



Summer flower pulses: Catkin litter processing in headwater streams

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With 5 figures and 4 tables

Abstract: The influences of inter- and intraspecific variation in leaf litter quality on aquatic food webs are well understood. However, the timing and type of organic matter inputs also varies within a species based on plant phenophase, including litterfall and flowering, which may influence instream communities and ecosystem processes including decomposition. We investigated how leaf and flower litters of a willow shrub species (*Salix sitchensis*) influenced decomposition and aquatic invertebrate colonization in a headwater stream. We collected abscised willow catkins and leaves from female plants, analyzed their initial chemistry, constructed litterbags, and incubated them at our stream site for five weeks. A subset of litterbags of each litter type was retrieved at 7, 22, and 37 days and analyzed for litter decomposition and invertebrate colonization. We found significant differences between catkins and leaves. Catkins had lower initial condensed tannin concentrations and higher invertebrate colonization compared to leaves. Initial leaching losses were faster for leaves, but overall decomposition rates were comparable. Catkin structural complexity supported higher invertebrate abundance and diversity as well as altered community composition, indicating their unique contribution to stream ecosystems. Although the ratio of reproductive to leaf inputs is low, we argue that the sequential availability of these two litter types may provide complementary energy inputs to consumers in headwater stream ecosystems dependent on organic matter.

Keywords: aquatic invertebrate; litter decomposition; litterfall; organic matter processing; phenology; reproductive litter; willow

Introduction

Seasonal organic matter (OM) pulses drive fundamental stream ecosystem processes such as nutrient transfer, carbon cycling, and secondary production. Headwater stream energy budgets are highly heterotrophic and depend on allochthonous OM inputs (Vannote et al. 1980; Wallace et al. 1997). After OM enters a stream, it alters the local and downstream nutrient pools through abiotic and biotic decomposition pathways: leaching,

microbial conditioning, detritivore shredding (Webster & Benfield 1986; Graça et al. 2015). Furthermore, OM processing rates span a continuum from slow- to fast-decomposing, depending on species-specific and intraspecific traits (Webster & Benfield 1986; LeRoy et al. 2007). Detritivorous aquatic invertebrates play a major role in OM processing in stream food webs by breaking down coarse OM inputs. These invertebrates prefer litter which is rich in nitrogen and low in phenolics (Graça 2001), and their presence, abundance and

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activity in detritus can thus vary among litter types, influencing the ecosystem process of OM decomposition.

Advances in our understanding of aquatic detritus processing have focused on leaf litter decomposition (Tank et al. 2010). Recently, stream ecologists have moved beyond autumn-shed leaves to assess other OM contributions including woody material (Spänhoff & Meyer 2004), pollen (Rosi-Marshall et al. 2007), periodical cicadas (Pray et al. 2009), maize debris (Griffiths et al. 2009), and herbivore-induced litter inputs (LeRoy et al. 2020a). However, little is known about other significant riparian contributions such as plant reproductive inputs, which may be seasonally important (Abelho 2001) and—for dioecious species—may differ between male and female plants (LeRoy et al. 2020b). Common reproductive inputs from riparian zones in the northern hemisphere include inflorescences, seeds, and fruits from the Salicaceae (willow, cottonwood) and Betulaceae (alder) families. In particular, willow catkins (i.e. reproductive structures) can be major biotic contributors to stream ecosystems at early successional stages of development (i.e. 5–50 years; Flory & Milner 1999; Milner et al. 2007). Examining the role of reproductive litters may further our understanding of OM processing in stream ecosystems.

Reproductive structures differ from leaves in their temporal, morphological, and chemical characteristics. First, in temperate headwater streams, reproductive detritus is typically deposited in early summer when naturally abscising leaf litter inputs are scarce. Because the timing of OM inputs can influence both leaf litter decomposition and the composition of associated invertebrate assemblages (Rodriguez-Cabal et al. 2017), reproductive structures may provide a temporally distinct input to streams. Second, reproductive structures tend to be more morphologically complex than leaves, providing a wider range of substrates for colonization. Aquatic invertebrates have been shown to prefer microhabitats created by such heterogeneous litter aggregates (Compson et al. 2013). Third, plants allocate more nutrients to complex flower parts (Wink 2010) compared to relatively nutrient-poor autumn-shed leaves, which may increase litter lability, microbial colonization, and detritivore consumption (Chomel et al. 2016). These three intrinsic characteristics of reproductive detritus may influence both its in-stream decomposition and its colonization by aquatic invertebrates.

We experimentally tested how summer-shed female catkins of the dioecious willow *Salix sitchensis*

Sanson ex. Bong differed from autumn-shed female willow leaves in terms of initial chemistry, decomposition rates, and invertebrate colonization. We chose female willow catkins and leaves because female willows are represented at a 2:1 female:male ratio at our study site (Che-Castaldo et al. 2015), female willows grow in closer proximity to streams at our study site (LeRoy et al. 2020b), and female catkins are roughly 1.5–2 times larger than male catkins (Fisher 1928). We tested three main hypotheses: 1) catkins have lower condensed tannin and higher nitrogen concentrations compared to leaves; 2) due to their increased lability, catkins decompose faster; and 3) aquatic invertebrates colonize catkins at greater abundances than leaves due to their higher litter quality and structural complexity, causing taxonomic and functional community composition to differ between the two OM types.

Methods

Site description

Geothermal-West (Geo-W) Creek is a tributary of one of five watersheds that formed on the north face of Mount St. Helens (Lawetlat'la in Cowlitz; Washington, USA; del Moral & Jones 2002) following its 1980 eruption (Blackman 2014). Our study site was a 30-m stream reach on Geo-W Creek, a first-order, sinuous, perennial stream in the central Cascade region (N 46° 15' 7.992", W 122° 10' 12.864", 1070 m elevation). Sitka alder (*Alnus viridis* [Chaix] DC. ssp. *sinuata* [Regel] A. Löve & D. Löve) and Sitka willow (*S. sitchensis*) are the dominant woody plant species along the creek, providing approximately 30 % canopy cover over the reach.

To characterize Geo-W Creek and provide a broader environmental context for our study, we measured a suite of physicochemical variables along the stream reach. Instream physicochemical characteristics were measured at the beginning of the study period and on days 7, 22, and 37 (Table 1). Water temperature (°C), specific conductivity ($\mu\text{S cm}^{-1}$), and dissolved oxygen (% and mg L^{-1}) were measured using a YSI multiprobe (Yellow Springs Instruments, Yellow Springs, OH, USA), and pH using a pH probe (Oakton, Melrose, MA, USA; Table 1). Chromophoric dissolved organic matter (CDOM, the fraction of dissolved organic matter that absorbs light in both the ultraviolet and visible ranges) was measured using a Cyclops-7 probe (Turner Designs, San Jose, CA, USA). Surveys of aquatic invertebrates and riparian plants in 2016 provided us with background information about the site (Claeson et al. 2021). The invertebrate community in Geo-W Creek

Table 1. Water chemistry measurements at Geo-W Creek on each litter collection date. DO, dissolved oxygen; Cond, conductivity; Temp, temperature; CDOM, chromophoric dissolved organic matter.

