Retesting a prediction of the River Continuum Concept: autochthonous versus allochthonous resources in the diets of invertebrates

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Abstract: The River Continuum Concept (RCC) predicts that food webs (and, in particular, invertebrates) of rivers in temperate, forested drainages should exhibit a longitudinal gradient from reliance on terrestrially derived organic matter (e.g., seasonally shed leaves) in the headwaters to autochthonous sources (e.g., algae) in the mid-orders, to suspended material in larger rivers. This prediction has been evaluated by longitudinal comparisons of macroinvertebrate communities in terms of functional feeding groups (FFGs), but such an approach yields only indirect evidence regarding actual food use. To retest this prediction, we investigated invertebrate diets from the Salmon River, Idaho, by examining the gut contents of archived specimens from the longitudinal set of sites sampled in the original 1976 RCC study and by collecting invertebrates from these same sites in 2009. We detected no apparent shifts in diets over the ~30-y time span. The importance of allochthonous materials in invertebrate diets differed significantly among sites along the longitudinal gradient. As predicted, it was greatest at the 2nd-order headwater site (30–42% of gut contents, on average) and decreased longitudinally to 10 to 20% at the most downstream site. However, invertebrates at the 2nd-order site also consumed large percentages of autochthonous materials (35–45%), and diets contained from 35 to 75% autochthonous resources across all sites. The shredder (*Yoraperla*) with the highest density at the most upstream 2nd-order site had gut contents composed of 52 to 81% diatoms depending on season, illustrating the importance of autochthonous resources in the headwaters. Our findings show that measuring shifts in FFG alone without examining actual food resources present in macroinvertebrate diets may lead to erroneous inferences regarding patterns of resource use by macroinvertebrates in food webs.

Key words: gut contents, functional feeding groups, diatoms, leaves

The River Continuum Concept (RCC; Vannote et al. 1980) provides a framework, rooted in application of energy equilibrium theory to fluvial systems (Leopold and Maddock 1953), that describes changes in ecological communities and ecosystem processes from headwaters to large rivers. Under this concept, these changes are tuned to a continuum of gradients in physical habitat conditions, organic-matter sources, and capitalization on products of upstream inefficiencies in organic-matter processing by downstream communities. A prediction derived from the RCC and depicted in its iconic figure is that, as streams increase in size, the availability of terrestrially vs riverine derived organic matter shifts (Vannote et al. 1980). The expected patterns are that the importance of terrestrially derived organic matter decreases in a downstream direction, whereas the organic matter in larger rivers is derived from the inefficiencies of upstream reaches and an increase in the relative importance of in situ primary production (Vannote et al. 1980, Minshall et al. 1983). As a result, the form of organic matter consumed by riverine animals is predicted to shift from primarily terrestrially derived coarse particulate matter in the headwaters to algae in the mid-orders to suspended fine particulate matter in larger-order rivers. In addition, the RCC posits that food webs in any given segment of a river network should be structured so as to make the most efficient use of the C resources available. Therefore, the RCC predicts that invertebrates that dwell in the headwaters should consume more coarse particulate C (e.g., leaves), whereas those that dwell in larger, downstream segments should consume primarily algae or fine particles.

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Research exploring the tenets of the RCC has been extensive, but investigators who have explored predictions regarding food resource use by macroinvertebrate communities typically have relied on analysis of shifts in the relative biomass or abundances of various functional feeding groups (FFGs). However, FFG classifications (e.g., shredders, scraper–grazers, collector–gatherers) are based on the organisms' mode of feeding (Cummins 1973), not on food resources consumed. The FFG composition at a site may shed some light on the size or form of food resources consumed, but FFGs are not a measure of the actual identity of food resources consumed (Mihuc and Minshall 1995, Rosi-Marshall and Wallace 2002), particularly for the many omnivorous invertebrates that compose most of the benthic animal biomass of riverine communities. In addition, many aquatic macroinvertebrate taxa undergo ontogenetic diet shifts (Merritt et al. 2008). Therefore, testing the RCC predictions regarding longitudinal shifts in resources supporting macroinvertebrate communities requires analysis of actual food consumed, information that can be obtained through gut-content analysis (Rosi-Marshall et al. 2016). Tests of these predictions by diet analysis are limited (but see Rosi-Marshall and Wallace 2002, Whiting et al. 2011).

In 1976, stream invertebrate samples were collected, specimens were classified to FFG, and availability of various food resources was measured at sites along the Salmon River, Idaho (USA). The amount of allochthonous C was highest in the headwaters, but the amount of periphyton chlorophyll *a* also was generally higher in the headwaters than at other sites in the Salmon River and in other river systems examined (Minshall et al. 1982, 1983). Metabolism measures indicated that these headwater stream ecosystems were consistently heterotrophic. Comparisons of FFG composition of communities across sites of sequentially larger stream order in the Salmon basin led investigators to interpret the patterns in FFG distribution (e.g., more shredders in headwaters and more scrapers and collector–filterers at downstream sites) as generally consistent with the shift in food resource use predicted by the RCC (Minshall et al. 1983).

