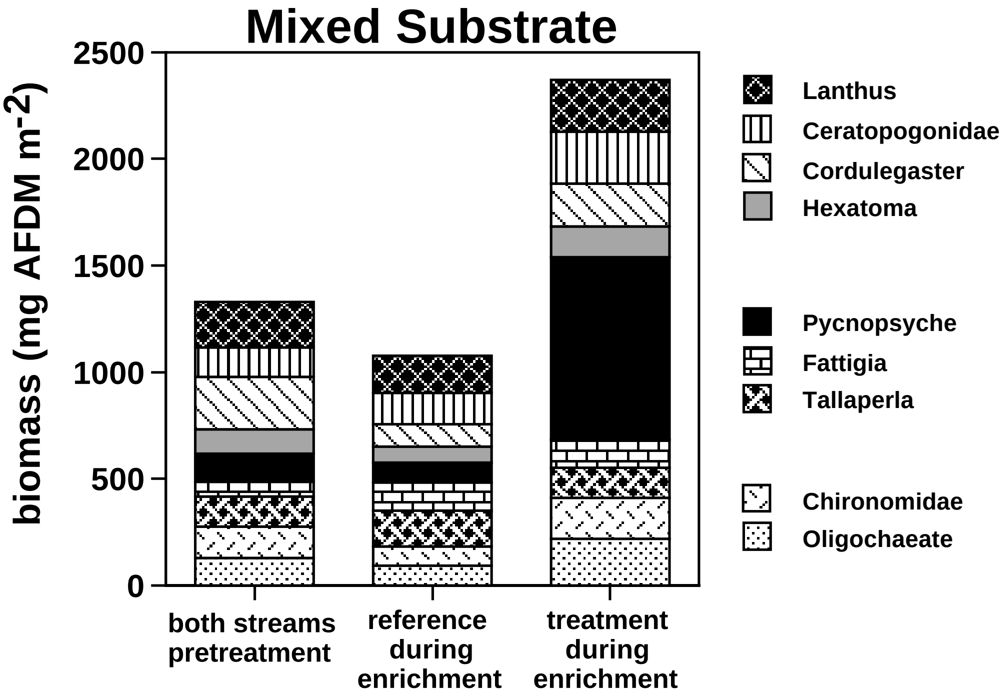


figure 4 (cont.)

Figure 4.5. Average biomass (mg AFDM m-2 y-1) of the 10 dominant taxa in mixed substrate and bedrock outcrop habitats in the reference and treatment stream before (both streams averaged) and during enrichment of the treatment stream (reference and treatment shown separately). Taxa in the legend are grouped by functional feeding group; in the mixed substrate habitat, the top 4 taxa are predators, the middle 3 taxa are shredders, and the bottom 2 taxa are gatherers. For bedrock outcrops, *Parapsyche* is a filterer, *Tallaperla* is a shredder, and the bottom 3 taxa are gatherers.


Mixed Substrate
biomass (mg AFDM m^{-2})
2500
2000
1500
1000
500
0
both streams pretreatment
reference during enrichment
treatment during enrichment
Lanthus
Ceratopogonidae
Cordulegaster
Hexatoma
Pycnopsyche
Fattigia
Tallaperla
Chironomidae
Oligochaeate

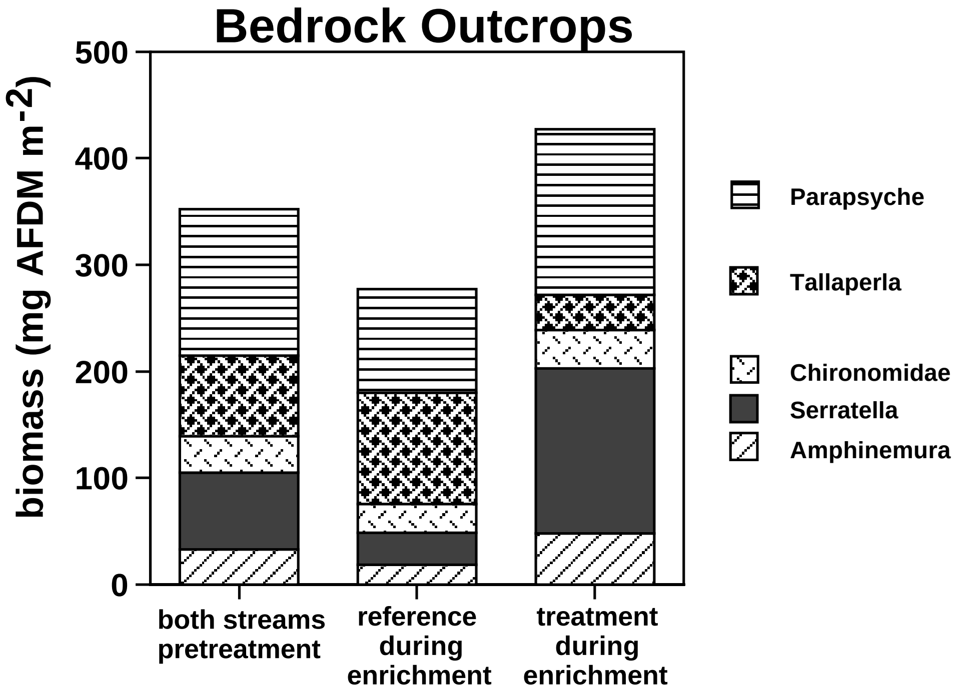

Bedrock Outcrops
biomass (mg AFDM m^{-2})
500
400
300
200
100
0
both streams pretreatment
reference during enrichment
treatment during enrichment
Parapsyche
Tallaperla
Chironomidae
Serratella
Amphinemura

Figure 4.6. Relationship between length of larval lifespan (days) and % increase in taxon-specific production in the treatment stream.

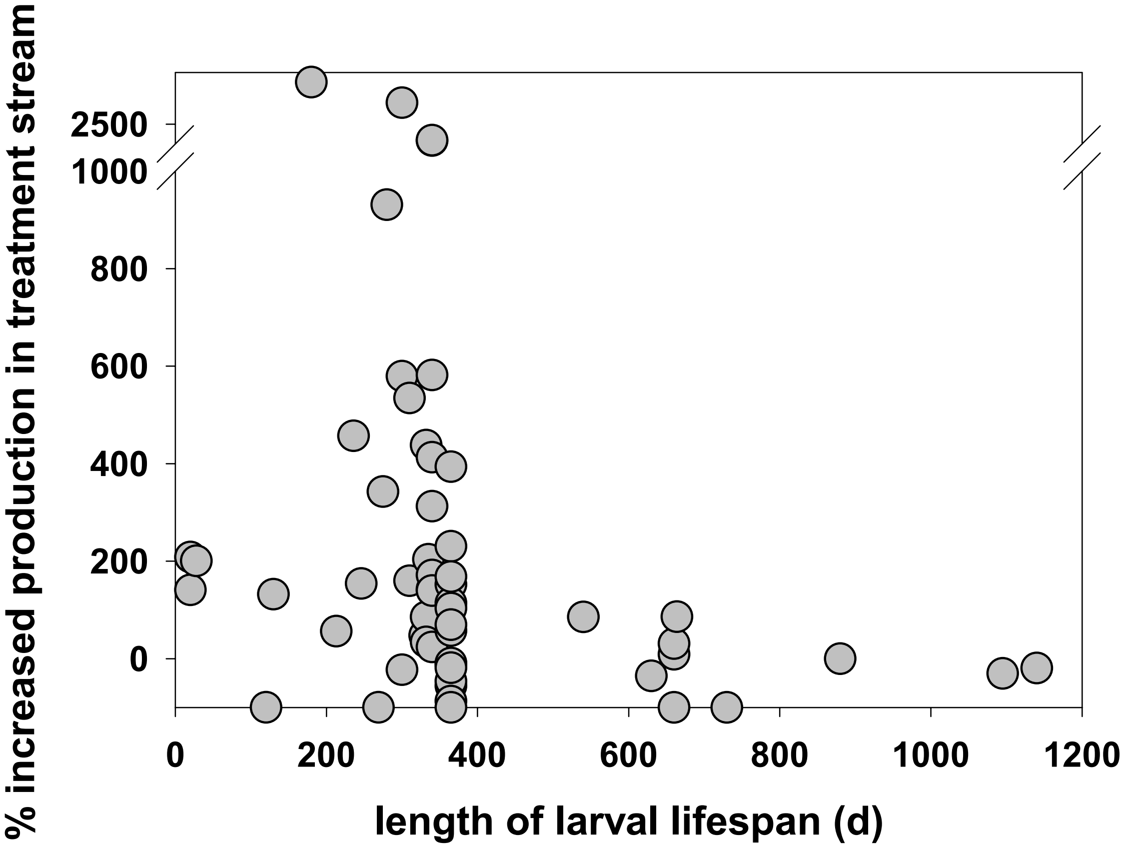

% increased production in treatment stream
2500
1000
800
600
400
200
0
0
200
400
600
800
1000
1200
length of larval lifespan (d)

Figure 4.7. Relationships between habitat-weighted primary consumer (grey circles) or total (white circles) secondary production (g AFDM m^{-2} y^{-1}) and predator production. These relationships include data from the reference stream and treatment stream during the 4 years of this study and 1 previous year (Lugthart and Wallace 1992) (total n = 10). Both linear regressions are statistically significant ($P < 0.001$). Equations for lines are reported on figure.

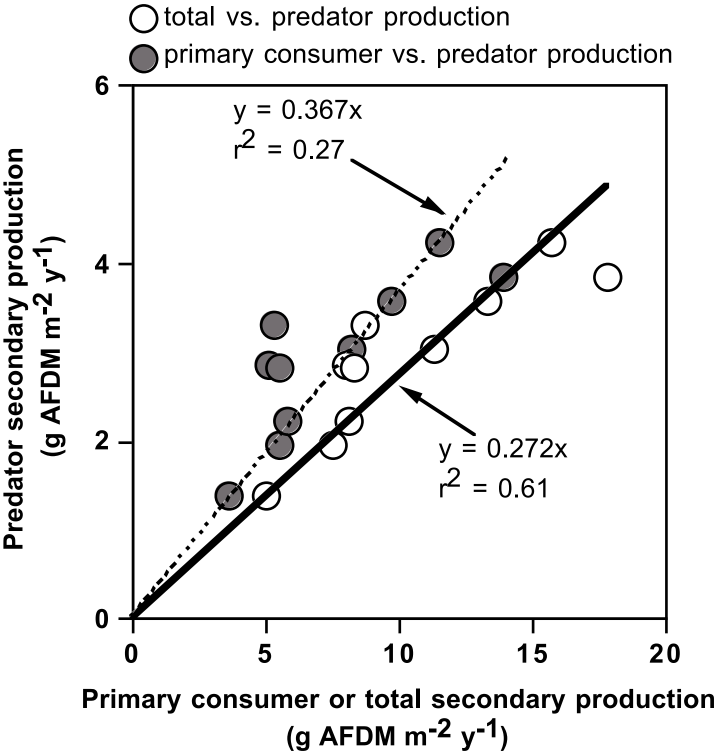
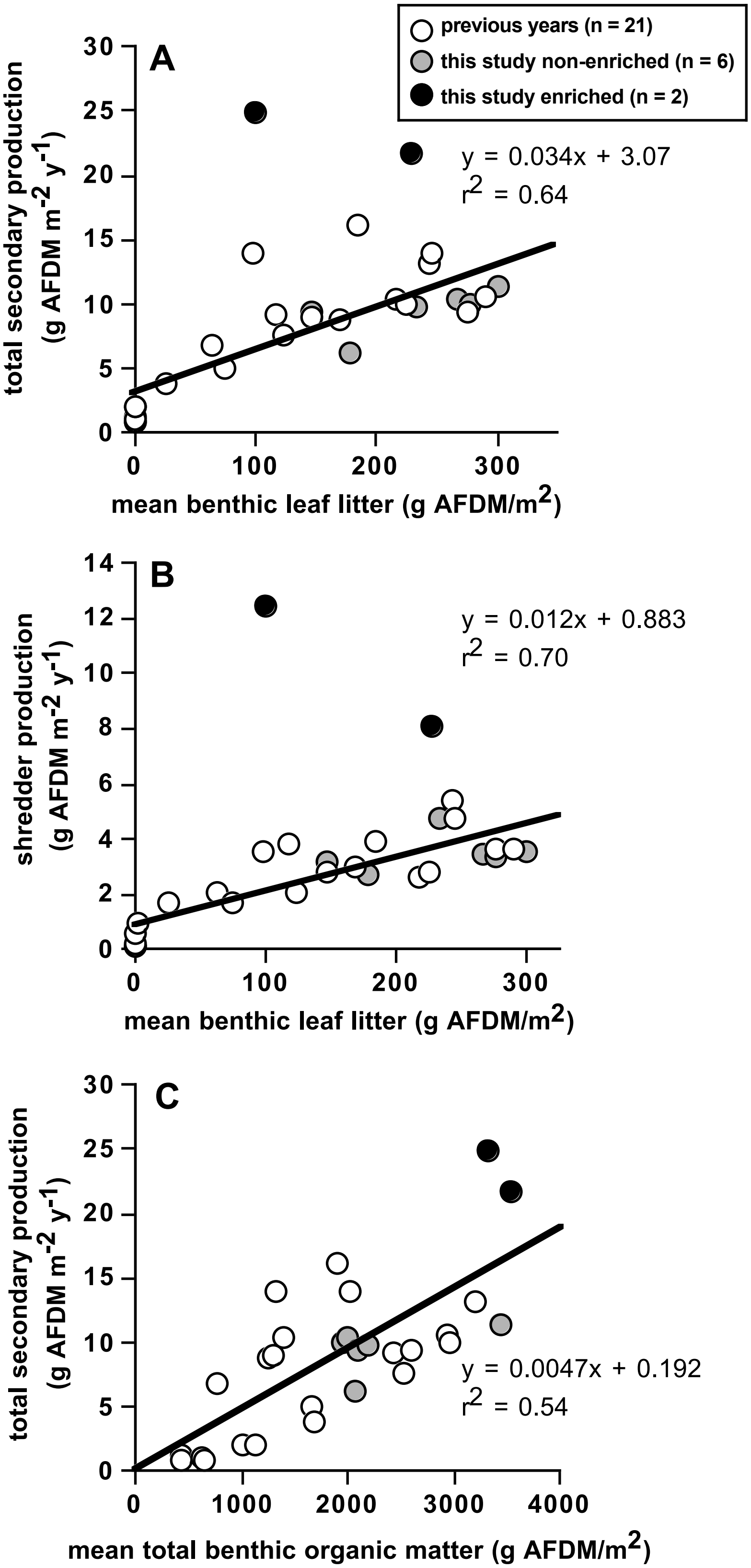

○ total vs. predator production
● primary consumer vs. predator production
y = 0.367x
r^2 = 0.27
y = 0.272x
r^2 = 0.61
Predator secondary production (g AFDM m^{-2} y^{-1})
Primary consumer or total secondary production (g AFDM m^{-2} y^{-1})
0
2
4
6
0
5
10
15
20

Figure 4.8. Long-term relationships between leaf litter standing crop (g AFDM m^{-2}) and (a) total secondary production (g AFDM m^{-2} y^{-1}) and (b) shredder secondary production (g AFDM m^{-2} y^{-1}) in the mixed substrate habitat. (c) Long term relationship between total benthic organic matter and total secondary production (g AFDM m^{-2} y^{-1}). Data include 29 years of production estimates from headwater streams at Coweeta, C53, C54, and C55 (Lughtart and Wallace 1992, Whiles and Wallace 1992, 1995, Wallace et al. 1999). The smallest 7 values are from a previous whole-stream experimental reduction of leaf litter in C55 (Wallace et al. 1999, J. B. Wallace and S. L. Eggert, unpublished data). White circles represent previous data from Coweeta, grey circles represent non-enriched years of this study, and black circles represent two years of experimental nutrient enrichment. The black circles are, from right to left, year 1 of enrichment and year 2 of enrichment.


previous years (n = 21)
this study non-enriched (n = 6)
this study enriched (n = 2)
A
total secondary production (g AFDM m^{-2} y^{-1})
y = 0.034x + 3.07
r^2 = 0.64
0
5
10
15
20
25
30
0
100
200
300
mean benthic leaf litter (g AFDM/m^2)
B
shredder production (g AFDM m^{-2} y^{-1})
y = 0.012x + 0.883
r^2 = 0.70
0
2
4
6
8
10
12
14
0
100
200
300
mean benthic leaf litter (g AFDM/m^2)
C
total secondary production (g AFDM m^{-2} y^{-1})
y = 0.0047x + 0.192
r^2 = 0.54
0
5
10
15
20
25
30
0
1000
2000
3000
4000
mean total benthic organic matter (g AFDM/m^2)

CHAPTER 5

NUTRIENT EFFECTS ON FOOD WEB DYNAMICS OF A DETRITUS-BASED STREAM[1]

---

[1] Cross, W. F., J. B. Wallace, A. D. Rosemond, and K. Suberkropp. To be submitted to *Oecologia*.

Abstract

Quantifying flows of organic matter and materials through food webs is critical for understanding the role of consumers in ecosystems, identifying functionally-key consumer taxa, and for assessing the influence of perturbations, such as nutrient enrichment, on food web dynamics. In detritus-based ecosystems, accurate estimates of organic matter flows require consideration of both detrital and microbial resources; this is especially true in comparative studies where among-system resource quality differs. We estimated the trophic basis of production and constructed explicit flow food webs for consumers (i.e., invertebrates and salamanders) in 2 detritus-based headwater streams. Flow webs were quantified before, and for 2 years during, an experimental whole-stream nutrient enrichment of 1 stream. Throughout the study, consumption by primary consumers was dominated by leaf litter (~50%) and amorphous detritus (~42%), but varied considerably among functional feeding groups. Secondary production of primary consumers was predominantly supported by amorphous detritus (45%), leaf litter (28%), and fungi (19%). Nutrient enrichment had little influence on the diets of consumers, yet total organic matter flows to consumers increased substantially (up to ~290%) during enrichment. Predators consumed a large proportion of prey production (~80%) under non-enriched conditions, but this proportion was reduced considerably (~40% – 50%) during enrichment. Invertebrate consumption of annual detrital and fungal inputs was relatively low, suggesting a potentially important top-down role of invertebrate predators in these streams. Our results suggest that nutrient enrichment had a large effect on the quality and assimilability of detrital resources, permitting increased flows of organic matter despite minimal changes in consumer dietary composition.

Introduction

A major goal of food web ecology is description and quantification of feeding relationships among consumers and resources (e.g., Pimm 1982, Cohen et al. 1993, Polis and Winemiller 1996). Although a variety of methodological approaches have been used to elucidate these relationships, consensus on the most informative approach is elusive, highly debated, and necessarily context-dependent (e.g., Paine 1988, Polis 1991, 1994). Typically, food webs have been described by assessing the presence or absence of feeding links and reporting various characteristics based on these links, such as connectivity, predator-prey ratios, or food chain length (e.g., Cohen et al. 1990, Martinez 1991, Pimm et al. 1991, Thompson and Townsend 1999). While these descriptive methods are informative for understanding food web structure or theoretical stability, they generally lack any quantitative analysis of material flows between consumers and resources or any attempt at understanding the importance of individual feeding links (but see Baird and Ulanowicz 1989). A significant development in this regard has been the use of experimental consumer manipulations to assess per capita interaction strengths (e.g., Paine 1992, Wooton 1994). However, this method is logistically problematic because of the difficulties associated with manipulating all major consumers in the food web or those that are physically too small to manipulate (but see Wootton 1997 and Hall et al. 2000 for an alternative method).

In the past few decades, it has become increasingly apparent that a thorough understanding of food web dynamics requires actual quantification of energy or material flows between consumers and resources (e.g., Polis 1994). One successful method developed for quantifying such flows is the trophic basis of production (Benke and Wallace 1980, 1997). Traditionally, this quantitative approach requires knowledge of consumer secondary production, diet, and assimilation efficiencies for major food types (e.g., leaves, wood, diatoms, animal, etc.). More recently, stable isotope analysis has been used in combination with secondary production estimates to quantify organic matter flows and trophic support of consumers (Hall et al. 2000, Lewis et al. 2001, McCutchan and Lewis 2002). The trophic basis of production method enables an estimation of a) how much of a consumer's production can be attributed to various food resources, and b) how much of each food type is consumed over the duration of the study (i.e., energetic or material flows). Results from recent studies indicate that this technique can shed considerable light on the magnitude and importance of food web linkages (Benke and Wallace 1997, Rosi-Marshall and Wallace 2002), as well as the response of food web dynamics to perturbations such as heavy metal contamination (Carlisle and Clements 2003), nutrient pollution (Shieh et al. 2002), or resource base reduction (Hall et al. 2000, Eggert 2003).

One limitation of the trophic basis of production method has been the reliance of flow estimates on a single literature-based assimilation efficiency for each food category (e.g., Benke and Wallace 1997). For example, leaf litter and animal prey have generally been assigned assimilation efficiency values of 0.1 and 0.8, respectively, based on few actual measurements (but see Eggert 2003). While these estimates are valuable for incorporating inherent food quality differences among major food categories, they are limited in that they do no account for subtle differences in nutritional quality within a given food type. For example, the quality of leaf litter for a consumer may vary depending on the amount of nutrient-rich microbial biomass associated with leaf litter (e.g., Arsuffi and Suberkropp 1984, 1985), leaf nutrient content (e.g., Iversen 1974), or leaf condition (e.g., Hutchens et al. 1997); generally, assimilation efficiency is assumed to be equivalent for all leaves. Such limitations make comparisons of trophic basis of production among systems difficult, especially when differences are manifested in the quality of food types as opposed to their quantity.

In detritus-based stream food webs, the dominant basal resource for invertebrate detritivores is a complex of detritus and associated heterotrophic microbes (i.e., bacteria and fungi) (e.g., Wallace et al. 1999, Hall et al. 2000, 2001). Many invertebrates indiscriminately ingest this resource, and assimilate carbon from both detrital and microbial sources (e.g., Findlay et al. 1984, 1986a, b). However, quantifying the flows of these separate resources to consumers, and the fraction of invertebrate production derived from each resource, is somewhat problematic. Traditional gut content analysis does not accurately assess the contribution of bacteria, fungi, or microbial exopolymers because these components are difficult to see under typical magnification (e.g., 400X), and the bulk of these microbes are bound to, or concealed within, detrital particles (i.e., leaves, amorphous detritus). Although some researchers have successfully used stable isotope tracer additions to assess the contribution of bacteria (Hall and Meyer 1998) or total microbes (e.g., Mulholland et al. 2000) to invertebrate production, realistic estimates of fungal contribution have not been achieved (but see Findlay et al. 1986a, b). Because leaf and wood-associated fungi comprise a significant proportion of the microbial biomass in detritus-based headwater streams (e.g., Findlay et al. 2002), their importance in supporting invertebrate production has likely been underestimated.

In this study we had two main objectives. The first was to explicitly estimate flows of detrital, microbial, and animal food resources to invertebrate consumers in 2 detritus-based headwater stream ecosystems. We aimed to overcome limitations of the trophic basis of production method by combining traditional gut content analyses with empirical measurements (and estimates) of detritus-associated microbial biomass. Our second objective was to assess the effects of experimental nutrient enrichment on trophic basis of production and organic matter flows to stream consumers. We compared food web flows in an un-manipulated reference stream and an adjacent treatment stream that was experimentally enriched with nitrogen (N) and phosphorus (P) for 2 years.

Study sites

This study was conducted in two adjacent headwater streams at the Coweeta Hydrologic Laboratory, Macon County, North Carolina, USA. Coweeta is a large (2185-ha) heavily forested basin located in the Blue Ridge physiographic province of the southern Appalachian Mountains (see Swank and Crossley 1988). Forest vegetation is dominated by mixed hardwoods (primarily oak, maple, and poplar) and a dense understory of *Rhododendron maximum* which shades the streams throughout the year. Headwater streams at Coweeta are extremely heterotrophic, and allochthonous inputs of detritus provide >90% of the energy base for microbial and invertebrate production (Wallace et al. 1997b, Hall et al. 2000). In-stream primary production is low (ca. 4 – 8 g C m^{-2} y^{-1}) and constitutes < 1% of the total carbon entering these streams (Wallace et al. 1997a).

The two streams used for this study drain catchments (C) 53 and 54. These streams have very similar physical and chemical characteristics (Table 5.1), but differ (since July 2000) in their concentrations of dissolved N and P as a result of our experimental nutrient enrichment of C54 (Table 5.1).

Starting in July 2000, nitrogen (NH_4NO_3) and phosphorus (K_2HPO_4 and KH_2PO_4) were dripped continuously into C54 to increase concentrations of dissolved inorganic N and P to ≈ 6-15 X background levels (Table 5.1). Nutrient solution was added along 140 m of C54 with a solar powered metered-dose pump (LMI, Acton, Massachusetts, USA) connected to a streamwater-fed plastic pipe laid along the streambed. The plastic pipe was fitted with garden irrigation valves every 10 m to evenly distribute nutrients along the length of the stream. Stream water nutrient concentrations were held relatively constant across a range of discharge by connecting the pump to a discharge data logger (Isco Inc., Lincoln, NE, USA) located at the base of the stream; the pump was engaged every time a known volume of water (generally 50-100 liters) passed through the downstream flume. Concentrations of $(NO_3 + NO_2)$-N, NH_4-N, and soluble reactive phosphorus (SRP) were measured biweekly at the flume of each study stream (APHA 1998). Nutrient concentrations in the enriched stream (Table 5.1) were well within the range of natural concentrations of streams in the region (Scott et al. 2002), and thus provided a realistic assessment of moderate enrichment effects.

Methods

We used the trophic basis of production method (Benke and Wallace 1980, 1997) to quantify a) the proportion of macro-consumer production derived from dominant basal food resources (i.e., leaf detritus, wood detritus, amorphous detritus, fungi, diatoms, and animals), and b) annual organic matter flows (i.e., mg ash-free dry mass [AFDM] m^{-2} y^{-1}) of these resources to consumers. Trophic basis of production was calculated for 22 primary consumer taxa and 8 predatory taxa in both study streams during three distinct time periods: before nutrient enrichment (September 1998 – August 2000), year 1 of enrichment (September 2000 – August 2001), and year 2 of enrichment (September 2001 – August 2002). The trophic basis of production calculation requires knowledge of invertebrate secondary production, proportion of food resources consumed, and assimilation efficiencies for each food resource. Methods used to obtain these data are described below.

Secondary production

Annual secondary production of invertebrates was estimated from benthic samples collected monthly in both study streams between September 1998 and August 2002 (Cross Chapter 4). Each month, 7 benthic samples were taken from two dominant stream habitats: mixed substrates (i.e., mixed cobble, pebble, gravel, sand, and silt, n = 4) and moss-covered bedrock outcrops (n = 3). Mixed substrate samples were collected at random locations in each stream with a stove pipe core sampler (400 cm^2). Bedrock samples (15 cm x 15 cm) were collected at random locations by brushing and scraping moss and associated particles from a known area (15 cm x 15 cm) into a plastic bag and Hess Sample net (250 μm mesh size) pressed flush against the bedrock.

Benthic samples were brought to the laboratory, refrigerated, and processed within 24 hours. Invertebrates were separated into 2 size fractions using nested 1 mm and 250 μm metal sieves and preserved in formalin solution (6-8%) with Phloxine B dye to aid in sorting. All invertebrates were removed from the > 1 mm fraction by hand picking under a dissecting microscope at 15x magnification. The smaller size fraction (<1mm >250μm) was generally subsampled (1/8 to 1/64 of whole samples) with a sample splitter (Waters 1969), and animals were removed from subsamples with a dissecting microscope at 15x magnification.