Days	Water chemistry					
	DO (mg L ⁻¹)	DO (%)	Cond (μ S cm ⁻¹)	Temp (°C)	pH	CDOM (ppb)
0	7.0	63.9	274	11.1	7.1	18.4
7	6.1	60.0	351	14.1	7.0	13.5
22	5.5	54.5	349	14.7	7.0	23.9
37	6.0	58.9	438	13.7	7.1	22.2
Mean	6.2	59.3	353	13.4	7.1	19.5

includes approximately 60 taxa, which—reflecting the stream's early successional stage—include many insects with aerial dispersal abilities (Claeson et al. 2021). Canopy cover was measured at the top, middle, and bottom of the reach in the four cardinal directions using a convex densiometer (Forestry Suppliers Inc., Jackson, MS, USA).

Litter collection

Litter samples were collected during the natural senescence period for each litter type (September–November for autumn-shed leaves and June–July for female catkins). Female Sitka willows were identified based on catkin morphology and tagged in May 2018. Naturally senescent leaves were collected from female plants in September–November 2018 and air-dried. Female catkins were collected in early July 2019 and oven-dried at 50 °C for 48 h, to attain comparable dryness to the air-dried leaves.

Litter chemistry

Subsamples of each litter type ($n=20$ pooled catkins, $n=7$ leaf samples from individual willows) were ground to a homogeneous composition prior to initial chemistry analysis. Carbon (%C) and nitrogen (%N) were determined by combustion of 2-mg subsamples ($n=5$ catkin, $n=5$ leaf) in tin capsules in an elemental analyzer (2400 CHNS/O Series II System, Perkin Elmer, Waltham, MA, USA). Condensed tannins (CT) were extracted from 25-mg subsamples ($n=9$ catkin, $n=7$ leaf) into a 70 % acetone and 10 mM ascorbic acid solution using ultra-sonication (Sonicor, West Babylon, NY, USA). We used the butanol-HCl method (Porter et al. 1985) to determine initial CT concentrations using a UV-Vis spectrophotometer (Spectramax 384, Molecular Devices, San Jose, CA, USA) and read absorbance values at 550 nm. A standard curve was generated from a *S. sitchensis* CT standard extracted following Hagerman & Butler (1989).

Litter decomposition

The decomposition experiment was conducted in Geo-W between 24 July 2019 and 30 August 2019. Litterbags were made by placing 1 ± 0.05 g samples into tagged, 1-mm mesh bags (15 × 15-cm). Five replicate bags were made for each litter type (catkin, leaf) and for each of three collection dates (i.e. 30 bags total). Handling losses were determined by transporting litterbags in individual paper bags and weighing any litter fragments in each bag after deploying litterbags. Each replicate litterbag ($n=5$) was placed in a blocked design across five pools in Geo-W Creek and secured individually with cable ties to ropes attached to anchored rebar posts. Litterbags were incubated in the stream in summer to match the period in which the stream receives natural female catkin inputs, and were collected after 7, 22, and 37 days. Autumn-shed litter remains on the surface of the riparian soil and enters the stream via lateral transport over the course of the year. On each collection date, one replicate of each litter type was removed from each of the five pools (with care taken to minimize loss of litter fragments and invertebrates), placed in polyethylene zipper bags, and transported to the laboratory on ice (Benfield et al. 2017).

In the laboratory, each litterbag sample was thoroughly rinsed with deionized water (over a tub to catch all litter particles) and litter fragments were placed in paper envelopes. Litter samples were frozen at -80 °C, freeze-dried (Millrock Technology, Kingston, NY, USA) for 60 h, weighed to the nearest 0.001 g, and handling loss weights subtracted to determine final dry weights. Ash-free dry mass (AFDM) for the freeze-dried litterbag material was determined by combusting 0.25-g subsamples of freeze-dried ground material in a muffle furnace at 550 °C for 1 h. Percent AFDM remaining was determined by subtracting the final AFDM from the initial AFDM and dividing by initial AFDM for each litterbag. We calculated

decomposition rates (k) using the negative exponential decay equation:

$$M_t = M_0 e^{-kt}$$

in which M_t is the mass remaining at time t , M_0 is the initial mass, e is Euler's number, and k is the decomposition rate. Decomposition rates (k) were determined by linear regression of the $\ln$ -transformed %AFDM remaining for each litter type against time (0, 7, 22, and 37 days in stream), with the slope of the equation providing an estimate of the exponential decomposition rate constant ($k \text{ day}^{-1}$, with associated R^2 , p , and standard error values for each regression). We calculated decomposition rate constants for both the entire experiment (days 0–37) and the leaching phase (days 0–7), due to strong differences in decomposition patterns between litter types.

Aquatic invertebrates

Aquatic invertebrates rinsed from litter samples were sieved through a 250- μm net and preserved in 85% ethanol prior to identification. Aquatic insects were identified to family or genus and non-insects were identified to a coarser resolution, typically class (Merritt et al. 2019), using a dissecting microscope. Each taxon was counted to determine the number of individuals per litterbag. Invertebrate taxa were classified into functional feeding groups (FFG), i.e. collector-gatherer, collector-filterer, scraper, shredder, parasite, piercing-herbivore, and predator (Merritt et al. 2019); EPT (Ephemeroptera, Plecoptera, Trichoptera) taxa; and insects/non-insects, to allow comparison of structural and functional differences in communities across litter types.

Data analysis

For all univariate data, Levene's and Shapiro-Wilks tests were used to determine if parametric test assumptions of homogeneity of variance and normality were met. Litter chemistry of catkins and leaves was compared with Student's t -tests (%CT, %C, C:N) and a Welch's test (%N, due to unequal sample variances). We compared the decomposition rates of catkins and leaves using analysis of covariance (ANCOVA) with $\ln$ -transformed %AFDM remaining as the response variable, time (number of days) as a continuous covariate, and litter type (catkin or leaf) as the factor. We interpreted a significant interaction term as indicating differences among slopes (i.e. decomposition rates). Invertebrate total abundance, taxa richness,

Shannon's diversity index (H), and taxa evenness (J ; $H/\ln[\text{richness}]$) were calculated for each litterbag. We used a two-way ANOVA to analyze how community metrics responded to litter type, collection date (as a categorical variable, 1–3), and their interaction. Analyses were performed with the statistical software R (R Core Development Team 2020), using the basic functions `lm()` to run the ANCOVAs, `aov()` for the two-way ANOVAs, and the `ggplot2` package (Wickham 2016) to plot graphics.

