
SHORT COMMUNICATIONS

Allochthonous and Autochthonous Food Sources for Zoobenthos in a Forest Stream

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The river continuum concept (RCC), one of the best known elements in the theory of aquatic ecology [1], declares the prevalence of allochthonous over autochthonous organic matter in the upper reaches of forest rivers and streams. This concept is based on the facts that there is a large amount of leaf litter on the bottom and that this litter is actively consumed by benthic animals (shredders). However, due to the development of biochemical and physicochemical methods of research on trophic webs, evidence has been obtained that (1) not only the amount but also the quality of organic matter—namely, its biochemical and elemental composition (stoichiometry)—is important for the functioning of trophic links [2, 3], and (2) by no means all objects ingested by consumers are then assimilated by them and become real food components [4, 5]. The use of biochemical markers and stable isotope analysis in studies on trophic chains in the upper reaches of some rivers and streams as shown that the biochemical and elemental composition of leaf litter make it a low-value food for aquatic animals, with its contribution to trophic webs being negligible, compared to that of autochthonous production of periphytic microalgae [6–10].

On the one hand, the data obtained by modern methods of biochemical markers and stable isotope analysis are difficult to interpret, which is especially true of compound specific isotope analysis (CSIA) [11]; on the other hand, it is obvious that benthic invertebrates are active consumers of leaf litter. In view of these facts, it is necessary to continue research aimed at defining more exactly the actual contribution of abundant allochthonous organic matter flux to the functioning of ecosystems in the upper reaches of rivers. A promising approach to this task is to comparatively ana-

lyze the relevant data obtained in different ecoregions (biomes). The purpose of this study was to assess the roles of leaf litter and periphyton in the feeding of benthic invertebrates in a small forest stream flowing in the ecoregion “Ussuri temperate broadleaf and mixed forests” of the “Temperate and Broadleaf Forests” biome (<https://ecoregions2017.appspot.com/>).

This stream, named Turova Pad’, is a right tributary of the Komarovka River in the Razdol’naya River basin (43°38’ N, 132°19’ E; Ussuriysky district, Primorsky krai). The study area is in the territory of Ussuriysky State Nature Reserve affiliated to the Federal Scientific Center of the East Asia Terrestrial Biodiversity (Far Eastern Branch, Russian Academy of Sciences). Samples from the stream were collected on October 3 and 14, 2019 in a stretch of rapids about 6 m long, 2 m wide, and 0.15 – 0.20 m deep with a current speed of up to 0.3 m/s and the bottom covered by stones and boulders of different sizes. The periphyton formed a thin coat on the boulders, and the leaf litter was piled up into bundles between them. These bundles were populated by amphipods *Gammarus coreanus* and a small number of pediciid larvae (*Pedicia* sp.). Objects sampled to analyze for stable isotope and fatty acid (FA) composition were as follows (figures in parentheses are the respective numbers of samples): leaves of linden *Tilia amurensis* (5 and 5), poplar *Populus maximowiczii* (8 and 8), and maple *Acer mono* (5 and 5); periphyton (5 and 5); gammarids (10 and 9); and pediciid larvae (7 and 6). Methods for collecting and processing samples of periphyton and benthic invertebrates and the procedure of analysis of stable isotope and FA composition are described in detail elsewhere [12, 13].