All invertebrates were counted, identified, and their body lengths measured to the nearest mm with a dissecting microscope and a graduated stage. Most invertebrates were identified to genus or species level; exceptions included chironomids which were identified as either

Tanypodinae (predatory) or non-Tanypodinae (non-predatory), and some non-insect taxa (e.g., Oligochaeta) which were identified to the ordinal level or higher. Taxa were assigned to functional feeding groups according to Merritt and Cummins (1996) and our knowledge of the local fauna. In this study, all functional feeding group designations (i.e., shredder, collector-gatherer [=gatherers], scraper, collector-filterer [=filterers], and predator) follow Wallace et al. (1999). Biomass (AFDM) of individuals was obtained using previously established length-weight regressions for invertebrates at Coweeta, or, for a few taxa, nearby North Carolina streams (Benke et al. 1999). Annual secondary production was estimated for most taxa using the size-frequency method (Hamilton 1969) corrected for the cohort production interval (CPI, Benke 1979, Wallace et al. 1999). Annual production of non-Tanypodinae chironomids and *Tallaperla* spp. (Plecoptera: Peltoperlidae) was estimated using the community-level instantaneous growth method (Huryn and Wallace 1986, Huryn 1990, Cross Chapter 3). All secondary production estimates were weighed by the relative proportions of mixed substrate and bedrock habitat in each stream. For a complete description of secondary production results see Cross (Chapter 4).

Gut content analysis

Gut content analysis was used to estimate the relative proportions of dominant food items in the guts of consumers (Cummins 1973, Benke and Wallace 1980). Invertebrate consumers were collected roughly seasonally from both streams between July 1999 and July 2002 (n = 10 collection dates). Five to 10 individuals of each of the dominant taxa were collected from each stream on each date, preserved in Kahle’s solution, and brought to the laboratory. Non-Tanypodinae chironomids were removed from collections of fine benthic organic material with a dissecting microscope in the laboratory.

Gut contents were teased from invertebrate guts into water, sonicated, and filtered onto 0.45 μm membrane filters. Filters were air-dried, placed on glass slides, cleared with immersion oil, covered, and sealed with clear nail polish. For most taxa, gut contents of 1-2 individuals were used for each slide. For smaller taxa (e.g., chironomids), we used a greater number of individuals (2-10). During each season, between 1-4 slides were made for each taxon; however, individuals from each taxon were not found during all seasonal collections. Approximately 50 food particles from each slide were identified and their area measured using a phase-contrast compound microscope (400x) equipped with a digital video camera and image analysis software (Image Pro 3.0).

The proportional area of each food type (i.e., leaf, wood, amorphous detritus, diatoms, fungi, and animal) was calculated for each individual. Because leaf- and wood-associated fungi is not accurately accounted for with this method, we further partitioned leaf and wood detrital particles into non-living detritus and fungi. We did this by multiplying leaf and wood percentages in the gut by the average contribution of fungi to leaf or wood biomass (K. Suberkropp, University of Alabama, unpublished data). Before nutrient enrichment, leaf litter and wood biomass contained 3.9% and 2.3% fungi, respectively; during enrichment fungal biomass increased to 5.3% of leaf litter and 10.2% of wood.

It was also necessary to account for differences in the quality of amorphous detritus between the reference and treatment stream during enrichment. We assumed that changes in C:N ratios of epilithon (Cross et al. 2003, Cross Chapter 2) reflected the relative increase in microbial contribution to amorphous detritus. Hence, we adjusted the assimilation efficiency of amorphous detritus (see below) during enrichment to reflect this difference.

Guts of the dominant predators were examined annually (R. O. Hall, University of Wyoming and W. F. Cross, unpublished data), and generally contained 100% animal material. For this study we assumed all flows to predators were animal prey.

Assimilation efficiencies and net production efficiency

Literature-based assimilation efficiencies were used for most food categories (Barlocher and Kendrick 1975, Slansky and Scriber 1982, Benke and Wallace 1980, 1997, Hall et al. 2000). An assimilation efficiency of 80% was used for animal material, 30% for diatoms, and 70% for fungi. Leaf and wood detritus were assigned an assimilation efficiency of 0.05%. This value is lower than the assimilation efficiency generally used for these detritus categories (10%) because we separately assessed flows of non-living leaf or wood detritus and detritus-bound fungi. An assimilation efficiency of 10% was used for amorphous detritus during non-enriched conditions (e.g., Benke and Wallace 1980, Hall et al. 2000). During nutrient enrichment we used an assimilation efficiency of 19% for amorphous detritus in the treatment stream because the C:N ratio of epilithon was 1.9-fold lower (i.e., nitrogen was roughly 1.9-fold higher) in the treatment stream than the reference stream during enrichment (Cross et al. 2003). Although nutrient enrichment may affect epilithic elemental ratios without altering the total microbial biomass (e.g., Stelzer and Lamberti 2001), this is only likely to occur when epilithon contains a large proportion of algae. In these streams, algal contribution to epilithic biomass is extremely low (Greenwood 2004), and changes in elemental content are more likely to reflect changes in microbial biomass. A net production efficiency of 0.4 was used for all calculations (e.g., Benke and Wallace 1997).

Trophic basis of production

Secondary production attributed to each food type, and the total amount of each food type consumed each year (i.e., flows) were quantified following the calculations of Benke and Wallace (1980, 1997). For each taxon, seasonal proportions of food types during a given year were averaged to obtain annual average proportions. The relative fraction of a consumer's annual production attributed to each food type was calculated as:

$$F_i = (G_i \text{ x } AE_i \text{ x } NPE)$$

where F_i = fraction of production attributed to food type i, G_i = percent of food type i in consumer's gut (mean annual percent), AE_i = assimilation efficiency for food type i, and NPE = net production efficiency. Each relative fraction was converted to a percentage of all food types (PF_i) as:

$$PF_i = F_i / \sum F_{(i+j+k \ldots n)} \text{ x } 100$$

To estimate the actual amount of consumer production attributed to these food types, percentages (PF_i) were multiplied by annual consumer secondary production (mg AFDM m^{-2} y^{-1}). Lastly, the amount of each food type consumed by a consumer was calculated as:

$$FC_i = (PF_i \text{ x } P) / (AE_i \text{ x } NPE)$$

where FC_i = amount of food type i consumed (mg AFDM m^{-2} y^{-1}), and P = secondary production of consumer (mg AFDM m^{-2} y^{-1}).

Statistical analyses

We compared consumer diet overlap between streams during each treatment period, and among treatment periods within each stream, for all functional feeding groups using proportional similarity analysis (Whittaker 1975). Proportional similarities were calculated for 3 separate time periods: before enrichment, year 1 of enrichment, and year 2 of enrichment as:

$$PS = \sum_{i = 1 \text{ to } n} (\text{between-stream } \min_i)$$

where i = a single food category (e.g., leaves), n = total number of food categories, and between-stream $\min_i$ = the minimum proportion of food type i between the reference and enriched streams. For this analysis we used the weighted-average proportions (weighted by secondary production) of food items across all taxa within a given functional feeding group. Mean and standard error proportional similarities for each time period were calculated from multiple collection dates within a time period. A PS of 1.0 is indicative of complete diet overlap.

Invertebrate consumption versus resource productivity

We calculated the average proportion of food resource productivity (i.e., inputs) consumed by invertebrates during each time period of the study (i.e., pretreatment and years 1 and 2 of enrichment). Average annual inputs of leaf litter (510 g AFDM m^{-2} y^{-1}) and small woody debris (160 g AFDM m^{-2} y^{-1}) were taken from long-term studies of headwater streams at Coweeta (Wallace et al. 1995, 2000). Fungal production per gram leaf litter was measured during each year of the present study (K. Suberkropp, University of Alabama, unpublished data); these values were multiplied by the average leaf litter standing crop during each year (Cross Chapter 4). We assumed that fungal production in fine organic sediments was negligible in comparison to that on leaf litter, and was therefore ignored (K. Suberkropp, unpublished data, Findlay et al. 2002). In lieu of estimates of fine benthic organic matter (FBOM) inputs, we used values of FBOM storage (Cross Chapter 4) to assess the proportion of this storage consumed by invertebrates.

Results

We examined the diet and estimated trophic basis of production for 22 primary consumer taxa. These taxa comprised 89% of the primary consumer community based on average annual secondary production. In addition, we estimated flows of prey to 8 dominant predatory taxa which comprised 76% of predator production. On average, our complete food webs represented 86% of the entire community secondary production.

Gut contents

As a group, gut contents of primary consumers in both streams were dominated by leaf litter (50%) and amorphous detritus (42%). Wood and fungi contributed 3.5% and 2.5% respectively, and the remaining 2% was split among diatoms and animal material.

Gut contents of individual functional feeding groups exhibited strong differences (Figure 5.1). Shredder guts were dominated by leaf litter (77%) with smaller contributions from amorphous detritus (12%), wood (6%), and fungi (4%). Diatoms and animal prey were rarely encountered in shredder guts. In contrast, gut contents of gatherers were dominated by amorphous detritus (76%) with the remainder consisting primarily of leaf litter (21%) (Figure 5.1). Filterer taxa had a more diverse diet with significant contributions from leaf litter (36%), amorphous detritus (46%), and animal prey (11%). Wood and fungi contributed an additional

4% and 2% to filterer gut contents respectively. Scrapers consumed primarily amorphous detritus (81%), and smaller amounts of diatoms (11%) and leaf litter (6%) (Figure 5.1).

Within each functional feeding group, noteworthy dietary differences existed among individual taxa (Appendix C). For the shredders, gut contents of *Leuctra* spp. and *Anchytarsus* sp. contained much higher amounts of amorphous detritus (45%) and lower amounts of leaf litter (47%) than the other dominant shredder taxa (i.e., *Fattigia pele*, *Lepidostoma* spp., *Pycnopsyche* spp., *Tallaperla* spp., and *Tipula* spp.). Among gatherers, Chironomidae, *Paraleptophlebia* sp., *Serratella* sp., and *Stenonema* sp. had gut contents dominated by amorphous detritus (84%). Gut contents of *Amphinemura* sp. and Oligochaetae were more evenly split among amorphous detritus and leaf litter, while Sciaridae guts were dominated by leaf litter (78%). Among filterers, animal material comprised a significant proportion of the diet of *Parapsyche cardis* (17%) and *Diplectrona* spp. (5%). The remainder for these taxa was split among leaf litter and amorphous detritus, with fungi and diatoms contributing very little to gut contents. In contrast, the filterers Simuliidae and *Wormaldia* spp. consumed primarily amorphous detritus (~88-98%). Scraper diets were consistently dominated by amorphous detritus (83%), however, small amounts of leaf litter (7%) and diatoms (8%) were found in the guts of *Baetis* sp., *Ectopria* sp., and *Epeorus* sp.. Diatoms made up a larger proportion (25%) of the diet of *Neophylax* sp. (Appendix C).

Nutrient enrichment had very little effect on the proportions of food resources in the guts of consumers (Figure 5.1). Proportional similarities of gut contents for each functional feeding group were consistently high between treatment periods within each stream (reference: 0.74 – 0.92, treatment: 0.63 – 0.92, Table 5.2) and between the reference and treatment streams during each time period (0.77 – 0.95, Table 5.2). There was, however, a small increase (~2%) in the proportion of fungi in the guts of shredders in the enriched stream (Figure 5.1). The lowest proportional similarity value (0.63) corresponded to higher amounts of leaf litter in the guts of gatherers in the enriched stream during the second year of enrichment (45%) versus the pretreatment period (12%). Other minor differences among treatment periods or between streams were idiosyncratic and did not appear to be related to nutrient enrichment effects (Figure 5.1).

Contribution of food resources to secondary production

Because food types are assimilated at different efficiencies, the fraction of invertebrate secondary production attributable to different food resources (i.e., trophic basis of production) contrasted with the raw proportions of food types in the guts (Appendix D). Overall, 32% of consumer production was attributed to amorphous detritus, while 31% and 20% of production was attributed to animal prey and leaf litter, respectively (Table 5.3). An additional 14% of consumer production was derived from fungi, and only ~3% came from wood detritus and diatoms. When only primary consumers were included in these estimates, 45% of total production was attributed to amorphous detritus, 28% was attributed to leaf litter, 19% was attributed to fungi, and only ~8% came from wood, diatoms, and animal prey (Table 5.3).

Nutrient enrichment had little effect on total invertebrate trophic basis of production. The percent contribution of food resources did not differ widely between the reference and treatment streams (Table 5.3). One exception was during year 1 of enrichment in which amorphous detritus contributed more and animal prey contributed less to consumer production in the treatment stream relative to the reference stream (Table 5.3).

On average, production of shredders in both streams was primarily attributed to leaf litter (46%), fungi (31%), and amorphous detritus (17%) (Table 5.4). Gatherer and scraper production were predominantly supported by amorphous detritus (81% and 73%, respectively), although ~23% of scraper production was attributed to diatoms (Table 5.4). Production of filterer taxa was largely attributed to amorphous detritus (38%) and animal prey (41%). The contribution of animal prey to filterer production declined in both streams during the 2 years of enrichment.

Although nutrient enrichment had little effect on invertebrate trophic basis of production, the absolute amount of production supported by food resources increased greatly in the treatment stream during enrichment (Figure 5.2, Table 5.4). For example, the amount of shredder production attributed to fungi increased from 603 mg AFDM m^{-2} y^{-1}before enrichment to ca. 2194 mg AFDM m^{-2} y^{-1}. Among gatherers, production attributable to amorphous detritus increased from ca. 1293 mg AFDM m^{-2} y^{-1} before enrichment to ca. 3404 mg AFDM m^{-2} y^{-1}. Similar increases in trophic support were seen for most food types among all functional feeding groups during enrichment (Figure 5.2).

Organic matter flows

Using trophic basis of production values and resource-specific assimilation efficiencies, we calculated the absolute amount of each resource consumed by the entire invertebrate assemblage, individual functional feeding groups, and all dominant taxa during each treatment period. Overall, leaf (52%) and amorphous detritus (35%) dominated resource flows to the invertebrate community, followed by animal prey (5%), wood detritus (4%), fungi (3%), and diatoms (~1%) (Table 5.3). Absolute flows from the dominant basal resources to invertebrates were very similar in the reference stream and the treatment stream before enrichment (Tables 5.3

and 5.5). This was due to similar gut contents and secondary production values in both streams under non-enriched conditions. In contrast, during nutrient enrichment organic matter flows to consumers were ca. 1-10X higher in the treatment stream than in the reference stream (Tables 5.3 and 5.5). This difference was primarily driven by increased consumer secondary production in the treatment stream (Cross Chapter 4).

Flows of basal resources differed greatly among functional feeding groups, and key taxa dominated consumption within each group. Organic matter flows to shredders under non-enriched conditions (i.e., reference stream and pretreatment in the treatment stream) were dominated by consumption of leaf litter by *Pycnopsyche* spp. and *Tipula* spp. (Table 5.5, Figure 5.3). These flows were dominant because of low assimilation efficiency of leaves, and relatively high secondary production of these taxa. Nutrient enrichment greatly increased the magnitude of flows to shredders, with increased flows of detritus (particularly leaf litter and amorphous detritus) as well as microbes (i.e., fungi) (Table 5.5, Figure 5.3). Flows of amorphous detritus increased considerably during the second year of enrichment despite the higher assimilation efficiency assigned to this nutrient-enriched resource. Flows to other taxa, such as *Leuctra* spp. and *Lepidostoma* spp. became more prominent with nutrient enrichment (Figure 5.3).

Resource flows to gatherer taxa under non-enriched conditions were dominated by flows of amorphous detritus (Table 5.5, Figure 5.4). Chironomidae and Oligochaetae dominated gatherer consumption because of their relatively high production. However, these taxonomic groups are highly aggregated, and flows to individual species would be considerably lower. During nutrient enrichment, total flows to gatherers increased ~56% relative to pretreatment values; much of this increased consumption was of amorphous detritus and leaf litter (Table 5.5, Figure 5.4). Flows of leaf litter to *Amphinemura* sp. increased substantially during enrichment.

Flows to filterer and scraper taxa were low in comparison to other primary consumer groups (Table 5.5, Figures 5.5 and 5.6). Among filterers, dominant flows were from amorphous detritus, leaf litter, and animal prey to *Diplectrona* spp, and *Parapsyche cardis* (Figure 5.5). Nutrient enrichment slightly increased the magnitude of these flows (ca. 25%) (Table 5.5, Figure 5.5). Total flows to scrapers (predominantly amorphous detritus) were extremely low because of their minimal secondary production (Table 5.5, Figure 5.6). Nutrient enrichment increased flows to scrapers, with the largest increases occurring from amorphous detritus to *Epeorus* sp., and *Neophylax* sp. (Figure 5.6).

Flows of animal prey to predators were moderate because of the relatively high assimilation efficiency of animal material (Figure 5.7). The largest flows of prey during reference conditions were to Ceratopogonidae, *Hexatoma* spp., and *Lanthus* sp., although other predators consumed significant amounts of prey (Figure 5.7). On average, 82% of total invertebrate secondary production was consumed by predators during reference years. During nutrient enrichment, total flows nearly doubled, and consumption by *Rhyacophila* spp. became more prominent (Figure 5.7). Interestingly, the total amount of prey production consumed by predators declined during enrichment to 64% during the first year (Enr-1) and 47% during the second year (Enr-2).

Invertebrate consumption vs. basal resource production

We calculated the percentage of food resource productivity (i.e., inputs) consumed by invertebrates during the study. Our estimates demonstrate that during non-enriched years, invertebrates consumed a relatively small proportion of the annual inputs of leaf litter (15%), wood (4%), fungi (2%), FBOM (8%), and diatoms (4%) (Figure 5.8). In contrast, a relatively large proportion of available prey was consumed by predators (79%) during non-enriched years. During nutrient enrichment, consumption of leaf litter increased in the treatment stream to an average of ~32% of annual leaf inputs. In addition, consumption of diatom production went up to ~ 54% of inputs. Consumption of prey declined during the enrichment to ~56% of prey secondary production.

Discussion

Quantifying material flows through food webs is a critical step towards understanding dominant pathways of energy and nutrients through consumers, and for identifying key players in the cycling and processing of these materials (e.g., Benke and Wallace 1997). In addition, this approach can provide insight into mechanisms responsible for changes to ecosystem functions (e.g., secondary production) resulting from natural or human-induced perturbations. We utilized the trophic basis of production method to follow organic matter flows to consumers in a reference stream and an experimentally-enriched stream. Our intent was to account for nutrient-induced differences in food quality by a) utilizing empirical measurements of fungal biomass, and b) estimating between-stream differences in the quality of amorphous detritus. Importantly, we demonstrated that nutrient enrichment had a major positive effect on resource flows to consumers despite a lack of evidence for dietary shifts or changes in invertebrate trophic basis of production. These results lend support to the notion that nutrient-induced changes to detrital food quality are more complex than simple changes in biomass of detritus-associated microbes.

The importance of non-living detritus versus detritus-bound microbes to the nutrition of consumers has long been a topic of interest to aquatic ecologists (e.g., Cummins 1973). Findlay et al. (1986a ,b) demonstrated that leaf-associated fungi comprised a moderate component of the total carbon respired by a freshwater isopod (up to 57%) and a stonefly (25%). In contrast, bacteria associated with leaf litter were shown to be much less important for consumer nutrition, comprising only ~1 - 5% of total carbon respired by the isopod, the stonefly, and a cranefly larva (Findlay et al. 1986a, b). These results imply that although some nutrition is derived from microbes, a large proportion of respired carbon must necessarily come from the non-living leaf substrate. Indeed, some detritivores have adapted the capability to assimilate recalcitrant detrital compounds through high gut pH or microbial gut symbionts (Martin et al. 1981, Sinsabaugh et al. 1985). Our estimates of fungal contribution to the production of leaf-eating taxa (i.e., shredders) were comparable (ca. 31%) to estimates of Findlay et al. (1986 a, b), but considerably higher than most studies which used traditional gut content analysis (e.g., Smock and Roeding 1986, Benke and Wallace 1997, Rosi-Marshall and Wallace 2002). Knowledge of fungal biomass on leaf litter and wood throughout the study allowed us to account for the fungi which is typically not detected with standard gut content analysis. Our results, along with previous laboratory experiments (Findlay et al. 1986 a,b), likely provide a more realistic view of the importance of fungi to the nutrition of leaf-eating consumers.

The importance of fungi to stream consumers, however, goes far beyond their direct contribution to the diet of consumers. Aquatic hyphomycetes play a prominent role in the physical and chemical breakdown of leaf litter, and make large particles more available for further breakdown and consumption by bacteria and invertebrates (e.g., Barlocher 1985, Suberkropp 1992). In addition, fungal exoenzymes are critical for degrading complex carbohydrates, rendering these compounds more available for assimilation by invertebrates (e.g., Sinsabaugh et al. 1985). Leaf-associated fungi may also stimulate invertebrate feeding by increasing leaf palatability and affecting feeding preferences (Arsuffi and Suberkropp 1984, 1985). In forested headwater streams where allochthonous inputs of leaf litter fuel the majority of secondary production (e.g., Hall et al. 2000), fungi potentially play a 'keystone' role in driving the processing and consumption of organic material. Experimental manipulation of the presence of fungi in these systems (A. D. Rosemond, study in progress) will be instrumental in evaluating their role as a keystone component of headwater stream food webs.

In our study, the diets of most non-shredder primary consumer taxa contained a large percentage of amorphous detritus, and we estimated that a significant component of their production (~64%) was derived from this resource. The dominance of amorphous detritus as a food resource has been consistently reported for a wide range of taxa and aquatic ecosystem types (e.g., Smock and Roeding 1986, Wallace et al. 1987, Carlough 1994, Benke and Wallace 1997, Hall et al. 2000, Rosi-Marshall and Wallace 2002, Hart and Lovvorn 2003, Benstead and Pringle 2004). However, understanding the trophic significance of this resource has been somewhat elusive because of difficulty in quantifying the contribution of microbial vs. non-microbial components. Studies that have explicitly examined the contribution of amorphous detritus-associated bacteria have indeed shown that a large proportion of assimilated carbon may come from the microbial component. Edwards (1987) and Edwards and Meyer (1990) labeled sestonic bacteria with tritiated thymidine to show that snag-dwelling mayflies and blackflies in a large blackwater river derived between 20 and 93% of their carbon from bacteria. Moreover, Hall and Meyer (1998) used a whole-stream ^{13}C-tracer addition to demonstrate that a large fraction of invertebrate carbon was derived from bacteria (and exopolymers) associated with amorphous detritus. In our study, it is likely that production attributed to amorphous detritus is synonymous with microbial-based production. Because of the prominence of amorphous detritus in the guts of stream consumers, and the uncertainty of its composition and origins (e.g., Carlough 1994), further research on the dynamics, composition, and importance of amorphous detritus will greatly improve our general understanding of stream trophic dynamics and basis of production.

Overall, diatoms were of minimal significance to the trophic basis of primary consumers at Coweeta. This is not surprising considering the generally low levels of primary production and epilithic chlorophyll *a* at Coweeta and in other forested headwater streams (Haines 1981, Webster et al. 1983, Hall et al. 2001, Bernhardt and Likens 2004, Greenwood 2004). Nonetheless, because of this low level of primary production and a relatively high secondary production of some taxa that included small amounts of diatoms in their diets, we estimated that invertebrates were capable of consuming a significant proportion (up to 72%) of the annual inputs (Figure 5.8). This result underscores the paucity of algal resources available for consumers at Coweeta. Among epilithic specialists (i.e., scrapers), however, we found that diatoms supported a significant proportion (~25%) of secondary production. In fact, during spring months, gut contents of *Neophylax* sp. contained up to 90% diatoms (Appendix C). Mayer and Likens (1987) similarly demonstrated that production of the scraper *Neophylax aniqua* was predominantly (~75%) supported by algae in a small stream in Hubbard Brook, New Hampshire. In streams at Coweeta, scraper biomass tends to peak during each spring (Cross Chapter 4) suggesting the life-histories of scrapers are adapted to intra-annual variation in food supply.

In contrast to streams at Coweeta, others have shown a strong dependence of whole-assemblage consumer production on autochthonous primary production despite shaded and/or heterotrophic conditions. McCutchan and Lewis (2002) combined secondary production estimates with stable isotope analysis to demonstrate that ca. 40% of invertebrate production in predominantly shaded reaches of Colorado mountain streams was derived from algae. These small streams in Colorado, however, are much less shaded than those at Coweeta which have a thick understorey of *Rhododendon maximum*.

Nutrient enrichment had only minor effects on consumer diets, yet there were large increases in organic matter flows as a result of elevated secondary production. This suggests that enrichment led to changes in the food base that positively affected consumer growth or survival beyond simple changes in microbial content of detritus. For heuristic purposes, we examined whether increased production of shredders in the enriched stream (Cross Chapter 4) could be accounted for by simple differences in microbial content of leaf litter between the reference and treatment stream. Microbial contribution to leaf litter biomass increased 1.4% in the treatment stream (from 3.9% - 5.3%) during enrichment. If we assume a 5% assimilation efficiency for leaf detritus and a 100% (theoretical maximum) assimilation efficiency for microbes, the total amount of material assimilated by consumers would only increase by 15% in the treatment stream. However, organic matter flows (and production) of shredders increased, on average, 162% with enrichment (Figure 5.3, Chapter 4). This large discrepancy suggests that factors other than actual differences in microbial biomass led to increased production and organic matter flows to consumers. In the treatment stream, it is likely that a greater proportion of benthic leaf material was ‘primed’ for invertebrate consumption by microbial degradation of recalcitrant leaf compounds. This could have a strong positive effect on leaf palatability and/or assimilation efficiency of the non-living leaf substrate by making leaf carbon more available (i.e., digestible) to invertebrates (e.g., Sinsabaugh et al. 1985). Moreover, enrichment caused significant changes in the nutrient content and C:nutrient ratios of detritus (Cross et al. 2003), reducing elemental imbalances and potentially contributing to increased secondary production (e.g., Sterner et al.

1998). Nonetheless, pinning down the actual mechanism responsible for increased secondary production is difficult because production is influenced by a suite of factors including individual growth, survivorship, and reproduction, which all could have been positively affected by microbial-induced changes of stream detritus.

We demonstrated that invertebrates consumed a relatively small proportion of the annual inputs of detrital and fungal food resources (Figure 5.8) throughout the study. Excluding leaf litter during enrichment and diatoms, invertebrates generally consumed less than 10-15% of annual food resource inputs. Using a similar energetic budget approach, Huryn (1998) demonstrated that only a small proportion of primary production was consumed by invertebrates in a New Zealand trout stream. In this case, nearly all invertebrate production was necessary to meet the energetic requirements of trout, and the surplus of primary production was presumably the result of cascading top-down control by trout. In streams at Coweeta, a similarly high proportion of invertebrate production (~80%) was necessary to meet the requirements of primarily invertebrate predators (Figure 5.8). This suggests that invertebrate predators have potentially strong cascading top-down effects on detrital resources (see Malmqvist 1993). Interestingly, the increased consumption of leaf litter during enrichment (up to ~38%) coincided with a decreased proportion of prey consumed by predators (down to ~47%). Such an energetic budget approach (e.g., Allen 1951, Huryn 1998) may be useful for examining trophic dynamics in systems where experimental manipulation of food web components is not logistically possible.

In summary, we have shown that the effects of nutrient enrichment on a detritus-based stream food web were manifested as changes to total organic matter flows with little effect on consumer diet or trophic basis of production. Our results suggest that elevated invertebrate production (Cross Chapter 4) in response to nutrient enrichment was brought on by microbially-mediated changes to organic matter (i.e., increased carbon assimilability, or release of stoichiometric constraints), as opposed to direct changes in the consumption and assimilation of microbes. Future research focused on species-specific nutritional requirements will undoubtedly add to our understanding of community-level responses to enrichment.

Acknowledgements

We thank Catherine Evans and James Norman for tireless assistance preparing slides. We also thank Sue Eggert, Jennifer Greenwood, Bob Hall, and Heidi Wilcox for help with invertebrate collections. Emma Rosi-Marshall and Jon Benstead gave valuable advice about identifying and digitizing gut contents. Funding was provided by NSF (DEB – 9806610) to Amy D. Rosemond, J. Bruce Wallace, Keller Suberkropp, and Pat J. Mulholland.

Literature Cited

Allen, K. R. 1951. The Horokiwi stream: a study of a trout population. Bull. Mar. New Zealand. Fish 10:1-238.

American Public Health Association. 1998. Standard Methods for the Examination of Water and Wastewater, 20th edn. American Public Health Association.

Arsuffi, T. L. and K. Suberkropp. 1984. Leaf processing capabilities of aquatic hyphomycetes - interspecific differences and influence on shredder feeding preferences. Oikos 42:144-154.

