# **Ecological Comparison of Forested and Agricultural Streams**

Final report to Buffalo Creek Watershed Alliance Projects conducted in fall 2007 by students in BIOL 334/634, Biology Department, Bucknell University

Projects completed under supervision of Dr. M. E. McTammany

### **Acknowledgments:**

Mr. Craig Ernst gave us access to his property along Conley Road in order to study the agriculture-affected stream. Shanon Burkland (Watershed Specialist, Union County Conservation District) directed us to potential sites for our study. Buffalo Creek Watershed Alliance inspired the projects and shared their enthusiasm for the subject of study. Bucknell University provided resources for sample collection and analysis and a forested reference stream on their property in Cowan, PA.

For information about projects, contact:
Matthew E. McTammany, Ph.D.
Biology Department
Bucknell University
Lewisburg, PA 17837
570-577-3975
mmctamma@bucknell.edu

### **Table of Contents**

Executive Summary	3
Physical and Chemical Characteristics	6
Algal Biomass and Productivity	12
Stream Metabolism	16
Nutrient Uptake	22
Benthic Macroinvertebrates	28
Leaf Breakdown	36
Professional Commentary	41

#### **Executive Summary**

Buffalo Creek watershed encompasses approximately 120 square miles, primarily in rural Union County. Several tributaries of Buffalo creek flow through varied habitats such as forests. pastures, and agricultural lands. In central Pennsylvania, small streams (first and second order) in their natural state generally exhibit certain characteristics based on their forested habitat. The presence of trees is important to stream ecosystems because it limits the amount of light reaching the stream and provides a supply of organic matter (primarily leaves) to support organisms in the stream. Vegetation also decreases erosion and acts as a buffer against sediment and high nutrient inputs to the stream. The combination of low light, sediments, and nutrients results in low primary productivity and cold, clear, relatively pollution-free water, which is important to the survival of many organisms. In contrast, agricultural streams typically lack streamside woody vegetation and suffer from erosion and nutrient loading, which results in higher primary production and potentially nuisance algae blooms. Increased light also leads to higher temperatures, which combined with sediment deposition in stream substrates, causes major ecological changes in agricultural streams. Agriculture is of major economic importance in central Pennsylvania and comprises a large portion of the Buffalo Creek watershed. The Buffalo Creek watershed is part of the larger Susquehanna and Chesapeake Bay watersheds, so changes in Buffalo Creek and its tributaries may have significant effects on downstream ecosystems

In order to assess the impact of agricultural land use on stream ecosystems, Professor McTammany's BIOL 334 class from Bucknell University studied various components (physical, chemical, biological) in 2 first-order (headwater) tributaries of Buffalo Creek near Cowan, PA. Stony Run watershed is approximately 68 % forested (100 % along the stream) and was used as a non-impaired reference site. Conley Run, similar to Stony Run in basic physical characteristics (discharge, watershed area, elevation, and slope), is listed impaired by PA-DEP and contains 31 % forest and 62 % agriculture in its watershed. Trees are absent from much of Conley Run's valley, and the stream runs through pasture where cattle have unrestricted access to the stream. The objectives of this study not only include assessing impacts of agriculture on this stream, but also to collect data that may help with management practices throughout the Buffalo Creek watershed. Six individual studies were conducted during fall 2007 that each focused on one aspect of stream ecology: physical and chemical characteristics, algal biomass, nutrient uptake, metabolism, macroinvertebrate community, and leaf litter breakdown.

As a baseline for all other projects, physical (light, temperature, discharge, and substrate size) and chemical (pH, conductivity, ionic concentrations, and TSS) characteristics were measured to determine whether Stony Run and Conley Run were in fact different. Temperature, discharge, and substrate size of the two streams were not significantly different. However, Conley Run had substantially higher light reaching the stream, increased bank erosion, and higher total suspended solids than Stony Run. While pH was not different between the streams, specific conductance (a measure of total dissolved solids) and concentrations of all measured ions, with the exception of phosphorus, were significantly higher in Conley Run, which suggests that there is a large influx of nutrients to Conley Run.

