International Review of Hydrobiology 2013, *98*, 132–140 DOI 10.1002/iroh.201301524

RESEARCH PAPER

The quality and availability of fine particulate organic matter for collector species in headwater streams

Marcos Callisto[1] *and Manuel A. S. Graça*[2]

[1] Lab. Ecologia de Bentos, Depto. Biologia Geral, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, Belo Horizonte, MG, Brazil
[2] IMAR-CMA and Depto. Life Sciences, University of Coimbra, Portugal

Received: August 2, 2012
Revised: February 12, 2013
Accepted: February 22, 2013

Fine particulate organic matter (FPOM) is abundant in rivers, but the quality and quantity of FPOM in low-order streams have been investigated less frequently than the dynamics of coarse particulate organic matter (CPOM). We (i) assessed the quantity and quality of FPOM in several low-order mountain streams and the relationship between the quantity of FPOM and invertebrate abundance and (ii) evaluated the importance of microbial colonization to the growth of the FPOM consumer *Chironomus riparius*. FPOM availability ranged from 0.3 to 25.9 g ash-free dry mass (AFDM)/m^2 and was significantly higher during summer than during autumn. The density of invertebrates (10–13,500 individuals/m^2) and FPOM were correlated ($R = 0.74$). Chironomids were the most abundant organisms in the depositional zones where FPOM accumulated. Typically, FPOM nitrogen content was $<1\%$, C:N ranged from 21 to 30%, and lignin ranged from 44 to 66%. Nitrogen content was generally lower during the autumn than summer. The ergosterol content of the FPOM was significantly lower (18 μg/g AFDM) than that determined for leaves, from either water or soil. Chironomids fed with FPOM obtained from milled oak leaves, natural FPOM and sterile FPOM had 80, 45, and 0% emergence, respectively. We concluded that FPOM varies seasonally in quantity and quality in low-order streams and is a poor food resource relative to CPOM. Microbial colonization of FPOM could be important for the trophic ecology of collector species in low-order streams.

Keywords:
FPOM / Low-order streams / Food quality / Chironomidae

1 Introduction

Headwater streams receive organic matter from both upstream reaches and lateral input [1]. Terrestrial-aquatic linkages have been studied by investigating the breakdown of coarse particulate organic matter (CPOM), primarily leaves from riparian vegetation (see review by [2]). The physical fragmentation of leaves [3] and the joint action of decomposers and detritivores result in the production of fine particulate organic matter (FPOM; 0.45 μm–1.0 mm), carbon respiration and carbon incorporation into secondary production [4].

Although the breakdown of CPOM to FPOM is well documented, the dynamics of FPOM in headwater streams and the availability and quality of FPOM to collectors are less well known (but see [5, 6]). FPOM is abundant in streams [7, 8] and in headwaters it can exceed the standing stock of CPOM [2, 9, 10]. The conversion of CPOM into FPOM most likely decreases the quality of organic matter, with a predominance of refractory compounds such as lignin and cellulose that are not easily digested by macroinvertebrate detritivores [11, 12]. The slow decomposition of FPOM [12] and its short retention time suggest that a large portion of FPOM is exported to downstream reaches, estuaries, and coastal zones before being mineralized or buried within sediments. In contrast,

Handling Editor: Helge Norf

Correspondence: Marcos Callisto, Lab. Ecologia de Bentos, Depto. Biologia Geral, Instituto de Ciências Biológicas, Universidade Federal de Minas Gerais, CP 486, CEP 30161-970, Belo Horizonte, MG, Brazil
E-mail: callistom@ufmg.br
Fax: +55-31-3409-2569

Abbreviations: AFDM, ash-free dry mass; **CPOM**, coarse particulate organic matter; **FPOM**, fine particulate organic matter.

as seen from the high abundance of sediment-feeding and filtering invertebrates in headwater streams (e.g. [13, 14]), FPOM most likely represents an important food source for stream consumers, and a large proportion of FPOM can be processed in streams.

Important consumers of FPOM are early stages of many stream invertebrates, including shredders (e.g. [15]). Chironomids are potentially important consumers of FPOM given their high densities in many headwaters [4, 16, 17], including organically enriched stretches [18].

Because it is probable that decomposers and detritivores use the high-quality substrate promptly as stated above, highly refractory compounds are expected to be present in FPOM as a result of biological decomposition. However, given its large surface area, FPOM is also susceptible to heavy colonization by microorganisms [19], which have a relatively high nitrogen and phosphorus content. Moreover, FPOM also includes algal cells and invertebrate fecal pellets. These components can represent a good-quality food source for consumers due to the low C:N ratio of living biological material.

We quantified the availability and the quality of FPOM in low-order deciduous forest streams during summer (before the litter fall) and autumn (during the litter fall peak). We also evaluated the importance of microbial decomposers to the growth of the FPOM consumer *Chironomus riparius*, a model consumer. We hypothesized that (i) the abundance and quality of FPOM will be high during the high-litterfall period and (ii) given the low quality of FPOM substrates, *Chironomus riparius* growth will be positively affected by the presence of microbial decomposers.