Aquatic invertebrate community composition in litterbags was compared between litter types, collection dates (1–3), and their interaction using two-way permutational multivariate analysis of variance of $\log_{10}$ -transformed abundance data (PERMANOVA; Anderson 2017). Differences in community structure were visualized using non-metric multidimensional scaling (NMDS) ordination of $\log_{10}$ -transformed taxa abundance data based on Bray-Curtis distances. Abundance data were $\log_{10}$ transformed for multivariate community analysis to minimize the influence of highly abundant species. We used Pearson correlation coefficients to characterize relationships between NMDS axis scores and community metrics, FFGs, days and %AFDM remaining, considering $r > 0.5$ as indicative of meaningful correlations. We represented these correlations as linear vectors through NMDS ordination space. Community analyses were performed in PC-ORD v.7 (MjM Software, Gleneden Beach, Oregon, USA).

Results

Litter chemistry

Initial chemistry differed between litter types. Catkins had lower CT concentrations than leaves ($t_{(14)} = 8.96$, $p < 0.001$; Fig. 1a), whereas %C was higher in catkins than leaves ($t_{(8)} = 7.36$, $p < 0.001$; Fig. 1b). Percent N and C:N ratios were similar between litter types (%N: $F_{1,4} = 0.05$, $p = 0.84$; C:N: $t_{(8)} = 0.68$, $p = 0.52$); Fig. 1c–d).

Litter decomposition

Percent AFDM decreased between days 0–37 for both catkins ($k = -0.011$, $R^2 = 0.78$, $p < 0.001$) and leaves ($k = -0.012$, $R^2 = 0.55$, $p < 0.001$; Fig. 2). Overall decomposition rates did not differ between litter types (Table 2). However, during the leaching phase (days 0–7) the %AFDM decreased for leaves ($k = -0.061$, $R^2 = 0.98$, $p < 0.001$), but not significantly for catkins ($k = -0.029$, $R^2 = 0.47$, $p = 0.08$). As such, initial leach-

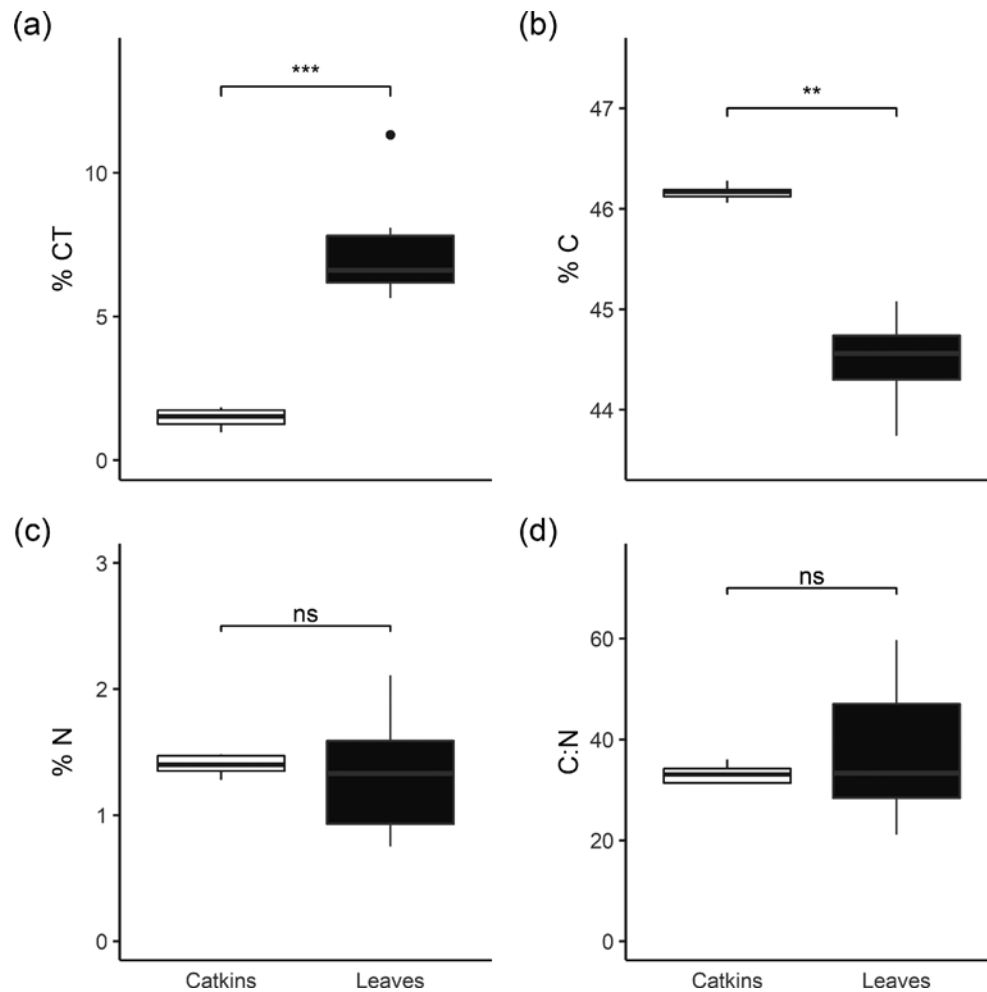


Fig. 1. Initial chemistry of willow catkins and leaves: **(a)** % condensed tannins (CT); **(b)** % carbon (%C); **(c)** % nitrogen (%N); and **(d)** C:N ratios. The lower and upper hinges correspond to the first and third quartiles, the central line indicates the median, the whiskers represent the largest and smallest value no further than 1.5 * the interquartile range from the upper and lower hinges respectively, and the point in panel a indicates an outlier. Asterisks indicate levels of significance: *** $p < 0.0001$, ** $p < 0.001$, ns = not significant at $\alpha = 0.05$.

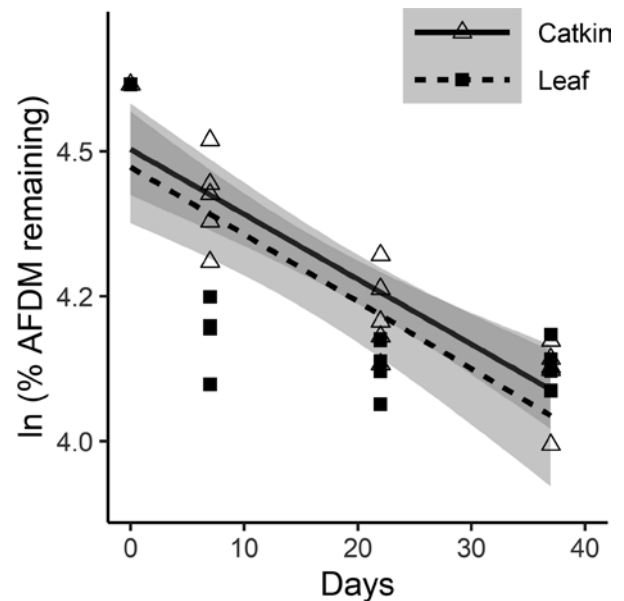


Fig. 2. Leaf litter mass remaining through time for willow catkins and leaves during the entire experiment (days 0–37); willow catkins ($k = -0.011 \pm 0.002$; $R^2 = 0.78$) and leaves ($k = -0.012 \pm 0.002$; $P^2 = 0.55$). Shaded regions show the 95 % confidence intervals.

