

Effect of macroinvertebrate colonization on decomposition rates of  
Speckled Alder (*Alnus incana*) leaves in northern Michigan streams

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### *Abstract*

Primary production by photosynthesis is limited in small, woodland streams; therefore energy must be derived from external sources. This allochthonous energy is primarily derived from coarse particulate organic matter (CPOM). Microbes and macroinvertebrates, particularly shredders, breakdown this CPOM into units accessible by other organisms and thus occupy an essential niche in the stream trophic chain. To investigate the influence macroinvertebrates have on decomposition, two streams (Brown Creek and Plum Creek) on the University of Notre Dame Environmental Research Center property in the Upper Peninsula of Michigan with similar nutrient and water chemistry were chosen as study sites. In response to lost data from past studies, a new frame system for litter bags was implemented. Decomposition and colonization rates were recorded between coarse mesh and fine mesh bags. Although statistically significant results for decomposition between streams was not found, an ANCOVA analysis of average number of shredders turned out statistically significant results for site ( $p$ -value=0.000,  $df$ =1,  $F$ -ratio=1.690), treatment ( $p$ -value=0.000,  $df$ =2,  $F$ -ratio=19.427) and site by treatment ( $p$ -value=0.474,  $df$ =2,  $F$ -ratio=3.143) indicating an interaction. The efficiency of shredder species of the streams may account for similar decomposition rates while maintaining statistically significant differences in shredder colonization. Because breakdown is essential to the food-web dynamics of many freshwater ecosystems, decomposition rates by macroinvertebrates can influence whole ecosystems.

### Introduction

Although primary production by autotrophs is the main energy source for most aquatic ecosystems, woodland streams employ a different energy-obtaining strategy. Because riparian vegetation blocks sunlight, of photosynthesis is limited. However, large amounts of non-living organic matter, or detritus, are deposited into the stream. Referred to as heterotrophic production, allochthonous energy is primarily derived from

external sources of coarse particulate organic matter (CPOM) such as leaf litter, wood, flowers and other plant parts, and animal inputs (Allan 1995). Most of this detritus enters the streams during autumnal dieback and are relatively nutrient-poor. The accepted sequence of leaf breakdown begins with the leaching of most of the remaining soluble nutrients, such as sugars and amino acids, into the water (Haapala 2001). The remaining lignin and cellulose are generally indigestible by animals and are then “microbially conditioned” by fungi and bacteria. This conditioning makes the remaining leaf material more palatable to shredder species (Benfield 1996) (Fig. 1). A key factor in the success of this heterotrophic energy pathway is the adequate and efficient breakdown of CPOM by macroinvertebrates into fine particulate organic matter (FPOM) (Roeding and Smock 1989). Through the feeding activities of shredders, CPOM is reduced in size by fragmentation into undigested pieces and by digestion into fecal pellets which then become a food source for other stream organisms (Benfield 1996). Numerous sources have classified benthic stream organisms into functional feeding groups, which include shredders. Common shredder taxa found in the Upper Peninsula are Isopoda, Amphipoda, and Trichoptera. When classified as shredders, Trichoptera are the most efficient in leaf breakdown from CPOM to FPOM followed by Amphipoda and Isopoda (Cummins and Klug 1979). Although most of

allochthonous matter is from autumnal die-back, summer leaf litter and other detritus provide a significant energy source when others are not readily available. A study by Maloney and Lamberti found that dried green leaves, which have a higher nutritional value than autumnal leaves, and warm, summer weather were more favorable to microbial conditioning and in turn had faster decomposition (1995). Several other factors affect rates of decomposition of leaves as well: chemical inhibitors, fiber content, and nutritional content, especially nitrogen. Higher concentrations of nitrogen have been associated with faster decomposition (Webster and Covich 1986). Of particular interest to this study, Alder (*Alnus sp.*) is known to have a symbiotic relationship with nitrogen fixing organisms which produce high nitrogen concentrations in the leaves resulting in consistently high rates of decomposition (Covich et al. 1999).