Specimens of a range of taxa collected in summer and autumn of 1976 were preserved, archived, and contained intact gut materials >30 y later. We examined the food resources actually consumed by these archived specimens and additional individuals newly collected in 2009 along these sites on the Salmon River. We first evaluated whether longitudinal changes in actual diets of all collected invertebrates agreed with patterns of resource use predicted based on FFG designations in the original RCC study. For example, we assessed the RCC prediction that a greater relative dominance of shredders in headwater communities indicates that the overall invertebrate community showed greater reliance on allochthonous resources at headwaters compared to downstream sites. We also classified individuals based on FFG and evaluated whether FFGs differed longitudinally in their reliance on allochthonous resources.

Given the lapse of time since the 1976 study, we had a limited opportunity to investigate the possibility of long-term shifts in resources consumed by invertebrates at these sites. Land use has not changed significantly in the Salmon basin since 1976 because it drains a large expanse of relatively intact wilderness. However, direct and indirect effects of changing climate have been documented, including increases in the severity and extent of wildfires, an outbreak of mountain pine beetle, and shifts in precipitation regime, any of which may have consequences for stream invertebrates and available organic-matter resources (reviewed by Davis et al. 2013). For instance, both beetle kill and wildfire may have opened the riparian forest canopy in some headwater locations, which might be expected to increase the importance of aquatic primary producers as food resources for stream-dwelling invertebrates. Therefore, we examined the diet composition of invertebrates collected in 1976 from the original RCC study sites along the Salmon River and compared them to diet composition of individuals collected in 2009. Thus, the final objective of our study was to test whether the proportion of individuals or FFGs more reliant on allochthonous resources in the 1970s may have declined by 2009.

METHODS

The original study sites were in the Upper Salmon River basin in the Sawtooth Mountain Range and Sawtooth National Recreation Area near Stanley, Idaho, USA. These 4 sites were selected and sampled in 1976, and the resulting data eventually were used in the initial testing of the RCC (Vannote et al. 1980, Minshall et al. 1982, 1983, 1992). We resampled the sites in 2009. The sites span ~65 km of river and are situated from the headwaters of the mainstem Salmon River to just upstream of its confluence with the Yankee Fork (Minshall et al. 1982). Detailed descriptions and maps of the study sites were published by Minshall et al. (1982, 1983). Camp Creek (site A; catchment area = 0.6 km^2, elevation ≈ 2500 m elevation), a 2nd-order stream, served as the forested, headwater site. Smiley, (site B; catchment area = 40.6 km^2, elevation = 2200 m), Obsidian (site C; catchment area = 478.1 km^2, elevation = 2000 m), and Casino (site D; catchment area = 846.3 km^2, elevation = 1900 m) were 4th-, 5th-, and 6th-order sites, respectively. In this area, snow is the main source of precipitation from November to March and peak flows occur from snowmelt from May to July (Minshall et al. 1983). Canopy cover was greatest at site A and decreased progressively downstream. The riparian vegetation of the Upper Salmon River basin is largely dominated by *Pinus contorta* (lodgepole pine), *Pseudotsuga menziesii* (Douglas fir), *Salix* sp. (willow), and *Populus tremuloides* (quaking aspen) in the headwaters, with sagebrush steppe plant community

dominating the riparian zones of the lower sites. The sites retained similar traits in 2009, with the principal exceptions that all sites (most notably B) had experienced some death of conifers from beetle kill, and all sites from C downstream were potentially influenced by a 2005 wildfire. The Valley Road Fire did not burn the watersheds of the study reaches themselves, but influenced substantial areas draining to these sites.

During the original RCC sampling period in 1976, invertebrates were collected from the Salmon River in summer (July) and late winter (March) and categorized by FFG. A few specimens were collected in September to November 1976. Aquatic invertebrates were sampled in 1976 as described by Minshall et al. (1982, 1983). Sampling was conducted on a single date at each site in each season with a modified kick-net (1-mm-mesh net) from pool and riffle habitats. Some of the specimens collected in summer and autumn were preserved in formalin and archived at Idaho State University in the Stream Ecology Center and were viable for gut-content analysis. Thus, whereas these invertebrates were collected from the Salmon River in 1976, their gut contents were not examined until 33 y later for our study in 2009. Only a limited number of individuals were archived, mainly from summer and autumn, so the number of taxa and individuals used for gut-content analysis was restricted to the specimens available from the archived collection. The number of archived taxa was highest for site A and was more limited at downstream sites. We analyzed diets from a total of 145 archived individuals, which included 80, 36, 23, and 6 individuals from sites A, B, C, and D, respectively. Not all taxa from sites A to D were consistently preserved, but individuals used in the gut-content analysis represented most of the macroinvertebrate assemblage (12–65% of the total community assemblage, based on 1976 abundance values). We used the samples collected in summer (25–29 July 1976) and autumn (23 September–20 November 1976) to assess longitudinal patterns in insect diets from the original RCC sampling sites. Those 2 periods were pre- and post-leaffall, so we were able to assess whether longitudinal patterns differed among seasons.

