Effects of Deployment Period on Decomposition and Colonization of Leaf Litter of Differing Quality by Invertebrates

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Abstract

Detritivorous invertebrates play major roles in organic matter processing and nutrient cycling in headwater streams. In this study, three common leaf species in upland Kenyan streams (Vernonia myriantha, Syzygium cordatum and the exotic *Eucalyptus globulus) were used to determine the influence of deployment period (14 vs 28 days) on relative decomposition rates and colonization by detritivorous invertebrates in headwater streams of the Nzoia River Basin. Leaf decomposition rates were measured by placing 216 litterbags made of coarse- and fine-mesh in six streams draining forested ($n =3$) and agricultural ($n = 3$) land-use during the dry months of February-March 2020. For each stream, physico-chemical water characteristics and habitat quality were determined. Measurements of electrical conductivity, pH, temperature, dissolved oxygen concentration and salinity were performed in situ using portable probes. There were no major differences in physical and chemical characteristics between forested and agricultural streams, except for significantly higher canopy cover ($p < 0.05$) in forested streams, and electrical conductivity and mean water temperature in agricultural streams. Decomposition rates were faster during the first 2 weeks (day 14), and differences between fine- and coarse-mesh litterbags were significant for* Vernonia *and* Syzygium, *but not for* Eucalyptus. *After 14 days, differences between microbial and shredder + microbial breakdown of leaves were clearer than after 28 days, suggesting that short deployment periods (14 days) are enough to establish relative roles of shredders and microbes in leaf litter decomposition experiments in tropical streams. There were inter-specific differences in colonization rates of the leaves by detritivores (shredders) with Vernonia having the highest number of shredder taxa and abundance followed by Syzygium and Eucalyptus. However, there were minimal differences in taxon richness and abundance of shredders and non-shredders between day 14 and day 28. Therefore, this study recommends shorter deployment periods of 14 days rather than long periods of one month or more when studying leaf litter decomposition and colonization by detritivores in tropical streams.*

Keywords*:* Organic Matter Processing, Tropical Streams, Litter Decomposition, Shredders, Microbial Processing

INTRODUCTION

Leaf litter decomposition is the process through which leaf material is broken down progressively into smaller particles (Cotrufo et al., 2010). This process is driven by the chemistry and intrinsic characteristics of leaf litter, activity and availability of detritivorous invertebrates and environmental conditions, such water temperature and availability of nutrients (Tank et al., 2010), as well as the result of physical abrasion by water currents (Graça et al., 2001; Wantzen et al., 2008).

In headwater streams, macroinvertebrate detritivores or shredders play a vital role in leaf litter decomposition by transforming coarse particulate organic matter to fine particulate organic matter (Vannote et al., 1980; Graça, 2017). By doing so, they release nutrients and provide food resources to other invertebrates (collectors) and microbiota (Patrick, 2013; Masese et al., 2014a, b). In low order streams, food webs are highly dependent on allochthonous energy flow through detrital pathways. Because of riparian shading that limits primary production in low order streams, allochthonous energy is usually more important than the amount generated through primary production (Vannote et al., 1980; Graça et al., 2015).

Determining the relative roles of microorganisms (microbes) and detritivorous invertebrates (shredders) in the decomposition of leaf litter is important as a measure of the ecological integrity and functioning of low order streams (Gessner and Chauvet, 2002; Young et al., 2008). Several factors determine the participation of microbes and shredders in the decomposition of leaf litter in aquatic ecosystems, including mean water temperature, nutrient concentrations (mainly the nitrate species), abundance, biomass and diversity of shredders and the quality of leaf litter (e.g., nutrient, lignin content and toughness) (Abelho, 2001; Boyero et al., 2021). The breakdown rates of leaf litter tend to be faster in tropical than in temperate streams owing to the higher mean water temperature and diversity of leaf-associated microbiota in tropical regions (Irons et al., 1994; Dobson et al., 2002; Gonçalves et al., 2006; Boyero et al., 2016). However, some tropical streams have slow decomposition rates (Ferreira et al., 2012) that is attributed to adverse environmental conditions and poor quality of tree species (González & Grácá, 2003; Graça et al., 2015).

Another important consideration in determining the relative rates of decomposition of leaf litter of differing quality is the deployment period. During the initial stages of litter decomposition, microbes mobilize nutrients from the environment to increase the amount available from the litter with a net increase in microbial pools of N and P (i.e., nutrient immobilization), but in later stages, the microbes lose the nutrients (i.e., nutrient mineralization) (Webster and Benfield, 1986; Manzoni et al., 2010). Because of the different rates of nutrient immobilization and mineralization by different leaf species depending on their quality (e.g., C: N: P stoichiometry and lignin content), it takes different periods of time for the leaves to be colonized by microbiota and detritivores (Webster and Benfield, 1986; Wantzen et al., 2008). If the leaves are of high quality, with a low C: N and/or C: P ratio and low concentration of inhibitory compounds and chemicals (Zhang et al., 2019), they will be colonized faster and decomposition rates will be higher compared with poorer quality leaves. Therefore, if the deployment period is short, high-quality leaf litter may decompose faster than poor quality leaf litter because of faster colonization by microbiota and invertebrates. However, longer deployment periods may minimize differences in decomposition rates among leaves of different quality because differences in nutrient immobilization and mineralization would be reduced with time.