The amount of algal productivity is tightly linked with the physical and chemical characteristics of the stream. Increased nutrient loading and increased light were found in the first project, which potentially could lead to increased algal production. Nutrient diffusing artificial substrates (terra cotta pots) containing either nitrogen, phosphorus, a combination of

both, or neither were placed in both streams and collected after 9, 19, and 26 days. Algae was scraped from the substrates and analyzed for chlorophyll a. Stony Run had very little algal growth while Conley Run had very high amounts of algal growth on all substrate types, indicating that algal growth was not limited by any nutrient in Conley Run.

In a related study, stream metabolism, a measure of the rates of photosynthesis and respiration is tightly linked to algal productivity. Dissolved oxygen was measured using sondes over a six-day period in both streams to calculate gross primary production (GPP), respiration, the ratio of photosynthesis to respiration (P/R), and net ecosystem production (NEP). GPP, NEP, and the P/R ratio were all significantly higher in Conley Run. Since algal biomass was higher in Conley Run, it makes sense that this stream had higher GPP, a measure of photosynthetic rate. However, respiration rates were similar in both streams. R was likely supported by heterotrophic activities (e.g., leaf decomposition) in Stony Run, as evidenced by P/R ratio close to zero, and autotrophic activities (e.g., algal respiration) in Conley Run (P/R close to 1). NEP, carbon available for storage in the ecosystem, was negative in both streams indicating that carbon sources from outside the streams themselves are important. However, NEP was significantly higher in Conley Run, which suggests that in-stream productivity is also extremely important in supporting respiration. In all, Conley Run appears to be suffering from eutrophication (excessive production) as a result of high nutrient loads and light availability.

The lack of nutrient limitation of algal growth observed in the second project suggests that Conley Run has nutrient concentrations exceeding biological demand. If this is the case, inorganic nutrients in Conley Run will not be taken up quickly by organisms. Nutrient uptake rates and lengths were measured in both streams by adding dissolved phosphorus and measuring concentrations at 10-meter intervals downstream from the introduction site. Uptake rates were slower in Conley Run, producing longer uptake lengths. In addition, mass flux of phosphorus (amount of P taken up per unit area of stream bottom per hour) was much lower in Conley Run. Combined, these results indicate that Conley Run did not retain nutrients and exported nutrients added to the stream. In contrast, Stony Run retained larger amounts of nutrients and slowed or prevented these nutrients from reaching downstream ecosystems.

Changes in physical and chemical characteristics of streams can also affect the invertebrate community. Quantitative and qualitative benthic samples were taken from each stream, and benthic macroinvertebrates were sorted and identified to family in the lab. Taxonomic information was used to calculate several biotic indices to compare relative abundance and types of organisms present in each stream based on their sensitivity to water quality. Total diversity of macroinvertebrates was similar in the 2 streams, but aquatic insect groups comprised almost all macroinvertebrates in Stony Run, while macroinvertebrates in Conley Run were primarily non-insect taxa (e.g., flatworms, crustaceans). Both the family-level Hilsenhoff biotic index (FBI) and % EPT (mayflies, stoneflies, and caddisflies) indicate that there are more pollution-sensitive species in Stony Run and more pollution-tolerant species in Conley Run. Shredders (organisms that eat large organic matter) were more abundant in Conley Run, most likely due to high numbers of herbivorous amphipods feeding on algae. Conley Run supported fewer filterers, most likely due to high amounts of suspended solids, which can damage filtering structures and inhibit their feeding.