For comparison, we collected invertebrate samples from each site in summer (16 July–1 August) and after leaffall (29 September–2 October) in 2009 with a coarse-mesh kick net (800–900 μm). We preserved invertebrates in Kahle's solution (Pennak 1978) in the field and held them in Kahle's solution until gut-content analysis. We chose taxa for diet analysis to provide a comparison to the 1976 invertebrates and a longitudinal representation of present-day diets. We identified all individuals to genus and classified them to FFG based on Merritt et al. (2008). In 2009, we analyzed 245 individuals including 69, 54, 59, and 63 individuals from sites A, B, C, and D, respectively.

Overall, we analyzed 6 shredder taxa, 11 collector–gatherer taxa, 2 collector–filterer taxa, and 3 scraper–grazer

taxa. In total, we analyzed gut contents from 22 taxa and 390 individuals preserved or collected from sites along the longitudinal gradient. We excluded predator taxa collected in 1976 and 2009 from this analysis because our main question concerned longitudinal patterns in allochthonous vs autochthonous organic matter use, and predators were presumed to have eaten predominantly animal material (Minshall et al. 1982, Rosi-Marshall and Wallace 2002).

We conducted gut-content analysis on individuals collected in 1976 and 2009 (Cummins 1973, Benke and Wallace 1980, Rosi-Marshall et al. 2016). We analyzed the foreguts of 1 to 15 individuals/taxon depending on availability of samples. At times, the amount of material in individual guts was limited and we had to pool 1 to 5 individuals on a slide. Therefore, the total number of individuals dissected for this study was actually greater than the number of replicates included in the statistical analysis. We used a compound microscope at 100× to identify food particles and assigned them to the categories: allochthonous, autochthonous (diatoms and filamentous algae), amorphous detritus, fungal, or animal material (Benke and Wallace 1980, Rosi-Marshall and Wallace 2002). We photographed gut contents with a camera attached to the microscope and measured the areas of food particles. For each slide, we measured the area of ≥50 identified particles along random transects with ImageJ (National Institutes of Health, Bethesda, Maryland; http://imagej.nih.gov/ij/), and we calculated the percentages of food items in the gut(s) of those insect(s).

To assess whether the relative contribution of all food resources in the diets of all sampled individuals varied longitudinally and agreed with predictions based on the RCC (i.e., research objective 1), we compared patterns in resource consumption for all sampled individuals as a whole, followed by a more focused analysis targeting longitudinal changes in allochthonous resource consumption. We first combined diet data for all individuals collected at a given site, and then qualitatively assessed whether the percentages of the various resource types in gut contents differed among sites. We applied a 2-way analysis of variance (ANOVA) (SYSTAT, version 12; Systat Software, San Jose, California) to assess how consumption of allochthonous resources differed among sites and season. This analysis was focused only on those individuals collected in 2009 because sample sizes of specimens from 1976 were too small for their statistical analysis. We were primarily concerned with differences between allochthonous and autochthonous consumption, but we focused the statistical analyses on allochthonous consumption because the percentages of the 2 types of resource are directly related to one another. We included site, season, and their interaction in the 2-way ANOVA. Season was included in this analysis because relative contributions of allochthonous resources to invertebrate diets can differ between pre- and post-leaffall (Hall et al. 2000, Rosi-Marshall and Wallace 2002). Thus, this

2-way ANOVA assessed whether the entire assemblage of sampled invertebrates exhibited longitudinal differences in their reliance on allochthonous resources that agreed with RCC predictions and whether those differences varied between seasons (pre- and post-leaffall).

To assess whether longitudinal patterns in diet differed among FFGs (research objective 2) and agreed with tenets of the RCC, we grouped invertebrates by FFG (shredders, collector–gatherers, collector–filterers, and scraper–grazers) and qualitatively compared the percentages of all resource types in invertebrate gut contents at each sampling site and year. We then applied a 2-way ANOVA specifically to compare the % contribution of allochthonous resources to the diets of each FFG in 2009. We used separate 2-way ANOVAs, which included the main effects of site and season and their interaction, for each FFG in 2009 because we lacked seasonal data from site B for shredders and collector–filterers (see Results), which would have necessitated the exclusion of those FFGs from a combined ANOVA. We excluded site B from the ANOVA comparing collector–filterers and shredders.