In this study, we aimed at assessing the effects of the deployment period on rates of decomposition and colonization by invertebrates of leaf litter differing in quality. We hypothesized that high-quality leaves would be colonized faster by microbes and invertebrates and hence display faster decomposition rates if deployment periods are shorter. However, with longer deployment periods the differences in decomposition rates would be reduced because nutrient immobilization would diffuse nutrient limitation of microbes and increase colonization by detritivorous invertebrates.

MATERIALS AND METHODS

Study area

This study was done during the dry months of February – March 2020, in six headwater streams of Nzoia River Basin (NRB), Lake Victoria Basin (LVB), Western Kenya (Figure 1). Three streams (Kipsinende, Mlango and Seger) were in agricultural areas and the other three (Sabor, Kipkarren and Chepkoilel) in forested land. The Nzoia River has its origin in the southeast part of Mt. Elgon and the Western slopes of Cheranganyi Hills (Nyadawa & Mwangi, 2010). the upper reaches are characterized by a tropical humid climate, having a mean annual rainfall ranging from 900 mm to 2200 mm and temperature ranges of 13 °C to 25 °C which are influenced by elevation (Wabusya et al., 2015). The catchment exhibits a bimodal rainfall pattern characterized by a period of long rains between March and June, and short rains between August and October (Nyadawa & Mwangi, 2010). The upper NRB has experienced rapid human population growth over the years (Wabusya et al., 2015). This has exerted pressure on existing natural resources as the demand for food and fibre has increased leading to deforestation, crop farming. Introduction of exotic eucalypt and other tree species along riparian corridors of streams, conversion of wetlands to farmlands and livestock grazing, sand whining from streams and brick-making (Masese et al., 2009). These activities have resulted in channel modification, loss of indigenous riparian vegetation and land degradation. The in-stream water of the Nzoia River also has declined over because of soil erosion and nutrient loading from converted lands (Wabusya et al., 2015; Sitati et al., 2021).

The sites for this study were selected from either forested or agricultural land-uses that were defined as either forested (n = 3) draining catchments with >60% forest cover or agricultural (n = 3) draining catchments with >60% agricultural land-use. At each site, data physical and chemical characteristics of the streams, including stream size variables were collected for comparison of forested and agricultural streams.

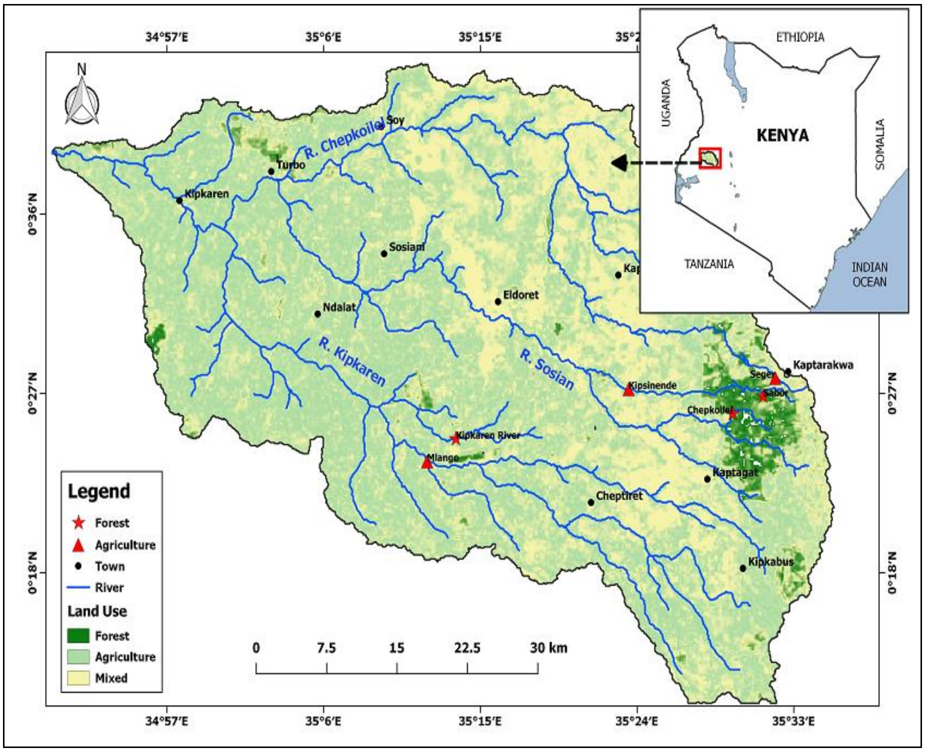

Figure 1: Map of Nzoia River Basin showing the locations of study sites in the six streams considered for this study.

Physical and chemical characteristics of streams

Velocity, water depth, stream width, discharge and per cent canopy cover were determined within a 100 m long stream reach of each stream. Flow velocity was measured by using a velocity plank and the velocity-area method was subsequently used to determine stream discharge (Wetzel & Likens, 2000). Stream width was measured at several points along the river reach using a measuring tape while water depth was measured across the river using a 1 m standard ruler. A YSI multi-probe water quality meter (556 MPS, Yellow Springs Instruments, Ohio, USA) was used to measure temperature (°C), total dissolved solids (TDS), dissolved oxygen concentration (DO), electrical conductivity (EC), pH, and salinity *in situ*.