2 Methods

2.1 FPOM availability and abundance of collectors

FPOM was sampled in eight 1st–2nd order streams located on Lousã Mountain, central Portugal (40°04′02″ to 40°05′49″N; 8°11′41″ to 8°13′33″W; Fig. 1) once in June (summer) and again in October (autumn) 2010; all samples in a season were taken in a same date. The mountain is heavily forested with mixed forest, composed mainly by chestnuts, pines, oaks, and acacias along roads. All streams were completely covered by canopy and with a high slope. At each stream, we measured the depth and discharge, pH, conductivity, and dissolved oxygen with field probes, and we collected 1 L of water for further laboratory analysis. Nitrates were determined with ion chromatography. Phosphorus (SRP) was determined with the ascorbic acid method [20]. FPOM was sampled from depositional areas with a device consisting of an inverted funnel (15 cm diameter) connected by a plastic tube (1.5 cm diameter) to a manual pump. The funnel was placed over the streambed and water and FPOM was pumped into 5 L plastic bottles (four replicates/stream/season). In preliminary tests in which five sequential samples were retrieved from the same place, we calculated that ~44% of the estimated FPOM per area was collected in the first bottle (negative exponential regression of FPOM over sequential samples, $r^2 = 0.91$).

In the laboratory, the samples were vigorously shaken, and a known amount of water with FPOM was filtered (Whatman 0.45 μm GFC pre-burned, pre-weighed filters) until the filter was partially blocked by the FPOM. The filters containing FPOM were dried (60°C, 72 h) and ignited (550°C, 4 h) to compute ash-free dry mass (AFDM; difference in mass between dry and ignited filters). The results were expressed in mg AFDM of FPOM/m^2 by extrapolating the values for the filters to the total volume of the sample, the funnel area and the extraction efficiency.

The remaining unfiltered material from each sample was allowed to settle. The excess water was removed with a syringe, and the sample was allowed to dry at room temperature and then freeze-dried. This material was used in other analyses (see below). The invertebrates in the samples (including the filters) were sorted and identified before the sample was dried.

2.2 Quality of benthic fine particulate organic matter

Samples of freeze-dried FPOM (see previous section) were ground into a homogeneous powder and sieved through a 106 μm mesh. Total carbon and total nitrogen concentrations were quantified in a CHNS/O analyzer (Fisons Instruments Model EA 1108, Waltham, MA, USA), and lignin was determined gravimetrically with acid-detergent fiber analysis [21]. Values were expressed as a percentage of AFDM.

In one of the streams (Candal, stream #1), we collected five FPOM samples from different pools to measure fungal biomass. For comparative purposes, we also randomly collected five replicate leaf samples from trees (two dominant species, *Quercus robur* and *Castanea sativa*), from the litter bed at the riparian zone and from the stream for fungal biomass measurements (ergosterol). The FPOM samples were transported in an ice chest to the laboratory and allowed to settle. The supernatant was eliminated, and the decanted material was freeze-dried and the ergosterol analyzed as described by Gessner [21].

2.3 Microbial respiration on FPOM

To estimate microbial respiration, extra samples from stream #1 (Candal) were transported to the laboratory in an ice chest. On the same day, 3-mL subsamples (containing ca. 580 mg) were placed in closed chambers (Strath-

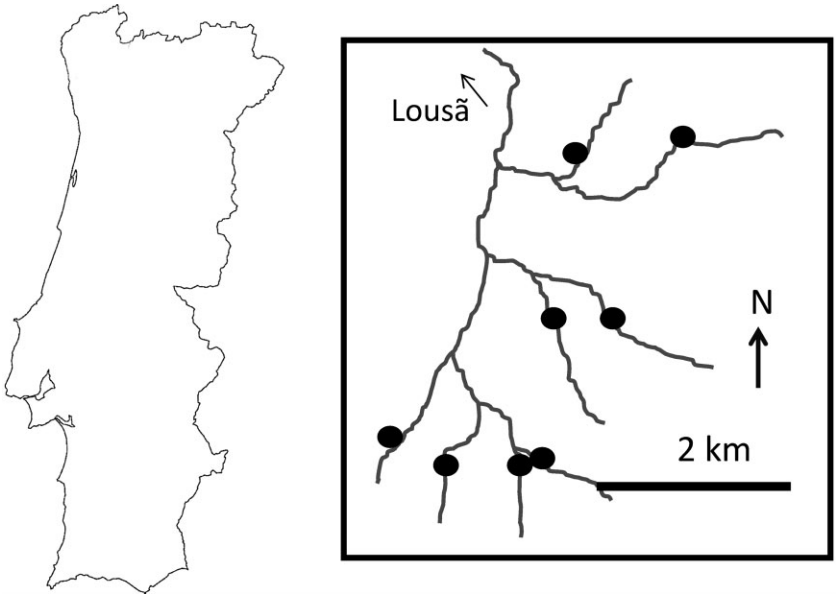

Figure 1. Location of the eight streams sampled in Lousã, central Portugal.

Kelvin 928 System, North Lanarkshire, Scotland) in a respirometry system in which oxygen was continuously measured. Three chambers without FPOM were used as controls. Oxygen consumption rates were calculated as the difference between the oxygen concentration in the sample and the control over a 20 min interval and corrected for the chamber volume. As an additional control, oxygen consumption was also measured on sterilized (autoclaved) samples. After the measurements were made, the AFDM of the FPOM samples was calculated as described above. The results were expressed as mg O_2/g AFDM/h.

2.4 FPOM as a food source for the collector *Chironomus riparius*

To evaluate the quality of natural FPOM as a resource for collectors and the importance of microbes for sediment-feeding invertebrates, we performed a laboratory experiment with a model consumer, *Chironomus riparius* (Meigen) 3rd instar larvae (10 days old from a laboratory culture). The experiments were conducted at 20 ± 2°C with a light/dark regime of 14:10 h under incandescent light. The larvae were obtained from at least ten egg masses of laboratory cultures (procedures described in Soares et al. [22]).