We were not able to compare dietary shifts statistically between 1976 and 2009 because of limited sample sizes of archived specimens. Thus, we were able to draw only limited inferences regarding how invertebrate diets may have changed over time at these sites (i.e., research objective 3). Assessments of statistical significance were based on $\alpha = 0.05$ for all comparisons and appropriate transformations (i.e., $\arcsin\sqrt{[x]}$) were applied to meet statistical assumptions when necessary.

RESULTS
All invertebrates

The % contribution of the various resource types varied among the 4 sites in the longitudinal analysis of gut contents of all invertebrates. In 2009, the % allochthonous material in all invertebrate guts differed among sites ($F_{3,174} = 5.29$, $p = 0.002$), and was greatest at site A (30–42%) and decreased longitudinally (to ≤20%) (Fig. 1A). Diets did not differ between pre- and post-leaffall samples. Season ($F_{1,174} = 0.49$, $p = 0.487$) and the site × season interaction ($F_{3,174} = 1.029$, $p = 0.381$) did not affect % allochthonous material in guts. Invertebrates at site B consumed more allochthonous material than did invertebrates at sites C and D, but they also consumed large percentages of autochthonous materials (Fig. 1B). Autochthonous material was an important resource and made up 40 to 75% of the material in guts from invertebrates at all sites in both years. Percent amorphous detritus was similar in guts of invertebrates collected at sites B to D. Gut contents of invertebrates collected from site A in autumn 1976 had the lowest % amorphous detritus (10%) (Fig. 1C). Visual comparison of trends in diet composition between 1976 and 2009 did not reveal apparent differences in relative consumption of allochthonous vs autochthonous resources (Figs 1A, B, 2A–D).

Shredders

Shredders consumed the most allochthonous material (20–100% of diets) relative to other FFGs, but they relied substantially on autochthonous material (10–70%). These trends were similar for individuals collected in 1976 and 2009 (Fig. 2A). In 2009, reliance on allochthonous material did not differ between seasons ($F_{1,19} = 3.81$, $p = 0.062$) or among sites ($F_{2,19} = 0.499$, $p = 0.687$), and the season × site effect was not significant ($F_{2,19} = 0.111$, $p = 0.895$). Furthermore, ≥½ of the guts examined contained both autochthonous material and amorphous detritus, and this pattern was consistent across sampling sites.

The relative reliance on allochthonous and autochthonous material varied among taxa. Allochthonous material made up >⅔ of gut contents of *Zapada*, *Eucapnopsis*, and *Capnia* in 1976 and 2009 at site A. However, *Yoraperla* consumed primarily algal material (Appendix S1). *Yoraperla* is a dominant shredder at these sites and made up 56 to 87% of the shredder abundance in summer and autumn 1976, respectively (Minshall et al. 1982; note: *Peltoperla* was divided and is now called *Yoraperla* [Stark and Nelson 1994]). In 1976, autochthonous material accounted for 69% and allochthonous material accounted for 20% of *Yoraperla* gut contents in summer. In 2009, autochthonous material accounted for 67% and allochthonous material accounted for 21% of *Yoraperla* gut contents at site A (mean of summer and autumn). *Pteronarcys*, the only shredder taxon found at sites C and D in 2009, consumed 48 and 68% allochthonous material at each site, respectively. Animal material made up a portion of *Pteronarcys* diets, particularly at site C (29%).

Collector–gatherers

Collector–gatherers consumed primarily amorphous detritus and autochthonous material (60–98%) (Fig. 2B), and only limited percentages of allochthonous material. This overall trend did not appear to differ between 1976 and 2009 (Fig. 2B). Seasonal differences were not detected in the gut contents of collector–gatherers sampled in 2009 ($F_{1,69} = 2.92$, $p = 0.092$), nor was the season × site effect significant ($F_{3,69} = 0.927$, $p = 0.432$). However, % allochthonous material differed among sites. Percent allochthonous materials was higher at site A than at downstream sites ($F_{3,69} = 4.72$, $p = 0.005$), but % amorphous detritus and % autochthonous material in gut contents was similar among sites.