Leaf litter decomposition experiment

A leaf-litter decomposition experiment was conducted involving leaves from three common riparian tree species: an indigenous tough species *Syzygium cordatum* (Family *Myrtaceae*), a soft indigenous species, *Vernonia myriantha* (Family *Asteraceae*) and a tough exotic species *Eucalyptus globulus* Labill (Family *Myrtaceae*). *V. myriantha* (hereafter *Vernonia*) represented high-quality leaves, whereas *S. cordatum* (hereafter *Syzygium*) and *E. globulus* (hereafter *Eucalyptus*) leaves are tough and smooth and were of poorer quality than *Vernonia* leaves. Recently abscised leaves of the three species were collected and air-dried for two weeks under a shade at room temperature until a constant mass was attained. Four (4) g of each species were then weighed and put into two litterbag types: fine-mesh (0.5 mm mesh size) and coarse-mesh (10-mm mesh) size litterbags. Fine-mesh litterbags were used to exclude macroinvertebrate access and allow only microbial leaf decomposition whereas coarse-mesh litterbags were used to allow both macroinvertebrate- and microbial-mediated processing of leaf litter.

A total of 228 litterbags were used. Of these, 216 litterbags were divided into 3 replicates per litterbag type (fine- vs. coarse-mesh) per leaf species (*Vernonia*, *Syzygium* and *Eucalyptus*) in each site (3 forested and 3 agricultural), for two deployment periods (Day 14 and Day 28). The extra 12 litterbags were used to determine the initial ash-free-dry mass (AFDM) conversion for each leaf species (4 replicates each).

Before deployment, the litterbags were tied along nylon lines to secure them and avoid any overlaps, as well as for easy retrieval. The litterbags were finally deployed on March 18, 2020, at the 6 study sites by attaching the nylon lines to metal stakes that were hammered into the streambed. Litterbags were set far apart to prevent overlapping and transfer of macroinvertebrates colonizing the bags from one bag to the other. The fine- and coarse- litterbags were retrieved two times on the 14th and 28th days to study macroinvertebrates colonization.

During retrieval, litterbags were carefully enclosed in a 300 mm mesh sampling net and placed in zip-lock plastic bags to avoid loss of invertebrates and leaf fragments. After each collection, samples were placed in plastic bags, preserved in 5% formalin solution and stored in a cooler box for transportation to the laboratory for processing. In the laboratory, litterbags were gently rinsed with tap water and sieved with a 250 mm mesh-size sieve to isolate macroinvertebrates from leaf litter. To obtain the remaining dry mass, litterbag leaf fragments were rinsed and oven-dried at 105°C for 48 hours, then ashed at 550°C for 4 hours and reweighed to calculate AFDM. One set of litterbags was lost at Seger, Kipkaren and Kipsinende streams due to human tampering and sedimentation. This reduced the number of analyzed replicates in these streams to five per leaf species per mesh size, but we considered this to be a minor infraction that would not affect the results.

Macroinvertebrate community composition

Macroinvertebrates were collected from coarse-mesh litterbags and preserved in vials in 70% ethanol until enumeration and identification. Macroinvertebrates were identified mainly to the genus level, with the aid of keys in several guides (Day et al., 2002; Merritt et al., 2008). Macroinvertebrates were classified into four major functional feeding groups (FFGs; scrapers, shredders, collectors and predators) according to Dobson et al. (2002), Merritt et al. (2008) and Masese et al. (2014b). Taxonomic richness and abundance of different FFGs were recorded per litterbag per site.

Data analysis

Estimation of leaf decomposition rates was done using an exponential decay model (Boulton & Boon,1991): $Wt = Wo\,e^{-kt}$, where Wt = the remaining AFDM at time t (14 and 28 days); W_0 = initial AFDM, -*k* = decay rate. Ash free dry mass (AFDM) was calculated from organic matter (OM) which was itself determined following the formula by Benfield (1996):

$$\mathrm{OM} = \left(\frac{(\mathrm{DM}_{\mathrm{sample}} - \mathrm{AM}_{\mathrm{sample}})}{\mathrm{DM}_{\mathrm{sample}}}\right) \mathrm{x}\ 100$$

where $\mathrm{DM}_{\mathrm{sample}}$ is the sample dry mass and $\mathrm{AM}_{\mathrm{sample}}$ is the sample ash mass. The OM values were later converted to AFDM using the following equation: $\mathrm{AFDM} = DM\ x\ \%OM$.

Two-sample t-tests were used to compare differences in physical stream characteristics and *in situ* water quality variables between forested and agricultural streams. The structural and functional composition of macroinvertebrate assemblages in coarse-mesh litterbags were described in terms of abundance and taxon richness. Two-sample t-tests were also used to test for differences in total abundance and richness of shredders and non-shredder taxa in coarse-mesh litterbags samples between the two sampling dates (deployment periods).

Differences in decomposition rates (-*k*) were tested using three-way ANOVA whereby leaf species (*Vernonia*, *Syzygium* and *Eucalyptus*), land-use (forested and agricultural), and mesh size (coarse- and fine-mesh), including interactions, were treated as the main factors and leaf litter decomposition rates (-*k*) as the response variable. The analysis was performed separately after 14 and 28 days of deployment. Where there was no significant interaction between land-use and both leaf species and mesh sizes, two-way ANOVA was re-run separately for each land-use with leaf species and mesh size including interactions, as the main factors affecting decomposition rates.