Arsuffi, T. L. and K. Suberkropp. 1985. Selective feeding by stream caddisfly (Trichoptera) detritivores on leaves with fungal-colonized patches. Oikos 45:50-58.

Baird, D. and R. E. Ulanowicz. 1989. The seasonal dynamics of the Chesapeake Bay ecosystem. Ecological Monographs 59:329-364.

Barlocher, F. 1985. The role of fungi in the nutrition of stream invertebrates. Botanical Journal of the Linnean Society 91:83-94.

Barlocher, F., and B. Kendrick. 1975. Assimilation efficiency of *Gammarus pseudolimnaeus* (Amphipoda) feeding on fungal mycelium or autumn-shed leaves. Oikos 26:55-59.

Benke, A. C. 1979. A modification of the Hynes method for estimating secondary production with particular significance for multivoltine populations. Limnology and Oceanography 24:168-171.

Benke, A.C., A. D. Huryn, L. A. Smock, and J. B. Wallace. 1999. Length-mass relationships for freshwater macroinvertebrates in North America with particular reference to the southeastern United States. Journal of the North American Benthological Society 18: 308–343.

Benke, A. C. and J. B. Wallace. 1980. Trophic basis of production among net-spinning caddisflies in a southern Appalachian stream. Ecology 61:108-118.

Benke, A. C. and J. B. Wallace. 1997. Trophic basis of production among riverine caddisflies: implications for food web analysis. Ecology 78:1132-1145.

Benstead, J. P and C. M. Pringle. 2004. Deforestation alters the resource base and biomass of endemic stream insects in eastern Madagascar. Freshwater Biology 49:490-501.

Berhardt, E. S. and G. E. Likens. 2004. Controls on periphyton biomass in heterotrophic streams. Freshwater Biology 49:14-27.

Carlisle, D. M. and W. H. Clements. 2003. Growth and secondary production of aquatic insects along a gradient of Zn contamination in Rocky Mountain streams. Journal of the North American Benthological Society 22:582-597.

Carlough, L. A. 1994. Origins, structure, and trophic significance of amorphous seston in a blackwater river. Freshwater Biology 31:227-237.

Cohen, J. E., R. A. Beaver, S. H. Cousins, D. L. DeAngelis, L. Goldwasser, K. L. Heong, R. D. Holt, A. J. Kohn, J. H. Lawton, N. Martinez, R. O'Malley, L. M. Page, B. C. Patten, S. L. Pimm, G. A. Polis, M. Rejmanek, T. W. Schoener, K. Schoenly, W. G. Sprules, J. M. Teal, R. E. Ulanowicz, P. H. Warren, H. M. Wilber, and P. Yodzis. 1993. Improving food webs. Ecology 74:252-258.

Cohen, J. E., F. Briand, and C. M. Newman. 1990. Community Food Webs: Data and Theory. Springer, Berlin.

Cummins, K. W. 1973. Trophic relations of aquatic insects. Annual Review of Entomology 18:183-206.

Edwards, R. T. 1987. Sestonic bacteria as a food source for filtering invertebrates in 2 southeastern blackwater rivers. Limnology and Oceanography 32:221-234.

Edwards, R. T. and J. L. Meyer. 1990. Bacterivory by deposit-feeding mayfly larvae (*Stenonema* spp.). Freshwater Biology 24:453-462.

Eggert, S. L. 2003. Resource use by detritivorous macroinvertebrates in southern Appalachian headwater streams. Dissertation, University of Georgia, Athens, GA, USA.

Findlay, S., J. L. Meyer, and P. J. Smith. 1984. Significance of bacterial biomass in the nutrition of a freshwater isopod (Lirceus sp.). Oecologia 63:38-42.

Findlay, S., J. L. Meyer, and P. J. Smith. 1986a. Contribution of fungal biomass to the diet of a fresh-water isopod (*Lirceus* Sp). Freshwater Biology 16:377-385.

Findlay, S., J. L. Meyer, and P. J. Smith. 1986b. Incorporation of microbial biomass by *Peltoperla* sp. (Plecoptera) and *Tipula* sp. (Diptera). Journal of the North American

Benthological Society 5:306-310.

Findlay, S., J. Tank, S. Dye, H. M. Valett, P. J. Mulholland, W. H. McDowell, S. L. Johnson, S. K. Hamilton, J. Edmonds, W. K. Dodds, and W. B. Bowden. 2002. A cross-system comparison of bacterial and fungal biomass in detritus pools of headwater streams. Microbial Ecology 43:55-66.

Greenwood, J. 2004. The response of heterotrophic and autotrophic resources to a long-term nutrient enrichment in an Appalachian headwater stream ecosystem. Dissertation. University of Georgia, Athens, GA, USA.

Hains, J. J. 1981. The response of stream flora to watershed perturbation. MS Thesis. Clemson University, Clemson, SC.

Hall, R. O., G. E. Likens, and H. M. Malcom. 2001. Trophic basis of invertebrate production in 2 streams at the Hubbard Brook Experimental Forest. Journal of the North American Benthological Society 20:432-447.

Hall, R. O. and J. L. Meyer. 1998. The trophic significance of bacteria in a detritus-based stream food web. Ecology 79:1995-2012.

Hall, R. O. J., J. B. Wallace, and S. L. Eggert. 2000. Organic matter flow in stream food webs with reduced detrital resource base. Ecology 81:3445-3463.

Hamilton, A. L. 1969. On estimating annual production. Limnology and Oceanography 14:771-782.

Hart, E. A. and J. R. Lovvorn. 2003. Algal vs. macrophyte inputs to food webs of inland saline wetlands. Ecology 84:3317-3326.

Huryn, A. D. 1990. Growth and voltinism of lotic midge larvae: patterns across an Appalachian mountain basin. Limnology and Oceanography 35:339-351.

Huryn, A. D. 1998. Ecosystem-level evidence for top-down and bottom-up control of production in a grassland stream system. Oecologia **115**:173-183.

Huryn, A. D. and J. B. Wallace. 1986. A method for obtaining in situ growth rates of larval Chironomidae (Diptera) and its application to studies of secondary production. Limnology and Oceanography 31:216-222.

Hutchens, J. J., E. F. Benfield, and J. R. Webster. 1997. Diet and growth of a leaf-shredding caddisfly in southern Appalachian streams of contrasting disturbance history. Hydrobiologia 346:193-201

Iversen, T. M. 1974. Ingestion and growth in *Sericostoma personatum* (Trichoptera) in relation to the nitrogen content of ingested leaves. Oikos 25:278-282.

Lewis, W. M., S. K. Hamilton, M. A. Rodriguez, J. F. Saunders, and M. A. Lasi. 2001. Foodweb analysis of the Orinoco floodplain based on production estimates and stable isotope data. Journal of the North American Benthological Society 20:241-254.

Malmqvist, B. 1993. Interactions in stream leaf packs – effects of a stonefly predator on detritivores and organic matter processing. Oikos 66:454-462.

Martin, M. M., J. J. Kukor, J. S. Martin, D. L. Lawson, and R. W. Merritt. 1981. Digestive enzymes of larvae of 3 species of caddisflies (Trichoptera). Insect Biochemistry 11:501-505.

Martinez, N. D. 1991. Artifacts or attributes? Effects of resolution on the Little Rock Lake food web. Ecological Monographs 61:367-392.

McCutchan, J. H. and W. M. Lewis. 2002. Relative importance of carbon sources for macroinvertebrates in a Rocky Mountain stream. Limnology and Oceanography 47:742-752.

Merritt, R.W. and K. W. Cummins. 1996. An Introduction to the Aquatic Insects of North America. 3rd edn., Kendall/Hunt Publishing Co., USA.

Mulholland, P. J., J. L. Tank, D. M. Sanzone, W. M. Wollheim, B. J. Peterson, J. R. Webster, and J. L. Meyer. 2000. Food resources of stream macroinvertebrates determined by natural abundance stable C and N isotopes and a 15N tracer addition. Journal of the North American Benthological Society 19:145-157.

Paine, R. T. 1988. Food webs - road maps of interactions or grist for theoretical development. Ecology 69:1648-1654.

Paine, R. T. 1992. Food-web analysis through field measurement of per capita interaction strength. Nature 355:73-75.

Pimm, S. L. 1982. Food Webs. Chapman and Hall, New York.

Pimm, S. L., J. H. Lawton, and J. E. Cohen. 1991. Food web patterns and their consequences. Nature 350:669-674.

Polis, G. A. 1991. Complex trophic interactions in deserts: an empirical critique of food-web theory. American Naturalist 138:123-155.

Polis, G. A. 1994. Food webs, trophic cascades and community structure. Australian Journal of Ecology 19:121-136.

Polis, G. A. and K. O. Winemiller. 1996. Food webs: Integration of Pattern and Dynamics. Chapman and Hall, New York.

Rosi-Marshall, E. J. and J. B. Wallace. 2002. Invertebrate food webs along a stream resource gradient. Freshwater Biology 47:129-141.

Scott, M. C., G. S. Helfman, M. E. McTammany, E. F. Benfield, and P. V. Bolstad. 2002. Multiscale influences on physical and chemical stream conditions across blue ridge landscapes. Journal of the American Water Resources Association 38:1379-1392.

Shieh, S-H., J. V. Ward, and B. C. Kondratieff. 2002. Energy flow through macroinvertebrates in a polluted plains stream. Journal of the North American Benthological Society 21:660-675.

Sinsabaugh, R. L., A. E. Linkins, and E. F. Benfield. 1985. Cellulose digestion and assimilation by 3 leaf-shredding aquatic insects. Ecology 66:1464-1471.

Slansky, F. and J. M. Scriber. 1982. Selected bibliography and summary of quantitative food utilization by immature insects. Bulletin of the Entomological Society of America 28:43-55.

Smock, L. A. and C. E. Roeding. 1986. The trophic basis of production of the macroinvertebrate community of a southeastern U.S.A. blackwater stream. Holarctic Ecology 9:165-174.

Stelzer, R. S. and G. A. Lamberti. 2001. Effects of N:P ratio and total nutrient concentration on stream periphyton community structure, biomass, and elemental composition. Limnology and Oceanography 46:356-367.

Sterner, R. W., J. Clasen, W. Lampert, and T. Weisse. 1998. Carbon: phosphorus stoichiometry and food chain production. Ecology Letters 1:146-150.

Suberkropp, K. 1992. Interactions with invertebrates. *In*: Barlocher, F. (ed). The Ecology of Aquatic Hyphomycetes. Springer-Verlag. New York.

Swank, W.T. and D.A. Crossley. 1988. Forest Hydrology and Ecology at Coweeta. Ecological Studies Series Volume 66, Springer-Verlag, New York.

Thompson, R. M. and C. R. Townsend. 1999. The effect of seasonal variation on the community structure and food-web attributes of two streams: implications for food-web science. Oikos 87:75-88.

Wallace, J. B., A. C. Benke, A. H. Lingle, and K. Parsons. 1987. Trophic pathways of macroinvertebrate primary consumers in subtropical blackwater streams. Archiv fur Hydrobiologie, Supplement 74:423-451.

Wallace, J. B., T. F. Cuffney, S. L. Eggert, and M. R. Whiles. 1997a. Stream organic matter inputs, storage, and export for Satellite Branch at Coweeta Hydrologic Laboratory, North Carolina, USA. Journal of the North American Benthological Society 16:67-74.

Wallace, J. B., S. L. Eggert, J. L. Meyer, and J. R. Webster. 1997b. Multiple trophic levels of a forest stream linked to terrestrial litter inputs. Science 277:102-104.

Wallace, J. B., S. L. Eggert, J. L. Meyer, and J. R. Webster. 1999. Effects of resource limitation on a detrital-based ecosystem. Ecological Monographs 69:409-442.

Wallace, J. B., M. R. Whiles, S. L. Eggert, T. F. Cuffney, G. J. Lugthart, and K. Chung. 1995. Long-term dynamics of coarse particulate organic matter in three Appalachian Mountain streams. Journal of the North American Benthological Society 14:217-232.

Wallace, J. B., J. R. Webster, S. L. Eggert, and J. L. Meyer. 2000. Small wood dynamics in a headwater stream. Verh. Internat. Verein. Linmol. 27:1361-1365.

Waters, T. F. 1969. Subsampler for dividing large samples of stream invertebrate drift. Limnology and Oceangraphy 14:813.

Webster, J. R., M. E. Gurtz, J. J. Hains, J. L. Meyer, W. T. Swank, J. B. Waide, and J. B. Wallace. 1983. Stability of stream ecosystems. Pages 355-395. *In*: Barnes, J. R. and G. W. Minshall (eds). Stream Ecology. Plenum Press, New York.

Whittaker, R.H. 1975. Communities and ecosystems. 2nd ed. Macmillan Pub. Co., New York.

Wootton, J. T. 1994. Predicting direct and indirect effects: an integrated approach using experiments and path analysis. Ecology 75:151-165.

Wootton, J. T. 1997. Estimates and tests of per capita interaction strength: diet, abundance, and impact of intertidally foraging birds. Ecological Monographs 67:45-64.

Table 5.1. Physical and chemical characteristics of headwater streams draining catchments 53 (reference) and 54 (enriched) at the Coweeta Hydrologic Laboratory. Elevations were measured at the gauging weirs.

Variable	**Reference (C53)**	**Treatment (C54)**
Catchment		
Area (ha)	5.2	5.5
Elevation (m asl)	829.0	841.0
Channel		
Gradient (cm m^{-1})	27.0	33.0
Length (m)	145.0	282.0
Bankfull Area (m^2)	327.0	443.0
Discharge (l s^{-1})¶		
Average	0.3	0.5
Maximum	3.8	4.8
Temperature (°C)¶		
Annual average	12.0	12.0
Minimum	2.6	4.8
Maximum	18.6	16.7
Water chemistry (range)		
pH	6.6 (6.2-7)	6.9 (6.6-7.9)
(NO_3-N + NO_2)-N (μg l^{-1})		
pretreatment: 1999 – 2000	15.4 (9.4-25.8)	18.8 (4-39.5)
enrichment: 2000 – 2002	16.9 (bd-151)	308.9 (11-1711)
NH_4-N (μg l^{-1})		
pretreatment: 1999 – 2000	9.4 (bd-30)	9.9 (bd-25)
enrichment: 2000 – 2002	10.4 (bd-76)	105.5 (6-566)
SRP‡ (μg l^{-1})		
pretreatment: 1999 – 2000	7.6 (bd-20)	8.8 (bd-22)
enrichment: 2000 – 2002	3.7 (bd-17)	51.2 (bd-268)

¶September 1998 – August 2002. Temperature was measured with Optic Stowaway temperature probes (Onset Computer Corp., Pocasset, MA, USA). ‡Soluble reactive phosphorus. bd = below detection limit. For nutrient data: reference stream - pretreatment n = 5, treated stream - pretreatment n = 12, reference stream - enrichment n = 33, treated stream - enrichment n = 44.

Table 5.2. Proportional similarity of diet between treatment periods or between streams for primary consumer functional feeding groups in the reference stream (C53) and treatment stream (C54). Pre = before nutrient enrichment (September 1998 - June 2000), Enr-1 = year 1 of enrichment (July 2000 – August 2001), Enr-2 = year 2 of enrichment (September 2001 – August 2002). Values are means ± SE for multiple seasons within each comparison. Food resource categories included leaves, wood, fungi, amorphous detritus, diatoms, and animal material.

	Reference stream			Treatment stream			Between-stream comparisons
	Pre	Enr-1	Enr-2	Pre	Enr-1	Enr-2	
Shredders							
Pre	---	---	---	---	---	---	0.95 ± 0.02
Enr-1	0.84 ± 0.05	---	---	0.88 ± 0.01	---	---	0.88 ± 0.03
Enr-2	0.88 ± 0.06	0.88 ± 0.07	---	0.84 ± 0.05	0.92 ± 0.02	---	0.93 ± 0.03
Gatherers							
Pre	---	---	---	---	---	---	0.90 ± 0.08
Enr-1	0.88 ± 0.08	---	---	0.88 ± 0.06	---	---	0.91 ± 0.04
Enr-2	0.86 ± 0.03	0.92 ± 0.03	---	0.63 ± 0.13	0.80 ± 0.13	---	0.83 ± 0.08
Filterers							
Pre	---	---	---	---	---	---	0.86 ± 0.02
Enr-1	0.81 ± 0.12	---	---	0.82 ± 0.06	---	---	0.85 ± 0.04
Enr-2	0.74 ± 0.10	0.83 ± 0.02	---	0.85 ± 0.03	0.84 ± 0.06	---	0.78 ± 0.08
Scrapers							
Pre	---	---	---	---	---	---	0.93 ± 0.04
Enr-1	0.85 ± 0.02	---	---	0.74 ± 0.15	---	---	0.77 ± 0.06
Enr-2	0.77 ± 0.16	0.81 ± 0.04	---	0.80 ± 0.06	0.80 ± 0.10	---	0.86 ± 0.08

Table 5.3. Fraction of total invertebrate production attributed to food resources and total flows from food resources to the invertebrate communities in the reference (C53) and treatment (C54) streams. Pre = pretreatment period (September 1998 – August 2000), Enr-1 = year 1 of enrichment (September 2000 – August 2001), and Enr-2 = year 1 of enrichment (September 2001 – August 2002). For fractions, values in parentheses only include primary consumers (i.e., no predators). For total organic matter flows, values in parentheses are percentages of the total.

Fraction of total secondary production derived from food resources

	Pre		Enr-1		Enr-2	
	reference	treatment	reference	treatment	reference	treatment
Leaf detritus	0.19 (0.27)	0.17 (0.26)	0.20 (0.30)	0.16 (0.21)	0.25 (0.34)	0.22 (0.28)
Wood detritus	0.02 (0.03)	0.03 (0.04)	0.02 (0.02)	0.00 (0.01)	0.01 (0.01)	0.01 (0.01)
Amorphous detritus	0.32 (0.45)	0.31 (0.46)	0.29 (0.45)	0.40 (0.54)	0.30 (0.42)	0.32 (0.40)
Fungi	0.12 (0.16)	0.11 (0.16)	0.12 (0.18)	0.13 (0.18)	0.14 (0.20)	0.19 (0.24)
Diatoms	0.03 (0.04)	0.01 (0.01)	0.01 (0.01)	0.03 (0.04)	0.01 (0.02)	0.01 (0.02)
Animal prey	0.33 (0.05)	0.37 (0.07)	0.37 (0.03)	0.28 (0.02)	0.28 (0.02)	0.24 (0.04)

Total organic matter flows from food resources (g AFDM m^{-2} y^{-1})

	Pre		Enr-1		Enr-2	
	reference	treatment	reference	treatment	reference	treatment
Leaf detritus	74.9 (47)	59.7 (45)	78.8 (50)	124.4 (53)	100.6 (57)	199.3 (64)
Wood detritus	8.3 (5)	9.2 (7)	6.4 (4)	3.7 (2)	3.5 (2)	9.8 (3)
Amorphous detritus	62.8 (40)	52.8 (40)	58.3 (37)	82.2 (35)	61.3 (35)	74.3 (24)
Fungi	3.2 (2)	2.6 (2)	3.4 (2)	7.4 (3)	4.2 (2)	12.4 (4)
Diatoms	1.8 (1)	0.4 (<1)	0.6 (<1)	4.2 (2)	0.9 (1)	2.1 (1)
Animal prey	8.0 (5)	8.0 (6)	9.1 (6)	13.6 (6)	7.1 (4)	13.2 (4)
Total	159.1	132.7	156.6	235.5	177.5	311.2

Table 5.4. Total annual production (g AFDM m^{-2} y^{-1}) attributable to each food type (percent of total in parentheses) for primary consumer functional feeding groups in the reference and treatment stream during the pretreatment period (average of September 1998 – August 1999 and September 1999 – August 2000), year 1 of enrichment (September 2000 – August 2001), and year 2 of enrichment (September 2001 – August 2002). Values in parentheses are percentages of the total.

	Pretreatment		(year 1) Nutrient Enrichment		(year 2) Nutrient Enrichment	
Functional group/ Food type	Reference	Treatment	Reference	Treatment	Reference	Treatment
Shredders						
Leaf detritus	1.022 (51)	0.944 (50)	1.260 (45)	1.915 (38)	1.598 (52)	3.159 (40)
Wood detritus	0.126 (6)	0.131 (7)	0.106 (4)	0.065 (1)	0.057 (2)	0.173 (2)
Amorphous detritus	0.250 (12)	0.208 (11)	0.631 (23)	1.015 (20)	0.484 (16)	1.632 (21)
Fungi	0.624 (31)	0.582 (31)	0.753 (27)	1.616 (32)	0.929 (30)	2.771 (35)
Diatoms	0.002 (<1)	0.007 (<1)	0.024 (1)	0.366 (7)	0.015 (<1)	0.156 (2)
Animal	0.000 (0)	0.000 (0)	0.005 (<1)	0.000 (0)	0.008 (<1)	0.004 (<1)
Total	2.025	1.873	2.780	4.978	3.092	7.897
Gatherers						
Leaf detritus	0.142 (10)	0.108 (6)	0.157 (10)	0.284 (6)	0.188 (11)	0.528 (13)
Wood detritus	0.006 (<1)	0.026 (2)	0.009 (1)	0.001 (<1)	0.000 (0)	0.007 (<1)
Amorphous detritus	1.140 (78)	1.445 (86)	1.234 (81)	3.828 (87)	1.366 (80)	2.980 (72)
Fungi	0.083 (6)	0.070 (4)	0.093 (6)	0.226 (5)	0.107 (6)	0.428 (10)
Diatoms	0.110 (7)	0.026 (2)	0.026 (2)	0.063 (<1)	0.052 (3)	0.063 (2)
Animal	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (0)	0.109 (3)
Total	1.483	1.674	1.520	4.402	1.714	4.116

Table 5.4 (cont.)

Functional group/ Food type	Pretreatment		(year 1) Nutrient Enrichment		(year 2) Nutrient Enrichment	
	Reference	Treatment	Reference	Treatment	Reference	Treatment
Filterers						
Leaf detritus	0.037 (8)	0.054 (9)	0.044 (14)	0.063 (9)	0.064 (17)	0.082 (9)
Wood detritus	0.008 (2)	0.012 (2)	0.004 (1)	0.002 (<1)	0.006 (2)	0.008 (1)
Amorphous detritus	0.111 (25)	0.140 (25)	0.129 (39)	0.344 (50)	0.195 (51)	0.368 (38)
Fungi	0.024 (5)	0.034 (6)	0.027 (8)	0.053 (8)	0.039 (10)	0.078 (8)
Diatoms	0.004 (1)	0.008 (1)	0.009 (3)	0.008 (1)	0.008 (2)	0.005 (1)
Animal	0.265 (60)	0.317 (56)	0.115 (35)	0.215 (31)	0.070 (18)	0.427 (44)
Total	0.447	0.564	0.327	0.686	0.275	0.968
Scrapers						
Leaf detritus	0.000 (<1)	0.000 (7)	0.001 (4)	0.001 (1)	0.000 (1)	0.001 (1)
Wood detritus	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (4)	0.000 (0)
Amorphous detritus	0.031 (77)	0.009 (72)	0.020 (71)	0.068 (81)	0.010 (54)	0.053 (80)
Fungi	0.000 (<1)	0.001 (4)	0.001 (2)	0.000 (1)	0.000 (2)	0.001 (1)
Diatoms	0.009 (22)	0.002 (16)	0.006 (22)	0.015 (18)	0.007 (39)	0.012 (18)
Animal	0.000 (0)	0.000 (0)	0.000 (<1)	0.000 (0)	0.000 (0)	0.000 (0)
Total	0.041	0.012	0.029	0.084	0.018	0.066

Table 5.5. Annual organic matter flows in g AFDM m^{-2} y^{-1} (percent of total in parentheses) from each food type to primary consumer functional feeding groups in the reference and treatment stream during the pretreatment period (average of September 1998 – August 1999 and September 1999 – August 2000), year 1 of enrichment (September 2000 – August 2001), and year 2 of enrichment (September 2001 – August 2002). Values in parentheses are percentages of the total.

	Pretreatment		(year 1) Nutrient Enrichment		(year 2) Nutrient Enrichment	
Functional group/ Food type	Reference	Treatment	Reference	Treatment	Reference	Treatment
Shredders						
Leaf detritus	51.126 (78)	47.221 (77)	63.022 (72)	95.726 (79)	79.911 (81)	157.963 (79)
Wood detritus	6.280 (10)	6.597 (11)	5.308 (6)	3.261 (3)	2.842 (3)	8.664 (4)
Amorphous detritus	6.270 (10)	5.192 (8)	15.779 (18)	13.358 (11)	12.104 (12)	21.478 (11)
Fungi	2.229 (2)	2.077 (3)	2.690 (3)	5.772 (5)	3.319 (3)	9.898 (5)
Diatoms	0.013 (<1)	0.062 (<1)	0.199 (<1)	3.053 (3)	0.126 (<1)	1.305 (1)
Animal	0.000 (0)	0.000 (0)	0.016 (<1)	0.000 (0)	0.024 (<1)	0.013 (<1)
Total	65.919	61.150	87.016	121.170	98.330	199.322
Gatherers						
Leaves	7.135 (19)	5.381 (12)	7.899 (20)	14.227 (22)	9.440 (21)	26.376 (39)
Wood	0.289 (1)	1.285 (3)	0.464 (1)	0.034 (<1)	0.000 (0)	0.369 (1)
Amorphous detritus	28.508 (77)	36.126 (84)	30.842 (78)	50.365 (76)	34.140 (77)	39.211 (57)
Fungi	0.297 (1)	0.249 (1)	0.332 (1)	0.807 (1)	0.384 (1)	1.530 (2)
Diatoms	0.921 (2)	0.218 (1)	0.220 (1)	0.524 (1)	0.431 (1)	0.523 (1)
Animal	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (0)	0.000 (0)	0.342 (1)
Total	37.151	43.260	39.757	65.956	44.395	68.351

Table 5.5 (cont.)

Functional group/ Food type	Pretreatment		(year 1) Nutrient Enrichment		(year 2) Nutrient Enrichment	
	Reference	Treatment	Reference	Treatment	Reference	Treatment
Filterers						
Leaf detritus	1.843 (31)	2.679 (34)	2.211 (36)	3.159 (36)	3.207 (36)	4.117 (37)
Wood detritus	0.398 (7)	0.586 (7)	0.204 (3)	0.099 (1)	0.302 (3)	0.395 (4)
Amorphous detritus	2.767 (47)	3.498 (44)	3.216 (52)	4.521 (52)	4.878 (55)	4.848 (44)
Fungi	0.084 (1)	0.123 (2)	0.095 (2)	0.190 (2)	0.138 (2)	0.277 (3)
Diatoms	0.030 (1)	0.064 (1)	0.074 (1)	0.070 (1)	0.064 (1)	0.041 (<1)
Animal	0.827 (14)	0.990 (12)	0.358 (6)	0.673 (8)	0.218 (2)	1.335 (12)
Total	5.948	7.940	6.158	8.713	8.806	11.013
Scrapers						
Leaf detritus	0.004 (<1)	0.045 (16)	0.059 (9)	0.030 (3)	0.008 (2)	0.037 (4)
Wood detritus	0.000 (0)	0.001 (<1)	0.000 (0)	0.000 (0)	0.037 (10)	0.000 (0)
Amorphous detritus	0.785 (91)	0.221 (77)	0.512 (82)	0.897 (85)	0.238 (69)	0.696 (83)
Fungi	0.000 (<1)	0.002 (1)	0.002 (<1)	0.002 (<1)	0.001 (<1)	0.002 (<1)
Diatoms	0.076 (8)	0.016 (6)	0.054 (9)	0.124 (12)	0.058 (17)	0.100 (12)
Animal	0.000 (0)	0.000 (0)	0.000 (<1)	0.000 (0)	0.000 (0)	0.000 (0)
Total	0.865	0.285	0.628	1.052	0.342	0.834

Figure 5.1. Proportion (%) of food resources consumed by invertebrate functional feeding groups in the reference stream (ref) and the treatment stream (tmt) during the pretreatment period (pre = average of September 1998 – August 1999 and September 1999 – August 2000), year 1 of enrichment (enr-1 = September 2000 – August 2001), and year 2 of enrichment (enr 2 = September 2001 – August 2002).