The final project looked at the breakdown of leaf litter, which is a combination of microbial decomposition, mechanical fragmentation, and macroinvertebrate feeding. Mesh bags containing 7 grams of sugar maple leaves were placed in each stream and retrieved every 2 weeks for 6 weeks to determine mass loss over time and to calculate rates of leaf litter

breakdown. Statistical analysis found no significant difference in breakdown rates between the two streams. Stony Run contains more macroinvertebrates specializing on consuming leaf detritus, so we expected faster breakdown in this stream. However, Conley Run contained high numbers of generalist shredders, like amphipods, that may have consumed leaf detritus from our packs. In addition, higher temperatures and nutrient availability in Conley Run may have supported faster decomposition of leaves by bacteria and fungi.

Based on results from all six studies, we conclude that agricultural land use has significantly impacted Conley Run. Most of the problems or differences we observed between these streams were the result of excessive nutrients, high bank erosion and sedimentation, and elevated amounts of light in Conley Run. To address these issues and hopefully improve the integrity of Conley Run, the agricultural sections of Conley Run should have some basic management installed. At the very least, fences should be installed to exclude cattle from having unrestricted, direct access to the entire stream channel. Planting woody vegetation along the stream would also benefit Conley Run by reducing light, filtering nutrient and sediments, and reducing bank erosion. Other tributaries of Buffalo Creek likely suffer from similar impairments, which will magnify agricultural effects on Buffalo Creek and possibly downstream systems, including Susquehanna River and Chesapeake Bay. Continued monitoring of Conley Run and other agriculturally impacted streams in this area is of great importance to assess the degree of impact and to establish management practices that may alleviate some of these problems.

# Agricultural Effect on the Physical and Chemical Characteristics of Streams in the Buffalo Creek Watershed

H. E. Bulle, E. M. Ercolano, L. J. Muli

#### Introduction

Water quality in streams is greatly dependent on watershed land use, especially agriculture. Agriculture has many detrimental effects on streams. Streams located in agricultural areas exhibit enhanced erosion from loss of streamside vegetation (Burcher and Benfield 2006). Increased contaminants including sediment, nutrients and pesticides are also observed in agricultural streams and are the largest source of water quality degradation (Osborne and Kovacic 1993). Many of these problems are caused by loss of riparian zones and vegetation around the stream. Therefore, integrating vegetation into stream banks reduces erosion as well as increases uptake of nutrients and is one way to improve water quality in agricultural streams (Karr 1978). Agricultural streams tend to have increased temperatures, higher sediment inputs, increased nutrient concentrations, and lower inputs of leaf detritus (Scott et al. 2002).

The objective of our study was to determine if agriculture in the watershed had an effect on physical and chemical characteristics of streams. We studied a forested stream, Stony Run, and an agricultural stream, Conley Run, in the Buffalo Creek watershed to determine differences in streams caused by land use. Stony Run is a forested stream with rocky sediment and limited light because of tree canopy. Conley Run is an agricultural stream with no woody streamside vegetation. We expected Conley Run to exhibit many of the typical characteristics of agricultural streams, including greater suspended solids, higher light, increased temperature, and increased concentration of nutrients. While organic matter tends to be lower in agricultural streams because of loss of allochthonous inputs from leaves and woody debris, we expected organic matter to be relatively high in Conley Run due to organic inputs from cow excrements.

#### Methods

Site description

The study region is located in central Pennsylvania. The two streams are comparable in size, watershed area, slope, elevation, as well as bedrock composition (Table 1, Figure 1). The riparian zone immediately adjacent to Stony Run is a mature hardwood forest. The stream therefore exhibits reduced light and increased allochthonous material from the tree canopy. The riparian zone of Conley Run is used intensively for agriculture. Conley Run has increased light and highly eroded sides. The sediment in both streams is rocky, but Conley Run has greater levels of silt and clay covering the rocks. Land use for these two streams was measured with a geographic information system (GIS). Stony Run's watershed area was predominantly forest and Conley Run's watershed area was significantly agricultural. The percentage of land utilized by forest and agriculture was flipped in the two watershed areas (Table 1). The human activities which occur in the watersheds, especially agriculture cause Conley Run to have very different physical and chemical characteristics than Stony Run.