For this study, I wished to investigate the role macroinvertebrates play in the decomposition rates of the University of Notre Dame Environmental Research Center streams. I hypothesized that the rates of decomposition will vary between streams based on resident macroinvertebrates. I further predicted that streams with higher average counts of shredder species would have more rapid decomposition.

## Methods and Materials

**Study Site-**The study was conducted in the summer of 2008 in Plum Creek and Brown Creek on the University of Notre Dame Environmental Research Center property in Gogebic county Michigan, USA. Both are low gradient streams located on the southeast corner of the property. There has also been previous research showing similar water chemistry and other abiotic factors making them ideal study sites.

**Experimental Procedure-** Fresh, green, Speckled Alder leaves were collected by hand 26<sup>th</sup>-30<sup>th</sup> of May 2008 from live trees along the riparian zone of Plum Creek, with care being taken to collect healthy leaves. The leaves were placed in a drying oven at 60 degrees Celsius for 48 hours. In past studies at Brown Creek and Plum Creek, inadequate data due to loss by burial has occurred. Mesh bags staked into the stream bottom were covered with substrate material, terminating the ability to study decomposition processes. Using suggestions from past students, my Graduate student advisor, Christopher Patrick, designed a new way to set up this study. Using PVC piping, a U-shaped frame was constructed for mesh bags to hang from. Coarse mesh bags filled with equal amounts of leaves as those on the frames were staked to the substrate in the traditional way to compare the effectiveness of the new frame system. Ninety-six coarse mesh produce bags and forty-eight 500  $\mu\text{m}$  fine mesh

leaf packs were constructed. Two produce bags, one within the other, were filled with a standard mass of 3 grams (sd  $\pm 0.007120334$ ) of dried leaves. As outlined by Benfield in 1996, the bags were then closed with a zip tie bearing a metal number code, and rewetted to reduce breakage. The fine mesh bags were constructed by folding a 36 x 46 centimeter section of mesh in half and stapling the doubly folded edges on three sides. The bags were then marked with a letter code and filled with the standard mass of leaves. The top was stapled shut to form a 15 x 20 centimeter bag. Twelve U-shaped frames were constructed using 1.27cm PVC piping. The two supporting bars measured 61cm in height, with the top cross bar measuring 122cm. A fair coin was used to determine the random placement of four coarse mesh and four fine mesh packs on the frame. The frames were then marked with the stream and a left and right orientation. Six frames were placed in each stream about 8cm from the substrate. Four additional coarse mesh packets were staked to the substrate in alignment with the frames.

### **DATA COLLECTION**

**Organic Material**-One frame, plus the associated staked packs, per stream per week were collected. The first collection date was two days after introduction. Each pack was placed into an individual zip-loc bag marked with the name of the stream. Subsequent frames and packs were

picked up every seven days after the previous collection. If immediate processing could not be conducted, samples were placed into the refrigerator. Each pack was processed within 5 days of retrieval. In the lab, the contents of each bag were gently rinsed with tap water over an 850  $\mu\text{m}$  sieve. Leaf fragments were placed into tin cups marked with tin weight, date of collection, site, and ID number or letter(s). The remaining material (including macroinvertebrates) was rinsed into a container filled with 70% ethanol and marked with site and ID number or letter(s). The leaf tins were placed into a dryer at 60 degrees Celsius and allowed to dry for at least 48 hours. After 48 hours or more had elapsed, the dry weight of the leaves was recorded and the tins covered with tinfoil that had been perforated for ventilation. To account for any inorganic material deposited into the bags while in the stream, the tins were placed into a muffle furnace at 550 degrees Celsius for 2 hours in order to obtain the ash-free dry matter (AFDM). When the samples had cooled, the AFDM weight was recorded and the tins and their contents properly disposed of.