Breakdown rates for coarse-mesh litterbags (k_c) and fine–mesh litterbags (k_f) were calculated separately. The coefficients k_c/k_f were then calculated for each site to determine the effect of excluding potential macroinvertebrate shredders from fine-mesh litterbags on breakdown rates (Gessner & Chauvet, 2002; Masese et al., 2014b). All statistical analyses were performed using Minitab software (Version 18) and PAST software (version 3.21), while figures were created in MS Office Excel (2016).

RESULTS

Physico-chemical water quality parameters

There were minor differences in physical characteristics and water quality between forested and agricultural streams (Table 1). Only per cent canopy cover and electrical conductivity significantly differed between forested and agriculture streams, with forested streams recording higher per cent canopy cover and agricultural streams recording higher electrical conductivity values (Table 1). Differences in mean water temperature were marginal ($p = 0.073$), indicating the potential for warmer agricultural streams.

Leaf litter decomposition

Decomposition rates were faster during the first phase of the experiment (day 14; Figure 2a) compared to the second phase (day 28; Figure 2b). During the first 14 days, there were differences in decomposition rates between fine- and coarse-mesh litterbags in *Vernonia* and *Syzygium,* but not with *Eucalyptus.* During the second phase, differences in decomposition rates between fine- and coarse-mesh litterbags significantly reduced, and only *Eucalyptus* and *Syzygium* showing differences in forested and agricultural streams, respectively.

Trends in leaf litter decomposition rates *(-k*) were the same during day 14 and day 28. V*ernonia* had the fastest decomposition rates in coarse- and fine-mesh litterbags, followed by *Eucalyptus* and *Syzygium* which largely had similar decomposition rates (Figure 2). There was a significant interaction between deployment period and leaf quality on decomposition rates, with the high-quality *Vernonia* decomposing faster than the other two leaf species by Day 14 (Figure 2a). At day 14, *Vernonia* had the highest decomposition rates in both forested and agricultural streams in both coarse- and fine-mesh litterbags, and decomposition rates were faster in coarse- than in fine-mesh litterbags. However, there were no significant differences in decomposition rates of *Eucalyptus and Syzygium* between coarse- and fine-mesh litterbags irrespective of catchment land use and deployment period. At day 28, differences in decomposition rates driven by microbes versus microbes + macroinvertebrate shredders seemed to have disappeared and both coarse- and fine-mesh litterbags having the same decomposition rates for all leaf species.

Differences in decomposition rates were significant between leaf species in both forested and agricultural sites (Table 2). Since there was no significant interaction between land-use and both leaf species and mesh sizes, two-way ANOVA was run separately for each land-use. Interestingly, overall decomposition (after 28 days) rates were not influenced by mesh-size nor land-use, but during the first phase (0-14 days), mesh-size (role of shredders) played a major role in leaf litter decomposition in both forested and agricultural sites, but only forested sites having a significant leaf species × mesh-size interaction (Table 2). In agricultural sites, significant differences were only obtained between leaf species, while differences between mesh sizes were only marginal ($p = 0.06$).

Table 1: Mean (±SD) values for physico-chemical and stream size variables, organic matter and nutrients concentrations in agricultural and forested streams in the headwaters of the Nzoia River Basin, Kenya

Physico-chemical variables	Agriculture	Forest	t-value	*p*-value
Stream characteristics				
% Canopy cover	37.8±6.8	66.4±2.8	4.303	**0.044**
Depth (m)	0.3±0.03	0.4±0.1	4.303	0.687
Width (m)	6.3±0.8	5.5±1.4	12.706	0.529
Velocity (m s^{-1})	0.6±0.1	0.6±0.05	3.182	0.990
Discharge (m^3 s^{-1})	2.1±0.4	2.2±0.4	4.303	0.808
Water quality variables				
Temperature (^{o}C)	16.8±0.6	15.3±1	3.182	0.073
DO (mg L^{-1})	5.0±0.3	6.2±1.7	4.303	0.435
pH	7.1±0.1	7.1±0.01	4.303	0.818
TDS (mg L^{-1})	0.03±0.01	0.02±0.01	12.706	0.983
EC (µS cm^{-1})	56.9±9.2	29.7±1.3	4.303	**0.043**
Salinity (ppt)	0.02±0.01	0.01±0.01	4.303	0.220

NB: p -values in bold are significant at $p \leq 0.05$

Table 2: Results of two-way ANOVA exploring variation in leaf litter decomposition rates (-*k*) with leaf species (*Vernonia*, *Syzygium* and *Eucalyptus*) and by mesh size (coarse- and fine-mesh) during the first 14 days and after 28 days of deployment in agricultural and forested sites. Degrees of freedom (df), sums of squares (SS), *F* - statistic and *p* - values are shown. Significant values at *p* < 0.05 are shown in bold