Despite overall similarities in the combined gut contents, diets did vary at the genus level. *Ephemerella* guts contained the greatest % allochthonous materials at site A (40% in autumn 2009, 69% in summer 2009; Appendix S1). At the downstream sites (B–D), the closely related
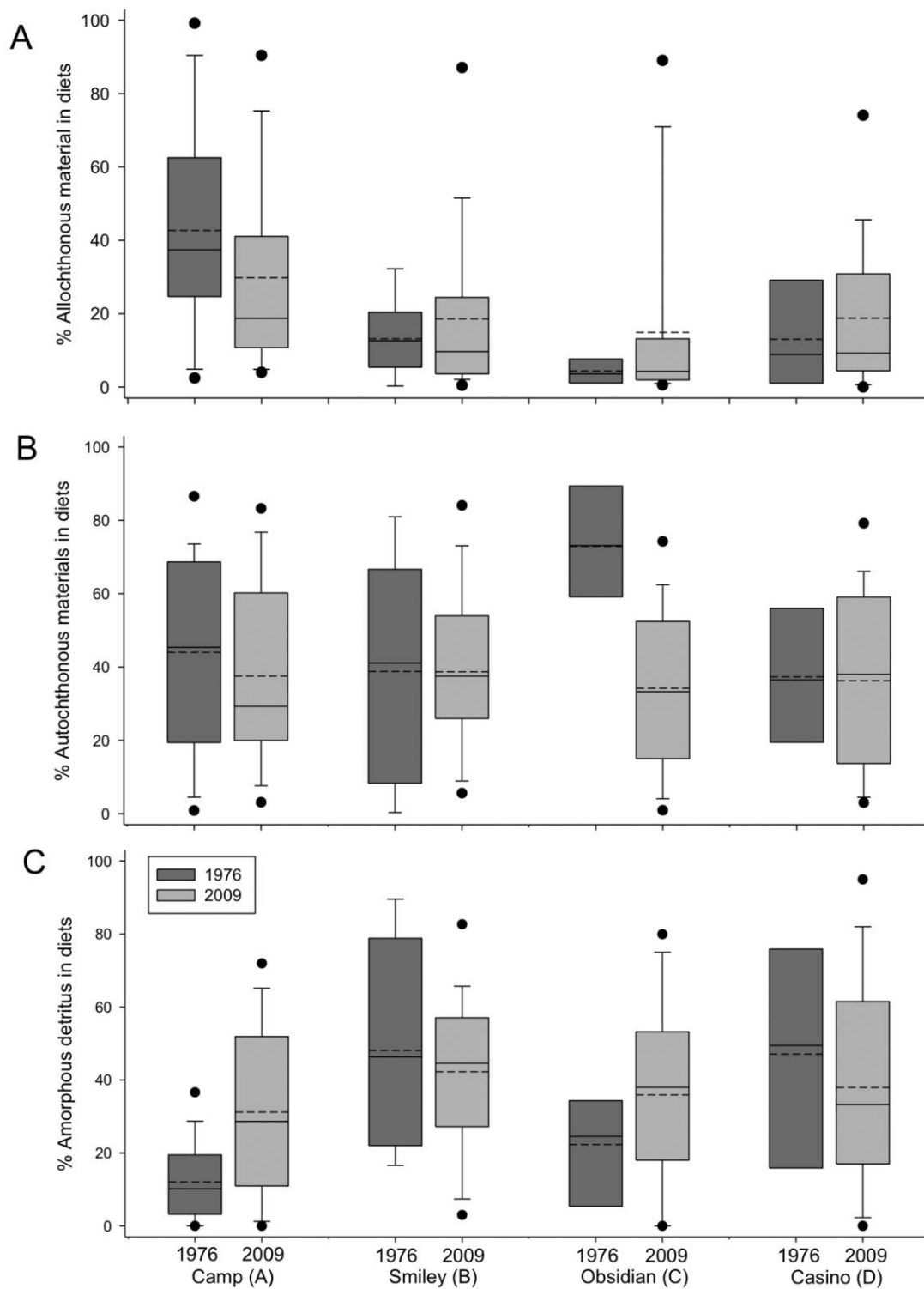
Figure 1. Box-and-whisker plots for % allochthonous materials (A), % autochthonous materials (B), and % amorphous detritus (C) in gut contents of individual invertebrates at sites along the longitudinal gradient of the Salmon River, Idaho, in 1976 and 2009. Sites are arranged from up- to downstream (Camp = A, 2nd order; Smiley = B, 4th order; Obsidian = C, 5th order; Casino = D, 6th order). Solid lines in boxes are medians, dotted lines in boxes are means, box ends are quartiles, dots indicate 5th and 95th percentiles, and whiskers show the range. The absence of whiskers or dots indicates insufficient data.
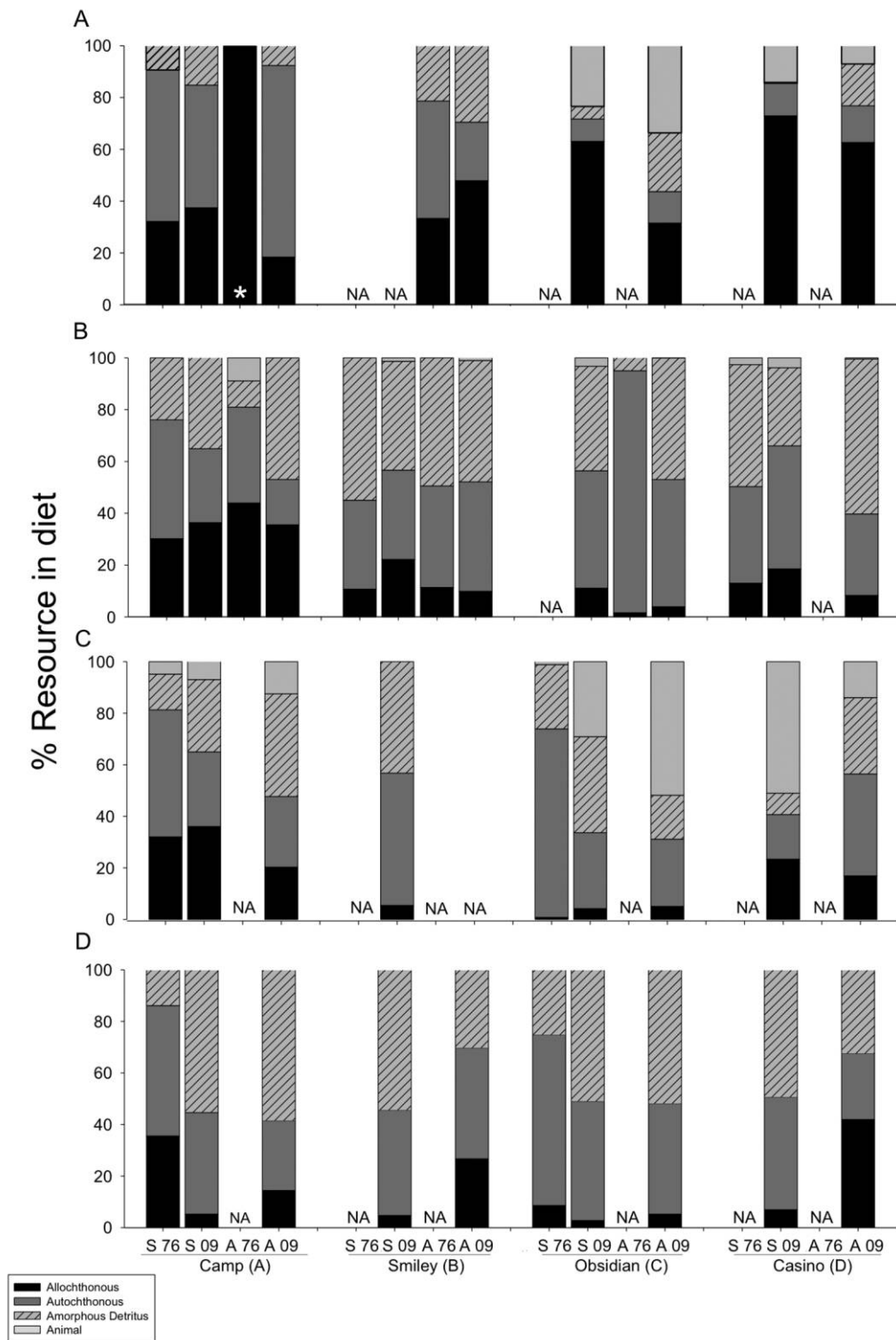
Figure 2. Percentage of food resources found in invertebrate gut contents at sites along a longitudinal gradient of the Salmon River in summer (S) 1976 (76) and summer and autumn (A) 2009 (09) for gut contents pooled by functional feeding group. A.—Shredders. B.—Collector–gatherers. C.—Collector–filterers. D.—Scraper–grazers. NA = no samples available for diet analysis, * = the diet of 1 individual from the genus *Capnia*.