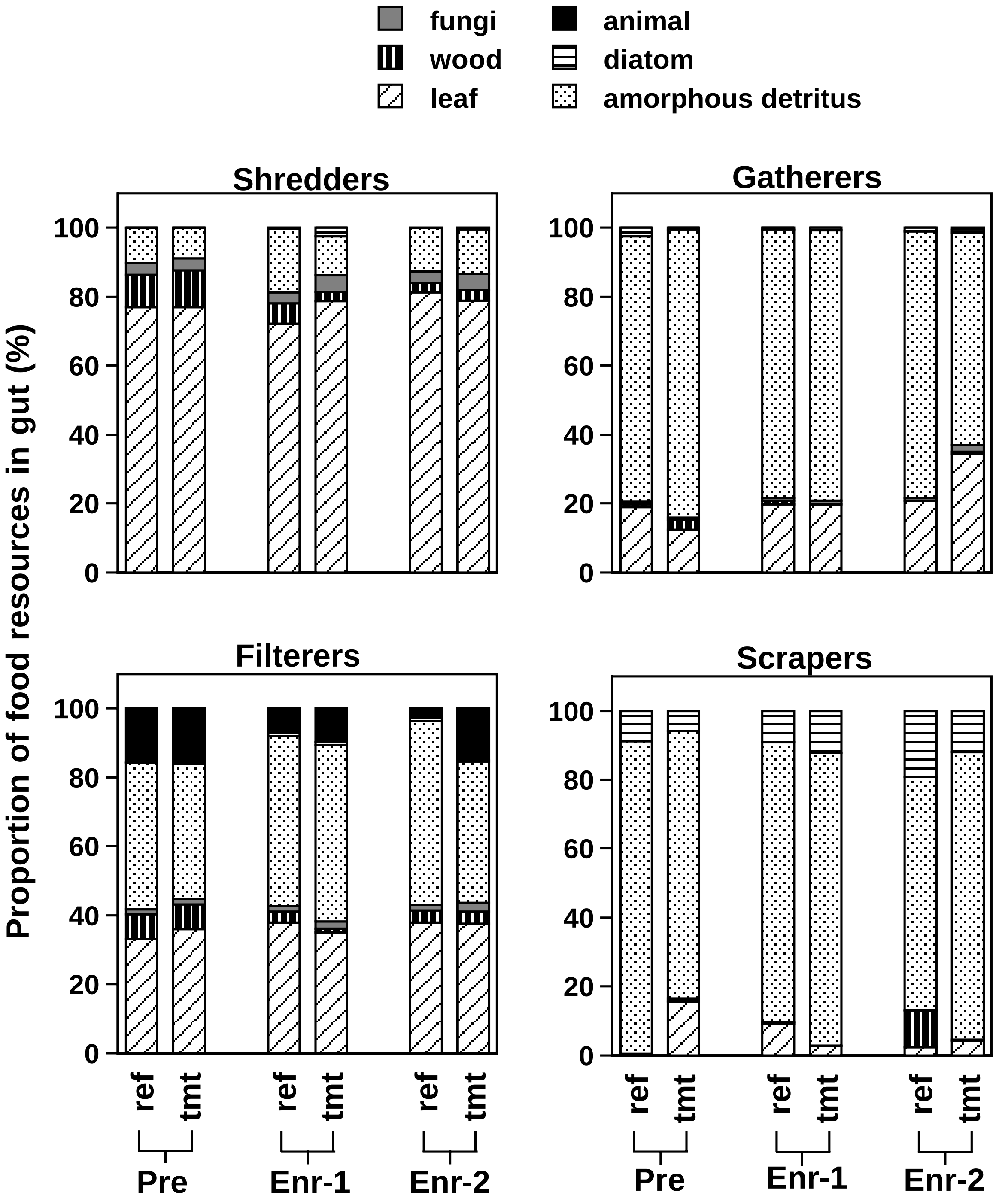

fungi
wood
leaf
animal
diatom
amorphous detritus
Shredders
Gatherers
Filterers
Scrapers
Proportion of food resources in gut (%)
0
20
40
60
80
100
ref
tmt
Pre
Enr-1
Enr-2

Figure 5.2. Invertebrate secondary production (mg m^{-2} y^{-1}) attributed to food resources among functional feeding groups in the reference stream (ref) and treatment stream (tmt) during the pretreatment period (pre = average of September 1998 – August 1999 and September 1999 – August 2000), year 1 of enrichment (enr-1 = September 2000 – August 2001), and year 2 of enrichment (enr 2 = September 2001 – August 2002). Note difference in scale among functional groups.

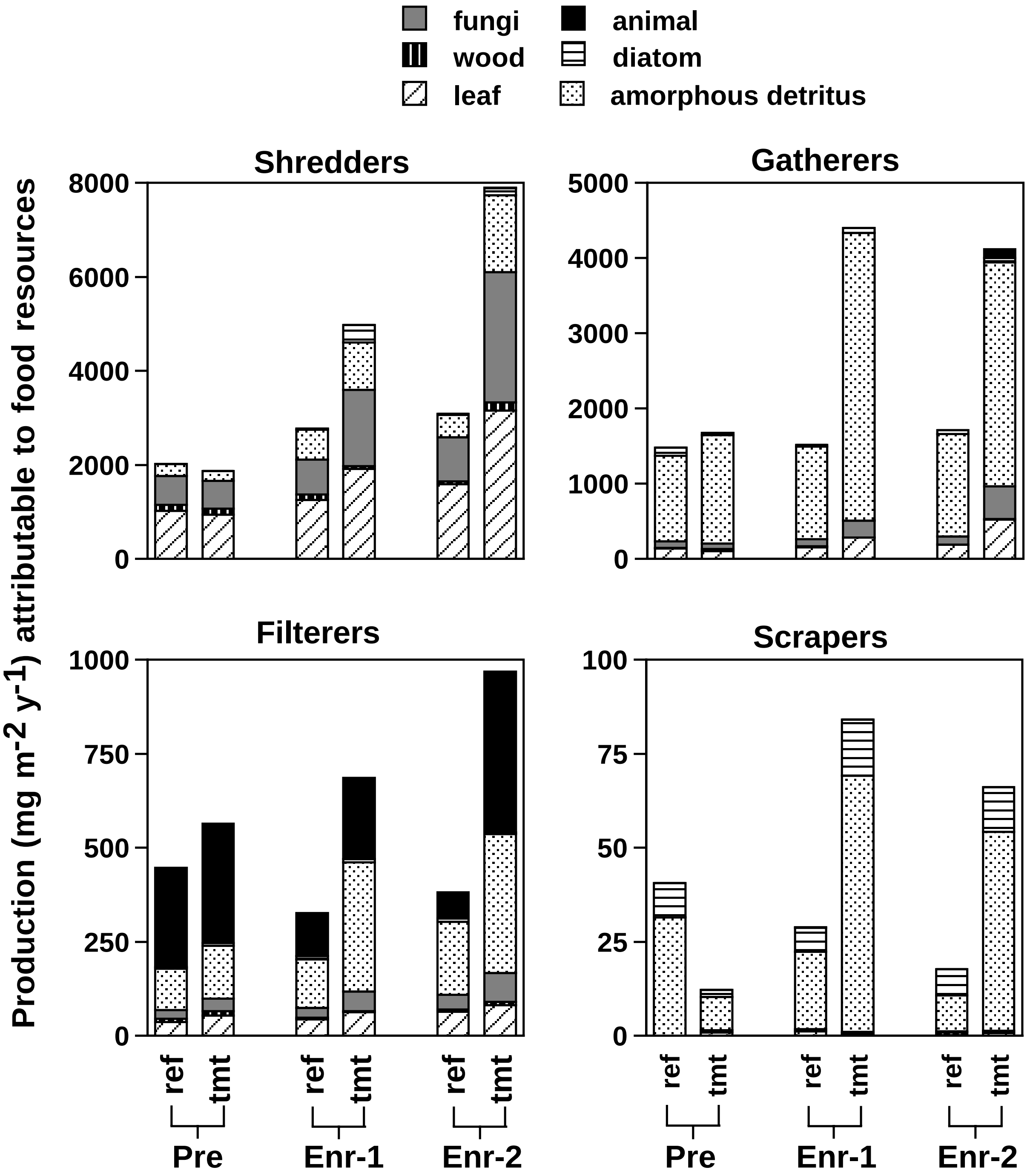
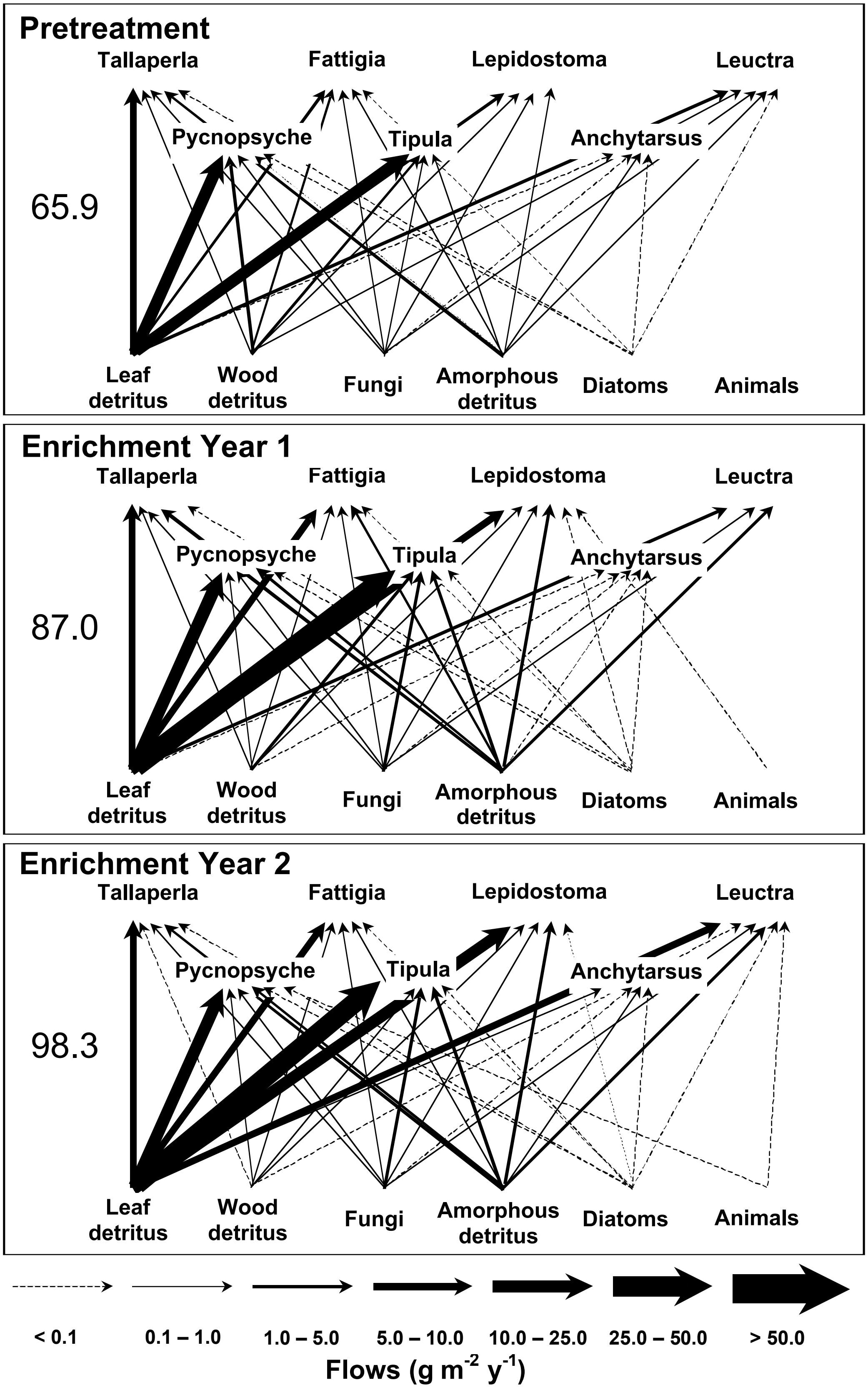

fungi
wood
leaf
animal
diatom
amorphous detritus
Shredders
Gatherers
Filterers
Scrapers
Production (mg m-2 y-1) attributable to food resources
0
2000
4000
6000
8000
0
1000
2000
3000
4000
5000
0
250
500
750
1000
0
25
50
75
100
ref
tmt
Pre
Enr-1
Enr-2

Figure 5.3. Organic matter flows (g AFDM m^{-2} y^{-1}) from basal food resources to dominant invertebrate shredder taxa in the reference stream and the treatment stream during each time period of the study. Numbers on the left side of each graph indicate the total amount of flows from basal resources to these shredder taxa. The thickness of arrows is proportional to the magnitude of flows (see flow key on figure).

Shredders (reference stream)



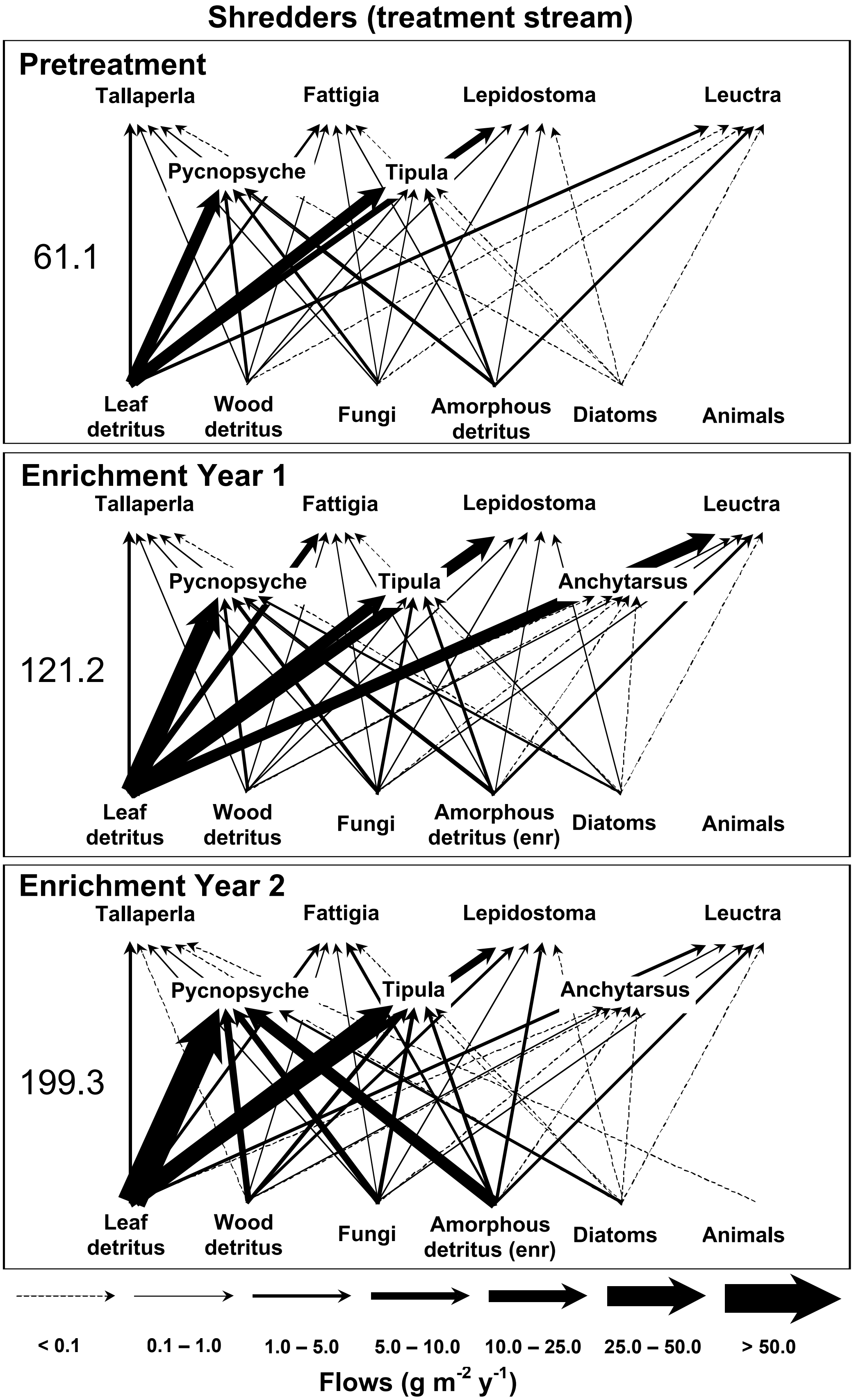
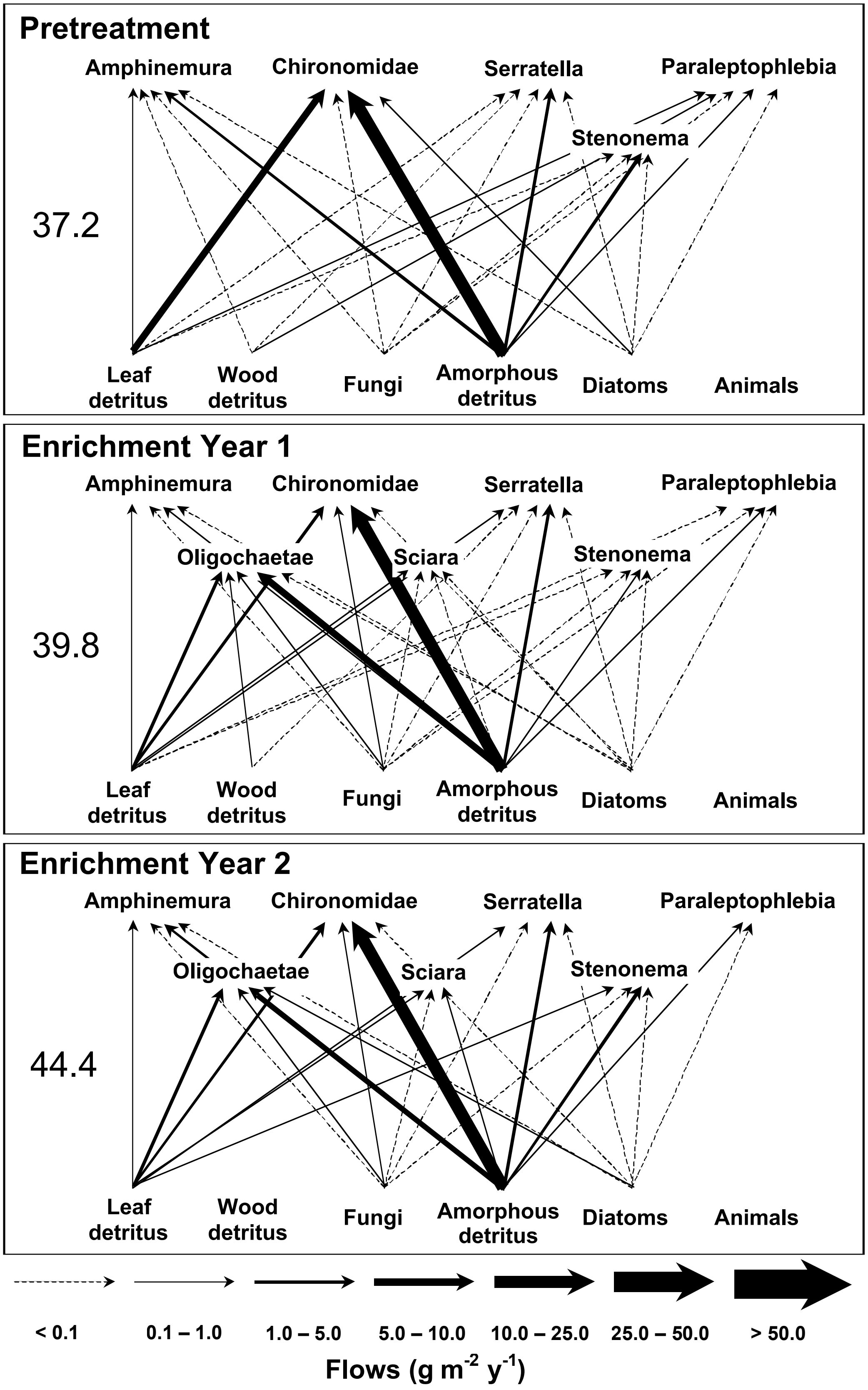

Shredders (treatment stream)
Pretreatment
Tallaperla
Fattigia
Lepidostoma
Leuctra
Pycnopsyche
Tipula
61.1
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
Enrichment Year 1
Tallaperla
Fattigia
Lepidostoma
Leuctra
Pycnopsyche
Tipula
Anchytarsus
121.2
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
Enrichment Year 2
Tallaperla
Fattigia
Lepidostoma
Leuctra
Pycnopsyche
Tipula
Anchytarsus
199.3
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m^{-2} y^{-1})

Figure 5.4. Organic matter flows (g AFDM m^{-2} y^{-1}) from basal food resources to dominant invertebrate gatherer taxa in the reference stream and the treatment stream during each time period of the study. Numbers on the left side of each graph indicate the total amount of flows from basal resources to these gatherer taxa. The thickness of arrows is proportional to the magnitude of flows (see flow key on figure).

Gatherers (reference stream)



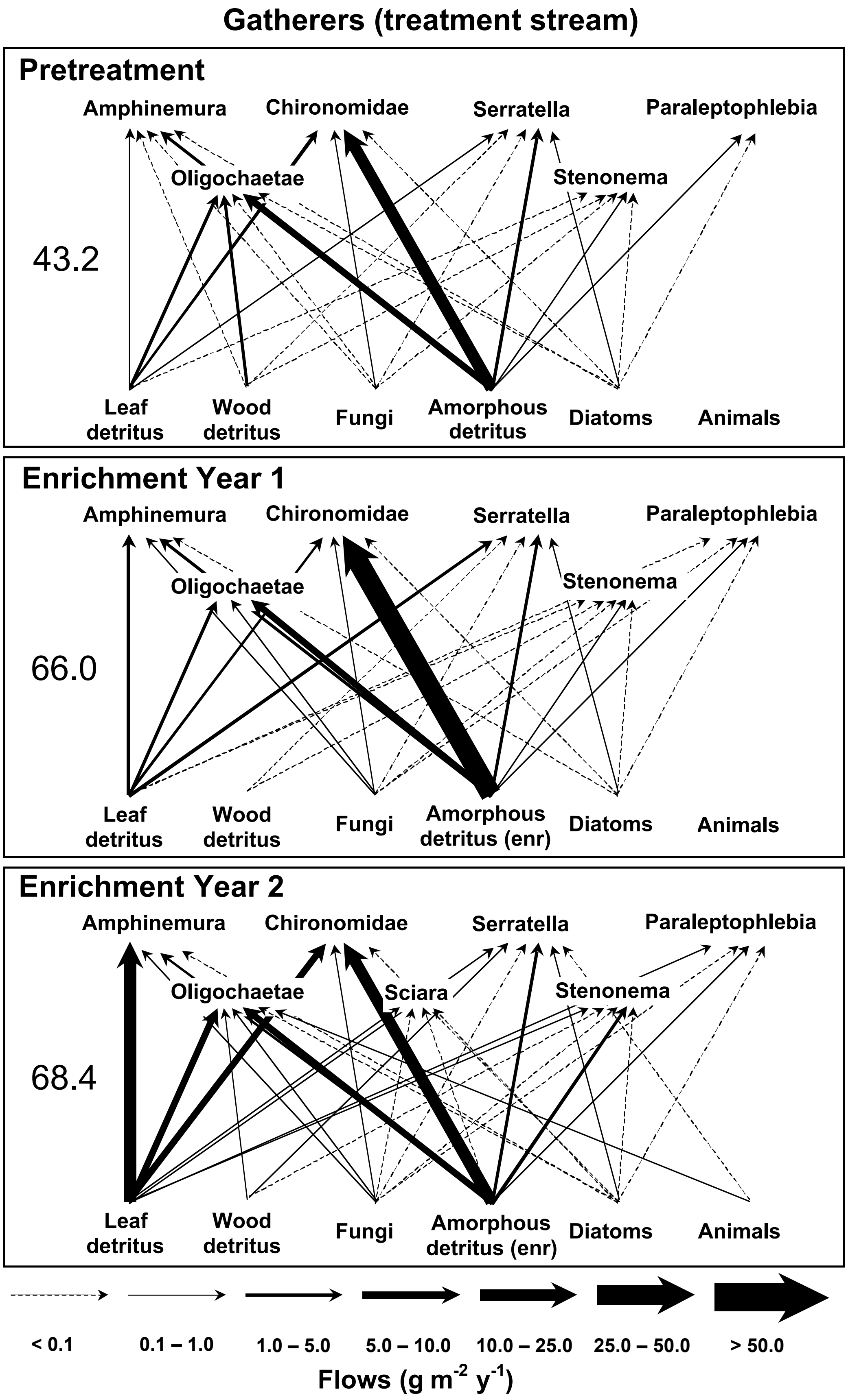
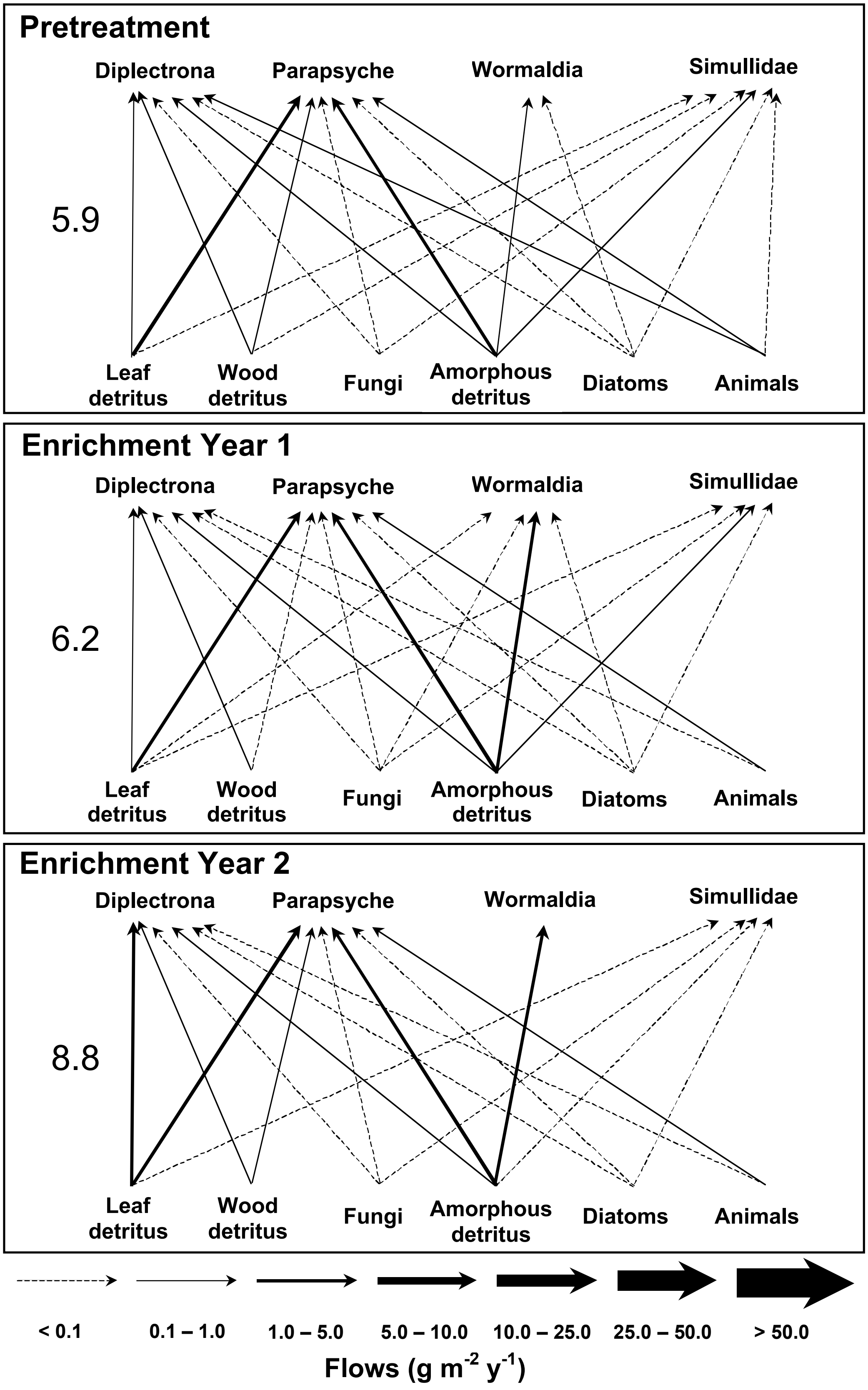

Gatherers (treatment stream)
Pretreatment
Amphinemura
Chironomidae
Serratella
Paraleptophlebia
Oligochaetae
Stenonema
43.2
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
Enrichment Year 1
Amphinemura
Chironomidae
Serratella
Paraleptophlebia
Oligochaetae
Stenonema
66.0
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
Enrichment Year 2
Amphinemura
Chironomidae
Serratella
Paraleptophlebia
Oligochaetae
Sciara
Stenonema
68.4
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m-2 y-1)

Figure 5.5. Organic matter flows (g AFDM m^{-2} y^{-1}) from basal food resources to dominant invertebrate filterer taxa in the reference stream and the treatment stream during each time period of the study. Numbers on the left side of each graph indicate the total amount of flows from basal resources to these filterer taxa. The thickness of arrows is proportional to the magnitude of flows (see flow key on figure).