	Day 0-14				**After 28 days**			
Sources of variation	**df**	**SS**	**F**	***p* -value**	**df**	**SS**	**F**	***p* - value**
Forested sites								
Leaf sp.	2	0.017	26.7	**<0.001***	2	0.0073	39.4	**<0.001***
Mesh size	1	0.002	6.6	**0.013***	1	0.00008	0.87	0.36
Leaf sp.*Mesh size	2	0.004	5.5	**0.007***	2	0.00002	0.09	0.92
Error	52	0.037			37	0.0103		
Agricultural sites								
Leaf sp.	2	0.013	47.9	**<0.001***	2	0.013	17.6	**<0.001***
Mesh size	1	0.001	3.68	0.06	1	0.00001	0.03	0.86
Leaf sp.*Mesh size	2	0.0002	0.89	0.42	2	0.00003	0.04	0.96
Error	47	0.02			47	0.027		

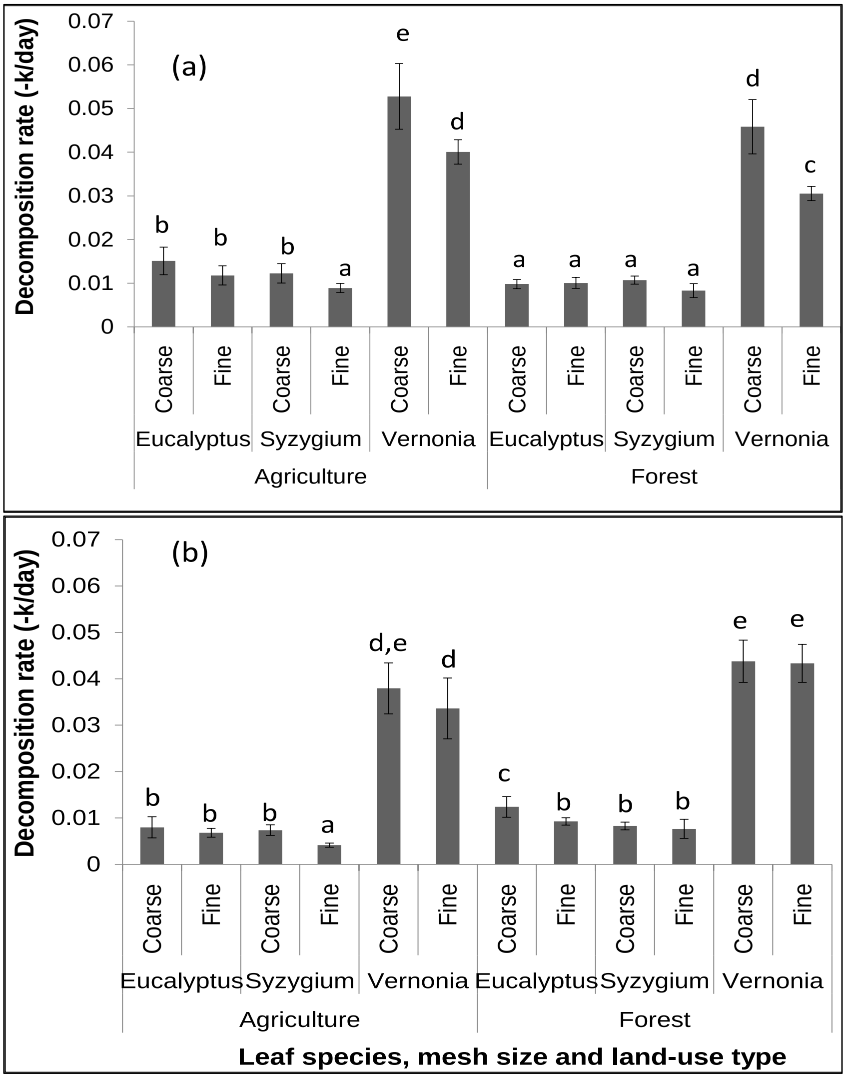

Figure 2: Leaf decomposition rates of *Vernonia*, *Eucalyptus* and *Syzygium* leaves in forested and agricultural streams at day 14 (a) and day 28 (b) in the headwaters of the Nzoia River Basin, Kenya. Different letters on top of bars represent significant *post hoc* differences in means (one-way ANOVA)

The role of shredder in decomposition rates depended on leaf species (litter quality) (Table 3). For all leaf species and during both deployment periods, decomposition rates were higher in coarse- (shredder + microbial decomposition) than in fine-mesh (microbial decomposition) litterbags (kc/kf >1) (Table 3). Except for *Eucalyptus* in forested sites decomposition rates in fine-mesh litterbags were higher on Day 28 than on Day 14, indicating a delay in microbial colonization during the first phase of the decomposition process, but an increase during the second phase (Day 14-28). In forested sites, mean kc/kf coefficient was highest for *Vernonia* during the short (kc/kf = 2.28) and long (kc/kf =1.38) (Table 3) deployment periods. However, in agricultural sites, kc/kf coefficients were highest for *Syzygium* during both the short (kc/kf = 1.48) and long (kc/kf =1.84) deployment periods. *Eucalyptus* had the lowest decomposition rates attributable to shedders (lowest kc/kf values) in both forested and agriculture sites.

