

Leaf breakdown and retention of four different sized leaf species in Rutanga Stream, Gombe Stream National Park, Tanzania

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Introduction

The relationship between terrestrial and aquatic ecosystems is particularly important in small, forested streams (Benfield 1996, Wallace *et al.* 1999). All stream organisms obtain their energy from either autochthonous sources such as photosynthetic algae or from allochthonous sources such as the surrounding riparian vegetation. Small, forested streams rely heavily on allochthonous inputs of leaves and woody debris as the primary source of organic matter instead of algae due to light-limitation (Benfield 1996). In order for this organic matter to be utilized, it must first be retained in the stream and then broken down. Leaf litter breakdown is an integral part of carbon cycling, nutrient uptake, and more specifically, the food web of stream ecosystems.

Leaves enter the stream following abscission and accumulate on rocks, twigs, and stream margins to form debris dams (Mathooko 2001). These debris dams provide food and habitat for macroinvertebrate shredders such as insects and crabs as well as microbes that colonize the leaves. Microbes use enzymes to obtain nutrients from the leaves while softening them in preparation for macroinvertebrate shredding. Together, microbes and macroinvertebrates break the leaves down from complete leaves to fine particulate organic matter (FPOM) that becomes suspended and carried downstream and is eventually broken down to inorganic nutrients. The length of time of leaf breakdown, from abscission to complete decomposition, varies widely, providing streams with a constant supply of nutrients during the months without leaf litter inputs (Benfield 1996). Many studies conducted in the temperate zone have shown a stream-specific continuum of leaf processing ranging from slow (more than a year) to fast (a month) (e.g. Peterson and Cummins 1974). In tropical streams, however, very little is known about leaf breakdown rates.

Previous studies have identified many factors affecting the rate at which leaves are processed. It remains unclear which of these factors is the most influential. The physical and hydrological characteristics of the stream are important for initial leaf retention, while characteristics such as temperature, velocity, ambient nutrients, and pH affect breakdown rate by altering the abundance and communities of organisms present (e.g. Ardòn *et al.* 2006, Irons *et al.* 1994).

In addition to the importance of stream characteristics, leaf characteristics are also crucial in determining breakdown rate. Leaf quality varies based on the amount and form of carbon molecules and the concentration of nutrients such as nitrogen and phosphorus. High quality leaves contain higher concentrations of small labile carbon molecules such as simple sugars that are quickly and easily broken down by microbes. Low quality leaves, however, contain higher concentrations of larger carbon molecules, such as lignin, cellulose, and hemicellulose that are slower and more difficult to break down since microbes must use specialized enzymes to process them. Without taking other factors into consideration, there is evidence that leaf quality and nutrient content (N and P) are positively correlated to breakdown rate (Ardòn *et al.* 2006). Essentially, microbes obtain carbon, nitrogen and phosphorus from the leaves, although it is undetermined to what extent they also rely on ambient nutrients for N and P uptake (e.g. Royer and Minshall 2001, Ardòn *et al.* 2006). Leaf quality differs based on leaf species but may also be affected by precipitation, soil quality, age of leaf, access to light (either via the leaf's position on the tree or the tree's position in the canopy), and possibly leaf size.

This study examines the retention and breakdown rates of four different leaf species in a tropical stream. Tree species that bear large leaves may devote more energy to the production of structural components such as lignin, resulting in lower quality leaves. Thus, large-leaved species may break down at a slower rate than small-leaved species as a result of differences in quality. On the other hand, it is possible that large leaves are more likely to be retained in the stream and thus breakdown more rapidly due to a pre-existing assemblage of organisms.

Methods

Site Description

The study was conducted in a 60-meter reach of Rutanga stream in Gombe Stream National Park, Tanzania, a third-order, forested stream that empties into Lake Tanganyika. The riparian community consists of many different species of trees, shrubs, and overhanging vines that heavily shade the stream and contribute to the leaf litter and woody debris entering the stream.

Three streams were surveyed on July 13th to determine the four most dominant and easily accessible tree and shrub species that appeared to be contributing significantly to the allochthonous organic matter input. Of

the four species, *Monanthotaxis poggei* and species “B” were designated as bearing small leaves and *Ficus vallis-choudae* and *Tabernaemontana pachysiphon* were large-leaf species. *M. poggei* is a riparian shrub with the smallest leaves in this study, with an average surface area of $52 \pm 4 \text{ cm}^2$ (mean \pm 1SE) based on ten haphazardly chosen leaves. Species B, *F. vallis-choudae*, and *T. pachysiphon*, are riparian tree species bearing leaves with average surface areas of $83 \pm 7, 211 \pm 18$, and $368 \pm 45 \text{ cm}^2$ (mean \pm 1SE) respectively, following the same method.