Table 2. ANCOVA results comparing effects of time (number of days in stream), litter type (catkins, leaves), and their interaction on decomposition rates. Days 0–37 and 0–7 represent the entire experiment and its leaching phase, respectively. Significant interaction terms indicate differences in decomposition rates, which are exponential slope constants and provided in the text. Bold values indicate statistical significance at $\alpha = 0.05$; df, degrees of freedom; F, F-statistic; p , p -value.

Source	Days (0–37)			Days (0–7)		
	df	F	p	df	F	p
Time	1	56.69	<0.001	1	169.41	<0.001
Litter type	1	0.80	0.38	1	37.42	<0.001
Time $\times$ litter type	1	0.01	0.90	1	13.10	0.003
Residuals	34			14		

Table 3. Aquatic invertebrate taxa and functional feeding groups (FFG) collected from willow catkin and leaf litterbags ($n = 15$), including total abundance (individuals per litterbag) and sample frequency (number of litterbags with taxa present, out of 15). CF, collector-filterer; CG, collector-gatherer; PA, parasite; PH, piercer-herbivore; PR, predator; SC, scraper; SH, shredder.

Major taxon	Family	Genus	FFG	Abundance		Samples	
				Catkin	Leaf	Catkin	Leaf
Coleoptera	Dytiscidae		PR	1	1	1	1
	Lampyridae		PR	0	1	0	1
Diptera	Chironomidae		CG	144	78	15	15
	Dixidae	<i>Dixa</i>	CG	0	1	0	1
	Empididae	<i>Clinocera</i>	PR	12	4	6	4
	Psychodidae	<i>Maruina</i>	SC	1	0	1	0
	Simuliidae	<i>Simulium</i>	CF	1	0	1	0
	Pediciidae	<i>Dicranota</i>	PR	1	0	1	0
Ephemeroptera	Baetidae	<i>Baetis</i>	CG	1	0	1	0
	Leptophlebiidae	<i>Neoleptophlebia</i> / <i>Paraleptophlebia</i>	CG	69	57	14	14
Plecoptera	Capniidae	<i>Mesocapnia</i>	SH	37	28	11	10
	Nemouridae	<i>Malenka</i>	SH	271	67	15	15
Trichoptera	Hydroptilidae	<i>Hydroptila</i>	PH	105	59	11	9
	Lepidostomatidae	<i>Lepidostoma</i>	SH	21	6	7	5
	Limnephilidae		SH	28	18	6	6
Copepoda			CF	177	4	12	3
Ostracoda			CG	546	100	15	14
Trombidiformes			PA	22	27	8	10
Oligochaeta			CG	1	3	1	2
Gastropoda			SC	4	5	3	3

ing decomposition rates in the first seven days were significantly faster for leaves compared to catkins, as shown by a significant interaction between litter type and time (Table 2).

Aquatic invertebrates

A total of 20 invertebrate taxa were identified from catkin and leaf litterbags. The most abundant taxa were ostracods, *Malenka* sp. stoneflies, and chironomids (Table 3). *Malenka*, chironomids, ostracods, and leptophlebiid mayflies were the most frequent taxa, occurring in 28–30 of 30 litterbags. As such, collec-

tor-gatherers (represented by ostracods, chironomids, and leptophlebiids) were the most common FFG, followed by shredders including *Malenka*.

Total invertebrate abundance (individuals per litterbag), taxa evenness, and diversity were higher in catkin litterbags compared to leaf litterbags ($p < 0.001$; Table 4; Fig. 3a–c), whereas taxa richness did not differ between litter types ($p = 0.13$; Table 4; Fig. 3d). The effects of collection date and litter type did not interact for any community metric, indicating comparable temporal changes in catkins and leaves ($p > 0.05$; Table 4).

Table 4. Results of two-way ANOVA and PERMANOVA tests comparing effects of collection date (day 7, 22, 37), litter type (catkins, leaves), and their interaction. Bold values indicate statistical significance at $\alpha = 0.05$; df, degrees of freedom; F, F-statistic; p , p -value.

Factor	df	Two-way ANOVA								PERMANOVA	
		Abundance		Richness		Evenness		Shannon		Composition	
		F	p	F	p	F	p	F	p	F	p
Collection date (CD)	2	0.29	0.75	6.04	0.01	3.35	0.05	3.03	0.07	6.34	<0.001
Litter type (LT)	1	27.09	<0.001	2.43	0.13	18.62	<0.001	27.47	<0.001	6.49	<0.001
CD $\times$ LT	2	0.25	0.78	0.35	0.71	1.71	0.20	2.46	0.11	1.13	0.31
Residuals	24										