Table 3: Means of k_c/k_f coefficients for leaf litter decomposition rates between coarse-mesh (kc) and fine-mesh (kf) litterbags in impacted and reference streams

Leaf Species	Deployment period (days)	k_c/k_f *values*	
		Forested streams	**Agricultural streams**
Vernonia myriantha	14	2.28	1.31
Syzygium cordatum	14	1.36	1.48
Eucalyptus globulus	14	0.98	1.28
Vernonia myriantha	28	1.38	1.28
Syzygium cordatum	28	1.10	1.84
Eucalyptus globulus	28	1.36	1.10

Macroinvertebrate community composition

A total of 1463 macroinvertebrate individuals were collected on day 14 and 1888 on day 28 from the coarse-mesh litterbags. There were no significant differences in the total number of taxa and non-shredder taxa between deployment periods (day 14 and day 28) (Figure 3). Although not statistically significant, there were high richness and abundance of shredders and non-shredders during the first 14 days of the experiment than after 28 days, indicating that prolonged deployment reduces the number of taxa colonizing the leaves. Per leaf species, *Vernonia* recorded the highest number of macroinvertebrates (43.1%), followed by *Syzygium* (31.3%) then *Eucalyptus* (25.6%). *Vernonia* also had the highest number of shredders colonizing the litterbags, followed by *Syzygium* and *Eucalyptus* (Figure 4). There were no significant differences in taxon richness and abundance of shredders colonizing the leaves between day 14 and 28.

DISCUSSION

There were minimal differences in in-situ water quality and habitat quality characteristics between forested and agricultural streams, hence the effect of land use on the patterns of leaf litter decomposition in this study have not been given further consideration. However, there were interesting patterns in decomposition rates of the three species of leaves deployed in agricultural and forested streams for 14 days (short period) and 28 days (long period). As hypothesized, the higher quality *Vernonia* displayed faster rates of both microbial (fine-mesh litterbags) and shredder + microbial (coarse-mesh litterbags) mediated decomposition during the shorter (14 days) deployment periods than the two poor-quality leaves (*Syzygium* and *Eucalyptus*). However, with a long deployment period, interspecific differences in decomposition rates were still maintained, while differences between fine- and coarse-mesh litterbags disappeared irrespective of leaf quality. Therefore, the shorter deployment period was better for discriminating inter-specific differences in decomposition rates due to differences in litter quality.

The trends of leaf litter decomposition on days 14 and 28 were all similar. In the first 14 days, *Vernonia*, which exhibited soft leaves of the highest quality among the three leaves, recorded the highest decomposition rate in coarse- and fine-mesh litterbags. For Vernonia, the decomposition rate was ultimately higher in coarse-mesh litterbags than in fine-mesh litterbags, but this only happened during the short deployment period. For the other leaf species (*Syzygium* and *Eucalyptus*), differences in decomposition rates between fine- and coarse-mesh litterbags were idiosyncratic but much lower than decomposition rates achieved by Vernonia. Leaves of *Syzygium* and the exotic *Eucalyptus* are tough (both belong to family Myrtaceae) and have high amounts of tannins and secondary compounds, which reduce palatability for shredders

(Gonçalves et al., 2006; Reis et al., 2018). This affects decomposition by microbes and detritivores (Graca et al., 2001; Janke & Trivinho-Strixino (2007).

The low decomposition rates of the poor-quality leaves of *Syzygium* and *Eucalyptus* indicate that microbes were potentially nutrient-limited, and nutrient immobilization during the earlier stages of the decomposition process (14 days) did not immobilize enough nutrients to diffuse the shortage. In comparison, the high-quality *Vernonia* leaves likely supplied enough nutrients to microbes and detritivores/ shredders, which led to higher decomposition rates, which were more than twice the rates achieved by *Syzygium* and *Eucalyptus* in both fine- and coarse litterbags.