Filterers (reference stream)



Filterers (treatment stream)

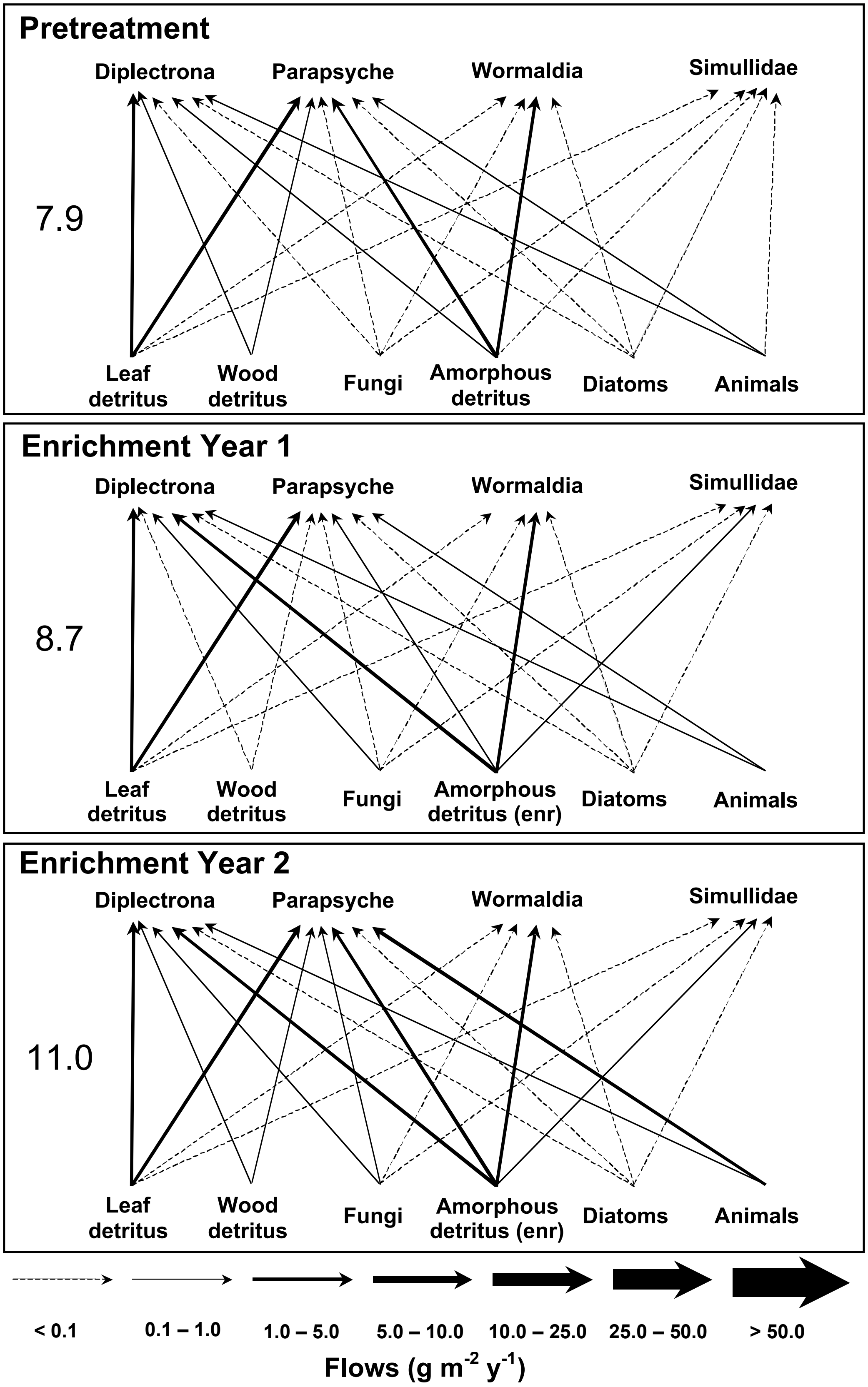
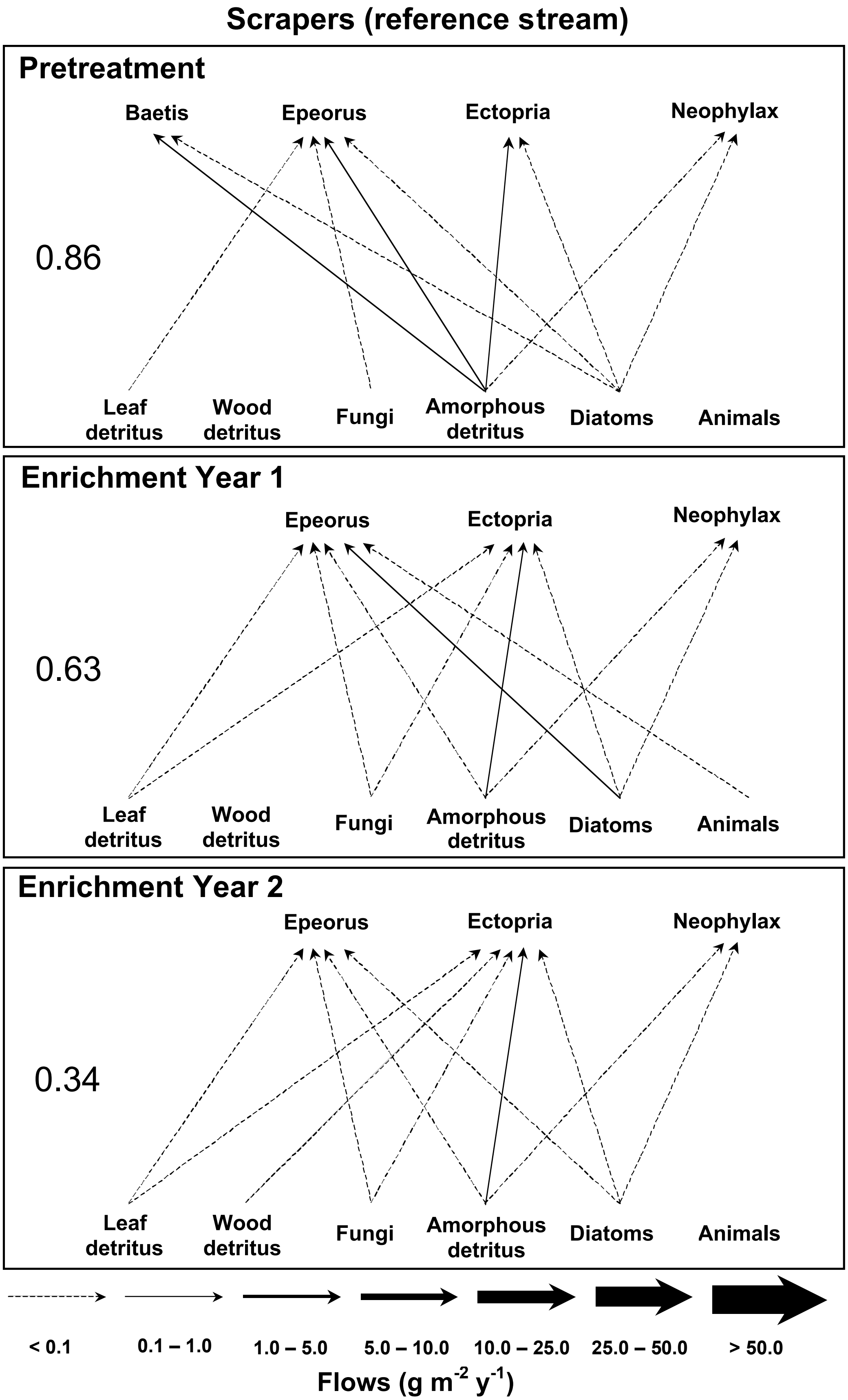


Figure 5.6. Organic matter flows (g AFDM m^{-2} y^{-1}) from basal food resources to dominant invertebrate scraper taxa in the reference stream and the treatment stream during each time period of the study. Numbers on the left side of each graph indicate the total amount of flows from basal resources to these scraper taxa. The thickness of arrows is proportional to the magnitude of flows (see flow key on figure).


Scrapers (reference stream)
Pretreatment
Baetis
Epeorus
Ectopria
Neophylax
0.86
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
Enrichment Year 1
Epeorus
Ectopria
Neophylax
0.63
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
Enrichment Year 2
Epeorus
Ectopria
Neophylax
0.34
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m-2 y-1)

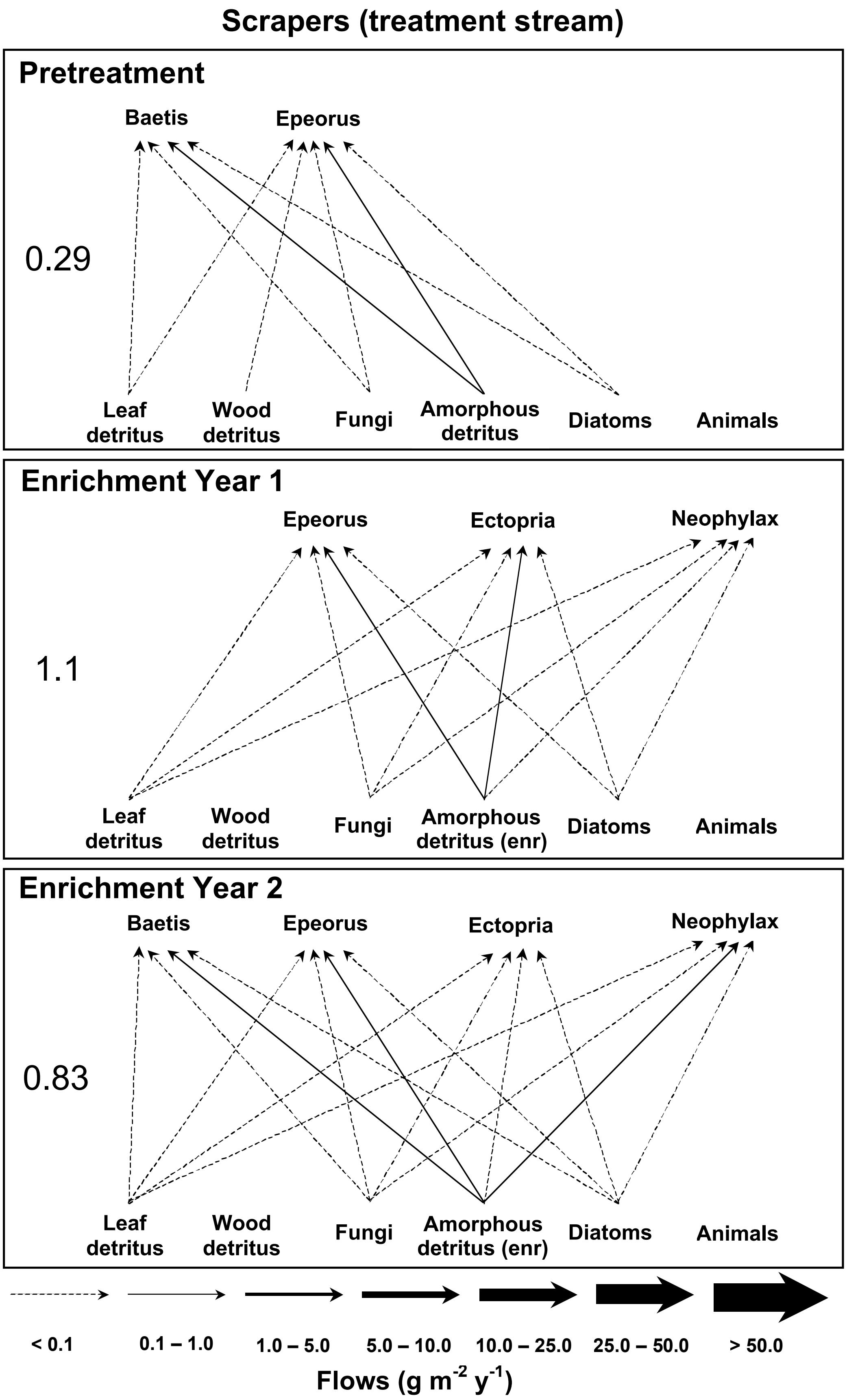
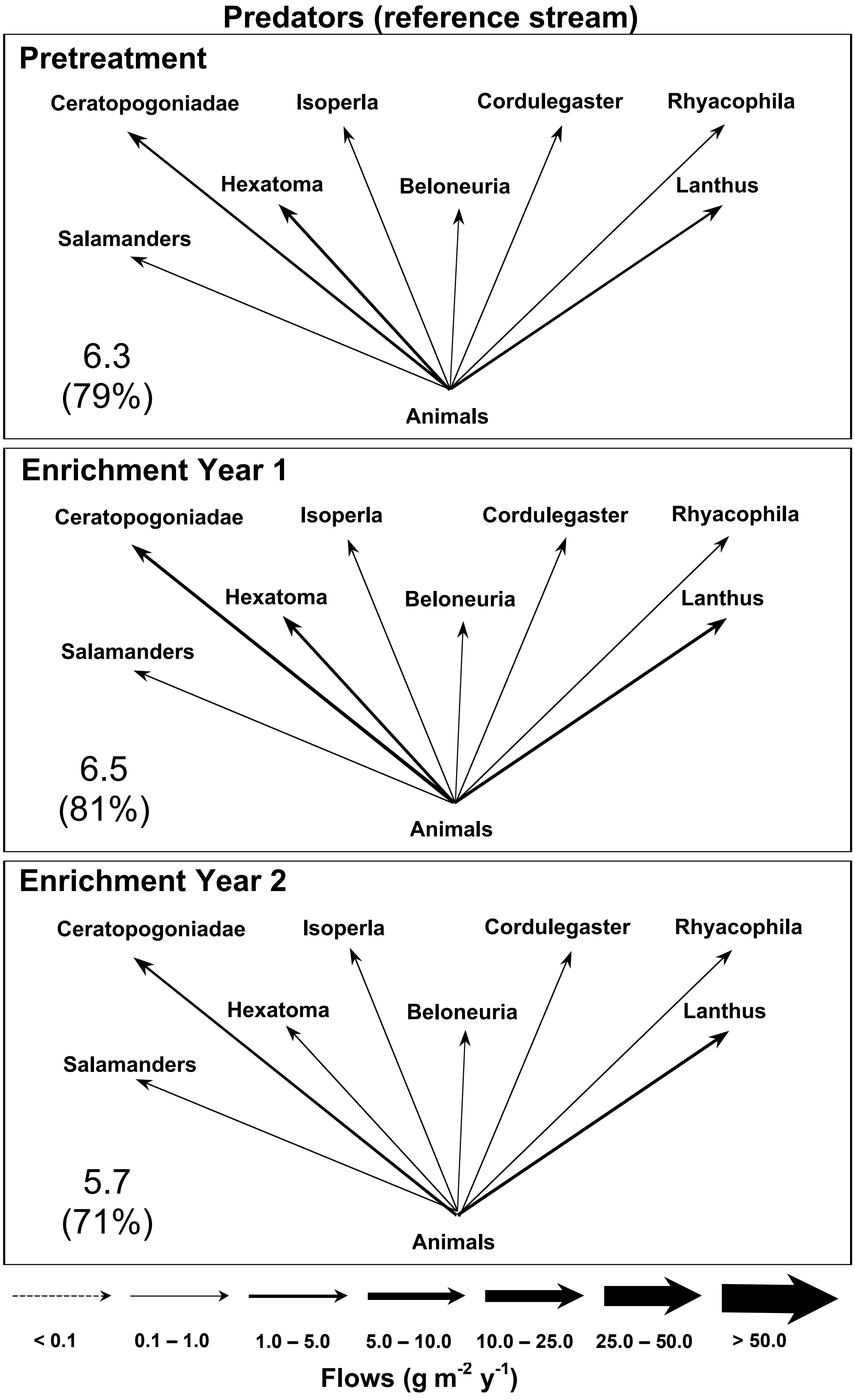

Scrapers (treatment stream)
Pretreatment
Baetis
Epeorus
0.29
Leaf detritus
Wood detritus
Fungi
Amorphous detritus
Diatoms
Animals
Enrichment Year 1
Epeorus
Ectopria
Neophylax
1.1
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
Enrichment Year 2
Baetis
Epeorus
Ectopria
Neophylax
0.83
Leaf detritus
Wood detritus
Fungi
Amorphous detritus (enr)
Diatoms
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m-2 y-1)

Figure 5.7. Organic matter flows (g AFDM m^{-2} y^{-1}) from animal prey to dominant predator taxa in the reference stream and the treatment stream during each time period of the study. Numbers on the left side of each graph indicate the total amount of flows from basal resources to these scraper taxa; numbers in parentheses indicate the percentage of total prey production consumed by these predators. The thickness of arrows is proportional to the magnitude of flows (see flow key on figure).


Predators (reference stream)
Pretreatment
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
6.3
(79%)
Animals
Enrichment Year 1
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
6.5
(81%)
Animals
Enrichment Year 2
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
5.7
(71%)
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m-2 y-1)

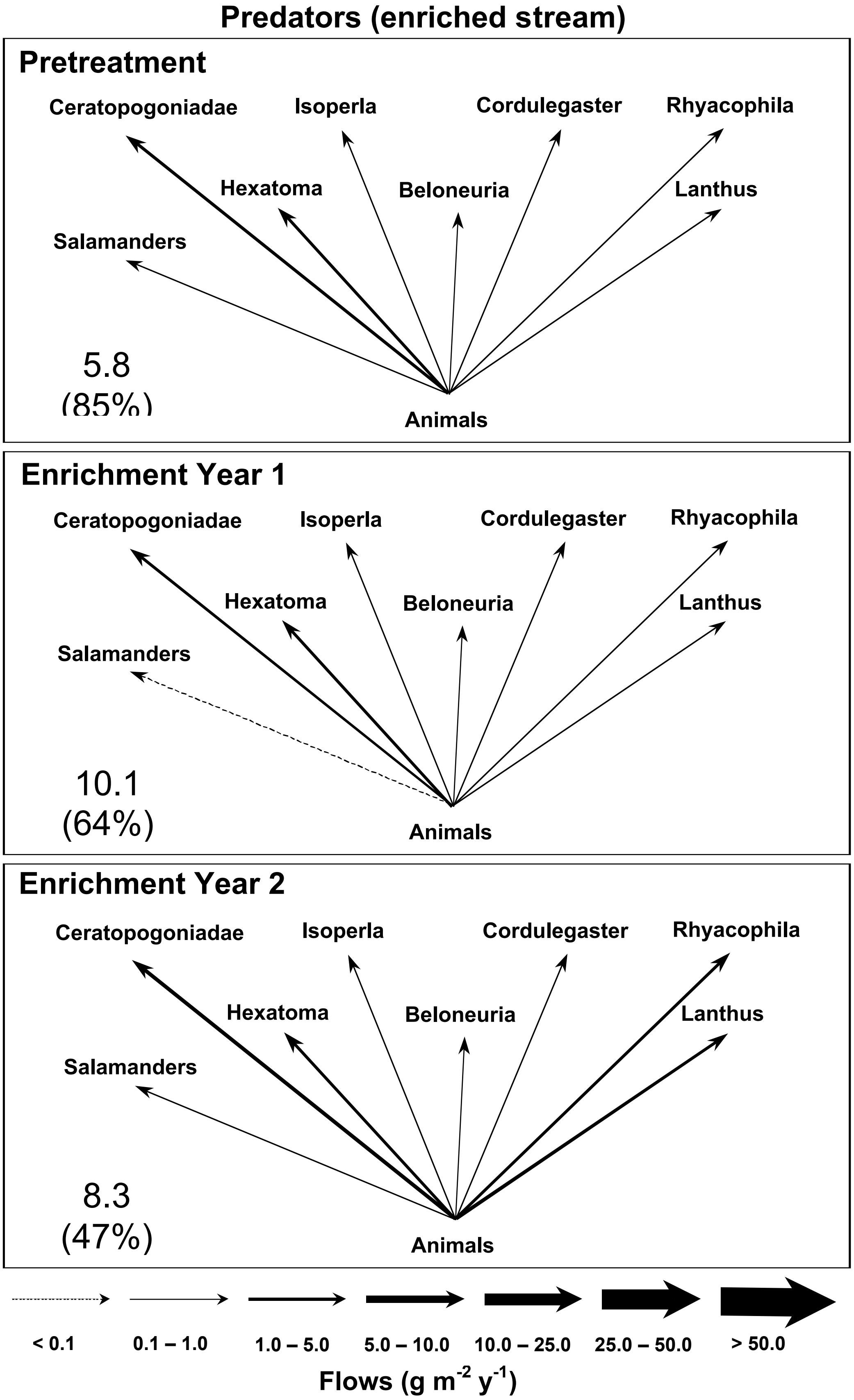
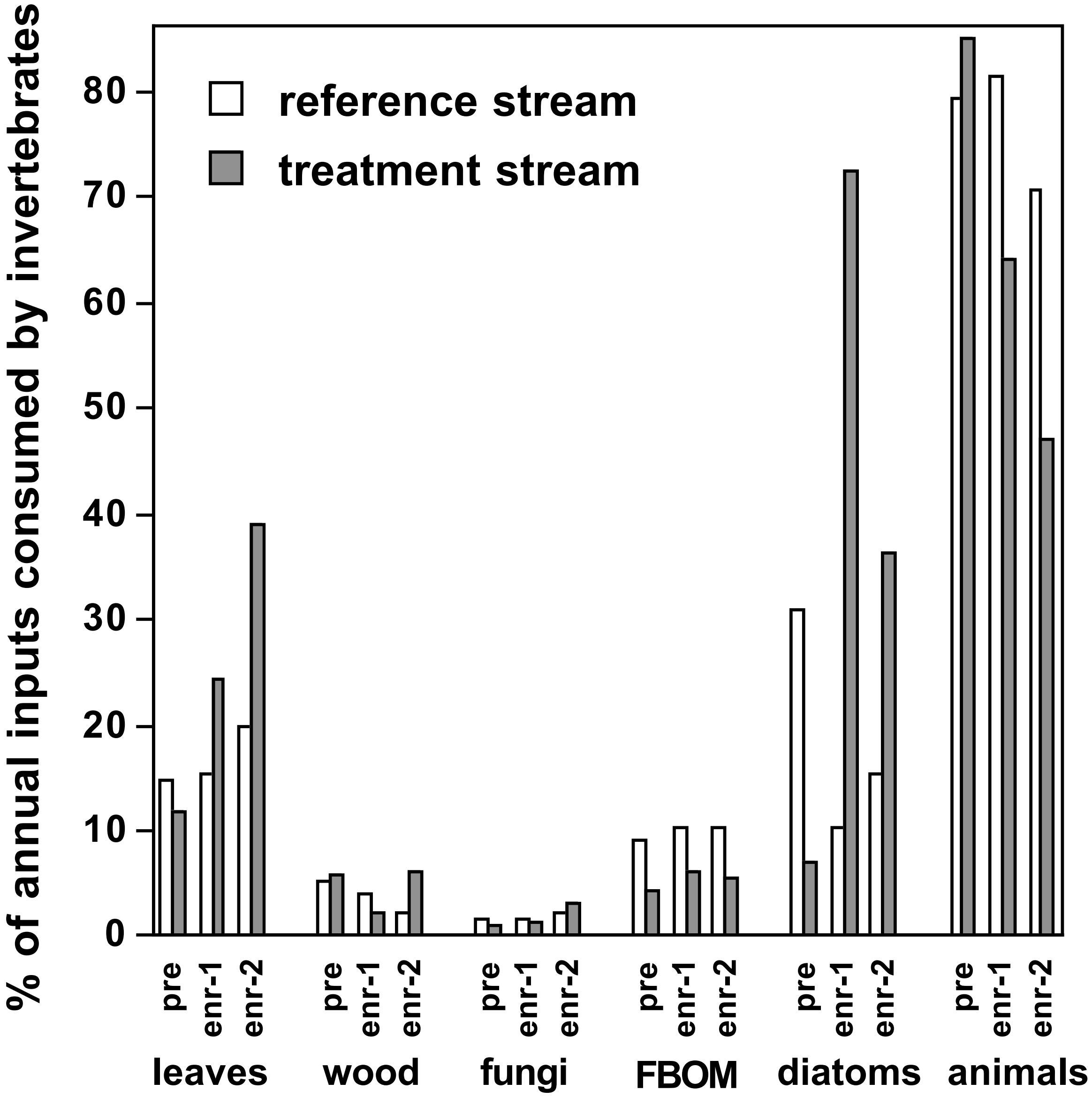

Predators (enriched stream)
Pretreatment
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
5.8
(85%)
Animals
Enrichment Year 1
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
10.1
(64%)
Animals
Enrichment Year 2
Ceratopogoniadae
Isoperla
Cordulegaster
Rhyacophila
Hexatoma
Beloneuria
Lanthus
Salamanders
8.3
(47%)
Animals
< 0.1
0.1 – 1.0
1.0 – 5.0
5.0 – 10.0
10.0 – 25.0
25.0 – 50.0
> 50.0
Flows (g m-2 y-1)

Figure 5.8. Percent of annual inputs of leaf litter, wood, fungi, FBOM (standing stock), diatoms, and animals consumed by invertebrates in the reference stream (white bars) and treatment stream (grey bars) during each time period of the study. pre = average of September 1998 – August 1999 and September 1999 – August 2000, enr-1 = September 2000 – August 2001, enr 2 = September 2001 – August 2002).


% of annual inputs consumed by invertebrates
0
10
20
30
40
50
60
70
80
reference stream
treatment stream
pre
enr-1
enr-2
leaves
wood
fungi
FBOM
diatoms
animals

CHAPTER 6

GENERAL CONCLUSIONS

Until recently, next to nothing was known about the effects of nutrient enrichment on consumers in detritus-based stream ecosystems. Although considerable previous research has examined effects of enrichment at the basal resource level (i.e., detritus and associated microbes), few attempts have been made to link basal resource changes to population/community dynamics and energetics of macro-consumers (Rosemond et al. 2001, 2002, Ramirez and Pringle, in review). Moreover, to this point, studies addressing this issue have been relatively short in duration (e.g., Pearson and Connolly 2000, Rosemond et al. 2001). This dissertation was an attempt to examine in detail the response of detritus-based stream invertebrates to nutrient enrichment using a variety of methodological approaches. All dissertation research fell within the context of a large-scale experimental manipulation of a headwater stream at Coweeta Hydrologic Laboratory. It was my hope that multiple studies at a variety of hierarchical scales would help clarify mechanisms by which individuals, populations, and communities may be affected by enrichment. My specific objectives were to examine effects of nutrient enrichment on 1) elemental composition and stoichiometric relationships of consumers and basal resources (Chapter 2), 2) individual growth rates and production of 2 dominant detritivores (Chapter 3), 3) whole-community invertebrate secondary production and organic matter dynamics (Chapter 4), and 4) trophic basis of production and organic matter flow food webs (Chapter 5). Each of these studies provided a unique 'lens' through which to examine enrichment effects, and all led to new insight as well as important questions for future study.