We tested three food treatments: (i) natural benthic FPOM; (ii) sterile FPOM (as in treatment (i), but sterilized in an autoclave; 120°C, 1 h); and (iii) FPOM obtained from milled oak leaves. The stream benthic FPOM was collected as previously described in a 50 mL volume. Senescent oak leaves taken from Lousã Mountain in autumn 2010 were mill-ground and passed through a 0.50-mm mesh net.

Twenty *C. riparius* larvae were randomly allocated to each treatment (total $= 20 \times 4 = 80$ larvae); an extra set of 20 individuals was sacrificed to calculate the initial mass of the cohort. The specimens were placed individually in 7-cm diameter Petri dishes containing 10 mL of reconstituted hard water [23]. Food was added ad libitum from an original decanted aliquot (approx. 5 mL, 10 mg dry mass). Every 2 days, food and water were replaced, up to a maximum of 19 days, when all larvae, pupae, males, and females were individually dried, weighed, oven-dried and reweighed, to determine AFDM and the time required for pupation was recorded.

2.5 Statistical data analysis

A two-way analysis of variance was used to test for seasonal differences in FPOM quantity (log $\times$ transformed data, Holm–Sidak test for multiple comparisons) and differences across streams. The same analysis was used to test for differences in C, N, C:N, and lignin (C:N values were square-root transformed, whereas absolute values were used for other parameters). The ergosterol content in the different organic substrates was also compared with an ANOVA (after rank transformation to correct for the non-normal distribution). A Pearson correlation was used to test if total invertebrate numbers and chironomid collectors were related to the abundance of organic matter. The microbial respiration during the two seasons was compared with a t test. The quality of the three-tested foods for *C. riparius* was investigated with a two-way ANOVA (to incorporate sex differences) of the individual adult weight. All calculations were run in SigmaStat 3.5 (Systat Software Inc.).

3 Results

3.1 FPOM availability

All streams were similar in environmental parameters, and the seasonal variation was also quantitatively low (Table 1). Only phosphates and discharge were consistently higher during autumn than during summer. FPOM levels in the eight studied headwater streams ranged from 5.7 to 25.9 g AFDM/m^2 during summer and 0.3 to 5.5 g AFDM/m^2 during autumn (Fig. 2). FPOM levels were significantly lower during autumn ($F_{1,46} = 5.967$; $p < 0.001$) and differed significantly among streams ($F_{1,46} = 271.383$; $p < 0.001$).

The invertebrate density in the samples ranged from approx. 100 to 3,500 individuals/m^2 in summer and 10 to 150 individuals/m^2 in autumn. Chironomids contributed to 33–81% of the total macrofauna (15% Chironomini, 36% Tanytarsini, 38% Tanypodinae, and 11% Orthocladiinae). Other invertebrates were the plecopterans *Leuctra* and *Nemoura* (12%), the ephemeropteran *Paraleptophlebia* (10%), the coleopterans *Hydroglyphus* and *Deronectes* (2%) and others (<1%), such as trichopterans (*Allogamus*), megalopterans (*Sialis*), Odonata (*Cordulegaster*), and Lumbriculidae.

The total abundance of invertebrates and the chironomid abundance in the streams were correlated with FPOM quantity (correlation coefficients = 0.738 and 0.605; $p = 0.001$ and 0.013, respectively; pooled summer and autumn data).

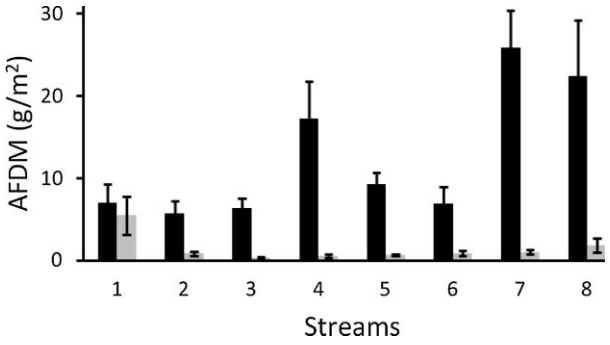

Figure 2. Abundance of FPOM (g AFDM/m^2, mean ± SE) in depositional areas of eight streams in the Lousã, Portugal. Dark columns, summer; gray columns, autumn.

3.2 Chemical quality of FPOM

Typically, N was <1% of the FPOM mass (AFDM basis). On the same basis, carbon represented 15–25% of the FPOM mass, lignin represented 44–66% of the FPOM mass and C:N ranged from ~20 to 30% (Fig. 3). The FPOM quality differed significantly among streams in terms of

Table 1. Selected chemical and physical characteristics of eight Lousã Mountain streams, central Portugal, where FOPM was sampled