#### Physical and chemical measurements

Physical and chemical features of the streams were measured during October 2007. Light, temperature, conductivity, pH, dissolved oxygen, and discharge were measured using

calibrated digital probes placed in flowing sections of the stream during baseflow. Conductivity can change as temperature fluctuates, so specific conductance was measured, which corrects for temperature by a standardization to 20°C. Substrate size was characterized by a gravelometer. One hundred particles were randomly selected from each of the stream beds and their size was recorded. Particle size data was then used to calculate substrate size composition (% different particle types) and median particle size (mm).

Concentrations in each stream were measured by collecting stream water and analyzing the samples back at the lab. An ion chromatograph was used to measure cations (sodium, potassium, magnesium, calcium) and anions (chloride, nitrate, sulfate) in parts per million. Ammonium and phosphate are also measured by ion chromatography, but the detection limits with our equipment are relatively high for natural concentrations in most surface water. Therefore, phosphorus concentrations were determined using a spectrophotometric assay (APHA 1998).

Total suspended solids (TSS) are organic particles and inorganic sediments in the water column. Total suspended solids were measured by filtering 1-liter samples through pre-weighed glass fiber filters (1.0-µm mesh). Filters were then dried and weighed to determine total weight. Filters were combusted at 550°C for 1 hour, and the ash was weighed to calculate inorganic and organic fractions of total suspended solids.

#### **Results**

Stony Run and Conley Run exhibit similarities in several physical and chemical properties, as confirmed by two-sample *t*-test statistics (Table 2). Temperature, dissolved oxygen concentrations, pH, discharge, substrate size, and phosphorus concentration showed no significant difference between the two streams based on our single point measurements made during stream visits.

However, differences in other characteristics were also observed. Light, with limited data collected, was much higher at 607.5 umol m<sup>-2</sup> sec<sup>-1</sup> for Conley Run than the mere 23.97 umol m<sup>-2</sup> sec<sup>-1</sup> for Stony Run. Specific conductivity was also statistically higher for Conley Run (t-test, p<0.05). Total suspended solids, averaging 0.18  $\pm$  0.145 mg/L in Stony Run and 31.83  $\pm$  12.0 mg/L in Conley Run, were significantly different (t-test, p<0.05). Sediments from Stony Run were almost entirely organic in composition, while suspended solids from Conley Run were 69.2  $\pm$  12.3 % inorganic. It cannot be concluded that there is a statistically significant difference between these two streams due to the p-value of 0.541.

Both anions and cations (excluding phosphorus) were significantly higher in Conley Run. Considerable differences were noticed between the two streams in calcium, nitrate and sulfate concentrations. For calcium, the concentration in Stony Run was  $20.670 \pm 2.052$  mg/L while in Conley Run, it was  $45.594 \pm 3.041$  mg/L (t-test, p<0.05). Nitrate concentration in Stony Run was  $3.546 \pm 0.532$  mg/L and in Conley Run, it was  $12.853 \pm 0.511$  mg/L (t-test, p<0.01). In Stony Run, the sulfate concentration was  $6.879 \pm 0.192$  mg/L, whereas in Conley Run, it was  $19.486 \pm 1.706$  mg/L (t-test, p<0.05). Phosphorus concentrations were not statistically different, but were very low for both streams, at averages of  $7 \pm 0.1$  µg/L for Stony Run and  $7 \pm 0.3$  µg/L for Conley Run.

Particle size of substrate was not significantly different between the two streams. Median particle size in both streams was 90 mm, but Stony Run contained more particles classified as

"cobble" and fewer classified as "gravel" than Conley Run. Substrate in Stony Run therefore appears to be dominated by slightly larger particles.

#### **Discussion**

The data collected supported the hypothesis that there would be a significant difference in the two streams because of differences in land use and vegetation in the riparian zone. Light measured at the surface of the stream was much higher for Conley Run than for Stony Run (Table 2). This difference is due to the differences in vegetation. The dense tree canopy of Stony Run limited the availability of light that reached the surface of the water.