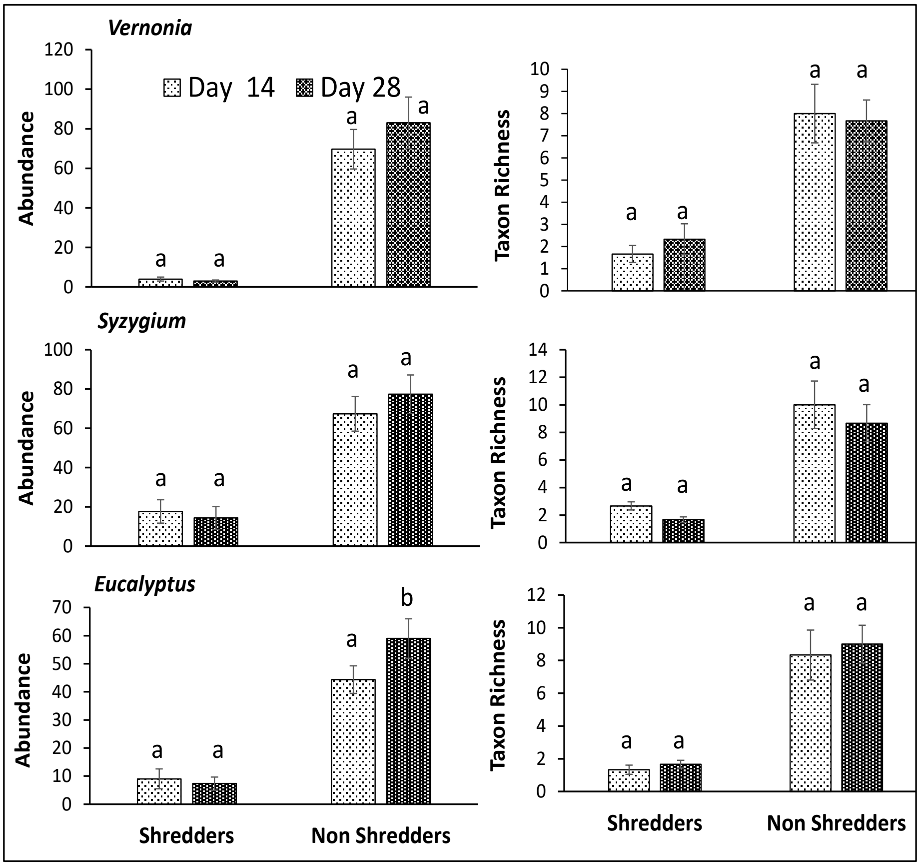

Figure 3: Mean (±SD, n=3) abundance and taxon richness (number of taxa) of shredders and non-shredders in coarse-mesh litterbags deployed in forested streams for 14 and 28 days. The different letters indicate significant differences among deployment periods (two-sample t-test). Note the differences in scale in the y-axes.

Lack of differences between the fine- and coarse-mesh litterbags, i.e., lack of differences between the role of shredders and microbes, in *Vernonia* after a long deployment period (28 days) and the idiosyncratic differences in *Syzygium* and *Eucalyptus* presents intriguing results. Lack of differences in decomposition rates between fine- and coarse-mesh litterbags shows that microbes played a greater role than shredders in the decomposition of leaf litter, but this role depended on litter quality. Indeed, microbes are thought to play major roles in the decomposition of leaf litter in tropical streams than in temperate streams because they are warmer (Irons et al., 1994). Some tropical streams also have a low diversity of shredders (Dobson et al., 2002), although a diverse shredder guild occurs in forested cooler high-elevation streams (Masese et al., 2014b and references therein). Shredders were not limited in this study (Figure 3), and both forested and agricultural streams have an abundant and diverse shredder guild (see Sitati et al., 2021). Differences between abundance and taxon richness of shredders in coarse-mesh litterbags between day 14 and day 28 (Figure 4) were minimal, suggesting that a short deployment period is just enough to study the colonization rate of different types of leaf litter.

CONCLUSION

This study shows that the deployment period of leaf litter has a significant influence on decomposition rates by determining the relative roles of microbes and shredders. Largely, inter-specific differences in decomposition rates between leaves of different quality were maintained during the short (14 days) and long (28 days) deployment periods. The short period was a better predictor of the relative roles of microbes and shredders in the decomposition of leaf litter differing in quality. The short period was also enough to study colonization rates of leaf litter by invertebrates, implying that long periods that are more than two weeks can be avoided in tropical streams. Short deployment periods will help cut costs and potential loss of litterbags to flooding or pilferage. Therefore, we recommend that 14 days are adequate to study leaf litter decomposition and colonization by invertebrates in tropical streams.

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REFERENCES

Abelho, M. (2001). From litter fall to breakdown in streams: A Review. The Scientific World. Journal, 1,656-680.

Benfield, E. F. (1996). Leaf breakdown in stream ecosystems. Methods in Stream Ecology, 579-590.

Boulton, A. J., & Boon, P. I. (1991). A review of methodology used to measure leaf litter decomposition in lotic environments: time to turn over an old leaf. Marine & Freshwater Research, 42(1), 1-43.

Boyero, L., Pearson, R. G., Hui, C., Gessner, M. O., Pérez, J., Alexandrou, M. A., & Barmuta, L. A. (2016). Biotic and abiotic variables influencing plant litter breakdown in streams: a global study. Proceedings of the Royal Society Biological Sciences, 14,283-296.

Boyero, L., López-Rojo, N., Tonin, A. M., Pérez, J., Correa-Araneda, F., Pearson, R. G., ... & Yule, C. M. (2021). Impacts of detritivore diversity loss on instream decomposition are greatest in the tropics. Nature Communications, 12(1), 1-11.

Cotrufo, M., Galdo, I., & Piermatteo, D. (2010). Litter decomposition: Concepts, methods and future perspectives. In W. Kutsch, M. Bahn, & A. Heinemeyer (Eds.), Soil Carbon Dynamics: An Integrated Methodology (pp. 76-90). Cambridge: Cambridge University Press.

Day, J. A., & I. J. de Moor. (2002a). Guides to the freshwater invertebrates of southern Africa. Volume 5: Non-arthropods (the protozoans, Porifera, Cnidaria, Platyhelminthes, Nemertea, Rotifera, Nematoda, Nematomorpha, Gastrotrichia, Bryozoa, Tardigrada, Polychaeta, Oligochaeta and Hirudinea). WRC Report No.TT 167/02. Water Research Commission, Pretoria, South Africa.

Day, J. A., & I. J. de Moor. (2002b). Guides to the freshwater invertebrates of southern Africa. Volume 6: Arachnida and Mollusca (Araneae, Water Mites and Mollusca). WRC Report No.TT 182/02. Water Research Commission, Pretoria, South Africa.

Dobson, M., Magana, A., Mathooko, J. M., & Ndegwa, F. K. (2002). Detritivores in Kenyan highland streams: more evidence for the paucity of shredders in the tropics? Freshwater biology,47(5), 909-919.

Ferreira, V., Encalada, A. C., & Graça, M. A. (2012). Effects of litter diversity on decomposition and biological colonization of submerged litter in temperate and tropical streams. Freshwater. Science, 31(3): 945-962.