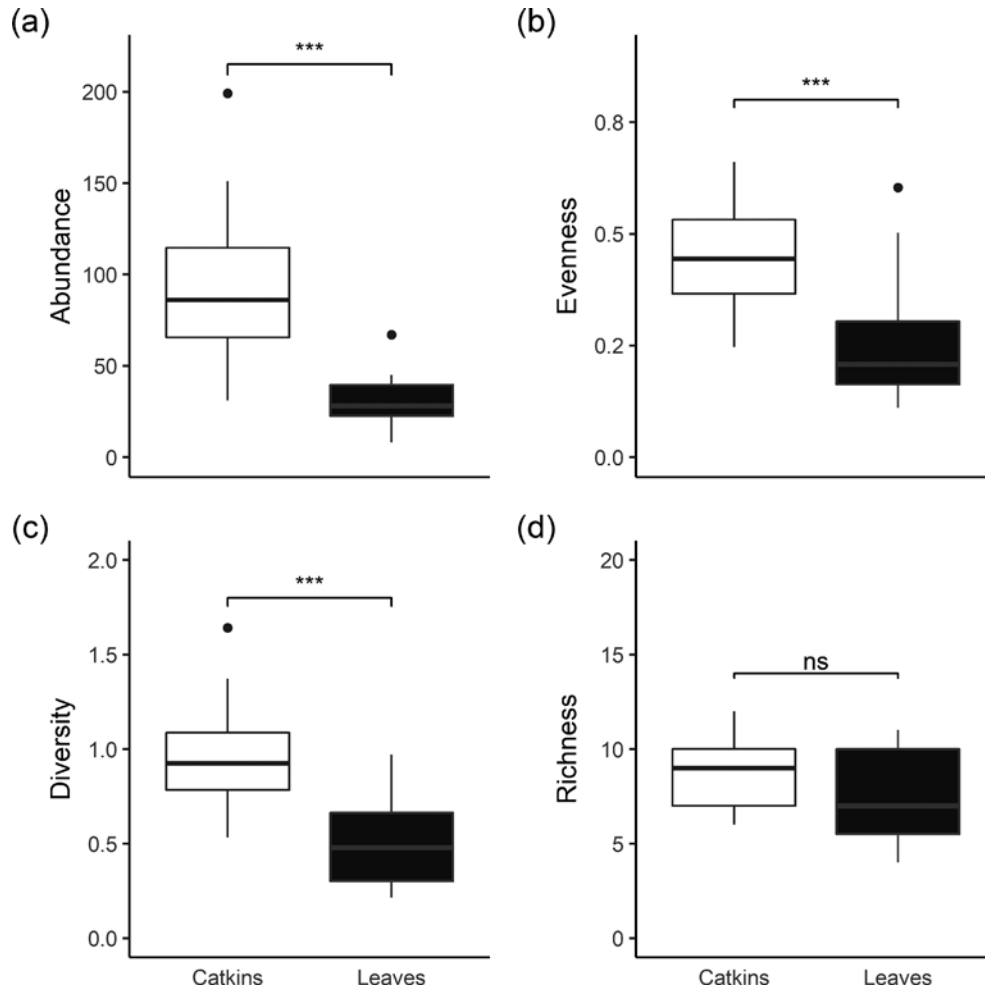


Fig. 3. Invertebrate community metrics in catkin and leaf litterbags ($n = 15$ per type): (a) total abundance (individuals per litterbag); (b) taxa evenness (J' ; $H'/\ln[\text{richness}]$); (c) diversity (Shannon's H'); and (d) taxa richness (taxa per litterbag). The lower and upper hinges correspond to the first and third quartiles, the central line indicates the median, the whiskers represent the largest and smallest value no further than $1.5 \times$ the interquartile range from the upper and lower hinges respectively, and the points in panels a–c indicate outliers. Asterisks indicate levels of significance: *** $p < 0.0001$, ** $p < 0.001$, ns = not significant (at $\alpha = 0.05$).

Community composition differed between litter types and among collection dates, but these factors did not interact (two-way PERMANOVA, Table 4). NMDS of the 20 aquatic macroinvertebrate taxa from 30 samples resulted in a 3-dimensional ordina-

tion (Fig. 4). Samples were distributed along NMDS1 in relation to collection date, NMDS2 differentiated between communities from catkins and leaves, and NMDS3 showed no association with either of these factors (Fig. 4). Pearson correlations with NMDS2

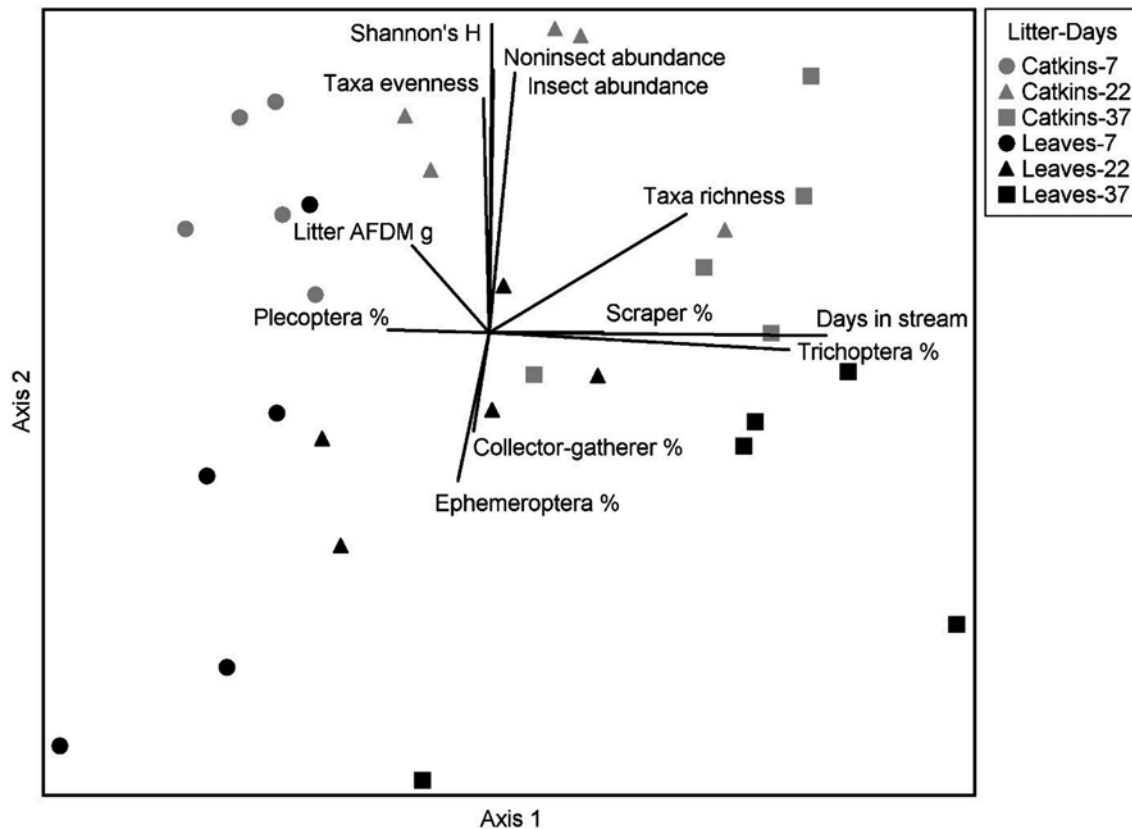


Fig. 4. NMDS ordination of $\log_{10}$ -transformed invertebrate taxa abundance in catkin and leaf litter samples (NMDS axes 1–2, axis 3 not shown, 3D stress = 0.096). Line vectors indicate the direction and magnitude of Pearson correlations ($r > |0.5|$) between NMDS axis scores and litter ash-free dry mass remaining, days in stream, community metrics, functional feeding groups, and different types of invertebrates (Ephemeroptera, Plecoptera, Trichoptera (EPT) taxa, and insects/non-insects).

show that catkins were associated with higher invertebrate abundance, taxa evenness, and diversity than leaves (Fig. 4). Correlations with NMDS2 also indicate that leaf litter had a greater proportion of leptohebiid mayflies and other collector-gathering taxa (Ostracoda and Chironomidae) than catkin litter. Correlations with NMDS1 indicate that stoneflies (which were shredders) were proportionately more abundant on the first collection date (day 7), whereas caddisflies (including piercers and shredders) and scrapers (which were largely represented by snails) were more abundant on the final collection date (day 37), regardless of litter type (Fig. 4; Table 3).