Specific conductivity was significantly higher in Conley Run (Table 2). Higher concentrations of dissolved ionic constituents in Conley Run caused this increase. Temperature was not statistically different between the two streams, but there was a slight increase in temperature in Conley Run (Table 2). This is a result of the increased exposure to light that this stream experienced. Based on daily fluctuations, temperature changes much more over 24 hours in Conley Run than in Stony Run, most likely due to exposure of stream to sunlight during day and being open to allow loss of heat during night.

Dissolved oxygen levels, while slightly higher in Conley Run, were not statistically different (Table 2). This increase is due to the abundance of algal biomass that was present in the stream causing an increase in photosynthetic rates. At night, a decrease in dissolved oxygen levels occurs because of higher saturation deficit due to increased biological activity of organisms and the lack of photosynthesis. Changes in dissolved oxygen could also have been affected by atmospheric diffusion and rapid water movement. Redox potential for both streams would be high during day when dissolved oxygen concentrations are high, but Conley Run might switch to a reducing condition at nighttime when oxygen is depleted. It is doubtful, however, that oxygen concentration drops low enough for major changes in redox chemical processes.

There was no statistical difference in pH between the two streams. The increase seen in Conley Run was due to the effects of photosynthesis during the day (Table 2). As carbon dioxide is consumed by organisms, the pH rises. In Stony Run, organic acids that accumulate from leafy debris and organic soils in the watershed lower the pH. Due to similar geology both streams maintain similar high buffering ability (Figure 1). However, pH shows dramatic patterns over 24-hour periods due to high photosynthetic consumption of CO<sub>2</sub> during day in Conley Run, which causes much higher pH.

Higher discharge levels indicate the ability to carry more sediment. Stony Run had greater discharge although it was not statistically different from Conley Run (Table 2). While the data show that both streams had similar substrate sizes, the stream beds were different (Table 2). Stony Run had a rocky stream bed with mostly cobble-sized stones. Conley Run also had gravel and stones, but had high levels of clay and silt covering the rocks which caused the water to have higher turbidity. It is possible that higher discharge in Stony Run has cleared out fine sediments from the channel, which results in larger substrate remaining, whereas Conley Run lacks the stream power to mobilize and transport its fine sediments. In addition, supplies of fine sediments from agriculture surrounding Conley Run likely offset any transport of sediments by this stream.

As we expected, total suspended solids were significantly higher in Conley Run than in Stony Run (Table 2). This is a direct result of runoff, leaching from soils and erosion because of minimal vegetation in the riparian zone, cow movement, and other allochthonous inputs from

plowing activity in the watershed (Burcher and Benfield 2006). Disturbances in the soil of the watershed, stream bank and stream bed were definite factors in the increase of total suspended solids in Conley Run. This increase in total suspended solids results in increased turbidity which affects the entire stream ecosystem. Although the data did not show statistical significance for the percentages of total inorganic solids between the two streams, it was evident that Conley Run had a drastically higher amount and with improvement to our methods, this can be confirmed.

Nutrient levels were significantly higher in Conley Run, which is expected of an agricultural stream (Table 2, Scott et al. 2002). Levels of sodium, potassium, magnesium, calcium, chloride, nitrate and sulfate were all statistically higher in Conley Run, which may indicate longer contact with limestone geologies in addition to the signature of land use. While both streams had relatively high nitrate concentrations, the difference in nitrate concentrations was due to runoff from agricultural products, as well as cow fecal matter. Sulfate was present in high amounts due to use of fertilizers. The cations, such as sodium and potassium, act with nitrate and phosphate as salts in the fertilizer or are added as nutrients themselves to the local watershed. Increased chloride concentrations are most likely a result of cow urine.