		Headwater streams							
Variables	Seasons	1	2	3	4	5	6	7	8
Temperature (°C)	Summer	12.4	11.2	12.7	13.5	13.4	14.0	15.4	16.1
	Autumn	10.9	10.1	10.1	10.3	10.4	11.2	11.9	12.1
Dissolved oxygen (mg/L)	Summer	8.30	9.52	8.44	8.42	8.83	8.15	7.74	8.15
	Autumn	8.60	8.50	8.90	8.70	8.80	8.40	7.50	8.60
pH	Summer	6.8	6.7	5.8	5.4	5.8	6.1	5.9	6.2
	Autumn	8.4	7.1	7.4	7.9	7.7	7.6	6.8	7.5
Conductivity (μS/cm)	Summer	25.0	28.0	29.0	35.0	33.0	25.0	53.0	46.0
	Autumn	28.8	26.7	30.9	29.3	36.6	43.7	54.2	45.6
Alkalinity ($CaCO_3$) (mg/L)	Summer	5.2	3.0	5.7	4.5	4.1	3.91	5.7	10.5
	Autumn	3.1	2.8	3.7	1.6	1.8	3.2	7.1	4.6
SRP (ppm)	Summer	<0.01	<0.01	0.012	<0.01	<0.01	<0.01	0.016	0.014
	Autumn	0.371	0.392	0.411	0.387	0.333	0.500	0.550	0.100
N-O3 (ppm)	Summer	0.244	0.110	0.223	0.953	0.873	0.226	0.299	0.285
	Autumn	0.145	0.442	0.460	0.159	0.766	0.163	0.204	0.210
Mean depth (cm)	Summer	30.2	5.0	10.0	10.0	15.0	15.0	15.0	10.0
	Autumn	35.0	15.0	12.0	9.0	15.0	27.0	15.0	23.0
Mean width (m)	Summer	1.8	1.0	1.1	1.5	2.0	1.5	1.0	1.0
	Autumn	1.9	1.0	1.4	1.9	2.0	2.8	1.5	2.5
Mean discharge (L/s)	Summer	0.014	0.016	0.015	0.010	0.013	0.020	0.012	0.022
	Autumn	0.105	0.098	0.104	0.099	0.120	0.105	0.103	0.084

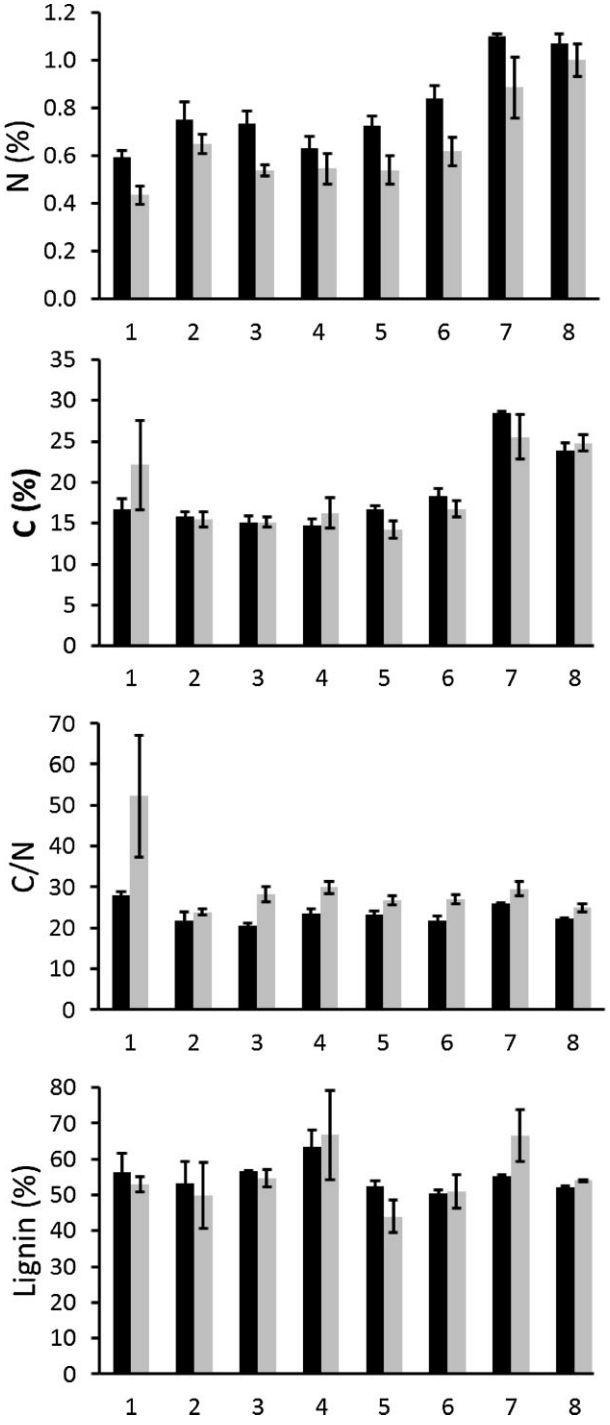

Figure 3. FPOM quality in eight headwater streams. Values are percentages of AFDM (mean ± SE). Dark columns, summer; gray columns, autumn.

carbon ($F_{7,47} = 17.791$; $p < 0.001$), nitrogen ($F_{7,47} = 19.199$; $p < 0.001$), C:N ($F_{7,47} = 8.589$; $p < 0.001$) and lignin ($F_{7,45} = 45.095$; $p < 0.001$). Nitrogen was significantly higher during summer than during autumn ($F_{1,47} = 28.848$; $p < 0.001$) and the opposite was true for C:N ($F_{1,47} = 62.126$; $p < 0.001$). No seasonal differences were observed in terms of carbon or lignin ($p > 0.05$).

3.3 Microbial quality of FPOM

The ergosterol content of the FPOM was significantly lower (18 μg/g AFDM) than litter from soil or water (95–54 μg/g AFDM) (Fig. 4; $H = 21.344$, $p < 0.001$). Leaf samples taken from trees had virtually no ergosterol.

Respiration on FPOM ranged from 16.47 to 34.31 mg O_2 g/AFDM/h (N = 9) during summer and 24.76 to 39.07 O_2 g/AFDM/h (N = 9) during autumn (Fig. 5). The respiration rates were significantly higher during autumn than during summer ($t = 2.672$; $p = 0.017$).