Before discussing the general conclusions of my dissertation, it is important to briefly mention major results from concurrent studies during the nutrient enrichment to provide context. Most noteworthy were nutrient-induced changes that occurred at the base of the food web. Enrichment led to significantly higher bacterial and fungal production on leaf litter, as well as increased leaf respiration rates (K. Suberkropp, University of Alabama, unpublished, Greenwood 2004). As well, enrichment accelerated decomposition rates of leaves and wood (Gulis and Suberkropp 2003, V. Gulis and K. Suberkropp, University of Alabama, unpublished, Greenwood 2004), and increased epilithic chlorophyll *a* levels during spring months (Greenwood 2004). Lastly, enrichment led to increased downstream export of fine particulate organic matter (A. D. Rosemond, unpublished data). Carbon budgets are currently being constructed for both streams and will shed considerable light on ecosystem-level effects of enrichment on carbon cycling in detritus-based streams. These changes to basal resources all had important implications for the structure and productivity of invertebrate communities.

In the first study chapter (2), I set out to test some of the basic tenets of ecological stoichiometry (e.g, Sterner and Elser 2002). This was a unique and timely study because virtually no previous work had addressed consumer-resource stoichiometry in detritus-based ecosystems. Additionally, very few studies had described the elemental composition of insect (as opposed to crustacean) consumers. This research led to a number of important results that provide a framework for further hypothesis testing. First, and least surprising, enrichment led to increased nutrient content of stream basal food resources (i.e., leaf litter, FBOM, and epilithon). However, contrary to my predictions, this effect appeared to be transferred to higher trophic levels. Carbon:P and N:P ratios of some consumers (primarily Trichoptera) were up to 4-fold lower in the treatment stream than in the reference stream during enrichment. This result demonstrated that some insects may be much more plastic (i.e., less homeostatic) with regard to their elemental composition (particularly P content) than other well-studied invertebrates such as cladocerans and copepods (e.g., Elser and Urabe 1999). Another surprise from this work was that average molar C:P and N:P ratios of invertebrates were much higher than any other invertebrate community reported in the literature. Interestingly, all other published values came from invertebrate communities based on plants or algae. An intriguing hypothesis is that low body P (or high C:P and N:P ratios) is an evolutionary adaptation of detritus-based consumers to low nutrient content of detritus. This idea will undoubtedly require much further testing.

Chapter 3 was aimed at understanding nutrient effects at the individual- and population-level by way of growth rate measurements and quantitative benthic sampling. The approach was to examine the response of taxa that occupy disparate ends of the ‘slow-fast’ life-history continuum (i.e., *r*- versus K-selected), and have different feeding behaviors (i.e., fine particulate organic matter vs. leaf litter). I examined field growth rates of 2 common detritivores (non-Tanypodinae chironomids and peltoperlid stoneflies) seasonally in both streams before and during the enrichment. By examining taxon-level responses (abundance, biomass and secondary production) on a per square meter basis, as well as on a per gram organic matter basis (i.e., per gram leaf litter or fine benthic organic matter), I was able to somewhat tease apart the effects nutrient-induced changes to food quantity versus food quality on consumer growth and production. Results from this study suggest an important interaction between larval lifespan and feeding behavior in determining species-specific responses to enrichment. Larval lifespan was apparently important in determining the response of leaf-eating consumers because long-lived taxa are highly susceptible to nutrient-induced food depletion. In contrast, larval lifespan was less important in determining the response of fine particulate feeders (i.e., gatherers) because their food supply is abundant and unaffected by enrichment.

Chapter 4 addressed community-level responses to nutrient enrichment as well as dynamics of benthic organic matter. This study, to my knowledge, is the first to quantify effects of enrichment on whole-assemblage secondary production in a detritus-based ecosystem. Here, I showed that total secondary production increased drastically in the treatment stream during enrichment, reaching the highest levels of secondary production recorded for streams at Coweeta. This response, however, was largely restricted to the dominant habitat (i.e., mixed substrate). Interestingly, production increased in the treatment stream despite a significant decline in benthic leaf litter (a dominant food resource), suggesting higher assimilation of leaf litter and other basal resources by detritivores during enrichment. Predator production also appeared to track increased availability of prey, but their ability to track prey production declined during the final year of study. Finally, years of enrichment were statistical outliers when plotted with long-term relationships between benthic organic matter and secondary production from streams at Coweeta. Important questions remain concerning the sustainability of this positive effect.

The final chapter (5) examined the effects of enrichment on invertebrate trophic basis of production and organic matter flow food webs. This chapter complemented the others by exploring the mechanistic basis for changes in secondary production through gut-content analysis. Originally, I hypothesized that consumer diets would shift to reflect changes in resource availability (e.g., increased fungi or algae, decreased leaf litter). Surprisingly, I found no large differences in the diets of primary consumers between streams before and during enrichment. Trophic support for primary consumers was predominantly based on amorphous detritus, leaf litter, and fungi and was unaffected by enrichment.

Although there was little effect of enrichment on the diets of consumers, I observed large effects on total organic matter flows to primary consumers. Were such large differences caused by relatively small between-stream differences in microbial biomass associated with detritus? Simple calculations revealed that small changes in microbial biomass could not possibly account for observed differences in secondary production and organic matter flows. This is an important result because it suggests that increased microbial activity and detrital degradation associated with nutrient enrichment increases the assimilability of detrital carbon for consumers. This is a slightly different mechanism than the commonly-held view that consumers primarily benefit from increased microbial biomass associated with detritus. These results, in combination with Chapter 2, also suggest that increased flows and secondary production may have been due to nutrient-induced reductions in consumer-resource elemental imbalances.

Taken together, the chapters of this dissertation demonstrate for the first time that nutrient enrichment can have strong effects on macro-consumers through changes in the quality and quantity of detrital resources. Results provide empirical support for conceptual theory that enrichment or increased resource productivity should positively influence consumers at all trophic positions (e.g., Polis and Strong). Nonetheless, interesting differences in response to enrichment occurred among taxa in relation to dominant life history characteristics. This dissertation should provide solid predictions for how landscape-scale nutrient enrichment may affect community structure, production, and material cycling. A productive avenue for future research will be testing whether results from this empirical study accurately predict the structure and production of invertebrate communities across a gradient of enrichment in forested streams.

Although the majority of nutrient effects on invertebrate productivity were positive, it is my hope that resource managers and policy makers will strongly consider potential long-term effects of enrichment on community structure (i.e., declines in biodiversity) and carbon loss from forested headwater streams.

Literature cited

Elser, J. J., and J. Urabe. 1999. The stoichiometry of consumer-driven nutrient recycling: theory, observations, and consequences. Ecology 80:735-751.

Greenwood, J. 2004. The response of heterotrophic and autotrophic resources to a long-term nutrient enrichment in an Appalachian headwater stream ecosystem. Dissertation. University of Georgia, Athens, GA, USA.

Gulis, V. and K. Suberkropp. 2003. Leaf litter decomposition and microbial activity in nutrient-enriched and unaltered reaches of a headwater stream. Freshwater Biology 48:123-134.

Hall, R. O. and J. L. Meyer. 1998. The trophic significance of bacteria in a detritus-based stream food web. Ecology 79:1995-2012.

Pearson, R. G. and N. M. Connolly. 2000. Nutrient enhancement, food quality and community dynamics in a tropical rainforest stream. Freshwater Biology 43:31-42.

Polis, G. A. and D. R. Strong. 1996. Food web complexity and community dynamics. American Naturalist 147:813-846.

Ramirez, A. and C. M. Pringle. In review. Response of detrital consumers to phosphorus levels in lowland tropical streams.

Rosemond, A. D., C. M. Pringle, A. Ramirez, and M. J. Paul. 2001. A test of top-down and bottom-up control in a detritus-based food web. Ecology 82:2279-2293.

Rosemond, A. D., C. M. Pringle, A. Ramirez, M. J. Paul, and J. L. Meyer. 2002. Landscape variation in phosphorus concentration and effects on detritus-based tropical streams. Limnology and Oceanography 47:278-289.

Sterner R.W. and J. J. Elser. 2002. Ecological Stoichiometry. Princeton University Press, Princeton, USA.

Appendix A. Growth rate data. Summary of growth data for non-Tanypodinae chironomid larvae and *Tallaperla* spp. in C53 (reference stream) and C54 (treatment stream) at Coweeta Hydrologic Laboratory, North Carolina, USA. Pretreat = before experimental enrichment began; Treatment = during enrichment of C54; T is the mean daily stream temperature during the incubation; (i) an (f) refer to the initial and final mean size of larvae (mm) or numbers of larvae (n) in incubation chambers. g is the instantaneous growth rate (mg mg^{-1} d^{-1}) of larvae during the incubation interval; % mortality is calculated as n(f)/n(i) * 100.

treatment	dates of incubation	mm (i)	mm (f)	T (°C)	degree days	g	n (i)	n (f)	% mortality	% mortality/d
Non-Tanypodinae chironomids										
Pretreat 53	7 Nov - 21 Nov 1999	1.25	2.25	11.3	169	0.116	30	18	40.0	2.9
Pretreat 53	7 Nov - 21 Nov 1999	1.91	2.30	11.3	169	0.037	50	12	76.0	5.4
Pretreat 53	7 Nov - 21 Nov 1999	2.92	3.09	11.3	169	0.015	19	4	78.9	5.6
Pretreat 53	21 Feb – 7 Mar 2000	1.18	2.01	9.6	154	0.098	51	27	47.1	3.4
Pretreat 53	21 Feb – 7 Mar 2000	1.87	2.42	9.6	154	0.048	94	21	77.7	5.5
Pretreat 53	21 Feb – 7 Mar 2000	2.87	3.50	9.6	154	0.036	29	8	72.4	5.2
Pretreat 53	25 May - 6 Jun 2000	1.96	2.48	14.9	193	0.066	59	16	72.9	6.1
Pretreat 53	25 May - 6 Jun 2000	2.81	3.19	14.9	193	0.030	26	15	42.3	3.5
Pretreat 54	7 Nov - 21 Nov 1999	1.20	1.78	12.1	181	0.086	30	13	56.7	4.0
Pretreat 54	7 Nov - 21 Nov 1999	2.83	3.45	12.1	181	0.040	20	3	85.0	6.1
Pretreat 54	21 Feb – 7 Mar 2000	1.21	2.02	9.5	152	0.094	53	22	58.5	4.2
Pretreat 54	21 Feb – 7 Mar 2000	1.92	2.60	9.5	152	0.057	71	31	56.3	4.0
Pretreat 54	21 Feb – 7 Mar 2000	2.93	3.29	9.5	152	0.027	29	13	55.2	3.9
Pretreat 54	25 May - 6 Jun 2000	1.27	1.72	14.2	184	0.077	40	5	87.5	7.3
Pretreat 54	25 May - 6 Jun 2000	1.97	2.32	14.2	184	0.043	59	8	86.4	7.2
Pretreat 54	25 May - 6 Jun 2000	2.85	3.71	14.2	184	0.059	30	4	86.7	7.2
Treatment 53	9 Sep - 17 Sep 2000	1.32	1.66	16.2	146	0.081	40	16	60.0	7.5
Treatment 53	9 Sep - 17 Sep 2000	1.94	2.59	16.2	146	0.107	82	10	87.8	11.0
Treatment 53	8 Dec - 20 Dec 2000	1.25	1.95	6.7	87	0.110	67	27	59.7	5.0
Treatment 53	8 Dec - 20 Dec 2000	1.91	2.44	6.7	87	0.063	73	11	84.9	7.1
Treatment 53	8 Dec - 20 Dec 2000	2.55	3.18	6.7	87	0.053	19	8	57.9	4.8
Treatment 53	17 Mar - 30 Mar 2001	1.32	1.85	8.5	119	0.074	61	20	67.2	5.2

Appendix A. (Continued)

treatment	dates of incubation	mm (i)	mm (f)	T (°C)	degree days	g	n (i)	n (f)	% mortality	% mortality/d
Treatment 53	17 Mar - 30 Mar 2001	2.82	3.33	8.5	119	0.038	31	16	48.4	3.7
Treatment 53	17 Jun - 27 Jun 2001	2.04	2.33	15.1	166	0.054	49	19	61.2	6.1
Treatment 53	17 Jun - 27 Jun 2001	3.46	3.84	15.1	166	0.030	15	2	86.7	8.7
Treatment 53	14 Aug - 22 Aug 2001	1.21	1.38	17.1	154	0.059	59	10	83.1	10.4
Treatment 53	14 Aug - 22 Aug 2001	1.89	2.04	17.1	154	0.030	40	21	47.5	5.9
Treatment 53	5 Dec - 20 Dec 2001	1.22	1.73	11.3	181	0.067	51	32	37.3	2.5
Treatment 53	5 Dec - 20 Dec 2001	1.83	2.19	11.3	181	0.035	68	27	60.3	4.0
Treatment 53	5 Dec - 20 Dec 2001	2.99	3.34	11.3	181	0.023	17	9	47.1	3.1
Treatment 53	20 Jun - 2 Jul 2002	1.23	1.80	15.6	203	0.086	50	3	94.0	7.8
Treatment 53	20 Jun - 2 Jul 2002	3.06	4.37	15.6	203	0.081	21	5	76.2	6.3
Treatment 54	9 Sep - 17 Sep 2000	1.25	1.92	15.5	140	0.146	41	4	90.2	11.3
Treatment 54	9 Sep - 17 Sep 2000	2.14	2.76	15.5	140	0.094	50	10	80.0	10.0
Treatment 54	9 Sep - 17 Sep 2000	2.93	3.50	15.5	140	0.061	42	14	66.7	8.3
Treatment 54	8 Dec - 20 Dec 2000	1.25	2.18	6.7	87	0.134	62	14	77.4	6.5
Treatment 54	8 Dec - 20 Dec 2000	1.86	2.62	6.7	87	0.083	86	28	67.4	5.6
Treatment 54	8 Dec - 20 Dec 2000	2.59	3.58	6.7	87	0.073	13	5	61.5	5.1
Treatment 54	17 Mar - 30 Mar 2001	1.30	1.96	8.2	115	0.092	50	15	70.0	5.4
Treatment 54	17 Mar - 30 Mar 2001	1.99	2.64	8.2	115	0.062	63	31	50.8	3.9
Treatment 54	17 Mar - 30 Mar 2001	2.86	3.55	8.2	115	0.047	44	19	56.8	4.4
Treatment 54	17 Jun - 27 Jun 2001	1.25	1.82	14.4	159	0.127	33	2	93.9	9.4
Treatment 54	14 Aug - 22 Aug 2001	1.21	1.86	16.2	146	0.153	70	4	94.3	11.8
Treatment 54	14 Aug - 22 Aug 2001	1.91	2.42	16.2	146	0.080	60	14	76.7	9.6
Treatment 54	14 Aug - 22 Aug 2001	3.18	3.87	16.2	146	0.066	21	3	85.7	10.7
Treatment 54	5 Dec - 20 Dec 2001	1.20	2.30	11.4	182	0.124	68	37	45.6	3.0
Treatment 54	5 Dec - 20 Dec 2001	1.80	2.64	11.4	182	0.075	80	12	85.0	5.7
Treatment 54	5 Dec - 20 Dec 2001	3.12	3.92	11.4	182	0.044	20	10	50.0	3.3
Treatment 54	20 Jun - 2 Jul 2002	1.95	2.86	15.0	195	0.088	32	3	90.6	7.6

Appendix A. (Continued)

treatment	dates of incubation	mm (i)	mm (f)	T (°C)	degree days	g	n (i)	n (f)	% mortality	% mortality/d
***Tallaperla* spp.**										
Pretreat 53	3 Nov 99 - 9 Jan 00	4.30	4.94	9.3	634	0.0031	30	22	26.7	0.4
Pretreat 53	3 Nov 99 - 9 Jan 00	8.30	8.52	9.3	634	0.0012	15	9	40.0	0.6
Pretreat 53	7 Feb 00 - 3 Apr 00	4.50	5.34	9.8	559	0.0085	40	21	47.5	0.8
Pretreat 53	7 Feb 00 - 3 Apr 00	6.20	6.86	9.8	559	0.0050	27	24	11.1	0.2
Pretreat 53	23 Apr 00 - 20 Jun 00	3.20	3.95	14	827	0.0103	36	33	8.3	0.1
Pretreat 53	23 Apr 00 - 20 Jun 00	5.30	6.23	14	827	0.0081	30	22	26.7	0.5
Pretreat 54	3 Nov 99 - 9 Jan 00	4.30	5.22	10.1	688	0.0087	30	10	66.7	1.0
Pretreat 54	3 Nov 99 - 9 Jan 00	8.20	8.24	10.1	688	0.0002	17	9	47.1	0.7
Pretreat 54	7 Feb 00 - 3 Apr 00	4.50	5.28	9.6	549	0.0083	40	27	32.5	0.6
Pretreat 54	7 Feb 00 - 3 Apr 00	6.20	7.02	9.6	549	0.0067	26	21	19.2	0.3
Pretreat 54	23 Apr 00 - 20 Jun 00	3.30	3.80	13.5	798	0.0068	35	33	5.7	0.1
Pretreat 54	23 Apr 00 - 20 Jun 00	5.20	5.98	13.5	798	0.0068	30	25	16.7	0.3
Treatment 53	26 Jul 00 - 29 Sep 00	2.80	3.62	16.7	1100	0.0118	20	7	65.0	1.0
Treatment 53	26 Jul 00 - 29 Sep 00	5.66	6.22	16.7	1100	0.0042	18	7	61.1	0.9
Treatment 53	26 Jul 00- 29 Sep 00	6.69	6.84	16.7	1100	0.0010	5	4	20.0	0.3
Treatment 53	6 Nov 00 - 10 Jan 01	4.72	5.44	6.2	412	0.0064	9	5	44.4	0.7
Treatment 53	6 Nov 00 - 10 Jan 01	6.25	6.54	6.2	412	0.0025	35	30	14.3	0.2
Treatment 53	6 Nov 00 - 10 Jan 01	9.13	9.32	6.2	412	0.0009	6	2	66.7	1.0
Treatment 53	10 Feb 01- 12 Apr 01	1.95	2.19	9.4	584	0.0052	39	23	41.0	0.7
Treatment 53	10 Feb 01 - 12 Apr 01	5.60	6.29	9.4	584	0.0054	25	23	8.0	0.1
Treatment 53	10 Feb 01 - 12 Apr 01	7.60	7.75	9.4	584	0.0009	10	7	30.0	0.5
Treatment 53	20 May 01 - 13 Jul 01	4.07	4.72	14.8	813	0.0077	30	13	56.7	1.0
Treatment 53	20 May 01 - 13 Jul 01	6.17	7.33	14.8	813	0.0092	20	8	60.0	1.1
Treatment 53	4 Sep 01 - 7 Nov 01	4.82	5.54	13.3	784	0.0061	20	14	30.0	0.5
Treatment 54	26 Jul 00 - 29 Sep 00	5.60	5.81	15.8	1046	0.0019	21	11	47.6	0.7
Treatment 54	6 Nov 00 - 10 Jan 01	6.34	6.76	6.4	422	0.0032	30	6	80.0	1.2
Treatment 54	10 Feb 01 - 12 Apr 01	1.95	2.56	8.9	551	0.0130	40	13	67.5	1.1

Appendix A. (Continued)

treatment	dates of incubation	mm (i)	mm (f)	T (°C)	degree days	g	n (i)	n (f)	% mortality	% mortality/d
Treatment 54	10 Feb 01 - 12 Apr 01	5.50	6.41	8.9	551	0.0069	25	12	52.0	0.9
Treatment 54	10 Feb 01 - 12 Apr 01	8.10	8.10	8.9	551	0.0000	14	12	14.3	0.2
Treatment 54	20 May 01 - 13 Jul 01	4.04	4.81	14.1	774	0.0094	30	8	73.3	1.4
Treatment 54	20 May 01 - 13 Jul 01	6.16	7.87	14.1	774	0.0107	20	7	65.0	1.2
Treatment 54	4 Sep 01 - 7 Nov 01	2.42	3.37	13.5	799	0.0151	27	3	88.9	1.4
Treatment 54	4 Sep 01 - 7 Nov 01	4.71	5.82	13.5	799	0.0096	20	10	50.0	0.8

Appendix B. Abundance, biomass, and secondary production data. Mean annual abundance (A, no./m^2), biomass (B, mg AFDM m^{-2}), and production (P, mg AFDM m^{-2} y^{-1}) of taxa in each functional feeding group in mixed substrate and bedrock outcrop habitats in the reference stream (C53) and the treatment stream (C54). Insect orders are as follows: E = Ephemeroptera, P = Plecoptera, T = Trichoptera, C = Coleoptera, O = Odonata, and NI = non-insect. CPI = cohort production interval (Benke 1979) in days or, where noted, the assumed annual P/B value that was used for production calculations. Years are as follows: Pre-2 = September 1998 – August 1999, Pre-3 = September 1999 – August 2000, Enr-1 = September 2000 – August 2001, Enr-2 = September 2001 – August 2002. Pre-1 data (October 1984 – September 1985) can be found in Lugthart and Wallace 1992.

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Scrapers										
Epeorus sp.	340	E	C53	Pre 2	1	7	13	6	1	8
				Pre 3	34	2	6	16	8	29
				Enr 1	0	0	0	14	5	41
				Enr 2	0	0	0	4	1	8
			C54	Pre 2	0	0	0	2	<1	6
				Pre 3	4	<1	1	11	11	23
				Enr 1	4	16	19	18	53	146
				Enr 2	0	0	0	32	30	127
Baetis sp.	120	E	C53	Pre 2	0	0	0	22	0	4
				Pre 3	0	0	0	132	2	36
				Enr 1	0	0	0	66	1	16
				Enr 2	17	<1	2	94	5	70
			C54	Pre 2	0	0	0	36	1	20
				Pre 3	9	<1	1	41	1	18
				Enr 1	0	0	0	23	<1	6
				Enr 2	0	0	0	77	2	25

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Hydroptila sp.	365	T	C53	Pre 2	0	0	0	26	4	9
				Pre 3	33	6	6	48	8	16
				Enr 1	4	1	2	58	10	19
				Enr 2	33	6	7	86	14	16
			C54	Pre 2	0	0	0	56	9	15
				Pre 3	1	<1	<1	123	21	30
				Enr 1	0	0	0	58	10	20
				Enr 2	0	0	0	10	2	1
Neophylax sp.	213	T	C53	Pre 2	0	0	0	13	0	2
				Pre 3	1	0	0	9	0	1
				Enr 1	1	<1	1	11	<1	1
				Enr 2	0	0	0	40	<1	4
			C54	Pre 2	0	0	0	23	<1	3
				Pre 3	1	<1	2	12	2	10
				Enr 1	2	<1	2	43	1	12
				Enr 2	1	<1	<1	171	3	28
Ectopria sp.	365	C	C53	Pre 2	7	2	12	26	5	25
				Pre 3	10	1	7	90	22	94
				Enr 1	6	1	3	61	16	54
				Enr 2	7	3	8	32	15	34
			C54	Pre 2	2	<1	2	10	2	8
				Pre 3	5	2	4	6	8	14
				Enr 1	11	<1	3	60	15	38
				Enr 2	0	0	0	15	3	8

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Elmidae	365	C	C53	Pre 2	0	0	0	20	1	3
				Pre 3	33	0	1	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	2	<1	<1	1	<1	<1
			C54	Pre 2	0	0	0	6	<1	<1
				Pre 3	9	<1	1	7	1	1
				Enr 1	0	0	0	7	<1	1
				Enr 2	11	<1	1	3	<1	<1
Total Scapers			C53	Pre 2	8	9	25	112	10	50
				Pre 3	112	9	19	294	39	175
				Enr 1	10	1	6	209	31	131
				Enr 2	60	8	16	257	36	131
			C54	Pre 2	2	0	2	133	13	52
				Pre 3	29	2	9	200	44	96
				Enr 1	17	17	24	211	79	222
				Enr 2	12	1	1	308	40	190
Leuctra spp.	340	P	C53	Pre 2	918	49	180	30	1	3
				Pre 3	1539	73	392	49	2	9
				Enr 1	1271	41	304	119	2	17
				Enr 2	1454	55	382	395	11	106
			C54	Pre 2	297	17	99	116	4	26
				Pre 3	750	31	120	78	5	28
				Enr 1	3847	145	1145	83	5	29
				Enr 2	1056	55	349	43	1	12

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Tallaperla spp.	540	P	C53	Pre 2	785	92	218	306	45	143
				Pre 3	1574	127	334	251	54	160
				Enr 1	573	161	442	465	96	275
				Enr 2	613	173	360	622	112	333
			C54	Pre 2	239	51	81	292	37	90
				Pre 3	511	107	222	295	50	135
				Enr 1	681	171	335	258	35	91
				Enr 2	385	113	227	191	32	80
Lepidostoma spp.	246	T	C53	Pre 2	196	19	144	19	<1	4
				Pre 3	592	46	387	6	<1	2
				Enr 1	607	39	381	13	1	7
				Enr 2	615	76	667	31	1	14
			C54	Pre 2	171	6	54	26	<1	4
				Pre 3	396	74	533	9	<1	2
				Enr 1	365	109	725	16	<1	3
				Enr 2	402	83	765	65	2	25
Pycnopsyche spp.	275	T	C53	Pre 2	323	63	895	47	6	94
				Pre 3	92	34	367	4	1	15
				Enr 1	318	95	706	15	<1	88
				Enr 2	263	88	680	72	1	61
			C54	Pre 2	520	137	1349	60	2	76
				Pre 3	213	192	1356	6	0	5
				Enr 1	775	557	3497	10	0.2	115
				Enr 2	567	1152	8482	44	4	111

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Fattigia pele	664	T	C53	Pre 2	112	158	233	0	0	0
				Pre 3	64	74	194	0	0	0
				Enr 1	95	133	306	0	0	0
				Enr 2	143	135	323	0	0	0
			C54	Pre 2	167	55	152	0	0	0
				Pre 3	105	111	263	0	0	0
				Enr 1	264	149	435	0	0	0
				Enr 2	136	116	336	0	0	0
Psilotreta sp.	335	T	C53	Pre 2	35	10	33	0	0	0
				Pre 3	15	7	26	0	0	0
				Enr 1	3	9	19	0	0	0
				Enr 2	5	5	20	0	0	0
			C54	Pre 2	2	1	3	1	<1	<1
				Pre 3	10	11	55	0	0	0
				Enr 1	16	15	77	0	0	0
				Enr 2	15	41	100	0	0	0
Molophilus sp.	365	D	C53	Pre 2	555	64	318	1	<1	1
				Pre 3	446	60	307	0	0	0
				Enr 1	189	33	179	1	1	4
				Enr 2	207	39	237	0	0	0
			C54	Pre 2	116	23	86	0	0	0
				Pre 3	199	55	223	10	<1	1
				Enr 1	327	97	455	0	0	0
				Enr 2	156	70	325	6	0	5

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Tipula sp.	310	D	C53	Pre 2	31	220	961	1	7	48
				Pre 3	45	190	1018	5	10	81
				Enr 1	55	230	1406	15	47	323
				Enr 2	67	261	1583	18	38	98
			C54	Pre 2	44	134	721	3	14	109
				Pre 3	39	98	512	5	11	83
				Enr 1	63	233	1382	1	4	16
				Enr 2	75	328	1818	4	8	77
Lipsothrix sp.	*5	D	C53	Pre 2	1	2	8	0	0	0
				Pre 3	4	6	28	0	0	0
				Enr 1	1	<1	1	0	0	0
				Enr 2	0	0	0	0	0	0
			C54	Pre 2	1	3	14	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	4	15	74	0	0	0
				Enr 2	0	0	0	0	0	0
Limonia sp.	340	D	C53	Pre 2	2	1	3	0	0	0
				Pre 3	1	<1	1	0	0	0
				Enr 1	12	6	25	0	0	0
				Enr 2	9	6	24	0	0	0
			C54	Pre 2	9	<1	2	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	7	3	10	0	0	0
				Enr 2	6	6	19	10	<1	2