Drunella and *Serratella* had primarily autochthonous material, 60 and 36%, respectively, and amorphous detritus, 30 and 54%, respectively, in their guts.

Collector–filterers

Collector–filterers had highly variable gut contents but, in general, had higher % animal material in their guts compared to invertebrates in other FFGs (Fig. 2C). We were unable to compare gut contents between years except at site A because few archived samples from 1976 were available. Composition of gut contents at site A in summer did not appear to differ between 1976 and 2009 (Appendix S1). Percent allochthonous material in gut contents did not differ between seasons in 2009 ($F_{1,20} = 0.792$, $p = 0.382$), and the site × season effect was not significant ($F_{2,20} = 0.324$, $p = 0.727$). Percent allochthonous material in gut contents differed among sites in 2009 ($F_{2,20} = 5.70$, $p = 0.004$). Percent allochthonous material was 20 to 35% in gut contents at site A and <20% in gut contents at downstream sites. At site A in 1976 and 2009, diets of *Simulium*, the only collector–filterer analyzed at this site, included 32 and 28% allochthonous materials, respectively (Appendix S1). In summer 2009, allochthonous material in simuliid gut contents did not differ among sites ($F_{3,24} = 2.30$, $p = 0.194$). *Simulium* gut contents contained some animal material, and *Brachycentrus* gut contents occasionally included high % animal material (up to 79%). Gut contents at site C differed between 1976 and 2009 (Fig. 2C). Autochthonous material accounted for 73% of gut contents in 1976, and animal material made up a substantial percentage of the gut contents in 2009. This difference may be a result of the high % animal material in *Brachycentrus* gut contents and the fact that *Simulium* was the only collector–filterer taxon available from site C for analysis of 1976 gut contents.