Discussion

Terrestrial leaves are an undisputed energy source that support headwater streams, and we contribute new evidence that reproductive litters including willow catkins also make important contributions to these ecosystems—during a season in which leaf litter is

scarce. We identified morphological as well as chemical differences that distinguished between catkin and leaf litters, potentially supporting their distinct, complementary seasonal roles in stream food webs.

Flower and leaf litters decompose at different rates

Leaves showed 3-fold higher condensed tannin (CT) concentrations than catkins, indicating variation in lability among willow litter types, and partially supporting hypothesis 1. Although decomposition is usually negatively correlated with CT concentrations (LeRoy et al. 2007; Schweitzer et al. 2008), we observed comparable decomposition rates in catkin and leaf litters, contradicting the faster catkin decomposition predicted by hypothesis 2. Both catkin and leaf litters are classified as fast-decomposing according to Petersen & Cummins (1974), however, had the study been longer, different patterns might have been found. In our study, autumn-shed willow leaves lost nearly half their mass during the first week of instream leaching,



Fig. 5. A *Lepidostoma* sp. (Lepidostomatidae: Trichoptera) larva (approximately 6 mm) collected from a *Salix sitchensis* (willow) flower litterbag, within a case constructed from willow flowers.

then mass loss was minimal for the rest of the experiment. This temporal pattern fits the classic exponential decay model used in leaf litter decomposition studies (Webster & Benfield 1986). In contrast, catkins had lower leaching losses and showed a steadier rate of decomposition throughout the experiment. The timing of catkin inputs to the detrital pool and their steadier decomposition rates may be seasonally important to stream communities that rely on allochthonous energy sources. Our findings of lower defense compounds (i.e. CT) and slower leaching for catkins suggest that these structurally complex reproductive inputs may provide important OM inputs to temperate streams, as also observed in the detrital pool of early successional glacial streams (Flory & Milner 1999).

The 2:1 female to male ratio of *Salix* willows in our study area (Che-Castaldo et al. 2015) and the tendency of female plants to grow closer to the stream edge than males (LeRoy et al. 2020b) may alter their contribution to stream OM budgets in the form of leaves, catkins, and woody material. Further studies could build on our findings by measuring the biomass contributions of male and female leaves and catkins in summer

and autumn seasons. Such research could add a seasonal dimension to existing OM processing studies as well as advancing understanding of the effects of plant dioecy on ecosystem functioning.

Litterbags provide a valid method of comparing litter types (Boulton & Boon 1991), but we may have underestimated decomposition rates by using fine mesh bags that restricted access by macroinvertebrates. However, catkins comprise numerous tiny parts that are prone to detachment, and a larger mesh size would have disproportionately increased catkin mass loss due to fragmentation and physical abrasion. In addition, many invertebrate taxa inhabiting Geo-W Creek occur as very small instars in July (i.e. the start of our experiment), facilitating their access to the litterbags (Claeson et al. 2021). Second, our two litter types received different initial drying treatments: catkins were oven-dried at higher temperatures (50 °C) compared to air-dried leaves (28 °C). These differences may have influenced recorded decomposition rates, with air-drying potentially slowing decomposition (Taylor 1998) whereas oven-drying litter can increase leaching (Canhoto & Graça 1996). However, both litter types

qualitatively appeared very dry at the start of the study, and oven-dried catkins had lower leaching losses than leaves.

Flower litters support abundant, diverse aquatic invertebrate communities

Aquatic invertebrate communities colonizing catkins were more abundant, taxonomically even and diverse than those colonizing leaves, supporting hypothesis 3, which predicted that community structure would differ between the two litter types. Invertebrates may be attracted to a wide range of chemical and morphological litter traits, including physical characteristics such as the complex structure of willow catkins, each of which comprises numerous flowers (Fisher 1928) and may provide an attractive habitat for colonization. Accordingly, we observed several instances of Trichoptera larvae including *Lepidostoma* sp. incorporating flower pieces into their cases (Fig. 5). Most research examining the role of aquatic invertebrates in OM decomposition has focused on invertebrate species traits and litter chemistry (Tank et al. 2010; Graça 2001), and better characterization of litter morphology could determine its influence on detrital systems.

Flower litters may provide key seasonal energy inputs to headwater streams

Despite a notable research focus on autumn-shed leaves, streams are enriched with terrestrially derived OM in multiple seasons, including fruits and flowers which are important nutrient sources in both tropical (Larned 2000; Schmitt & Perfecto 2020) and temperate systems (Scarsbrook et al. 2001; Medina-Villar et al. 2015). We found that nearly 2.5-fold more invertebrates colonized willow catkins compared to leaves, indicating that reproductive litters may provide seasonal energy inputs that support stream biota in early summer. Invertebrate production in headwater streams can be limited by the availability of palatable detrital resources (Wallace et al. 1997; Chadwick & Huryn 2007), especially after displacement of autumn-shed litter by high flows (Hall et al. 2001). The contribution of reproductive litters such as willow catkins to the detrital pool in early summer may thus be a critical seasonal driver of secondary production in temperate headwater streams. Further research is needed to quantify the timing and extent of flower litter contributions to the total annual OM budget of stream ecosystems.

Conclusions

Although our study advances understanding of interactions between summer flower inputs and aquatic invertebrate communities, other aspects of summer reproductive inputs to streams remain unclear, including litter morphological traits, microbial colonization patterns, dissolved organic carbon fluxes, and macro-nutrient pulses. The potential trophic effects of these summer litter pulses require further investigation as climate change alters plant phenology, litter chemistry, litter inputs, and stream flow regimes (Kominoski et al. 2013) all of which interact to influence ecosystem functioning (Salinas et al. 2018; Palmer & Ruhi 2019). Understanding the influences of emerging asynchronous patterns in aquatic–terrestrial systems requires a robust examination of the cascading effects of litterfall and phenophase timing on aquatic invertebrates and the higher trophic levels that depend on them.

Authors' contributions

IJG and CJL designed the study. IJG, AMFM, SMC, JMRH, and CJL assisted with fieldwork. IJG did the lab work. AMFM and SMC provided expertise on aquatic invertebrate identification. IJG, AMFM, SMC, JMRH, and CJL analyzed data. IJG, SHH, SMC, JMRH, and CJL drafted the manuscript and all others edited. CJL was the lead Principal Investigator and supervisor on the research grant.