3.4 *Chironomus riparius* feeding experiments

The initial biomass of *C. riparius* larvae used in the experiments was 0.26 ± 0.05 mg (N = 20). By the end of the 22-day feeding trial, mortality was 10, 35, and 30%, respectively, for sets of larvae fed *Quercus* FPOM, stream FPOM, and sterile FPOM. Adult emergence was 80, 45, and 0% of the initial population, respectively, for the same treatments (Fig. 6). Emergent females were heavier than males ($F = 16.825$, $p < 0.001$), and specimens fed *Quercus* FPOM were heavier than those fed with stream FPOM ($F = 8.513$, $p = 0.008$; Fig. 7). However, no differences in the time to adult emergence success were observed ($F = 2.143$, $p > 0.05$).

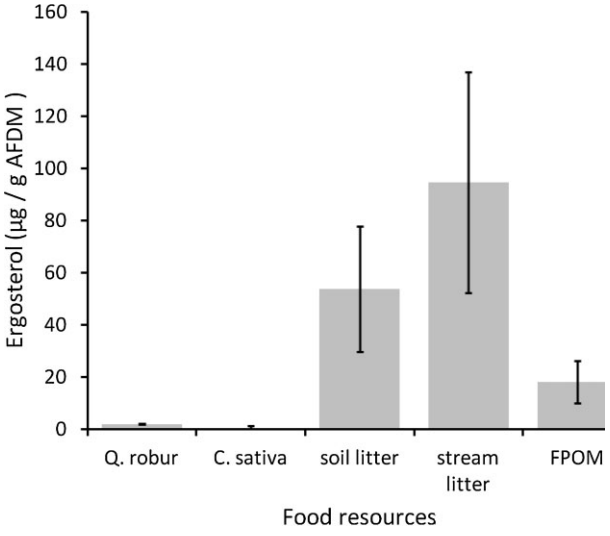

Figure 4. Ergosterol content in four organic substrates collected in autumn at stream #1 site (mean ± SE).

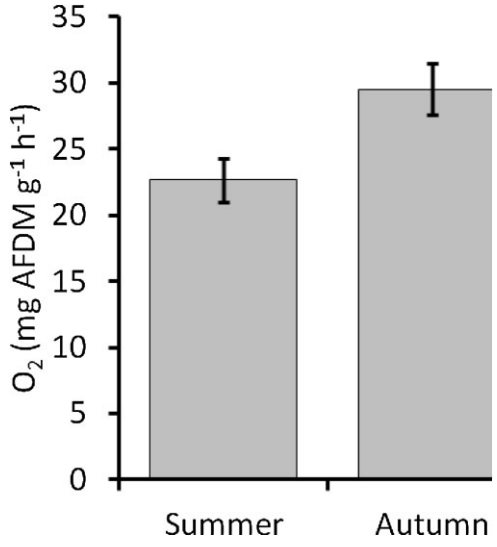

Figure 5. Microbial respiration on FPOM substrates from a stream during summer and autumn (mean ± SE).

4 Discussion

Our study showed that FPOM is abundant in depositional areas of low-order streams but that the quality of the FPOM is generally lower than that of the CPOM. As postulated in our initial hypothesis, the abundance of invertebrates (and deposit-feeding chironomids) was correlated with FPOM quantity. Finally, we showed that the quality of the FPOM affected the growth of *C. riparius*. We will first discuss the implications of the variation in the quantity of the FPOM for stream invertebrates and will then discuss the implications of the quality variations. In our comparisons with the literature (below) we used a conversion factor of 1 mg dry mass = 0.4 mg AFDM, which was an overall mean in our study.

The FPOM was quantitatively similar across the eight low-order streams. The values obtained (0.3–25.9 g AFDM/m^2) were within the range of values reported for low-order streams in other studies: 0.8 g/m^2 [24], 5.32 g/m^2 (annual mean; [9]); 750 g m^2 [25] and higher than the obtained for CPOM in a larger local stream [26]. However, given that we sampled exclusively depositional zones, the overall stream values were most likely lower than the values we reported here.

If FPOM is a product of CPOM consumption by decomposers and detritivores, physical abrasion [2, 3] and run-off from the riparian area, we could expect a higher FPOM density in autumn, the period during which the litterfall is more intense in the temperate zone (e.g. [27]). Our autumn samples coincided with the peak litter input to the streams, and leaves are the primary source of FPOM in many systems (e.g. [28]). However, the standing stock of FPOM was lower in autumn. This result may be related to the hydrological regime. The high level of precipitation in autumn (∼100–140 mm monthly means for October and November versus ∼10–40 mm means in June and July [29]) flushes the FPOM to the lower reaches, with only a small contribution from the upper reaches given the short stream lengths (1st and 2nd order). Other authors have shown that the transport of fine organic matter is strongly conditioned by discharge (e.g. [30]).