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Anchytarsus sp.	1095	C	C53	Pre 2	7	3	5	2	1	2
				Pre 3	23	7	13	0	0	0
				Enr 1	4	5	1	0	0	0
				Enr 2	13	10	14	0	0	0
			C54	Pre 2	6	1	4	0	0	0
				Pre 3	5	8	10	0	0	0
				Enr 1	57	10	1	0	0	0
				Enr 2	8	6	8	0	0	0
Cambarus bartoni	*5	NI	C53	Pre 2	4	349	203	0	0	0
				Pre 3	8	715	415	0	0	0
				Enr 1	6	118	68	0	0	0
				Enr 2	3	119	69	0	0	0
			C54	Pre 2	0	0	0	0	0	0
				Pre 3	2	249	144	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	0	0	0	0	0	0
Total Shredders		C	C53	Pre 2	2975	681	3198	406	59	407
				Pre 3	4393	623	3434	315	67	376
				Enr 1	3127	751	3327	628	147	438
				Enr 2	3387	849	4768	1139	164	1024
			C54	Pre 2	1572	428	2636	499	59	395
				Pre 3	2229	686	3531	403	67	395
				Enr 1	6403	1504	7802	368	44	163
				Enr 2	2805	1970	12805	362	47	410

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Paraleptophlebia sp.	340	E	C53	Pre 2	210	6	38	18	4	19
				Pre 3	265	15	109	11	1	8
				Enr 1	59	1	7	9	<1	1
				Enr 2	76	7	27	0	0	0
			C54	Pre 2	89	4	26	53	3	17
				Pre 3	140	12	65	86	3	27
				Enr 1	148	9	56	1	0	1
				Enr 2	142	13	56	29	1	4
Serratella sp.	330	E	C53	Pre 2	0	0	0	165	30	174
				Pre 3	129	4	51	99	24	154
				Enr 1	33	<1	9	156	28	218
				Enr 2	4	0	1	242	32	299
			C54	Pre 2	27	1	42	400	33	189
				Pre 3	61	1	10	383	93	541
				Enr 1	31	4	19	623	221	1216
				Enr 2	52	9	57	459	89	596
Stenonema sp.	340	E	C53	Pre 2	86	14	71	26	0	8
				Pre 3	129	21	120	1	<1	<1
				Enr 1	2	<1	3	44	11	43
				Enr 2	41	18	67	0	0	0
			C54	Pre 2	17	4	25	1	<1	1
				Pre 3	9	8	37	5	1	6
				Enr 1	18	23	84	0	0	0
				Enr 2	82	53	234	6	1	4

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Amphinemura sp.	300	P	C53	Pre 2	107	1	15	441	14	99
				Pre 3	211	10	66	297	25	136
				Enr 1	49	2	16	339	12	110
				Enr 2	188	5	79	479	25	283
			C54	Pre 2	45	1	10	746	27	169
				Pre 3	131	8	22	959	33	254
				Enr 1	327	26	139	1298	52	474
				Enr 2	918	97	975	1101	44	488
Soyedina sp.	300	P	C53	Pre 2	4	0	0	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	5	1	7	2	<1	3
				Enr 2	48	8	51	2	1	5
			C54	Pre 2	0	0	0	1	<1	0
				Pre 3	0	0	0	0	0	0
				Enr 1	42	3	15	0	0	0
				Enr 2	53	11	54	6	1	3
Lype diversa	332	T	C53	Pre 2	64	4	19	0	0	0
				Pre 3	258	12	71	1	<1	0
				Enr 1	58	7	48	4	<1	<1
				Enr 2	48	5	38	3	0	1
			C54	Pre 2	22	5	19	0	0	0
				Pre 3	41	4	24	0	0	0
				Enr 1	34	15	66	1	<1	1
				Enr 2	113	31	163	1	<1	2

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Chironomidae (non-Tanypodinae)	‡	D	C53	Pre 2	42495	109	1005	9164	18	293
				Pre 3	50931	134	1704	7997	18	322
				Enr 1	32079	83	1261	7422	19	333
				Enr 2	32143	98	1279	12751	35	582
			C54	Pre 2	19252	80	803	12393	30	480
				Pre 3	30658	194	1457	18214	43	854
				Enr 1	58493	204	3968	15240	44	1097
				Enr 2	41919	180	2817	11389	28	764
Leptotarus sp.	365	D	C53	Pre 2	13	107	536	0	0	0
				Pre 3	8	104	545	0	0	0
				Enr 1	2	162	1095	0	0	0
				Enr 2	0	0	0	0	0	0
			C54	Pre 2	2	28	95	0	0	0
				Pre 3	1	48	435	0	0	0
				Enr 1	6	40	205	0	0	0
				Enr 2	11	71	260	0	0	0
Nymphomyiidae	*5	D	C53	Pre 2	0	0	0	7	0	0
				Pre 3	0	0	0	4	<1	<1
				Enr 1	0	0	0	1	0	0
				Enr 2	0	0	0	6	<1	<1
			C54	Pre 2	3	<1	<1	102	1	3
				Pre 3	8	<1	<1	91	1	3
				Enr 1	6	<1	<1	90	1	3
				Enr 2	0	0	0	30	<1	1

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Ormosia sp.	*5	D	C53	Pre 2	4	1	3	0	0	0
				Pre 3	38	5	25	0	0	0
				Enr 1	3	3	14	0	0	0
				Enr 2	2	2	11	0	0	0
			C54	Pre 2	4	0	0	0	0	0
				Pre 3	1	0	1	0	0	0
				Enr 1	20	5	24	0	0	0
				Enr 2	3	3	13	0	0	0
Sciaridae	365	D	C53	Pre 2	547	11	56	3	<1	<1
				Pre 3	410	9	64	0	0	0
				Enr 1	259	8	43	0	0	0
				Enr 2	129	7	25	0	0	0
			C54	Pre 2	246	6	24	2	<1	<1
				Pre 3	302	17	71	1	<1	<1
				Enr 1	1355	33	187	1	<1	<1
				Enr 2	75	4	15	0	0	0
Copepoda	*18	NI	C53	Pre 2	22990	23	414	659	1	13
				Pre 3	33732	34	607	390	<1	7
				Enr 1	16441	17	297	1652	2	31
				Enr 2	20665	21	372	2850	3	52
			C54	Pre 2	12213	12	220	199	<1	4
				Pre 3	30563	31	551	287	<1	5
				Enr 1	59365	59	1069	1050	1	18
				Enr 2	43695	44	786	420	<1	7

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Nematoda	*5	NI	C53	Pre 2	14752	12	58	162	0	1
				Pre 3	13589	12	62	260	<1	2
				Enr 1	11117	12	62	266	<1	2
				Enr 2	10612	11	54	385	<1	2
			C54	Pre 2	2053	2	9	36	<1	<1
				Pre 3	13744	15	73	169	<1	1
				Enr 1	26327	29	144	223	<1	2
				Enr 2	20510	21	106	529	1	4
Oligochaeta	*5	NI	C53	Pre 2	6519	80	398	350	1	5
				Pre 3	7020	102	509	119	<1	1
				Enr 1	4905	96	482	225	1	3
				Enr 2	4239	88	439	461	1	3
			C54	Pre 2	1805	104	519	43	<1	1
				Pre 3	6201	153	767	192	1	7
				Enr 1	15636	201	1004	111	<1	2
				Enr 2	9849	235	1174	203	1	6
Total C-gatherers			C53	Pre 2	87778	261	3572	10993	68	704
				Pre 3	106711	357	3389	9180	69	629
				Enr 1	65011	230	2248	10119	74	743
				Enr 2	68194	269	2442	17180	97	1227
			C54	Pre 2	35777	220	1695	13976	94	863
				Pre 3	81858	442	3077	20388	175	1698
				Enr 1	161802	611	6775	18638	319	2813
				Enr 2	117410	700	6451	14173	165	1879

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Diplectrona modesta	332	T	C53	Pre 2	297	10	58	36	5	36
				Pre 3	430	21	187	7	3	23
				Enr 1	183	12	82	25	2	24
				Enr 2	232	15	106	10	3	20
			C54	Pre 2	128	18	108	62	10	48
				Pre 3	368	62	353	17	2	22
				Enr 1	517	59	320	1	1	2
				Enr 2	350	107	534	20	5	42
Diplectrona metaqui	332	T	C53	Pre 2	4	1	5	1	1	1
				Pre 3	0	0	0	0	0	0
				Enr 1	1	3	8	0	0	0
				Enr 2	2	3	8	10	5	28
			C54	Pre 2	0	0	0	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	1	1	5	0	0	0
				Enr 2	0	0	0	0	0	0
Parapsyche cardis	332	T	C53	Pre 2	68	1	20	904	135	871
				Pre 3	1	1	2	1217	157	1343
				Enr 1	1	<1	1	388	106	742
				Enr 2	1	<1	1	414	88	694
			C54	Pre 2	2	9	17	652	126	1000
				Pre 3	0	0	0	467	131	909
				Enr 1	0	0	0	404	112	759
				Enr 2	18	2	23	665	199	1422

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Wormaldia sp. (summer cohort)	130	T	C53	Pre 2	1	<1	3	6	1	12
				Pre 3	104	2	28	15	2	22
				Enr 1	31	5	42	7	2	22
				Enr 2	17	3	58	25	2	48
			C54	Pre 2	27	3	35	74	15	105
				Pre 3	15	4	29	14	3	24
				Enr 1	28	10	106	88	9	113
				Enr 2	16	3	41	23	1	28
Wormaldia sp. (winter cohort)	236	T	C53	Pre 2	3	0	1	7	1	3
				Pre 3	37	3	28	21	2	11
				Enr 1	19	1	13	13	2	11
				Enr 2	80	3	47	55	5	61
			C54	Pre 2	5	<1	2	36	5	31
				Pre 3	33	4	20	20	2	11
				Enr 1	106	14	94	92	5	46
				Enr 2	90	12	30	93	6	77
Dolophilodes distinctus	269	T	C53	Pre 2	1	1	1	0	0	0
				Pre 3	1	3	6	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	0	0	0	0	0	0
			C54	Pre 2	1	<1	2	1	<1	1
				Pre 3	0	0	0	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	0	0	0	10	6	28

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Simuliidae	180	D	C53	Pre 2	11	<1	3	250	2	35
				Pre 3	34	<1	3	276	5	57
				Enr 1	8	<1	1	205	3	39
				Enr 2	8	<1	1	62	1	13
			C54	Pre 2	0	0	0	125	2	19
				Pre 3	1	<1	<1	19	1	5
				Enr 1	38	2	16	109	4	46
				Enr 2	2	<1	2	53	3	26
Dixa sp.	365	D	C53	Pre 2	332	1	6	64	0	2
				Pre 3	273	2	12	26	1	2
				Enr 1	107	2	12	208	1	5
				Enr 2	205	3	23	81	1	7
			C54	Pre 2	42	<1	2	30	<1	1
				Pre 3	50	1	4	17	<1	1
				Enr 1	152	2	13	60	<1	2
				Enr 2	40	1	5	38	<1	3
Sphaeridae	280	NI	C53	Pre 2	63	4	18	0	0	0
				Pre 3	30	5	23	0	0	0
				Enr 1	23	2	20	0	0	0
				Enr 2	29	7	36	0	0	0
			C54	Pre 2	8	1	4	0	0	0
				Pre 3	18	2	14	0	0	0
				Enr 1	148	15	114	0	0	0
				Enr 2	106	9	70	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Total C-filterers			C53	Pre 2	778	18	114	1268	145	960
				Pre 3	909	37	289	1562	169	1459
				Enr 1	373	26	177	845	115	842
				Enr 2	573	33	279	657	104	871
			C54	Pre 2	212	32	170	979	158	1207
				Pre 3	484	72	420	554	138	972
				Enr 1	989	103	666	754	130	968
				Enr 2	621	134	706	902	220	1626
Cordulegaster sp.	1140	O	C53	Pre 2	63	99	161	0	0	0
				Pre 3	32	191	339	0	0	0
				Enr 1	86	88	158	0	0	0
				Enr 2	23	46	121	0	0	0
			C54	Pre 2	44	153	185	0	0	0
				Pre 3	79	151	370	0	0	0
				Enr 1	81	183	224	0	0	0
				Enr 2	27	218	223	0	0	0
Lanthus sp.	660	O	C53	Pre 2	42	115	277	1	0	3
				Pre 3	89	256	670	1	9	14
				Enr 1	62	185	488	0	0	0
				Enr 2	61	164	567	4	47	138
			C54	Pre 2	46	73	184	0	0	0
				Pre 3	159	272	674	1	<1	1
				Enr 1	65	191	439	0	0	0
				Enr 2	51	295	686	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Sweltsa sp.	630	P	C53	Pre 2	18	1	2	1	<1	<1
				Pre 3	138	2	6	0	0	0
				Enr 1	8	1	4	0	0	0
				Enr 2	4	2	4	0	0	0
			C54	Pre 2	16	1	1	0	0	0
				Pre 3	9	2	3	0	0	0
				Enr 1	2	3	2	0	0	0
				Enr 2	1	1	1	0	0	0
Beloneuria sp.	660	P	C53	Pre 2	32	33	80	6	10	43
				Pre 3	95	77	114	5	<1	<1
				Enr 1	20	75	190	6	1	6
				Enr 2	51	130	320	38	7	37
			C54	Pre 2	20	38	95	51	16	82
				Pre 3	179	182	566	233	13	61
				Enr 1	58	147	428	23	17	56
				Enr 2	29	117	290	16	6	24
Isoperla spp.	300	P	C53	Pre 2	310	8	81	76	7	42
				Pre 3	154	5	57	40	6	50
				Enr 1	147	8	79	36	1	15
				Enr 2	38	3	36	22	4	35
			C54	Pre 2	116	8	56	124	17	116
				Pre 3	149	12	36	184	27	125
				Enr 1	489	33	257	149	24	156
				Enr 2	271	51	368	188	26	198

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Malerikus hastatus	660	P	C53	Pre 2	1	<1	<1	1	1	<1
				Pre 3	0	0	0	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	0	0	0	0	0	0
			C54	Pre 2	5	2	7	2	1	2
				Pre 3	1	1	1	10	4	8
				Enr 1	0	0	0	0	0	0
				Enr 2	0	0	0	1	1	<1
Rhyacophila spp.	340	T	C53	Pre 2	158	12	74	20	6	49
				Pre 3	305	17	130	41	4	36
				Enr 1	47	15	103	78	4	64
				Enr 2	87	12	94	41	7	64
			C54	Pre 2	82	19	122	127	7	83
				Pre 3	71	36	177	122	29	178
				Enr 1	167	60	388	104	13	112
				Enr 2	176	57	424	180	20	169
Pseudogoera singularis	365	T	C53	Pre 2	3	1	3	131	3	29
				Pre 3	2	<1	<1	62	4	20
				Enr 1	17	<1	2	56	1	9
				Enr 2	2	<1	2	67	4	16
			C54	Pre 2	22	3	12	175	2	17
				Pre 3	14	15	43	168	9	90
				Enr 1	19	14	16	236	28	163
				Enr 2	44	23	132	274	19	101

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Ceratopognidae	365	D	C53	Pre 2	4410	132	865	108	7	32
				Pre 3	7052	233	1272	110	3	16
				Enr 1	4172	144	983	105	3	25
				Enr 2	3704	154	794	89	3	23
			C54	Pre 2	1651	78	427	106	9	35
				Pre 3	3909	194	1001	73	4	21
				Enr 1	7001	290	1850	51	2	13
				Enr 2	3568	195	1055	46	4	21
Hexatoma spp.	365	D	C53	Pre 2	865	82	472	0	0	0
				Pre 3	1048	106	522	10	<1	1
				Enr 1	597	108	703	18	2	16
				Enr 2	312	46	334	1	<1	2
			C54	Pre 2	454	67	364	6	1	7
				Pre 3	823	134	799	9	<1	18
				Enr 1	1087	183	1161	10	1	10
				Enr 2	348	108	666	6	<1	2
nr. *Pedicia* sp.	340	D	C53	Pre 2	78	15	84	0	0	0
				Pre 3	78	14	92	0	0	0
				Enr 1	6	35	684	0	0	0
				Enr 2	70	30	186	0	0	0
			C54	Pre 2	0	0	0	0	0	0
				Pre 3	5	21	62	0	0	0
				Enr 1	14	24	130	0	0	0
				Enr 2	3	3	20	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Pedicia sp.	365	D	C53	Pre 2	8	28	88	0	0	0
				Pre 3	8	38	14	3	<1	1
				Enr 1	10	26	84	0	0	0
				Enr 2	4	30	78	5	1	10
			C54	Pre 2	5	1	5	1	<1	<1
				Pre 3	5	52	172	0	0	0
				Enr 1	4	52	80	0	0	0
				Enr 2	6	50	64	0	0	0
Dicranota spp.	310	D	C53	Pre 2	32	17	79	66	1	12
				Pre 3	277	10	86	42	1	11
				Enr 1	85	5	42	22	1	7
				Enr 2	85	4	37	91	2	25
			C54	Pre 2	20	3	27	27	1	8
				Pre 3	77	7	36	97	1	21
				Enr 1	216	30	163	78	2	18
				Enr 2	142	37	235	132	4	44
Glutops sp.	365	D	C53	Pre 2	5	13	39	0	0	0
				Pre 3	2	11	28	0	0	0
				Enr 1	2	19	32	0	0	0
				Enr 2	4	15	51	0	0	0
			C54	Pre 2	17	42	148	0	0	0
				Pre 3	18	56	185	0	0	0
				Enr 1	11	53	155	0	0	0
				Enr 2	20	33	147	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Tanypodinae	340	D	C53	Pre 2	3765	4	58	186	1	7
				Pre 3	5435	12	111	160	<1	4
				Enr 1	1585	6	50	170	1	5
				Enr 2	1449	7	51	219	1	8
			C54	Pre 2	280	2	15	49	<1	3
				Pre 3	2038	15	97	233	1	6
				Enr 1	3057	18	125	177	1	7
				Enr 2	2477	22	143	45	<1	2
Empididae	340	D	C53	Pre 2	370	2	23	104	2	12
				Pre 3	93	1	5	108	3	21
				Enr 1	154	2	13	67	2	13
				Enr 2	90	2	12	119	1	9
			C54	Pre 2	79	<1	5	108	3	17
				Pre 3	156	2	16	185	5	28
				Enr 1	361	9	51	139	2	13
				Enr 2	141	7	35	75	1	7
Pilaria sp.	365	D	C53	Pre 2	3	0	1	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	1	<1	<1	0	0	0
				Enr 2	0	0	0	0	0	0
			C54	Pre 2	0	0	0	0	0	0
				Pre 3	0	0	0	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	1	<1	<1	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Pseudolimnolphila sp.	365	D	C53	Pre 2	6	4	11	1	1	3
				Pre 3	48	5	37	0	0	0
				Enr 1	89	14	81	2	<1	5
				Enr 2	153	20	102	3	<1	1
			C54	Pre 2	55	17	50	1	<1	<1
				Pre 3	109	41	175	0	0	0
				Enr 1	341	82	434	0	0	0
				Enr 2	339	182	676	10	<1	3
Rhabdomastix sp.	*5	D	C53	Pre 2	1	0	2	0	0	0
				Pre 3	3	2	9	0	0	0
				Enr 1	1	<1	2	0	0	0
				Enr 2	0	0	0	1	<1	1
			C54	Pre 2	1	<1	1	1	2	9
				Pre 3	3	8	41	0	0	0
				Enr 1	0	0	0	0	0	0
				Enr 2	7	16	82	0	0	0
Dolichopodidae	300	D	C53	Pre 2	10	9	33	0	0	0
				Pre 3	46	13	66	0	0	0
				Enr 1	7	6	21	0	0	0
				Enr 2	11	6	21	1	<1	3
			C54	Pre 2	28	2	14	0	0	0
				Pre 3	78	25	126	0	0	0
				Enr 1	63	23	83	0	0	0
				Enr 2	12	6	25	0	0	0

Appendix B (cont.)

					Mixed Substrate			Bedrock Outcrops		
Functional group or taxon	CPI	Order	Site	Year	A	B	P	A	B	P
Turbellaria	*5	NI	C53	Pre 2	38	0	2	34	0	2
				Pre 3	364	3	15	98	1	6
				Enr 1	49	<1	2	40	1	4
				Enr 2	46	1	4	93	1	7
			C54	Pre 2	61	1	4	16	<1	1
				Pre 3	28	<1	1	20	<1	2
				Enr 1	26	<1	2	12	<1	1
				Enr 2	60	1	5	21	<1	1
Acari	*5	NI	C53	Pre 2	2485	7	33	1726	5	23
				Pre 3	4820	13	64	1727	5	23
				Enr 1	1937	5	26	1130	3	15
				Enr 2	2118	6	28	750	2	10
			C54	Pre 2	767	2	10	485	1	7
				Pre 3	1446	4	19	693	2	9
				Enr 1	3653	10	49	755	2	10
				Enr 2	1602	4	21	641	2	9
Total invertebrate predators			C53	Pre 2	12703	582	2468	2461	44	257
				Pre 3	20088	1009	3635	2406	37	203
				Enr 1	9080	741	3745	1730	20	184
				Enr 2	8312	678	2839	1544	80	386
			C54	Pre 2	3769	512	1732	1280	60	385
				Pre 3	9353	1232	4598	2026	96	567
				Enr 1	16712	1404	6036	1734	91	559
				Enr 2	9325	1426	5297	1636	83	581

Appendix B (cont.)

Functional group or taxon	CPI	Order	Site	Year	Mixed Substrate A	Mixed Substrate B	Mixed Substrate P	Bedrock Outcrops A	Bedrock Outcrops B	Bedrock Outcrops P
Eurycea sp.	365	NI	C53	Pre 2	1	5	9	0	0	0
				Pre 3	2	11	40	0	0	0
				Enr 1	1	2	3	0	0	0
				Enr 2	1	2	8	0	0	0
			C54	Pre 2	2	17	24	0	0	0
				Pre 3	2	18	15	2	6	35
				Enr 1	0	0	0	0	0	0
				Enr 2	1	4	5	0	0	0
Desmognathus spp.	880	NI	C53	Pre 2	2	54	68	1	13	37
				Pre 3	3	68	120	2	47	56
				Enr 1	4	75	84	0	0	0
				Enr 2	2	55	119	0	0	0
			C54	Pre 2	1	53	46	2	44	171
				Pre 3	2	62	82	0	0	0
				Enr 1	2	30	38	0	0	0
				Enr 2	2	37	89	1	85	140
Total vertebrate predators (salamanders)			C53	Pre 2	3	59	77	1	13	37
				Pre 3	4	79	160	2	47	56
				Enr 1	4	77	88	0	0	0
				Enr 2	2	57	128	0	0	0
			C54	Pre 1	3	70	70	2	44	171
				Pre 2	4	81	97	2	6	35
				Enr 1	2	30	38	0	0	0
				Enr 2	3	41	94	1	85	140

Appendix C. Invertebrate gut contents (raw proportions). Mean percent (%) by area of food types in the guts of primary consumers (adjusted for fungal content) in the reference stream (C53) and treatment stream (54).