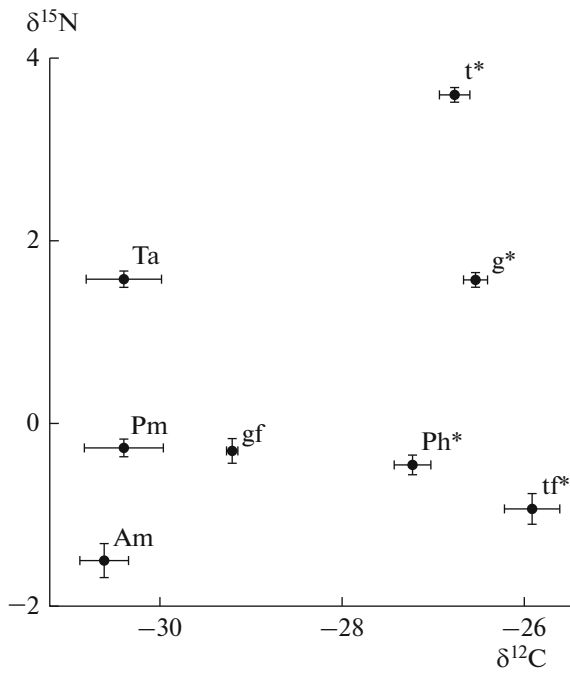


Fig. 1. Mean stable isotope ratios of nitrogen ($\delta^{15}\text{N}$, ‰) and carbon ($\delta^{13}\text{C}$, ‰) in components of the Turova Pad' stream ecosystem (October 3 and 14, 2019): Ta, linden (*T. amurensis*) leaves; Am, maple (*A. mono*) leaves; Pm, poplar (*P. maximowiczii*) leaves; Ph, periphyton; g, gammarids; t, *Pedicia* sp., gf, gammarid feces; tf, *Pedicia* feces. Asterisks indicate components in which $\delta^{13}\text{C}$ is significantly different from that in components without an asterisk ($p < 0.05$, one-way ANOVA with post hoc Tukey's test). Whiskers are standard errors of the mean.

The results of isotope analysis are shown in Fig. 1. On average, the carbon isotope ratios in gammarids and pediciid larvae are significantly different from those in litter leaves but almost coincide with these ratios in the periphyton. Obviously, benthic animals receive carbon from the periphyton rather than from the litter. The average nitrogen isotope ratio in pediciids is 2.03‰ higher than in gammarids (Fig. 1); i.e., taking into account the generalized isotopic fractionation factor for aquatic animals [14], it appears that the trophic position of pediciids is approximately one level higher than that of gammarids. Moreover, the average $\delta^{13}\text{C}$ in the feces of gammarids differ significantly from those in their bodies but are similar to these ratios in the leaves (Fig. 1). At the same time, $\delta^{13}\text{C}$ in the feces of predatory pediciids are almost equal to those both in their bodies and in their potential prey (gammarids). The above differences indicate that, although gammarids ingest fragments of the leaf litter, they do not assimilate but excrete them. Apparently, they assimilate only the periphyton (microalgae and bacteria) on the surface of these fragments, while organic matter contained in the leaves themselves passes in transit through the gut, as has been previously shown for some algae ingested by gammarids [5].

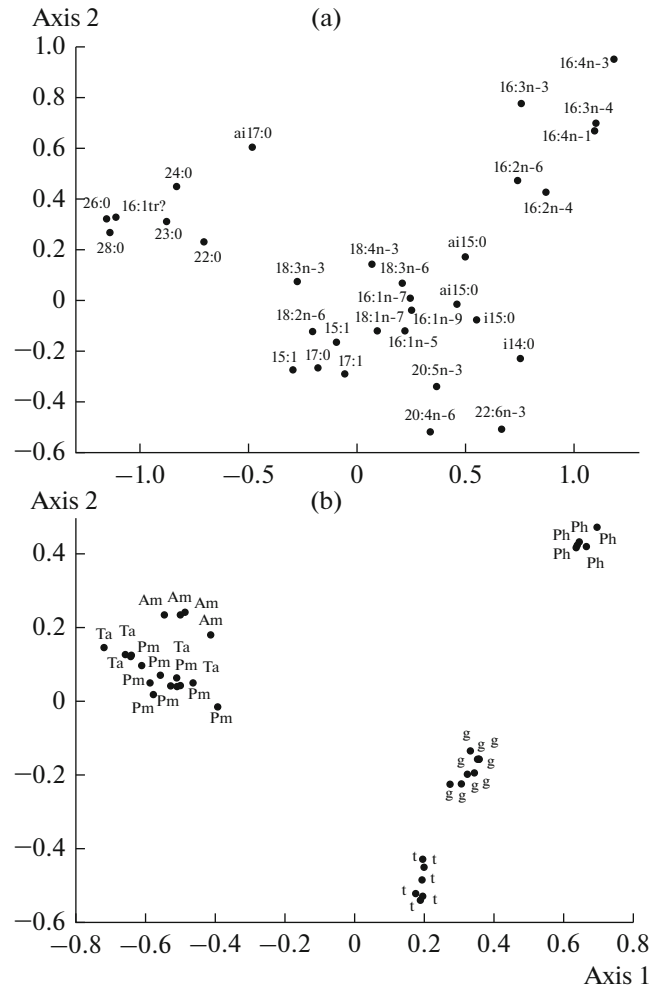


Fig. 2. Multivariate canonical correspondence analysis of arcsin-square-root transformed proportions of dietary fatty acids (a) in the biomass of components of the Turova Pad' stream ecosystem (b): Ta, linden (*T. amurensis*) leaves; Am, maple (*A. mono*) leaves; Pm, poplar (*P. maximowiczii*) leaves; Ph, periphyton; g, gammarids; t, *Pedicia* sp. Proportions of explained variance (inertia): along axis 1, 62.1%; along axis 2, 22.3%; $\chi^2 = 37.1$, d.f. = 1116.