We calculated that deposit-feeding chironomids attained densities up to >2100 individuals/m^2. This value is much less than the maximum of 50,000 reported by Romito et al. [17] for Appalachian headwater streams. Other small invertebrates sampled from the FPOM deposits included *Leuctra* sp., whose early instars have been reported to feed on fine particles of organic matter [15]. The total abundance of invertebrates in depositional areas of the benthos (c. 50% of total streams length) was correlated with FPOM availability across streams. This result could reflect two non-exclusive causes. First, invertebrate population levels could be determined by the availability of food resources. Second,

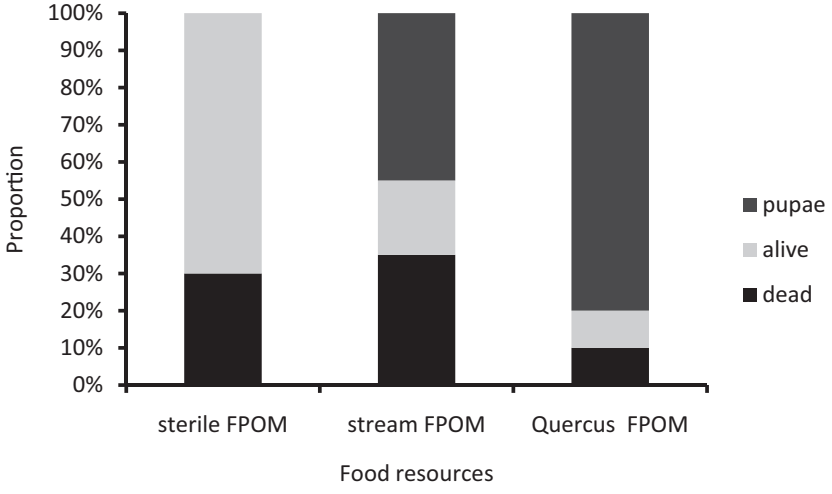

Figure 6. Proportion of *Chironomus riparius* entering pupation, dead and still alive as larvae after 18 days on three diet types.

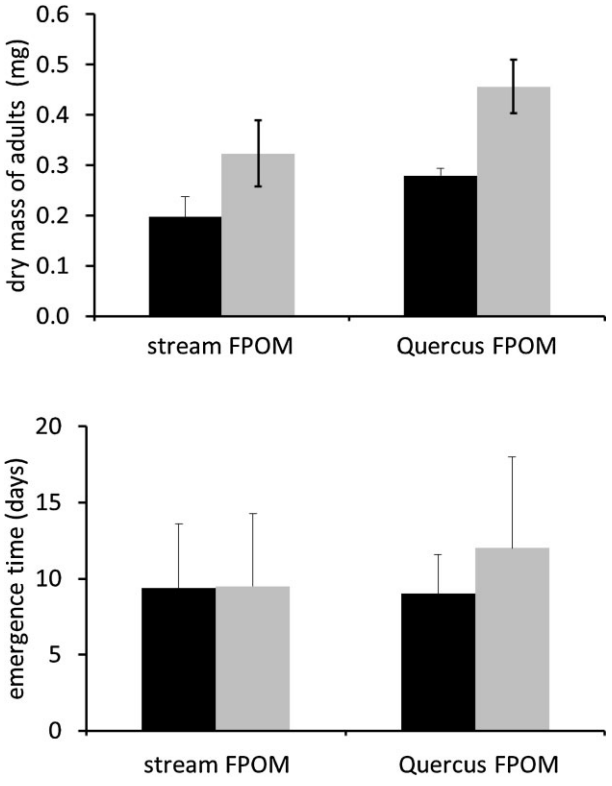

Figure 7. Adult size of male (black columns) and female (gray columns) *Chironomus riparius* and time to emergence for specimens fed two diets.

the same forces that sort and transport FPOM could remove small invertebrates, as well. Therefore, the abundance of FPOM may be related rather to hydrology (and its variation) than to production in the tested low-order streams. However, more studies are needed to allow generalizations because the data in the literature on this topic are contradictory. For example, a higher abundance of FPOM was reported for autumn than for summer in a Japanese study [9].

The analysis of FPOM quality showed a low N content and high C:N and lignin. For comparative purposes, the C:N range obtained here (21–30 and one value of 51) was within the range reported in the literature: 7–39 [8], 17–28 [5], and 10–34 [12] but greater than the value of 9.3 reported for headwaters in Japan [9]. Our N-values (0.44–1.10) were also within the higher range reported by other studies (0.07–1.10 [5]). Finally, we measured a lignin content of 44–67%. This value is also comparable to the values of 45% [31, 32] and 21–46% [12] reported in the literature. Overall, the quality of the FPOM was comparatively lower than that reported for decomposing leaves in streams [33].

N (and C:N) differed across seasons, but lignin did not. High quality was observed during summer, as reported by Akamatsu et al. [9]. However, the opposite pattern was reported by Bonin et al. [5] for C:N. The higher quality of summer samples could be related to a presumably higher residence time of particles and therefore to a longer period allowing higher microbial colonization and perhaps a higher abundance of algae during summer. However, Cummins and Klug [8] have suggested that the quality of FPOM decreases with time because easily digestible substrates are metabolized earlier. Differences between studies may reflect differences in forest cover [5], the location along the longitudinal gradient in the stream [9], the size of particles and their origin (e.g. fecal pellets, leaf fragments, and living cells). Future studies might consider the use of stable isotopes to evaluate the origin of the FPOM.

The higher FPOM quality during summer has ecological implications. During this season, we observed more invertebrates in the depositional areas of the FPOM. Therefore, FPOM could be relatively more important for consumers during a period in which leaves are not present in the stream.