Scraper–grazers

Scraper–grazer gut contents were composed predominantly of amorphous detritus and autochthonous materials in similar percentages, with limited contributions of allochthonous material (<42% and often <10%) (Fig. 2D). Comparison of gut contents between 1976 and 2009 was possible only at sites A and C. At site A in summer 1976 and 2009, scraper–grazer diets were composed primarily of autochthonous materials and amorphous detritus (Fig. 2D). At site C, % allochthonous material in the gut contents was similar in summer 1976 and 2009 (5 and 8%, respectively), but % autochthonous material and % amorphous detritus differed between the 2 summers (Fig. 2D). Consumption of allochthonous materials by scraper–grazers was highest at site A in summer 1976 (36%). The greatest % allochthonous materials was found in the gut contents of scraper–grazers at site D in autumn 2009 (42%). Con-

sumption of allochthonous materials did not differ among sites ($F_{3,41} = 2.30$, $p = 0.091$), and the site × season effect was not significant ($F_{3,41} = 0.241$, $p = 0.241$). However, % allochthonous material in gut contents did differ between seasons ($F_{1,41} = 4.96$, $p = 0.032$), with higher % allochthonous materials in gut contents in autumn.

DISCUSSION

Our results provide evidence that longitudinal patterns in the diets of macroinvertebrates of the Salmon River are generally consistent with the predictions of the RCC regarding longitudinal shifts in the relative importance of allochthonous vs autochthonous resources, but less so than would be expected based upon inferences drawn from comparison of FFG proportions alone. Invertebrates classified as shredders and collector–gatherers did consume proportionately more allochthonous resources at the headwater site than at downstream locations. In contrast, the reliance of a number of taxa, including shredders, on algal resources was high, possibly because this food resource is both abundant and of high quality even in the forested headwaters of the Salmon River. In addition, some shredders common at larger, downstream sites (e.g., *Pteronarcys*) ate primarily allochthonous material, although the RCC predicts that allochthonous resources should decline in importance in these larger river habitats. However, *Pteronarcys* are known to consume autochthonous resources (Plague et al. 1998, Rosi-Marshall and Wallace 2002) and their feeding probably is context specific. Previous work suggests that some shredders may be obligate consumers of allochthonous material, whereas others are facultative consumers of allochthonous or autochthonous resources (Rosi-Marshall and Wallace 2002). We also observed that consumption of allochthonous material by shredders was similar in the summer and autumn in 2009 (when sufficient numbers of individuals were available for statistical comparison). Minshall et al. (1982) suggested that the decline in shredder abundance along the Salmon River was caused by a decline in the availability of allochthonous resources at downstream sites. However, our observations demonstrated that shredders in the headwaters eat primarily algae and some shredders downstream eat principally allochthonous material. These findings do not support the notion that changes in invertebrate functional feeding group composition track the availability of food resources along a river continuum. Rather, our observations are consistent with insects exhibiting preference for high-quality resources and suggest that traits of particular taxa other than feeding mode may drive longitudinal patterns in resource use in rivers. These traits could include a wide range of characteristics, such as life-stage-specific nutritional requirements, stoichiometric imbalances between organisms and their resources, and interactions between habitat conditions and feeding behavior (i.e., changes in feeding arena).

Overall, our data demonstrate that FFGs are not reliable indicators of resources consumed, which has been shown elsewhere (e.g., Mihuc and Minshall 1995), and that different assessments of the RCC predictions arise when invertebrate diets are assessed directly. For example, whereas the original RCC studies identified a marked decline with increasing stream order in both the absolute and relative abundance of invertebrates belonging to the shredder FFG (Minshall et al. 1982, 1983, Bruns and Minshall 1985), we found a much less pronounced trend in the percentage of allochthonous material actually consumed by these organisms. Instead, the most striking pattern was the consistently high contribution of autochthonous resources to invertebrate diets across all sites and periods, regardless of FFG assignment. Thus, our results add to evidence from a growing array of studies (Minshall 1978, Mayer and Likens 1987, Rosenfeld and Roff 1991, Thorp and Delong 1994, 2002, McCutchan and Lewis 2002, McNeely et al. 2007) that the importance of autochthonous resources to stream animals may be underestimated, particularly in headwaters. Headwaters are presumed to have the largest amounts of detrital material available to the macroinvertebrate community (Vannote et al. 1980), and detrital material certainly supports macroinvertebrates in food webs of other rivers (e.g., Wallace et al. 1997, Rosi-Marshall and Wallace 2002, Cross et al. 2013). We think it probable that forested streams like those in eastern North America experience more extreme shading compared to the relatively open-canopied, headwater streams of the Salmon River, and algal production may be limited and not readily available as a food resource in headwaters like those in eastern North America. Moreover, the seasonally shed leaves of predominantly deciduous eastern forests probably provide allochthonous inputs in larger quantities compared to those of headwater streams in the mountainous west of North America. Regardless, whereas FFGs may be a useful index for some lines of inquiry regarding stream ecosystems, a more direct evaluation of what is actually consumed by aquatic invertebrates is required to measure the relative importance of different resources to supporting stream food webs. FFGs are not reliable predictors of food resources consumed for many aquatic invertebrates (Benke and Wallace 1997, Mihuc and Minshall 1995, Plague et al. 1998, Rosi-Marshall and Wallace 2002). In addition, although some invertebrates exhibit inflexible dietary preferences (Eggert and Wallace 2003), others can be opportunistic, exhibiting functional plasticity and capacity to switch resources in the face of changing availability, environmental perturbation, or disturbance (Mihuc and Minshall 1995, Dangles 2002).