**Macroinvertebrates**-The ethanol samples were analyzed under a dissecting microscope and macroinvertebrates were counted and identified to order according to Lehmkuhl (1979) and McCafferty (1981). The macroinvertebrates from each bag were then placed into separate

containers labeled with site and identification number or letter(s). All data were recorded in Excel and analyzed in SYSTAT 12.

## Results

Data was collected for day two, but was lost by entry error and unable to be retrieved. Analysis of Covariance (ANCOVA) on remaining data was determined as the most efficient statistical analysis with a  $p$ -value  $\leq 0.05$  indicating statistically significant results (Boulton and Boon 1991). When an ANCOVA was conducted on percent remaining leaf material, a statistically significant value ( $p$ -value=0.000,  $df=2$ ,  $F$ -ratio=14.077) was found only for treatment. Upon ANCOVA analysis of average colonization rates, statistically significant results were found for site ( $p$ -value=0.000,  $df=1$ ,  $F$ -ratio=23.408) and treatment ( $p$ -value=0.000,  $df=2$ ,  $F$ -ratio=24.849). ANCOVA analysis of average number of shredders turned out statistically significant results for site ( $p$ -value=0.000,  $df=1$ ,  $F$ -ratio=1.690), treatment ( $p$ -value=0.000,  $df=2$ ,  $F$ -ratio=19.427) and site by treatment ( $p$ -value=0.474,  $df=2$ ,  $F$ -ratio=3.143) indicating an interaction. Tukey's Honestly-Significant-Difference test was conducted for Brown Creek and found that there was a statistically significant difference between the treatments of coarse mesh bags and exclusion bags ( $p$ -value=0.000) and between exclusion bags and staked bags ( $p$ -



value=0.000). Plum Creek was also analyzed with Tukey's Honestly-Significant-Difference test and resulted only coarse mesh bags and exclusion bags being statistically significantly different (p-value=0.000). When average decomposition rates, colonization rates, and shredder densities were compared to days in stream, respectively, error bars indicate standard deviation (Figures 2, 3 & 4).

### Discussion

**Explanations-**The results of this study did not indicate statistically significant differences in decomposition in UNDERC streams based on resident macroinvertebrates. However, data did suggest that resident fauna are important role in stream function. A similar study was executed in the tropics to investigate macroinvertebrate exclusion and decomposition. Both coarse and fine mesh bags were filled with the equal amounts of leaf material and analyzed for decomposition rates. It was found that macroinvertebrates strongly influence leaf decomposition and the effects of exclusion can vary with leaf type (Wright and Covich 2005). Brown Creek was shown to have higher average colonization (Fig. 2) and shredder counts (Fig. 3), however did not have statistically significant rates of decomposition (Fig. 1). The dominant shredder found in Brown Creek is Isopoda while the dominant shredder found in Plum Creek was

Amphipoda. Since Amphipods are more efficient shredders in comparison to Isopods, this could attribute to statistically insignificant decomposition rates. Plum Creek also had a much higher average of Diptera (21.738 vs. 6.578). Although not identified to lower taxonomic ranks, many Diptera are included in the shredder functional feeding group and may have aided the Amphipod numbers to similarly compare to that of the Isopod shredders of Brown Creek (Table 1). In addition to excluding macroinvertebrates, fine mesh bags are less vulnerable to fragmentation by abrasion and rapid leaching rates. They may also limit gas and nutrient exchange within the bag, producing anerobic conditions (Webster 1986). Although shredder colonization of exclusion bags was statistically significant in Brown Creek, when comparing exclusion bags to staked bags in Plum Creek there was no statistically significant difference in colonization rates ( $p$ -value = .115). This could have been a result of insecurely stapled exclusion bags allowing macroinvertebrate species to enter. Dance et al observed in their similar study comparable colonization rates of exclusion and non-exclusion bags study and suggested that small invertebrates were able to invade the exclusion bags and may have contributed to their breakdown (1979).