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Reference stream							
Shredders							
Anchytarsus sp.	6/2000	21.6	14.4	1.2	62.4	0.4	0.0
	3/2001	20.7	0.0	0.8	78.3	0.1	0.0
	5/2001	32.1	7.1	1.5	57.4	1.8	0.0
	8/2001	54.7	6.1	2.4	36.8	0.0	0.0
	7/2002	40.0	0.0	1.6	58.1	0.2	0.0
Fattigia pele	6/1999	87.2	7.8	3.7	1.3	0.0	0.0
	3/2000	78.0	16.5	3.6	1.9	0.0	0.0
	6/2000	59.6	15.5	2.8	22.2	0.0	0.0
	11/2000	80.1	5.7	3.4	10.5	0.3	0.0
	3/2001	64.9	0.0	2.6	27.0	5.5	0.0
	5/2001	91.9	2.2	3.8	2.2	0.0	0.0
	8/2001	72.0	4.3	3.0	20.7	0.1	0.0
	12/2001	78.1	8.1	3.4	10.3	0.1	0.0
	3/2002	72.7	7.9	3.1	16.3	0.0	0.0
	7/2002	73.9	13.3	3.3	9.0	0.6	0.0
Lepidostoma spp.	6/1999	82.1	11.5	3.6	2.8	0.0	0.0
	3/2000	91.8	0.0	3.7	4.5	0.0	0.0
	6/2000	61.5	13.9	2.8	21.8	0.0	0.0
	11/2000	88.8	0.0	3.6	7.6	0.0	0.0
	3/2001	68.5	2.3	2.8	26.2	0.2	0.0
	5/2001	79.1	4.5	3.3	12.2	0.1	0.8
	8/2001	80.8	0.0	3.3	15.9	0.0	0.0
	12/2001	88.2	0.0	3.6	8.2	0.0	0.0
	3/2002	90.8	0.0	3.7	5.5	0.0	0.0
	7/2002	83.4	2.8	3.5	10.2	0.2	0.0
Leuctra spp.	3/2000	42.5	0.5	1.7	55.2	0.0	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Leuctra spp. (cont.)	5/2001	36.7	0.0	1.5	61.9	0.0	0.0
	8/2001	58.4	0.0	2.4	39.3	0.0	0.0
	12/2001	50.0	0.0	2.0	47.8	0.2	0.0
	3/2002	74.6	0.0	3.0	21.7	0.0	0.6
	7/2002	68.8	2.3	2.9	25.8	0.3	0.0
Pycnopsyche spp.	6/1999	55.4	41.2	3.2	0.1	0.1	0.0
	3/2000	92.8	3.0	3.8	0.3	0.0	0.0
	6/2000	91.4	3.5	3.8	1.4	0.0	0.0
	11/2000	82.6	1.9	3.4	11.5	0.5	0.0
	3/2001	86.7	3.5	3.6	6.0	0.1	0.0
	5/2001	78.6	10.0	3.4	7.7	0.2	0.0
	12/2001	84.5	0.8	3.5	11.1	0.0	0.1
	3/2002	85.3	2.4	3.5	8.7	0.0	0.0
	7/2002	74.7	10.6	3.3	11.4	0.0	0.0
Tallaperla spp.	3/2000	85.6	4.7	3.6	5.9	0.2	0.0
	6/2000	65.7	8.1	2.9	23.3	0.0	0.0
	11/2000	60.5	3.8	2.6	33.2	0.0	0.0
	3/2001	63.2	8.2	2.8	25.7	0.1	0.0
	5/2001	56.3	0.7	2.3	39.8	0.9	0.0
	8/2001	70.3	1.7	2.9	25.1	0.0	0.0
	12/2001	65.8	2.5	2.7	28.9	0.0	0.0
	3/2002	89.8	0.0	3.7	6.5	0.0	0.0
	7/2002	82.4	0.0	3.4	14.2	0.1	0.0
Tipula spp.	6/1999	89.7	4.0	3.7	2.5	0.0	0.0
	3/2000	83.0	12.8	3.7	0.5	0.0	0.0
	6/2000	86.2	5.1	3.6	5.0	0.0	0.0
	11/2000	83.4	3.9	3.5	9.1	0.0	0.0
	3/2001	57.6	22.9	2.9	16.6	0.0	0.0
	5/2001	74.1	3.0	3.1	19.7	0.2	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Tipula spp. (cont.)	8/2001	75.4	10.7	3.3	10.6	0.0	0.0
	12/2001	84.4	0.0	3.4	11.7	0.4	0.0
	3/2002	93.3	0.4	3.8	2.3	0.1	0.0
	7/2002	77.9	6.5	3.3	12.3	0.0	0.0
Enriched stream							
Shredders							
Anchytarsus sp.	3/2001	60.8	0.0	3.4	35.2	0.6	0.0
	5/2001	67.4	10.7	5.0	16.6	0.3	0.0
	7/2002	48.4	10.4	3.9	35.8	1.4	0.0
Fattigia pele	6/1999	81.9	10.2	3.6	4.3	0.0	0.0
	3/2000	86.4	4.1	3.6	5.8	0.0	0.0
	6/2000	77.6	11.0	3.4	8.0	0.0	0.0
	11/2000	81.8	9.1	5.6	3.5	0.0	0.0
	3/2001	85.8	0.0	4.8	6.1	3.3	0.0
	5/2001	83.0	3.0	5.0	8.9	0.0	0.0
	8/2001	64.5	7.3	4.5	23.7	0.0	0.0
	12/2001	80.0	3.8	4.9	11.3	0.0	0.0
	3/2002	69.7	0.0	3.9	26.4	0.0	0.0
	7/2002	63.2	5.4	4.2	26.6	0.6	0.0
Lepidostoma spp.	6/1999	87.9	5.9	3.7	2.0	0.5	0.0
	3/2000	85.0	1.7	3.5	9.9	0.0	0.0
	6/2000	74.4	11.1	3.3	11.0	0.1	0.0
	11/2000	83.9	5.7	5.4	5.0	0.1	0.0
	3/2001	82.3	0.0	4.6	11.5	1.6	0.0
	5/2001	80.7	2.7	4.9	8.7	3.1	0.0
	8/2001	84.1	3.7	5.2	4.9	2.1	0.0
	3/2002	74.3	3.7	4.6	17.4	0.0	0.0
	7/2002	67.6	16.4	5.7	10.3	0.1	0.0
Leuctra spp.	3/2000	79.5	4.6	3.3	12.4	0.1	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Leuctra spp. (cont.)	6/2000	10.9	0.0	0.4	86.1	2.6	0.0
	11/2000	58.1	0.0	3.3	38.6	0.0	0.0
	3/2001	71.3	0.0	4.0	23.4	1.3	0.0
	5/2001	54.7	7.4	3.9	33.6	0.4	0.0
	8/2001	79.6	3.2	4.9	12.3	0.0	0.0
	12/2001	52.9	0.0	3.0	44.0	0.1	0.0
	3/2002	51.7	12.1	4.3	31.7	0.2	0.0
	7/2002	43.0	0.0	2.4	54.6	0.0	0.0
Pycnopsyche spp.	6/1999	68.1	23.4	3.3	5.2	0.0	0.0
	3/2000	80.7	7.3	3.5	8.5	0.0	0.0
	6/2000	75.8	17.1	3.5	3.6	0.0	0.0
	11/2000	84.5	0.0	4.8	10.0	0.7	0.0
	3/2001	80.0	1.9	4.7	3.9	9.4	0.0
	5/2001	82.8	4.9	5.2	7.1	0.0	0.0
	12/2001	84.1	1.3	4.9	9.7	0.0	0.0
	3/2002	80.0	0.7	4.6	12.6	2.0	0.0
	7/2002	79.9	4.1	5.0	11.1	0.0	0.0
Tallaperla spp.	6/1999	91.3	0.0	3.7	4.8	0.3	0.0
	3/2000	84.1	0.4	3.4	12.0	0.1	0.0
	6/2000	67.5	9.8	3.0	19.7	0.1	0.0
	11/2000	83.6	3.0	5.1	8.4	0.0	0.0
	3/2001	74.4	0.0	4.2	21.0	0.4	0.0
	5/2001	74.3	9.4	5.3	10.6	0.4	0.0
	12/2001	82.3	1.8	4.8	11.1	0.0	0.0
	3/2002	87.4	0.0	4.9	7.4	0.2	0.0
	7/2002	82.8	2.1	4.9	10.2	0.0	0.0
Tipula spp.	6/1999	95.1	0.5	3.9	0.5	0.0	0.0
	3/2000	80.5	7.0	3.4	9.0	0.0	0.0
	6/2000	70.9	11.4	3.2	14.3	0.2	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Tipula spp. (cont.)	11/2000	88.1	0.3	5.0	6.0	0.6	0.0
	3/2001	62.8	2.1	3.8	18.2	13.1	0.0
	5/2001	81.0	2.1	4.8	11.9	0.1	0.0
	12/2001	79.0	6.2	5.2	9.6	0.0	0.0
	3/2002	73.2	6.4	4.9	15.5	0.1	0.0
	7/2002	75.5	5.4	4.9	14.0	0.2	0.0
Reference stream							
Gatherers							
Amphinemura sp.	3/2000	32.6	9.4	1.5	55.5	0.9	0.0
	6/2000	50.9	0.0	2.1	47.1	0.0	0.0
	11/2000	23.3	0.0	0.9	75.7	0.0	0.0
	5/2001	3.6	0.0	0.1	95.7	0.5	0.0
	12/2001	14.2	0.0	0.6	83.4	1.8	0.0
Chironomidae	6/1999	19.2	0.0	0.8	77.1	2.9	0.0
	11/2000	13.5	0.0	0.5	86.0	0.0	0.0
	3/2001	19.3	0.0	0.8	79.9	0.0	0.0
	5/2001	6.1	0.0	0.2	93.6	0.0	0.0
	8/2001	25.0	0.0	1.0	73.6	0.4	0.0
	12/2001	15.2	0.0	0.6	84.1	0.1	0.0
	3/2002	21.9	0.0	0.9	77.2	0.0	0.0
	7/2002	7.7	0.0	0.3	91.1	0.9	0.0
Oligochaetae	11/2000	28.0	13.8	1.5	55.0	1.8	0.0
	5/2001	31.9	0.0	1.3	66.8	0.0	0.0
	8/2001	19.6	0.0	0.8	79.7	0.0	0.0
	12/2001	34.0	0.0	1.4	63.2	1.4	0.0
	7/2002	58.3	0.0	2.4	36.8	2.6	0.0
Paraleptophlebia sp.	3/2000	0.0	0.0	0.0	99.6	0.4	0.0
	6/2000	59.9	17.3	2.8	19.9	0.0	0.0
	5/2001	6.8	0.0	0.3	92.8	0.2	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Paraleptophlebia sp. (cont.)	7/2002	0.0	0.0	0.0	92.9	7.1	0.0
Sciaridae	5/2001	83.3	0.0	3.4	6.1	7.2	0.0
	12/2001	52.4	0.0	2.1	45.5	0.0	0.0
	7/2002	76.7	0.0	3.1	20.2	0.0	0.0
Serratella sp.	3/2000	0.3	3.0	0.1	95.5	1.1	0.0
	11/2000	27.4	0.0	1.1	69.1	2.4	0.0
	3/2001	1.6	5.2	0.2	88.1	4.9	0.0
	5/2001	21.0	0.0	0.9	76.5	1.7	0.0
	12/2001	15.4	0.0	0.6	84.0	0.0	0.0
	3/2002	6.6	0.0	0.3	86.4	6.8	0.0
Stenonema sp.	6/1999	9.4	0.0	0.4	90.3	0.0	0.0
	3/2000	4.2	0.0	0.2	93.9	1.8	0.0
	6/2000	0.0	0.0	0.0	99.9	0.1	0.0
	11/2000	13.2	0.0	0.5	86.2	0.0	0.0
	3/2001	0.9	0.0	0.0	80.3	18.8	0.0
	5/2001	5.5	0.0	0.2	86.9	7.3	0.0
	12/2001	7.2	0.0	0.3	91.2	1.3	0.0
	3/2002	25.3	0.0	1.0	72.6	1.1	0.0
	7/2002	8.1	0.0	0.3	91.5	0.0	0.0
Enriched stream							
Gatherers							
Amphinemura sp.	3/2000	28.5	4.3	1.3	65.9	0.1	0.0
	6/2000	26.2	1.5	1.1	68.5	2.7	0.0
	11/2000	65.3	0.0	3.7	31.0	0.0	0.0
	3/2001	79.2	0.0	4.5	14.0	2.3	0.0
	5/2001	50.7	0.0	2.9	45.5	0.9	0.0
	12/2001	57.2	0.0	3.2	39.6	0.0	0.0
	3/2002	74.0	0.0	4.2	21.6	0.2	0.0
	7/2002	82.0	0.0	4.6	13.0	0.4	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Chironomidae	6/1999	11.8	0.0	0.5	87.7	0.1	0.0
	11/2000	9.7	0.0	0.5	89.4	0.3	0.0
	3/2001	13.5	0.0	0.8	85.7	0.0	0.0
	5/2001	7.0	0.0	0.4	92.6	0.0	0.0
	8/2001	14.7	0.0	0.8	84.5	0.0	0.0
	12/2001	14.0	0.0	0.8	85.3	0.0	0.0
	3/2002	17.6	0.0	1.0	80.9	0.4	0.0
	7/2002	22.1	0.0	1.2	76.7	0.0	0.0
Oligochaetae	3/2000	18.2	10.6	1.0	70.2	0.0	0.0
	6/2000	5.0	9.8	0.4	84.2	0.6	0.0
	11/2000	53.3	0.0	3.0	43.7	0.0	0.0
	5/2001	27.0	0.0	1.5	71.5	0.0	0.0
	7/2002	52.3	1.6	3.1	40.0	0.6	2.4
Paraleptophlebia sp.	3/2000	0.0	0.0	0.0	99.7	0.3	0.0
	11/2000	1.5	0.0	0.1	98.3	0.2	0.0
	5/2001	13.9	0.0	0.8	85.4	0.0	0.0
	12/2001	9.4	0.0	0.5	89.7	0.4	0.0
	3/2002	27.3	0.0	1.5	69.5	1.6	0.0
Sciaridae	7/2002	75.6	0.0	4.3	20.0	0.2	0.0
Serratella sp.	3/2000	9.2	0.0	0.4	84.8	5.7	0.0
	6/2000	20.0	4.4	0.9	73.3	1.3	0.0
	11/2000	21.4	0.0	1.2	74.9	2.5	0.0
	3/2001	14.1	0.0	0.8	68.9	16.2	0.0
	5/2001	32.1	1.4	2.0	64.4	0.1	0.0
	12/2001	10.6	7.4	1.4	69.0	9.8	1.7
	3/2002	13.0	0.0	0.7	75.7	10.6	0.0
Stenonema sp.	6/1999	12.4	0.0	0.5	84.4	2.7	0.0
	3/2000	0.7	1.4	0.1	96.9	1.0	0.0
	6/2000	3.6	0.5	0.2	94.0	1.7	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Stenonema sp. (cont.)	11/2000	5.6	0.0	0.3	94.0	0.2	0.0
	3/2001	0.3	1.5	0.2	86.2	11.8	0.0
	5/2001	9.7	0.0	0.5	88.5	1.3	0.0
	12/2001	7.4	3.2	0.8	88.6	0.0	0.0
	3/2002	25.1	0.7	1.5	71.0	1.8	0.0
Reference stream							
Filterers							
Diplectrona spp.	6/1999	43.2	8.6	2.0	23.4	1.7	21.2
	3/2000	61.7	5.5	2.6	24.6	0.4	5.2
	6/2000	37.6	11.1	1.8	38.7	0.0	10.8
	11/2000	39.9	7.2	1.8	48.6	1.0	1.6
	3/2001	34.4	20.9	1.9	40.9	0.0	1.9
	5/2001	42.4	2.0	1.8	47.0	0.7	6.1
	8/2001	80.7	0.3	3.3	14.1	0.1	1.5
	12/2001	50.1	5.6	2.2	41.2	0.4	0.5
Parapsyche cardis.	6/1999	34.7	6.6	1.6	35.5	0.3	21.4
	3/2000	33.0	11.7	1.6	45.6	0.0	8.0
	6/2000	30.0	4.8	1.3	37.2	0.2	26.5
	11/2000	40.7	1.2	1.7	40.1	0.9	15.4
	3/2001	32.9	5.2	1.5	56.5	2.9	1.1
	5/2001	38.3	2.0	1.6	47.2	0.6	10.4
	8/2001	68.3	2.5	2.8	13.3	0.1	13.0
	12/2001	32.9	8.6	1.5	47.5	0.4	9.0
	3/2002	63.9	0.6	2.6	25.3	2.3	5.3
	7/2002	66.2	4.7	2.8	22.6	1.4	2.2
Simuliidae	6/1999	20.1	9.4	1.0	68.2	0.0	1.3
	3/2000	0.2	0.0	0.0	98.3	1.5	0.0
	6/2000	9.0	0.0	0.4	90.5	0.1	0.0
	3/2001	0.0	0.0	0.0	95.7	4.3	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Simuliidae (cont.)	5/2001	9.7	0.0	0.4	87.9	2.1	0.0
	3/2002	10.6	0.0	0.4	86.1	2.8	0.0
Wormaldia spp.	3/2000	0.0	0.0	0.0	97.8	2.2	0.0
	6/2000	0.0	0.0	0.0	98.8	1.2	0.0
	11/2000	0.0	0.0	0.0	97.5	2.5	0.0
	5/2001	1.0	0.0	0.0	95.4	3.5	0.0
	8/2001	0.0	0.0	0.0	100.0	0.0	0.0
	12/2001	0.0	0.0	0.0	100.0	0.0	0.0
Enriched stream							
Filterers							
Diplectrona spp.	6/1999	66.3	2.4	2.8	21.1	1.0	6.4
	3/2000	35.4	21.9	2.0	27.5	0.2	13.1
	6/2000	27.1	16.5	1.5	51.6	0.0	3.3
	11/2000	59.4	0.0	3.4	37.2	0.0	0.0
	3/2001	52.4	4.5	3.5	36.0	1.8	1.9
	5/2001	60.2	3.6	3.8	20.1	0.2	12.1
	8/2001	65.7	0.0	3.7	28.1	0.2	2.2
	12/2001	49.6	6.9	3.6	39.6	0.1	0.2
	3/2002	46.3	6.8	3.4	39.5	0.5	3.5
	7/2002	33.5	0.0	1.9	54.1	0.8	9.7
Parapsyche cardis.	6/1999	20.0	0.4	0.8	41.2	0.8	36.6
	3/2000	56.5	10.3	2.5	23.3	0.2	7.2
	11/2000	43.5	1.8	2.7	38.5	0.4	13.2
	3/2001	49.4	2.8	3.1	18.6	0.3	25.7
	5/2001	32.7	0.0	1.8	37.7	0.1	27.7
	8/2001	46.6	0.0	2.6	28.8	0.5	21.5
	12/2001	45.5	5.9	3.2	27.4	0.5	17.4
	3/2002	45.0	0.0	2.5	23.3	0.1	29.1
	7/2002	30.2	4.2	2.2	34.2	0.1	29.2

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Simuliidae	6/1999	31.4	0.0	1.3	66.7	0.3	0.3
	6/2000	5.9	0.0	0.2	88.1	5.8	0.0
	3/2001	7.7	0.0	0.4	87.3	4.6	0.0
	5/2001	2.5	0.0	0.1	92.8	4.5	0.0
	3/2002	0.6	0.0	0.0	97.7	1.6	0.0
Wormaldia spp.	6/1999	6.3	0.0	0.3	93.4	0.0	0.0
	3/2000	2.3	0.0	0.1	93.2	4.5	0.0
	11/2000	0.7	0.0	0.0	98.9	0.4	0.0
	5/2001	0.0	0.0	0.0	97.1	2.9	0.0
	12/2001	4.6	0.0	0.3	94.9	0.2	0.0
	3/2002	0.0	0.0	0.0	99.6	0.4	0.0
Reference stream							
Scrapers							
Baetis sp.	3/2000	0.0	0.0	0.0	97.0	3.0	0.0
Ectopria sp.	3/2000	0.0	0.0	0.0	89.4	10.6	0.0
	3/2001	0.0	0.0	0.0	90.4	9.6	0.0
	8/2001	16.5	0.0	0.7	68.2	14.6	0.0
	3/2002	1.3	23.6	0.6	59.2	15.3	0.0
	7/2002	3.3	2.3	0.2	76.1	18.2	0.0
Epeorus sp.	6/1999	0.0	0.0	0.0	83.1	16.9	0.0
	3/2000	3.1	0.0	0.1	96.3	0.5	0.0
	11/2000	16.1	0.0	0.7	83.2	0.0	0.0
	3/2001	7.0	0.0	0.3	86.8	5.8	0.1
	12/2001	0.0	0.0	0.0	92.6	7.4	0.0
	3/2002	6.5	0.0	0.3	89.4	3.8	0.0
Neophylax sp.	3/2000	0.0	0.0	0.0	99.8	0.2	0.0
	3/2001	0.0	0.0	0.0	72.7	27.3	0.0
	3/2002	0.0	0.0	0.0	19.2	80.8	0.0

Appendix C (cont.)

Stream/Functional group/Taxon	Date	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Enriched stream							
Scrapers							
Baetis sp.	6/2000	28.1	0.0	1.1	64.3	6.5	0.0
	3/2002	1.7	0.0	0.1	93.7	4.6	0.0
Ectopria sp.	8/2001	10.6	0.0	0.6	75.9	12.9	0.0
	3/2002	27.5	0.0	1.6	52.9	18.0	0.0
Epeorus sp.	6/1999	0.0	0.0	0.0	94.2	5.8	0.0
	3/2000	0.0	2.3	0.1	93.3	4.3	0.0
	6/2000	0.2	0.0	0.0	95.2	4.5	0.0
	11/2000	0.7	0.0	0.0	90.7	8.5	0.0
	3/2001	0.0	0.0	0.0	76.4	23.6	0.0
	5/2001	3.6	0.0	0.2	93.3	2.9	0.0
	8/2001	0.0	0.0	0.0	98.6	1.4	0.0
Epeorus sp. (cont.)	12/2001	2.0	0.0	0.1	91.7	6.2	0.0
	3/2002	1.1	0.0	0.1	83.3	15.5	0.0
	7/2002	8.4	0.0	0.5	68.1	23.0	0.0
Neophylax sp.	3/2001	0.0	0.0	0.0	27.1	72.9	0.0
	5/2001	0.3	0.0	0.0	80.1	19.6	0.0
	3/2002	2.4	0.0	0.1	93.2	4.3	0.0

Appendix D. Contribution of food types to invertebrate secondary production. Percent (%) contribution in the reference stream (C53) and treatment stream (54) before (Pre) and during (Enr-1, Enr-2) nutrient enrichment. Pre = September 1998 – June 2000, Enr-1 = July 2000 – August 2001, Enr-2 = September 2001 – August 2002.

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Reference stream							
Shredders							
Anchytarsus sp.	Pre	12.0	8.0	9.5	69.2	1.4	0.0
	Enr 1	19.8	2.4	12.1	63.5	2.2	0.0
	Enr 2	22.2	0.0	12.7	64.5	0.6	0.0
Fattigia pele	Pre	49.2	8.7	30.9	11.1	0.0	0.0
	Enr 1	47.0	1.9	27.4	18.3	5.3	0.0
	Enr 2	48.1	6.3	29.5	15.2	0.9	0.0
Lepidostoma spp.	Pre	51.0	5.5	30.9	12.6	0.0	0.0
	Enr 1	49.2	1.1	28.4	19.2	0.3	1.9
	Enr 2	56.5	0.6	32.4	10.3	0.3	0.0
Leuctra sp.	Pre	22.9	1.9	13.7	61.5	0.0	0.0
	Enr 1	27.0	0.0	15.4	57.5	0.0	0.0
	Enr 2	37.9	0.4	21.7	37.3	0.6	2.0
Pycnopsyche spp.	Pre	54.0	10.7	34.3	0.8	0.1	0.0
	Enr 1	53.3	3.3	31.5	10.8	1.0	0.0
	Enr 2	51.6	3.6	30.6	13.6	0.2	0.4
Tallaperla spp.	Pre	48.2	4.1	28.8	18.6	0.4	0.0
	Enr 1	37.6	2.2	22.1	37.2	0.9	0.0
	Enr 2	50.0	0.5	28.6	20.8	0.1	0.0
Tipula spp.	Pre	57.3	4.9	34.3	3.5	0.0	0.0
	Enr 1	46.7	6.5	28.7	18.0	0.2	0.0
	Enr 2	54.8	1.5	31.7	11.3	0.7	0.0

Appendix D. (cont.)

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Treatment stream							
Shredders							
Anchytarsus sp.	Enr 1	27.9	2.3	25.8	42.8	1.2	0.0
	Enr 2	18.8	4.0	21.2	52.7	3.3	0.0
Fattigia pele	Pre	53.9	5.5	32.6	7.9	0.1	0.0
	Enr 1	39.7	2.4	35.2	20.2	2.5	0.0
	Enr 2	32.6	1.4	28.0	37.4	0.5	0.0
Lepidostoma spp.	Pre	53.5	4.0	31.8	9.9	0.8	0.0
	Enr 1	42.5	1.6	36.0	14.6	5.4	0.0
	Enr 2	34.5	4.9	35.0	25.6	0.1	0.0
Leuctra sp.	Pre	25.0	1.3	14.7	54.5	4.6	0.0
	Enr 1	28.7	1.2	24.5	44.6	1.1	0.0
	Enr 2	18.6	1.5	17.1	62.5	0.3	0.0
Pycnopsyche spp.	Pre	49.9	10.6	31.9	7.6	0.0	0.0
	Enr 1	41.2	1.1	34.3	13.2	10.2	0.0
	Enr 2	42.0	2.0	36.3	17.2	2.6	0.0
Tallaperla spp.	Pre	51.7	2.2	30.1	15.5	0.6	0.0
	Enr 1	39.4	1.8	34.0	24.2	0.6	0.0
	Enr 2	41.0	1.2	34.2	20.9	0.3	2.5
Tipula spp.	Pre	53.5	4.1	31.8	10.3	0.3	0.0
	Enr 1	37.0	1.3	31.3	20.6	9.8	0.0
	Enr 2	38.7	2.5	34.7	23.4	0.7	0.0
Reference stream							
Gatherers							
Amphinemura sp.	Pre	23.6	2.7	14.3	57.9	1.6	0.0
	Enr 1	6.2	0.0	3.5	89.3	1.0	0.0
	Enr 2	7.1	0.0	4.1	83.5	5.3	0.0

Appendix D. (cont.)

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Gatherers (cont.)							
Chironomidae	Pre	9.5	0.0	5.4	76.4	8.7	0.0
	Enr 1	8.4	0.0	4.8	86.5	0.3	0.0
	Enr 2	7.7	0.0	4.4	86.9	1.0	0.0
Oligochaetae	Pre	14.3	2.5	9.0	72.4	1.9	0.0
	Enr 1	25.0	0.0	14.3	54.3	6.5	0.0
Paraleptophlebia sp.	Pre	16.7	4.8	11.1	66.6	0.7	0.0
	Enr 1	3.4	0.0	2.0	94.1	0.5	0.0
	Enr 2	0.0	0.0	0.0	81.3	18.7	0.0
Sciara sp.	Pre	44.7	0.0	25.5	6.6	23.2	0.0
	Enr 1	38.7	0.0	22.0	39.3	0.0	0.0
Serratella sp.	Pre	0.1	1.5	0.6	94.4	3.4	0.0
	Enr 1	8.3	0.9	5.0	77.0	8.9	0.0
	Enr 2	5.3	0.0	3.0	81.9	9.8	0.0
Stenonema sp.	Pre	2.3	0.0	1.3	94.6	1.9	0.0
	Enr 1	2.8	0.0	1.6	73.0	22.6	0.0
	Enr 2	6.9	0.0	3.9	86.7	2.4	0.0
Treatment stream							
Gatherers							
Amphinemura sp.	Pre	14.4	1.5	8.7	71.0	4.3	0.0
	Enr 1	27.4	0.0	21.6	48.3	2.7	0.0
	Enr 2	32.0	0.0	25.2	42.3	0.5	0.0
Chironomidae	Pre	6.1	0.0	3.5	90.2	0.3	0.0
	Enr 1	3.2	0.0	2.5	94.2	0.1	0.0
	Enr 2	5.4	0.0	4.2	90.1	0.3	0.0
Oligochaetae	Pre	6.2	5.4	5.3	82.2	0.9	0.0
	Enr 1	13.8	0.0	10.9	75.3	0.0	0.0
	Enr 2	17.9	0.5	15.0	52.1	1.3	13.1

Appendix D. (cont.)

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Gatherers (cont.)							
Paraleptophlebia sp.	Pre	0.0	0.0	0.0	99.0	1.0	0.0
	Enr 1	2.1	0.0	1.7	96.1	0.2	0.0
	Enr 2	5.4	0.0	4.2	88.6	1.8	0.0
Sciara sp.	Enr 2	35.6	0.0	28.1	35.7	0.6	0.0
Serratella sp.	Pre	7.1	1.1	4.4	77.2	10.2	0.0
	Enr 1	6.6	0.1	5.4	76.9	11.0	0.0
	Enr 2	3.1	1.0	4.0	72.2	16.1	3.7
Stenonema sp.	Pre	2.7	0.3	1.7	90.0	5.4	0.0
	Enr 1	1.4	0.1	1.3	90.2	7.0	0.0
	Enr 2	4.7	0.6	4.7	88.5	1.6	0.0
Reference stream							
Filterers							
Diplectrona spp.	Pre	13.7	2.4	8.6	16.7	1.2	57.3
	Enr 1	23.5	3.6	14.6	35.9	1.3	21.1
	Enr 2	28.0	3.1	17.0	46.1	1.3	4.4
Parapsyche cardis	Pre	7.4	1.7	4.8	17.9	0.2	67.9
	Enr 1	14.1	0.8	8.3	24.6	2.1	50.0
	Enr 2	21.6	1.8	12.9	25.3	3.3	35.0
Simuliidae	Pre	4.9	1.6	3.3	85.2	1.7	3.5
	Enr 1	2.3	0.0	1.3	87.3	9.0	0.0
	Enr 2	5.1	0.0	2.9	83.7	8.3	0.0
Wormaldia spp.	Pre	0.0	0.0	0.0	95.1	4.9	0.0
	Enr 1	0.2	0.0	0.1	93.9	5.8	0.0
	Enr 2	0.0	0.0	0.0	100.0	0.0	0.0

Appendix D. (cont.)

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Treatment stream							
Filterers							
Diplectrona spp.	Pre	15.5	4.9	10.5	24.1	0.9	44.1
	Enr 1	20.1	0.7	17.0	39.1	1.2	21.9
	Enr 2	13.0	1.4	12.5	50.7	0.8	21.6
Parapsyche cardis	Pre	7.9	1.1	4.9	13.3	0.6	72.2
	Enr 1	7.8	0.2	6.5	21.3	0.3	63.9
	Enr 2	6.8	0.6	6.3	18.1	0.2	68.1
Simuliidae	Pre	9.1	0.0	5.2	75.7	8.9	1.0
	Enr 1	1.3	0.0	1.1	90.3	7.2	0.0
	Enr 2	0.2	0.0	0.1	97.1	2.6	0.0
Wormaldia spp.	Pre	2.1	0.0	1.2	90.3	6.5	0.0
	Enr 1	0.1	0.0	0.0	98.2	1.7	0.0
	Enr 2	0.6	0.0	0.5	98.5	0.4	0.0
Reference stream							
Scrapers							
Baetis sp.	Pre	0.0	0.0	0.0	91.5	8.5	0.0
Ectopria sp.	Pre	0.0	0.0	0.0	73.8	26.2	0.0
	Enr 1	3.4	0.0	1.9	64.9	29.7	0.0
	Enr 2	0.9	5.0	2.2	52.7	39.2	0.0
Epeorus sp.	Pre	0.7	0.0	0.4	76.7	22.3	0.0
	Enr 1	5.6	0.0	3.2	82.4	8.5	0.3
	Enr 2	1.5	0.0	0.8	82.5	15.1	0.0
Neophylax sp.	Pre	0.0	0.0	0.0	99.4	0.6	0.0
	Enr 1	0.0	0.0	0.0	47.1	52.9	0.0
	Enr 2	0.0	0.0	0.0	7.3	92.7	0.0

Appendix D. (cont.)

Stream/Functional group/Taxon	Treatment period	Leaves	Wood	Fungi	Amorphous	Diatoms	Animals
Treatment stream							
Scrapers							
Baetis sp.	Pre	13.3	0.0	7.6	60.8	18.3	0.0
	Enr 2	0.4	0.0	0.3	92.1	7.1	0.0
Ectopria sp.	Enr 1	2.8	0.0	2.2	74.9	20.2	0.0
	Enr 2	7.7	0.0	6.1	56.1	30.2	0.0
Epeorus sp.	Pre	0.0	0.4	0.1	86.1	13.4	0.0
	Enr 1	0.3	0.0	0.2	85.8	13.7	0.0
	Enr 2	0.9	0.0	0.7	76.2	22.1	0.0
Neophylax sp.	Enr 1	0.0	0.0	0.0	42.3	57.6	0.0
	Enr 2	0.6	0.0	0.5	92.2	6.7	0.0

* * *