Gessner, M. O., & Chauvet, E. (2002). A case for using litter breakdown to assess functional stream integrity. Ecological applications, 12(2), 498-510.

Gonçalves Jr, J. F., Graça, M. A., & Callisto, M. (2006). Leaf-litter breakdown in 3 streams in temperate, Mediterranean, and tropical Cerrado climates. Journal of the North American Benthological Society, 25(2), 344-355.

González, J. M., & Graça, M. A. S. (2003). Conversion of leaf litter to secondary production by a shredding caddis-fly. Freshwater Biology, 48(9), 1578-1592.

Graça, M. A. S. (2017). The Role of Invertebrates on Leaf Litter Decomposition in Streams, a review. International Review of Hydrobiology, 86:386-393.

Graça, M. A., 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.

Graça, M.A. S., Cressa, C. M. O. G., Gessner, T. O., Feio, M. J., & Callies, K. A. (2001). Food quality, feeding preferences, survival and growth of shredders from temperate and tropical streams. Freshwater. Biology, 46(7): 947-957.

Irons, J. G., Oswood, M. W., Stout, R. J., Pringle, C. M. (1994). Latitudinal patterns in leaflitter breakdown: is temperature really important? Freshwater Biology 32, 401–411.

Janke, H., & Trivinho-Strixino, S. (2007). Colonization of leaf litter by aquatic macroinvertebrates: a study in a low order tropical stream. Acta Limnologica Brasiliensia, 19(1), 109-115.

Manzoni, S., Trofymow, J.A., Jackson, R.B., Porporato, A. (2010). Stoichiometric controls on carbon, nitrogen, and phosphorus dynamics in decomposing litter. Ecological Monographs, 80, 89–106.

Masese, F. O., Kitaka, N., Kipkemboi, J., Gettel, G. M., Irvine, K., & McClain, M. E. (2014a). Litter processing and shredder distribution as indicators of riparian and catchment influences on ecological health of tropical streams. Ecological Indicators, 46, 23-37.

Masese, A. F. O., Kitaka, N., Kipkemboi, J., Gettel, G. M., Irvine, K. (2014b). Macroinvertebrate functional feeding groups in Kenyan highland streams: evidence for a diverse shredder guild. Freshwater Science, 33(2), 435-450.

Masese, F. O., Raburu, P. O., & Muchiri, M. (2009). A preliminary benthic macroinvertebrate index of biotic integrity (B-IBI) for monitoring the Moiben River, Lake Victoria Basin, Kenya. African Journal of Aquatic Science, 34(1), 1-14.

Merritt, R. W., Cummins, K. W., & Berg, M. B. (2008). An Introduction to the Aquatic Insects of North America. 4th (Edition). Kendall Hunt Publishing. Dubuque, Iowa, USA.

Nyadawa, M. O., & Mwangi, J. K. (2010). Geomorphologic characteristics of Nzoia River basin. Journal of Agriculture, Science & Technology, 12(2):145-161.

Patrick, C. J. (2013). The effect of shredder community composition on the production and quality of fine particulate organic matter. Freshwater Science,32 (3):10261035.

Reis, D. F., Machado, M. M. D., Coutinho, N. P., Rangel, J. V, Moretti, M. S., & Morais, P. B. (2018). Feeding preference of the shredder Phylloicus sp. plant leaves of Chrysophyllum oliviforme or Miconia chartacea after conditioning in streams from different biomes. Brazilian Journal of Biology, 79(1): 22-28.

Sitati, A., Raburu, P. O., Yegon, M. J., & Masese, F. O. (2021). Land-use influence on the functional organization of Afrotropical macroinvertebrate assemblages. *Limnologica*, *88*, 125875.

Tank, J. L., Rosi-Marshall, E. J., Griffiths, N. A., Entrekin, 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.

Vannote, R. L., Minshall, G. W., Cummins, K. W., Sedell, J. R., & Cushing, C.E. (1980). The river continuum concept. Canadian Journal of Fisheries & Aquatic Sciences, 37, 130–137.

Wabusya, M., Nyongesa, H., Konje M., & Agevi, H. M. T. (2015). Effects of land-use practices on soil organic carbon, nitrogen and phosphorus in river Nzoia drainage basin, Kenya, Agriculture, Forestry and Fisheries, 4(4): 153-158.

Wantzen, K.M., Yule, C.M., Mathooko, J.M., & Pringle, C.M. (2008). Organic matter processing in tropical streams. In: Dudgeon, D. (Ed.), Tropical Stream Ecology. Academic Press, London, pp. 44–65. Webster, J. R., & Benfield, E. F. (1986). Vascular plant breakdown in freshwater ecosystems. Annual review of ecology and systematics, 17(1), 567-594.

Wetzel, R. G., & Likens, G. E. (2013). Limnological analyses. Springer Science & Business Media.

Young, R.G., Matthaei, C.D., & Townsend, C.R. (2008). Organic matter breakdown and ecosystem metabolism: functional indicators for assessing river ecosystem health. J Journal of the North American Benthological Society 27, 605–625.

Zhang, M., Cheng, X., Geng, Q., Shi, Z., Luo, Y., & Xu, X. (2019). Leaf litter traits predominantly control litter decomposition in streams worldwide. Global Ecology and Biogeography, 28(10), 1469-1486.