**Importance-** The breakdown rates of streams by macroinvertebrates is a sensitive measurement and may be useful in monitoring the health of

aquatic ecosystems (Webster 1986) In reference the river continuum concept, Covich et al suggest that the loss of an aquatic keystone species, such as shredders, would alter food and nutrient availability, especially that of detrital carbon (1999). The loss of a pivotal invertebrate species may also affect larger aquatic systems because they accelerate nutrient availability of adjacent riparian zone and thus to the open waters of lakes (Covich et al 1999). The activities of benthic macroinvertebrates have been linked to terrestrial ecosystems as well. Because aquatic macroinvertebrates release bound nutrients to other organisms, such as bacteria and macrophytes by their feeding habits, this makes them a key player in plant growth.

**Future suggestions-**Many recent studies are now focusing on naturally submerged leaf packs to link riparian vegetation to the macroinvertebrate communities they sustain (Grubbs 1994). Because there is little knowledge of the meiofauna of aquatic ecosystems, I agree with many other studies suggestions that the influence of the organisms may be higher than suspected. The study of these organisms provides an important link between detritus and larger consumers, such as fish (Webster 1986).

## Figures

Figure 1. Diagram outlining the river-continuum concept of nutrient flow in a stream ecosystem (Cummins and Klug 1979).