Data availability statement

Data are available at Github: https://github.com/carrileroy/Garthwaite_et_al_FAL

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References

- Abelho, M. (2001). From litterfall to breakdown in streams: A review. *TheScientificWorldJournal*, 1, 656–680. <https://doi.org/10.1100/tsw.2001.103>

- Anderson, M. J. (2017). Permutational Multivariate Analysis of Variance (PERMANOVA). In N. Balakrishnan, T. Colton, B. Everitt, W. Piegorisch, F. Ruggeri & J. L. Teugels (Eds.), *Wiley StatsRef: Statistics Reference Online* (pp. 1–55). <https://doi.org/10.1002/9781118445112.stat07841>
- Benfield, E., Fritz, K., & Tiegs, S. (2017). Leaf-litter breakdown. In F. R. Hauer, & G. Lamberti (Eds.), *Methods in stream ecology, volume 2: Ecosystem function* (3rd ed., pp. 71–82). Burlington, MA: Elsevier. <https://doi.org/10.1016/B978-0-12-813047-6.00005-X>
- Blackman, T. E. (2014). Life-history strategies of rainbow trout (*Oncorhynchus mykiss*) across a volcanic disturbance gradient at Mount St. Helens, Washington. MSc thesis, Oregon State University, Corvallis.
- Boulton, A., & Boon, P. (1991). A review of methodology used to measure leaf litter decomposition in lotic environments: Time to turn over an old leaf? *Marine and Freshwater Research*, 42(1), 1–43. <https://doi.org/10.1071/MF9910001>
- Canhoto, C., & Graça, M. A. (1996). Decomposition of *Eucalyptus globulus* leaves and three native leaf species (*Alnus glutinosa*, *Castanea sativa* and *Quercus faginea*) in a Portuguese low order stream. *Hydrobiologia*, 333(2), 79–85. <https://doi.org/10.1007/BF00017570>
- Chadwick, M. A., & Huryn, A. D. (2007). Role of habitat in determining macroinvertebrate production in an intermittent-stream system. *Freshwater Biology*, 52(2), 240–251. <https://doi.org/10.1111/j.1365-2427.2006.01679.x>
- Che-Castaldo, C., Crisafulli, C. M., Bishop, J. G., & Fagan, W. F. (2015). What causes female bias in the secondary sex ratios of the dioecious woody shrub *Salix sitchensis* colonizing a primary successional landscape? *American Journal of Botany*, 102(8), 1309–1322. <https://doi.org/10.3732/ajb.1500143>
- Chomel, M., Guittonny-Larchevêque, M., Fernandez, C., Gallet, C., DesRochers, A., Paré, D., . . . Baldy, V. (2016). Plant secondary metabolites: A key driver of litter decomposition and soil nutrient cycling. *Journal of Ecology*, 104(6), 1527–1541. <https://doi.org/10.1111/1365-2745.12644>
- Claeson, S. M., LeRoy, C. J., Finn, D. S., Stancheva, R. H., & Wolfe, E. R. (2021). Variation in riparian and stream assemblages across the primary succession landscape of Mount St. Helens, USA. *Freshwater Biology*, 66(5), 1002–1017. <https://doi.org/10.1111/fwb.13694>
- Compson, Z. G., Adams, K. J., Edwards, J. A., Maestas, J. M., Whitham, T. G., & Marks, J. C. (2013). Leaf litter quality affects aquatic insect emergence: Contrasting patterns from two foundation trees. *Oecologia*, 173(2), 507–519. <https://doi.org/10.1007/s00442-013-2643-6>
- del Moral, R., & Jones, C. (2002). Vegetation development on pumice at Mount St. Helens, USA. *Plant Ecology*, 162(1), 9–22. <https://doi.org/10.1023/A:1020316503967>
- Fisher, M. J. (1928). The morphology and anatomy of the flowers of the Salicaceae I. *American Journal of Botany*, 15(5), 307–326. <https://doi.org/10.1002/j.1537-2197.1928.tb04904.x>
- Flory, E. A., & Milner, A. M. (1999). Influence of riparian vegetation on invertebrate assemblages in a recently formed stream in Glacier Bay National Park, Alaska. *Journal of the North American Benthological Society*, 18(2), 261–273. <https://doi.org/10.2307/1468464>
- Graça, M. A. S. (2001). The role of invertebrates on leaf litter decomposition in streams – A review. *International Review of Hydrobiology*, 86(4–5), 383–393. [https://doi.org/10.1002/1522-2632\(200107\)86:4/53.0.CO;2-D](https://doi.org/10.1002/1522-2632(200107)86:4/53.0.CO;2-D)
- Graça, M. A. S., Ferreira, V., Canhoto, C., Encalada, A. C., Guerrero-Bolaño, F., Wantzen, K. M., & Boyero, L. (2015). A conceptual model of litter breakdown in low order streams. *International Review of Hydrobiology*, 100(1), 1–12. <https://doi.org/10.1002/iroh.201401757>
- Griffiths, N. A., Tank, J. L., Royer, T. V., Rosi-Marshall, E. J., Whiles, M. R., Chambers, C. P., . . . Evans-White, M. A. (2009). Rapid decomposition of maize detritus in agricultural headwater streams. *Ecological Applications*, 19(1), 133–142. <https://doi.org/10.1890/07-1876.1>
- Hagerman, A. E., & Butler, L. G. (1989). Choosing appropriate methods and standards for assaying tannin. *Journal of Chemical Ecology*, 15(6), 1795–1810. <https://doi.org/10.1007/BF01012267>
- Hall, R. O., Jr., Likens, G. E., & Malcom, H. M. (2001). Trophic basis of invertebrate production in 2 streams at the Hubbard Brook Experimental Forest. *Journal of the North American Benthological Society*, 20(3), 432–447. <https://doi.org/10.2307/1468040>
- Kominoski, J. S., Shah, J. J. F., Canhoto, C., Fischer, D. G., Gilling, D. P., González, E., . . . Tiegs, S. D. (2013). Forecasting functional implications of global changes in riparian plant communities. *Frontiers in Ecology and the Environment*, 11(8), 423–432. <https://doi.org/10.1890/120056>
- Larned, S. T. (2000). Dynamics of coarse riparian detritus in a Hawaiian stream ecosystem: A comparison of drought and post-drought conditions. *Journal of the North American Benthological Society*, 19(2), 215–234. <https://doi.org/10.2307/1468066>
- LeRoy, C. J., Fischer, D. G., Schweitzer, J. A., & Bailey, J. K. (2020a). Aphid gall interactions with forest tree genotypes influence leaf litter decomposition in streams. *Forests*, 11(2), 182. <https://doi.org/10.3390/f11020182>
- LeRoy, C. J., Ramstack Hobbs, J. M., Claeson, S. M., Moffett, J., Garthwaite, I., Criss, N., & Walker, L. (2020b). Plant sex influences aquatic–terrestrial interactions. *Ecosphere*, 11(1), e02994. <https://doi.org/10.1002/ecs2.2994>
- LeRoy, C. J., Whitham, T. G., Wooley, S. C., & Marks, J. C. (2007). Within-species variation in foliar chemistry influences leaf-litter decomposition in a Utah river. *Journal of the North American Benthological Society*, 26(3), 426–438. <https://doi.org/10.1899/06-113.1>
- Medina-Villar, S., Castro-Díez, P., Alonso, A., Cabra-Rivas, I., Parker, I. M., & Pérez-Corona, E. (2015). Do the invasive trees, *Ailanthus altissima* and *Robinia pseudoacacia*, alter litterfall dynamics and soil properties of riparian ecosystems in Central Spain? *Plant and Soil*, 396(1–2), 311–324. <https://doi.org/10.1007/s11104-015-2592-4>
- Merritt, R., Cummins, K., & Berg, M. (2019). *An Introduction to the Aquatic Insects of North America* (5th ed.). Dubuque, IA: Kendall Hunt.
- Milner, A. M., Fastie, C. L., Chapin, F. S., Engstrom, D. R., & Sharman, L. C. (2007). Interactions and linkages among ecosystems during landscape evolution. *Bioscience*, 57(3), 237–247. <https://doi.org/10.1641/B570307>
- Palmer, M., & Ruhi, A. (2019). Linkages between flow regime, biota, and ecosystem processes: Implications for river restoration. *Science*, 365(6459), eaaw2087. <https://doi.org/10.1126/science.aaw2087>
- Petersen, R. C., & Cummins, K. W. (1974). Leaf processing in a woodland stream. *Freshwater Biology*, 4(4), 343–368. <https://doi.org/10.1111/j.1365-2427.1974.tb00103.x>