The biological quality of the FPOM was assessed in three ways: (a) microbial activity, measured as respiration rates; (b) fungal biomass; and (c) the capability of the FPOM (with and without microorganisms) to promote the growth of a consumer. In terms of respiration, our range of 23–30 mg O_2 g/AFDM/h was very high compared with the range of 0.3–1.4 O_2 g/AFDM/h (recalculated from C and organic matter) reported by Bonin et al. [5], the 0.06 O_2 g/AFDM/h reported by Webster et al. [34], and the range of 0.03–0.54 O_2 g/AFDM/h reported by Ward [31]. These differences in respiration could again be related to the origin of the FPOM, with different fractions differing in quality (e.g. [31]). Based on the chemical quality of the FPOM, we would expect higher respiration rates in summer than in autumn, but this was not the case. However, we did not compare microbial biomass and types of microorganisms (bacteria, fungi, algae) across seasons. Regardless of differences in biomass, microbial activity was higher in autumn.

The fungal biomass was lower in the FPOM than in the leaves collected from soil or water. This result was expected, to some degree, because the consumer:resource ratio may favor the growth of hyphae in larger rather than in smaller particles of organic matter (e.g. [35]). It could be argued that bacteria biomass (not measured in our study) is more important than fungal biomass (e.g. [8]) improving substrate quality for FPOM consumers. However, growth and survivorship of *Chironomus riparius* was a direct measure of quality in our study and it showed that FPOM had a relatively low quality.

The differences in the biological quality of the FPOM could be as important as differences in FPOM chemistry. Regardless of the identity of the microbes in the FPOM, it

was clear that their absence had costs in terms of adult success. Moreover, the smaller adult size of specimens grown on natural FPOM than in those grown on powdered *Quercus* leaves is an indication of the lower quality of the FPOM as a food resource for stream consumers. Fragments of leaves obtained from physical processes are a realistic source of FPOM [3]. Given the highly refractory nature of the FPOM, the dependence of FPOM consumers on microbes is not surprising. A similar observation of decreased growth was reported for *Simulium* fed with sterile FPOM [36]. In the real world, differences in the microbial biomass in the FPOM could be related to the nutrient load of the stream waters, FPOM chemistry, and grazing by protozoa.

In conclusion, the abundance of small invertebrates in streams was related to the abundance of FPOM, whose quality is lower than that of leaves and varies strongly across seasons. Microorganisms colonizing FPOM are very important for the trophic ecology of *C. riparius* and presumably for other stream collector consumers.

The study was supported by the IMAR-CMA, by the Brazilian Science Foundation CNPq, CAPES, and FAPEMIG. We thank Veronica Ferreira, Cristina Docal, and Elsa Rodrigues for their support in the laboratory and Débora Lobato, Franck Brites, Juliana França, Joseline Molozzi and Wander Ferreira for field support. Matilde Moreira-Santos and Rui Ribeiro provided specimens of C. riparius and provided assistance with laboratory experiments. Two anonymous referees and editor Helge Norf made detailed suggestions to improve the manuscript. MC acknowledges CAPES for a sabbatical fellowship (4959/09-4) at the IMAR, University of Coimbra, Portugal.

The authors have declared no conflict of interest.

5 References

[1] Gessner, M. O., Swan, C. M., Dang, C. K., Mckie, B. G. et al., Diversity meets decomposition. *Trends Ecol. Evol.* 2010, *25*, 372–380.

[2] Abelho, M., From litterfall to breakdown in streams: a review. *Scientific World J.* 2001, *1*, 656–680.

[3] Heard, S. B., Schultz, G. A., Ogden, C. B., Griesel, T. C., Mechanical abrasion and organic matter processing in an Iowa stream. *Hydrobiologia* 1999, *400*, 179–186.

[4] Wallace, J. B., Cuffney, T. F., Webster, J. R., Lugthart, G. J. et al., Export of fine organic particles from headwater streams – effects of season, extreme discharges, and invertebrate manipulation. *Limnol. Oceanogr.* 1991, *36*, 670–682.

[5] Bonin, H. L., Griffiths, R. P., Caldwell, B. A., Nutrient and microbiological characteristics of fine benthic organic matter in sediment settling ponds. *Freshwater Biol.* 2003, *48*, 1117–1126.

[6] Yoshimura, C., Fujii, M., Omura, T., Tockner, K., Instream release of dissolved organic matter from coarse and fine particulate organic matter of different origins. *Biogeochemistry* 2010, *100*, 151–165.

[7] Alvarez, S., Guerrero, M. C., Enzymatic activities associated with decomposition of particulate organic matter in two shallow ponds. *Soil Biol. Biochem.* 2000, *32*, 1941–1951.

[8] Cummins, K. W., Klug, M. J., Feeding ecology of stream invertebrates. *Ann. Rev. Ecol. Syst.* 1979, *10*, 147–172.

[9] Akamatsu, F., Kobayashi, S., Amano, K., Nakanishi, S., Oshima, Y., Longitudinal and seasonal changes in the origin and quality of transported particulate organic matter along a gravel-bed river. *Hydrobiologia* 2011, *669*, 183–197.

[10] Rodriguez-Barrios, J., Ospina-Torres, R., Gutierrez, J. D., Inputs and transport of organic matter in a tropical mountain stream, Bogota, Colombia. *Caldasia* 2008, *30*, 421–439.

[11] Meybeck, M., Carbon, nitrogen, and phosphorus transport by world rivers. *Am. J. Sci.* 1982, *282*, 401–450.

[12] Yoshimura, C., Gessner, M. O., Tockner, K., Furumai, H., Chemical properties, microbial respiration, and decomposition of coarse and fine particulate organic matter. *J. N. Am. Benthol. Soc.* 2008, *27*, 664–673.