In contrast to the diets of aquatic invertebrates in well-characterized food webs of streams in eastern deciduous forests (e.g., Hall et al. 2000, Rosi-Marshall and Wallace 2002), the diets of aquatic invertebrates collected in the headwaters of the Salmon River contained much autochthonous material. Proportional consumption of this resource often exceeded the proportion of allochthonous material consumed. This observation is consistent with the results of a coarse C budget of the Salmon River sites conducted at the time of the initial RCC studies, which suggested that the organic-matter resources in the headwaters were dominated by autochthonous production (Minshall 1978, Minshall et al. 1983). Previously, this pattern was viewed as an outlier, particularly because of the prevalence of shredding taxa in these headwaters. Our observation that these shredders tend to rely as much or more on algae as on leaves helps to resolve this apparent paradox. In addition, algae are typically a higher-quality food resource than detrital material (McNeely et al. 2007), and this quality probably would increase its contribution to the secondary production of macroinvertebrate communities. The differences in quality between leaves and algae and stoichiometric constraints on aquatic invertebrates (Cross et al. 2005) certainly suggest that stream invertebrates may be more likely to be nutrient- than C-limited. The RCC predicted that invertebrate food webs would exhibit efficient use of available food resources, but its authors did not consider how this use might be mediated by the relative differences in quality among food resources (Marcarelli et al. 2011). Such mounting evidence combined with studies on the importance of P (Frost and Elser 2002) and algae in determining invertebrate growth rates may require an update of the RCC that considers both availability of resources and their relative quality. Therefore, regardless of FFG classification, aquatic invertebrates may preferentially consume a high-quality food resource (e.g., algae) when available even if abundant allochthonous material is available. The shredder *Yoraperla* consumes detrital material (Dudley and Anderson 1982, Hughes et al. 1999), but our results suggest that they also are facultative algivores. The large amounts of diatoms present in *Yoraperla* gut contents may have been associated with leaves they were consuming, but the fact that the majority of their gut contents consisted of diatoms suggests that they were selectively foraging on autochthonous resources in these sites. More research that explores the range of invertebrate diets, especially in relation to food availability and quality, would help to shed light on the degree of opportunistic feeding on high-quality food resources that occurs in stream food webs.

The comparison of gut contents of invertebrates collected in 1976 and 2009 did not reveal significant differences in food resources consumed during these 2 snapshots separated by >30 y. However, the observation that food resources consumed by specific taxa do not appear to have shifted does not imply that no changes have occurred over this time span. Other significant ecological changes have occurred in the basin that may have consequences for stream-dwelling invertebrates or stream ecosystems in general (reviewed by Davis et al. 2013). Climate-

mediated fire-regime changes and forest-beetle outbreaks coupled with changes in precipitation regimes may alter riparian zones, with consequences for inputs of organic matter, light, and nutrients. These climate-mediated processes are predicted to increase in-stream primary productivity and reduce allochthonous inputs (Davis et al. 2013). Given that the region is expected to experience more dramatic change in the future, including dynamics that may influence invertebrate diets, the data we presented provide a benchmark for future comparisons.

Our study provides new insights into the food resources that support macroinvertebrate communities along the Salmon River, but these data test only a specific prediction derived from the RCC and not the RCC itself. The central tenet of the RCC is that communities in river ecosystems mirror their habitat and are adapted to use the energy resources available efficiently. This central tenet leads to the prediction of a continuum of macroinvertebrate community structure. To test this central tenet, information on food resources consumed, ideally gathered on the basis of an energetics approach that uses empirically derived rates of consumption (e.g., Benke and Wallace 1980, 1997, Rosi-Marshall and Wallace 2002) and data on production are needed. This approach would allow explicit estimates of trophic transfer efficiencies (Cross et al. 2013), which would enable one to test whether the food web shifts in structure to maintain consistent, efficient energy transfer as a stream increases in size. A substantial gap exists between this approach, which generally has not been used, and examinations of macroinvertebrate community composition or diet analyses like those presented here. Consequently, one might argue that the central tenet of the RCC remains untested.

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