In contrast, the carbon isotope ratio in predatory pediciids does not differ between their feces and bodies (Fig. 1). Our data shown that the larvae of *Pedicia* sp. selectively consume only animal food and easily assimilate it but apparently do not ingest inedible leaves.

The results of FA analysis generally coincide with those of isotope analysis. Indeed, multivariate canonical correspondence analysis of 32 dietary FAs has placed gammarids and pediciids closer to the periphyton than to the leaf litter (Fig. 2). Nevertheless, it cannot be excluded that absolutely essential polyunsaturated fatty acids (PUFA) such as linoleic (LA, 18:2n-6) and alpha-linolenic (ALA, 18:3n-3) are assimilated by gammarids from the leaf litter. The average LA levels in the leaf litter and biomass of zoobenthos are signifi-

Table 1. Levels of essential and conditionally essential polyunsaturated fatty acids (%) relative to the total FA content (Σ FA, mg/g wet weight) in the biomass of different components of the Turova Pad' stream ecosystem. Values with the same letter indices do not differ statistically from each other according to one-way ANOVA with post hoc Tukey's test

Acid	Linden	Maple	Poplar	Periphyton	Gammarids	Tipulids
18:2n-6	13.8 \pm 0.4 ^a	13.2 \pm 0.7 ^a	13.7 \pm 0.9 ^a	5.9 \pm 0.0 ^b	10.4 \pm 0.3 ^c	12.5 \pm 0.2 ^{a, c}
18:3n-3	20.0 \pm 1.7 ^a	32.7 \pm 2.3 ^b	20.0 \pm 3.1 ^a	13.6 \pm 0.1 ^a	9.3 \pm 0.2 ^c	9.7 \pm 0.5 ^c
20:4n-6	0.1 \pm 0.0 ^{a, b}	0.1 \pm 0.0 ^a	0.4 \pm 0.1 ^{a, b}	1.2 \pm 0.0 ^b	2.2 \pm 0.0 ^c	4.6 \pm 0.6 ^d
20:5n-3	0.4 \pm 0.1 ^a	0.2 \pm 0.1 ^a	1.2 \pm 0.2 ^a	6.5 \pm 0.1 ^b	4.5 \pm 0.1 ^c	9.5 \pm 0.9 ^d
22:6n-3	0.0 \pm 0.0 ^a	0.0 \pm 0.0 ^a	0.0 \pm 0.0 ^a	0.1 \pm 0.0 ^b	0.4 \pm 0.0 ^c	0.2 \pm 0.0 ^b
Σ FA	4.7 \pm 0.3 ^a	4.9 \pm 0.3 ^a	3.4 \pm 0.3 ^a	3.1 \pm 0.1 ^a	20.9 \pm 0.7 ^b	5.0 \pm 1.5 ^a

cantly higher than in the periphyton (Table 1). Large amounts of LA are synthesized by terrestrial plants, and this fatty acid is regarded as a marker of allochthonous organic matter in trophic webs of aquatic ecosystems [15], which is confirmed by our data. It is not excluded that benthic animals partially utilize LA from the leaf litter for synthesizing physiologically important arachidonic acid (20:4n-6, ARA), in a case when dietary ARA is in deficit (see Table 1). The levels of ALA (18:3n-3) in the leaf litter are also significantly higher than in the periphyton. However, it is the periphyton that has provided benthic animals with other physiologically important (conditionally essential) PUFAs such as eicosapentaenoic (EPA, 20:5n-3) and docosahexaenoic (DHA, 22:6n-3) (Table 1). Higher terrestrial plants do not synthesize EPA [16], and its traces in the leaf litter may be explained by the presence of periphyton that has not been completely brushed off during sample processing.

Thus, benthic invertebrates in a Far Eastern forest stream receive organic carbon mainly from the periphyton, an autochthonous source, rather than from the allochthonous leaf litter. This agrees with the results of studies performed by similar methods on streams from other biomes. However, it is the leaf litter that appears to provide benthic animals with one of dietary components, namely, essential linoleic acid 18:2n-6. Our data indicate the importance of assessing trophic chains for the sources of not only bulk organic carbon but also of individual essential substances. Moreover, we have found that gammarids ingesting fragments of leaf litter do not assimilate but excrete them.

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