- Porter, L. J., Hrstich, L. N., & Chan, B. G. (1985). The conversion of procyanidins and prodelphinidins to cyanidin and delphinidin. *Phytochemistry*, 25(1), 223–230. [https://doi.org/10.1016/S0031-9422\(00\)94533-3](https://doi.org/10.1016/S0031-9422(00)94533-3)
- Pray, C. L., Nowlin, W. H., & Vanni, M. J. (2009). Deposition and decomposition of periodical cicadas (Homoptera: Cicadidae: Magicicada) in woodland aquatic ecosystems. *Journal of the North American Benthological Society*, 28(1), 181–195. <https://doi.org/10.1899/08-038.1>
- R Core Development Team (2020). *R: A language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing. <https://www.R-project.org/>
- Rodriguez-Cabal, M. A., Barrios-Garcia, M. N., Rudman, S. M., McKown, A. D., Sato, T., & Crutsinger, G. M. (2017). It is about time: Genetic variation in the timing of leaf-litter inputs influences aquatic ecosystems. *Freshwater Biology*, 62(2), 356–365. <https://doi.org/10.1111/fwb.12872>
- Rosi-Marshall, E. J., Tank, J. L., Royer, T. V., Whiles, M. R., Evans-White, M., Chambers, C., . . . Stephen, M. L. (2007). Toxins in transgenic crop byproducts may affect headwater stream ecosystems. *Proceedings of the National Academy of Sciences of the United States of America*, 104(41), 16204–16208. <https://doi.org/10.1073/pnas.0707177104>
- Salinas, M. J., Casas, J. J., Rubio-Ríos, J., López-Carrique, E., Ramos-Miras, J. J., & Gil, C. (2018). Climate-driven changes of riparian plant functional types in permanent headwater streams: Implications for stream food webs. *PLoS One*, 13(6), e0199898. <https://doi.org/10.1371/journal.pone.0199898>
- Scarsbrook, M. R., Quinn, J. M., Halliday, J., & Morse, R. (2001). Factors controlling litter input dynamics in streams draining pasture, pine, and native forest catchments. *New Zealand Journal of Marine and Freshwater Research*, 35(4), 751–762. <https://doi.org/10.1080/00288330.2001.9517040>
- Schmitt, L., & Perfecto, I. (2020). Who gives a flux? Synchronous flowering of *Coffea arabica* accelerates leaf litter decomposition. *Ecosphere*, 11(7), e03186. <https://doi.org/10.1002/ecs2.3186>
- Schweitzer, J. A., Madritch, M. D., Bailey, J. K., LeRoy, C. J., Fischer, D. G., Rehill, B. J., . . . Whitham, T. G. (2008). From genes to ecosystems: The genetic basis of condensed tannins and their role in nutrient regulation in a *Populus* model system. *Ecosystems (New York, N. Y.)*, 11(6), 1005–1020. <https://doi.org/10.1007/s10021-008-9173-9>
- Spänhoff, B., & Meyer, E. I. (2004). Breakdown rates of wood in streams. *Journal of the North American Benthological Society*, 23(2), 189–197. [https://doi.org/10.1899/0887-3593\(2004\)0232.0.CO;2](https://doi.org/10.1899/0887-3593(2004)0232.0.CO;2)
- Tank, J. L., Rosi-Marshall, E. J., Griffiths, N. A., Entekin, S. A., & Stephen, M. L. (2010). A review of allochthonous organic matter dynamics and metabolism in streams. *Journal of the North American Benthological Society*, 29(1), 118–146. <https://doi.org/10.1899/08-170.1>
- Taylor, B. R. (1998). Air-drying depresses rates of leaf litter decomposition. *Soil Biology & Biochemistry*, 30(3), 403–412. [https://doi.org/10.1016/S0038-0717\(97\)00126-0](https://doi.org/10.1016/S0038-0717(97)00126-0)
- Vannote, R. L., Minshall, G. W., Cummins, K. W., Sedell, J. R., & Cushing, C. E. (1980). The River Continuum Concept. *Canadian Journal of Fisheries and Aquatic Sciences*, 37(1), 130–137. <https://doi.org/10.1139/f80-017>
- Wallace, J. B., Eggert, S. L., Meyer, J. L., & Webster, J. R. (1997). Multiple trophic levels of a forest stream linked to terrestrial litter inputs. *Science*, 277(5322), 102–104. <https://doi.org/10.1126/science.277.5322.102>
- Webster, J. R., & Benfield, E. F. (1986). Vascular plant breakdown in freshwater ecosystems. *Annual Review of Ecology and Systematics*, 17(1), 567–594. <https://doi.org/10.1146/annurev.es.17.110186.003031>
- Wickham, H. (2016). *ggplot2: Elegant graphics for data analysis* (pp. 1–260). New York: Springer. <https://doi.org/10.1007/978-3-319-24277-4>
- Wink, M. (2010). Introduction: Biochemistry, physiology and ecological functions of secondary metabolites. In M. Wink (Ed.), *Annual plant reviews volume 40: Biochemistry of plant secondary metabolism*. (2nd ed., pp. 1–19). Oxford, UK: Wiley-Blackwell. <https://doi.org/10.1002/9781444320503.ch1>

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