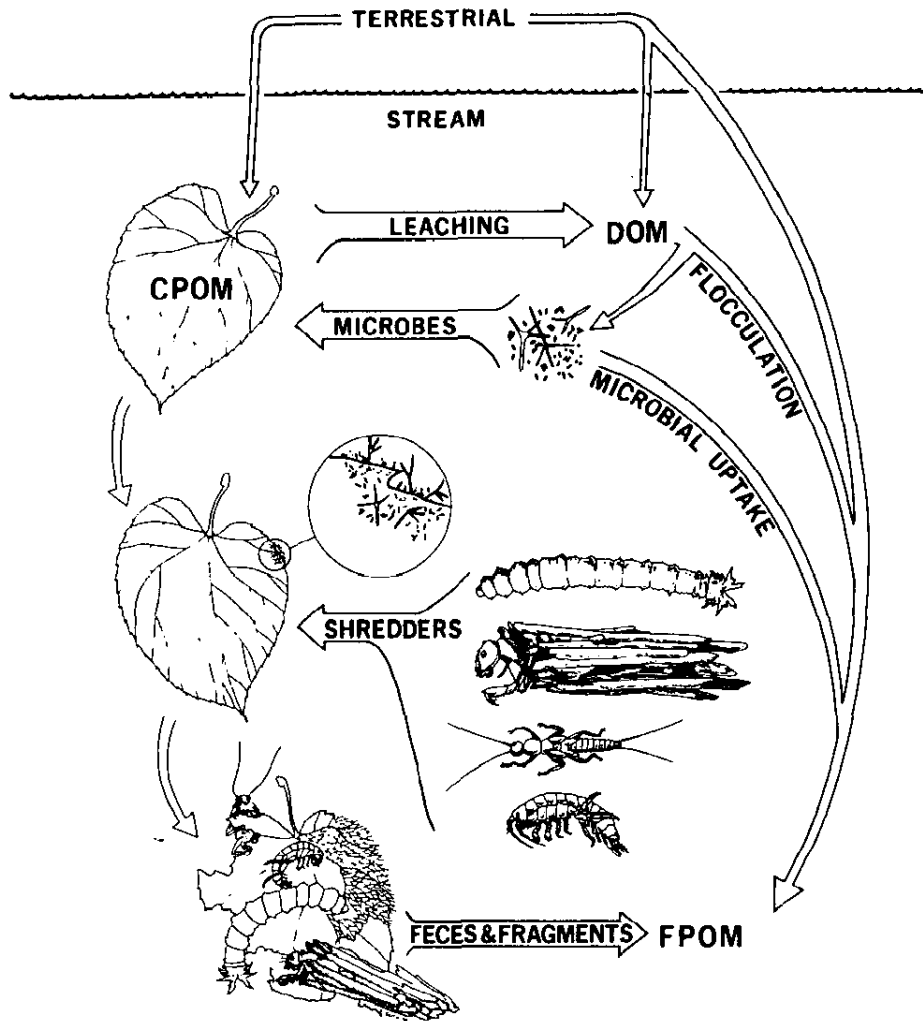


Figure 2-Average % remaining leaf matter across all pick-up dates

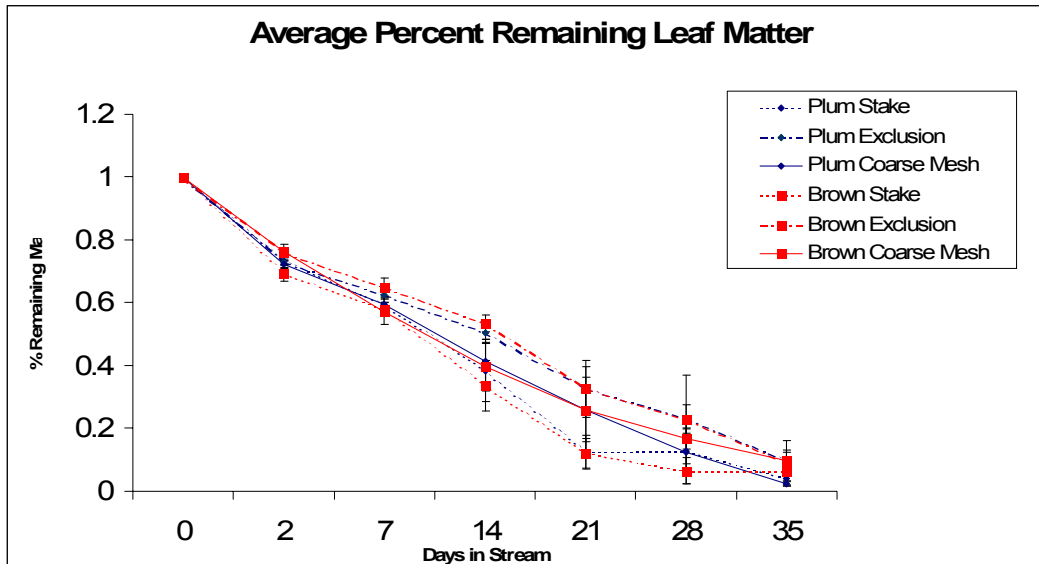


Figure 3-Colonization rates of all macroinvertebrates per treatment per site per day.