[13] Callisto, M., Moreno, P., Barbosa, F. A. R., Habitat diversity and benthic functional trophic groups at Serra do Cipó, Southeast Brazil. *Braz. J. Biol.* 2001, *61*, 259–266.

[14] Miserendino, L., Masi, C. I., The effects of land use on environmental features and functional organization of macroinvertebrate communities in Patagonian low order streams. *Ecol. Indic.* 2010, *10*, 311–319.

[15] Lopez-Rodriguez, M. J., de Figueroa, J. M. T., Bo, T. Z., Mogni, A., Fenoglio, S., Living apart together: on the biology of two sympatric *Leuctra* species (Plecoptera, Leuctridae) in an Apenninic Stream, Italy. *Int. Rev. Hydrobiol.* 2012, *97*, 117–123.

[16] Graça, M. A. S., Pinto, P., Cortes, R., Coimbra, N. et al., Factors affecting macroinvertebrate richness and diversity in Portuguese streams: a two-scale analysis. *Int. Rev. Hydrobiol.* 2004, *89*, 151–164.

[17] Romito, A. M., Eggert, S. L., Diez, J. M., Wallace, J. B., Effects of seasonality and resource limitation on organic matter turnover by Chironomidae (Diptera) in southern Appalachian headwater streams. *Limnol. Oceanogr.* 2010, *55*, 1083–1092.

[18] Del Arco, A. I., Ferreira, V., Graça, M. A. S., The performance of biological indicators in assessing the ecological state of streams with varying catchment urbanization levels in Coimbra, Portugal. *Limnetica* 2012, *31*, 141–154.

[19] Romani, A. M., Guasch, H., Munoz, I., Ruana, J. et al., Biofilm structure and function and possible implications for riverine DOC dynamics. *Microb. Ecol.* 2004, *47*, 316–328.

[20] American Public Health Association, Phosphorus, *Standard Methods for the Examination of Water and Wastewater*, 19th edn., in: Eaton A. E., Clesceri L. S., Greenberg A. E. (Eds.), Apha Press, Washington, DC 1995.

[21] Gessner, M. O., Proximate lignin and cellulose, in: Graça, M. A. S., Bärlocher F., Gessner, M. O. (Eds.), *Methods to Study Litter Decomposition. A Practical Guide*, Springer, New York 2005.

[22] Soares, S., Cativa, I., Moreira-Santos, M., Soares, A. M. V. M., Ribeiro, R., A short-term sublethal in situ sediment assay with *Chironomus riparius* based on post exposure feeding. *Arch. Environ. Cont. Toxicol.* 2005, *49*, 163–172.

[23] ASTM, *Standard Guide for Conducting Acute Toxicity Tests on Test Materials with Fishes, Macroinvertebrates, and Amphibians*, ASTM International, Philadelphia 2002.

[24] Cushing, C. E., Minshall, G. W., Newbold, J. D., Transport dynamics of fine particulate organic matter in two Idaho streams. *Limnol. Oceanogr.* 1993, *38*, 1101–1115.

[25] Iversen, T. M., Thorup, J., Skriver, J., Inputs and transformation of allochthonous particulate organic matter in a headwater stream. *Holar. Ecol.* 1982, *5*, 10–19.

[26] González J. M., Graça, M. A. S., Conversion of leaf litter to secondary production by a shredding caddis-fly. *Freshwater Biol.* 2003, *48*, 1578–1592.

[27] Graça M. A. S., Pozo, J., Canhoto, C., Elosegi, A., Effects of *Eucalyptus* plantations on detritus, decomposers and detritivores in streams. *Scientific World J.* 2002, *2*, 1173–1185.

[28] Peters, C. M., Gentry, A. H., Mendelsohn, R. O., Valuation of an Amazonian rainforest. *Nature* 1989, *539*, 655–655.

[29] Câmara Municipal de Arganil, 2008 *Plano Municipal de Defesa da Floresta Contra Incêndios [accessed 25-06-2012, available at* http://www.cm-batalha.pt/areas-de-intervencao/gabinete-tecnico-florestal/pmdfci] (in Portuguese).

[30] Webster, J. R., Benfield, E. F., Hutchens, J. J., Tank, J. L. et al., Do leaf breakdown rates actually measure leaf disappearance from streams? *Int. Rev. Hydrobiol.* 2001, *86*, 417–427.

[31] Ward, G. M., Lignin and cellulose content of benthic fine particulate organic matter (FPOM) in Oregon Cascade mountain streams. *J. N. Am. Benthol. Soc.* 1986, *5*, 127–139.

[32] Ward, G. M., Aumen, N. G., Woody debris as a source of fine particulate organic-matter in coniferous forest stream ecosystems. *Can. J. Fish. Aquat. Sci.* 1986, *43*, 1635–1642.

[33] Graça, M. A. S., Bärlocher, F., Gessner, M. O., *Methods to Study Litter Decomposition. A Practical Guide*. Springer, New York 2005.

[34] Webster, J. R., Benfield, E. F., Ehrman, T. P., Schaeffer, M. A. et al., What happens to allochthonous material that falls into streams? A synthesis of new and published information from Coweta. *Freshwater Biol.* 1999, *41*, 687–705.

[35] Bärlocher, F., Kendrick, B., Dynamics of the fungal population on leaves in a stream. *J. Ecol.* 1974, *11*, 761–791.

[36] Fuller, R. L., Fry, T. J., The influence of temperature and food quality on the growth of *Hydropsyche betteni* (Trichoptera) and *Simulium vittatum* (Diptera). *J. Freshwater Ecol.* 1991, *6*, 75–86.