Leaf Pack Preparation

On July 13th, leaves that appeared to be close to abscission were collected from multiple trees of each species and air-dried overnight. The following day, leaf packs were assembled and weighed. In order to simulate natural leaf pack composition, each pack was assembled with a minimum of three leaves. Due to the variation in leaf size, approximately 3 g of *M. poggei*, 5 g of species B, 10 g of *F. vallis-choudae*, 30 g of *T. pachysiphon* were used. A ~1-inch (2.25 cm) nylon mesh fish net was chosen for leaf pack bag construction so that macroinvertebrates could access the leaves, and non-sample leaf litter would not accumulate in the leaf packs. One bag of each species was tied together with fishing line to form a four species set. Twenty-eight sets were tied to roots along the stream margins on day 0, July 15, at locations suitable for natural leaf pack formation. Stream features such as roots, rocks, and natural debris dams were optimal locations for leaf packs (Mathooko et al. 2001).

On days 3, 6, 9, 12, 15, and 18, four species sets (16 leaf pack bags), were collected with ziplock bags. After transporting samples back to the lab they were rinsed over a 500 μm sieve and macroinvertebrates were stored in 70% ethanol for future analyses. Leaves from each pack were air-dried for a minimum of 24 hours and later weighed. A subsample of 0.25 g to 1.5 g, proportional to the leaf pack’s dry mass, was ashed at 500 °C for one hour to determine the ash-free dry mass (AFDM). The portion of sample remaining was stored in aluminum envelopes for leaf nutrient analyses. Four additional leaf packs of each species were constructed and used to measure handling losses and estimate initial AFDM as described by Benfield (1996). Since leaves were initially weighed only 24 hours after collection but reweighed at their constant dry mass, a conversion factor from wet to dry weight was determined and applied to the initial mass measurements.

Leaf Retention

A straight 60-meter reach (distinct from that used in the leaf breakdown experiment) of Rutanga stream was divided into 12 transects designated by flags at five-meter intervals. A ~1-inch (2.25 cm) mesh fishing net was tied across the width of the stream to catch non-retained leaves. Ten leaves of each species were haphazardly chosen and traced onto waterproof Rite-in-the-Rain[®] paper to facilitate retrieval during the experiment. On July 29th, a trial experiment was performed involving 150 “leaves” of each species. Due to the high volume of leaves and coincidental overlap between *M. poggei* and species B in terms of surface area, the experiment was altered and run three times with 20 leaves of uniform size of each species. In this way, the original categorization of *M. poggei* as the smallest, followed by species B, *F. vallis-choudae*, and *T. pachysiphon* largest was maintained. The leaves were released simultaneously. After 45 minutes, the net was emptied of all non-retained leaves and retained leaves were collected. Retention structure, distance traveled, and species were recorded for each leaf.

Statistical Analyses

Processing coefficients (k) were determined by fitting breakdown rates to an exponential regression. The ANOVA test of variance was used in conjunction with the Tukey-Kramer test of significance for both breakdown and retention rates.

Results

Leaf Breakdown Rate

All four species broke down at different rates according to processing coefficients, k, determined by the slope of the regression line (Figures 1-4, regression line unavailable for Figure 3). The largest leaf species, *T. pachysiphon*, decayed the fastest (Figure 4, $k=0.2392 \text{ day}^{-1}$), followed by the other large-leaf species, *F. vallis-choudae* (Figure 3, $k=0.1315 \text{ day}^{-1}$), species B ($k=0.0888 \text{ day}^{-1}$), and the smallest, *M. poggei*, with the slowest breakdown rate (Figure 1, $k=0.0314 \text{ day}^{-1}$). The analysis of variance (ANOVA) indicated that species, day, and their interaction all had a significant effect on breakdown rate ($df=3$, $F=28.57$, $P<0.001$; $df=1$, $F=111.32$, $P<0.001$; $df=3$, $F=14.462$, $P<0.001$ respectively). The Tukey-Kramer test for significance showed that the breakdown rate of *T. pachysiphon* was significantly faster than the others, which were not significantly different from each other.

Leaf Retention

The leaves of the four different species (sizes) were retained at similar rates with distance. All of the leaves were retained after 45 minutes. The ANOVA indicated that there was no significant effect of species (size) on retention with distance ($df=3$ $F=2.006$, $P=0.0902$). The only retention structures were rocks and pre-existing debris dams located in the stream channel. Leaves of all species (sizes) were more often retained on debris dams than rocks (Figure 5).

Discussion

Leaf Breakdown

All four species have processing coefficients that indicate relatively fast processing as determined by Peterson and Cummins (1974) who designated $k=0.01 \text{ day}^{-1}$ as the cut-off for fast and medium processors. The rates are not exceptionally fast for the tropics, although this has not been previously studied in Tanzania, and rarely in Africa. The processing coefficients in other studies performed in streams in Kenya, Costa Rica, Columbia, and Hawaii range from $0.001-0.559 \text{ day}^{-1}$ (Irons *et al.* 1994, Dobson *et al.* 2003, and others cited therein). This includes the range reported in this study of $0.031-0.239 \text{ day}^{-1}$, indicating that the chosen leaves did not breakdown abnormally fast for the latitudinal position of Rutanga stream in Tanzania.