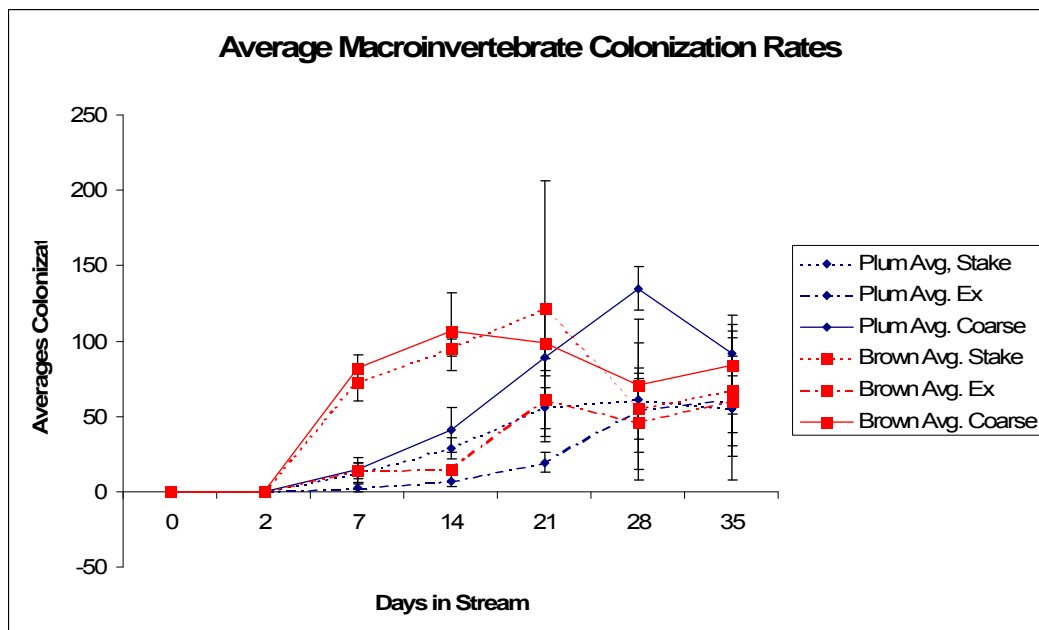
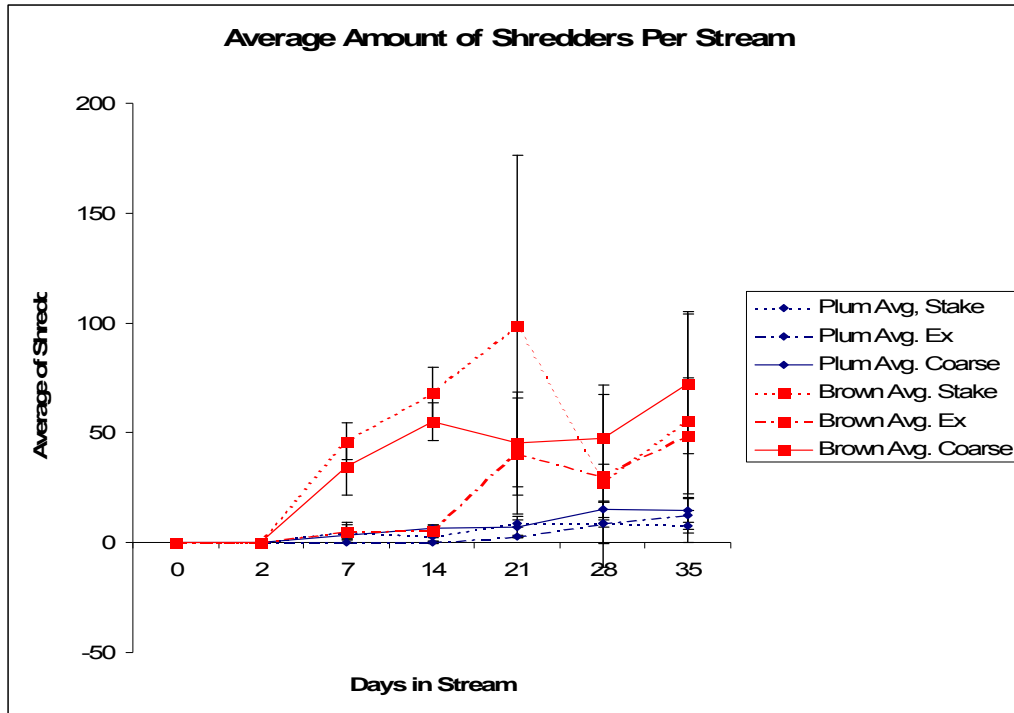


Figure 4-Average number of shredders (Amphipoda, Isopoda, and Trichoptera) per treatment per site per day.



## Table(s)

Table 1-Average number of macroinvertebrate colonization per creek over all six collection dates.

Taxa	Site	
	Brown Creek	Plum Creek
Amphipoda	0.984375	5.138462
Coleoptera	0.1875	1.076923
Decapoda	0	0.030769
Diptera	6.578125	21.73846
Ephemeroptera	7.046875	1.553846
Gastropod	7.234375	11.64615
Hirudinea	0.1875	0.169231
Isopoda	41.359375	0.061538
Odonata	0.078125	0.092308
Other	1.796875	2.938462
Plecoptera	0.015625	0.015385
Tricoptera	0.171875	0.307692

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