The most often cited reason for fast leaf breakdown in the tropics is warmer stream temperature, resulting in rapid microbial colonization, and more shredding activity (Benstead 1996). Another study of streams along a latitudinal gradient discovered that shredders expedite leaf breakdown in temperate latitudes, but are less influential in the tropics due to a paucity of macroinvertebrates. In the tropics, microbes may be more important to the speed of decomposition (Irons *et al.* 1994). In addition to temperature differences between the temperate and tropical zones, other environmental factors affect the chemical and physical characteristics of the leaves themselves that may be reflected in faster rates of decomposition (Benstead 1996).

It is surprising that leaves of *T. pachysiphon* broke down significantly faster than all other species based on the original hypothesis of this study that predicted smaller leaves breakdown faster due to higher quality and fewer structural carbon molecules. However, the results from Wilton's controlled lab experiment (this Nyanza Report), further corroborate these findings. He found that crabs prefer *T. pachysiphon* when given a choice between the four species in this study. In the field, only three crabs were found: two on leaf packs of *T. pachysiphon*, and one on species B. The role of microbes is difficult to discern since it was impossible to measure microbial respiration in this study. However, the strongest odor was noted from leaf packs of this species, implying high microbe concentrations. This microbial conditioning may play a particularly strong role in preparing leaves for crabs and speeding up breakdown (Wilton, this Nyanza Report).

Despite evidence supporting a faster breakdown rate of the largest leaves, *T. pachysiphon*, it is difficult to draw conclusions about the effect of the size of leaves on breakdown rate as there are many confounding factors such as inherent species differences. Since size was used as a proxy for leaf quality in the original hypothesis, future analyses of initial leaf quality and leaf nutrient content in terms of % lignin, N and P will be useful in analyzing the effect of size on leaf quality and its subsequent breakdown rate. If quality is the most important controlling factor, it could be a coincidence that the large-leaf species that were chosen, *F. vallis-choudae* and *T. pachysiphon*, happen to be of higher quality than the two smaller-leaf species B and *M. poggei*. Or, since the two large-leafed species in this study happened to be larger trees than species B and *M. poggei* (a shrub), the leaves of these species may photosynthesize at a higher rate due to greater light access, resulting in greater concentrations of simple sugars. Leaf quality may have also differed between species due to the age of the leaves on individual plants. Older leaves contain fewer nutrients than young leaves since prior to abscission trees reabsorb nutrients from leaves. An effort was made to take leaves that appeared to be senescing, but certain species may have been closer to abscission than others based on natural cycles of leaf fall.

Despite the importance placed on leaf quality in many studies, leaf quality alone may not effectively reflect breakdown rate due to pre-existing nutrient limitations. Since these streams are oligotrophic, microbes may be nutrient-limited on high quality leaves and carbon-limited on low quality leaves. Ardòn *et al.* (2006), supported this hypothesis in Costa Rican streams where low quality leaves were unaffected by enhanced nutrient conditions. In fact, Ardòn *et al.* (2006) concluded that ambient nutrients and leaf quality combined are more important than either factor alone. Therefore, leaf quality may be less important in streams that are nutrient-limited such as Gombe Stream National Park, which are potentially N-limited (Burce, Nyanza Report 2003).

Leaf Retention

The results from the leaf retention study indicate that size has no effect on retention in the reach studied. Similarly, Mathooko *et al.* (2001) found that leaf surface area was not correlated to rate of retention in another tropical stream.

This study indicates that leaves are very well retained in Rutanga stream (all trials with 100% retention) due to an abundance of woody debris already in the stream and the presence of large cobbles within the main

channel. Once again, this is consistent with Mathooko *et al.* (2001) who attributed 19 % of retention to debris dams in their study. However, Mathooko *et al.* (2001) found that stream margins were the next most important retention structure, unlike this study. Such high retention and abundance of debris dams, in addition to fast breakdown rates, suggest that leaf litter and woody debris are very important sources of organic matter in Rutanga stream.

Future Work

Leaf Breakdown

Future leaf breakdown experiments could include mixed species leaf packs, or a comparison between leaves from shrubs and vines to those of trees. The effect of stream velocity is widely debated and could clarify daily fluctuations in leaf breakdown rates. I would be interested to see an assessment of natural leaf pack composition in forested streams. Additional leaf breakdown experiments could investigate the importance of leaf breakdown by performing leaf litter exclusion experiments and measuring changes in secondary productivity following this exclusion (Webster *et al.* 2000). Finally, measurements of microbial respiration on leaf packs would be imperative to further understand the role of microbes in leaf breakdown in the tropics.

Leaf Retention

Future leaf retention studies should have a longer waiting period after release. Comparisons between retention rates in streams with different substrate compositions could be an interesting addition to this study. If the issue of size as a factor in retention could be furthered, a full size range of leaves should be used with many replicates.

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