Phosphorus, which is expected to be higher in an agricultural stream, was not statistically different between the two streams in this study. While the data does not show a difference, Conley Run likely has higher amounts of phosphorus in non-dissolved forms, which are not detectable by the spectrophotometric assay used. Phosphorus can be immobilized by microbes on organic matter (e.g., feces) and algae on rocks and may adsorb onto silt and clay substrate present in abundance in Conley Run, which removes phosphorus from solution.

Our results confirmed that the use of agriculture in the watershed, specifically in the adjacent riparian zones, causes significant changes in physical and chemical characteristics of a stream.

#### **Literature Cited**

- APHA (American Public Health Association, American Water Works Association, and Water Environment Federation). 1998. *Standard methods for the examination of water and wastewater*, 20<sup>th</sup> edition. American Public Health Association, Washington, DC.
- Burcher, C.L, and E.F. Benfield. 2006. Physical and biological responses of streams to suburbanization of historically agricultural watersheds. The Journal of North American Benthological Society 25(2):356-369.
- Karr, J.R, and I.J. Schlosser. 1978. Water resources and the land-water interface. Science 201(4352):229-234.
- Osborne, LL, and D.A. Kovacic. 1993. Riparian vegetated buffer strips in water-quality restoration and stream management. Freshwater Biology 29:243-258.
- Scott, M.C, G.S. Helfman, M.E McTammany, E.F. Benfield, and P.V. Bolstad. 2002. Multiscale influences on physical and chemical stream conditions across Blue Ridge landscapes. Journal of the American Water Resources Association 38(5):1379-1392.

### Index

TABLE 1. Basic Site Characteristics for Stony Run and Conley Run

Variable Description	Units	Stony Run	Conley Run	
Watershed Area	ha	326.69	403.12	
Site devation	m	158	170	
Latitude		40° 57.692' N	40° 57.749' N	
Longitude		77° 00.196' W	77° 01.820' W	
Slope	percentage	0.8	0.9	
Watershed Land Cover Forest	percentage	67.9	31.4	
Agriculture	percentage	26.1	61.7	
Urban	percentage	6.0	6.7	

FIGURE 1. Geology of Stony Run and Conley Run

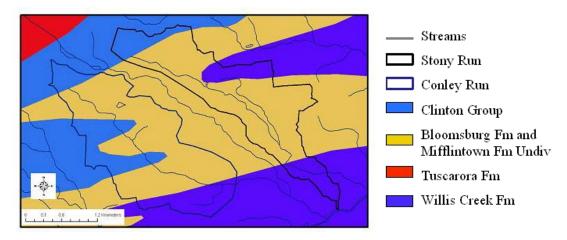


FIGURE 2. Land cover for Stony Run and Conley Run

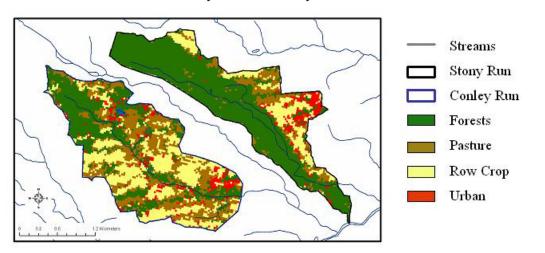


TABLE 2. Physical and Chemical Variables Measured within a 50m Stream Reach at Stony Run and Conley Run. SE = standard error of the mean value. t-stat values were determined from a two-sample statistical t-test.

Variable Description	Units	Data Source	Stony Run	SE	Conley Run	SE	⊭stat	p-value
Light	umol m <sup>-2</sup> sec <sup>-1</sup>	Calibrated digital probe	23.97	n/a	607.5	n/a	n/a	n/a
Specific Conductivity	uS cm <sup>-1</sup>	Calibrated digital probe	144.8	11.9	335.4	7.2	-13.77	0.046*
Temperature	°C	Calibrated digital probe	14.7	0.3	17.6	1.3	-2.23	0.269
Dissolved Oxygen	$mgL^{-1}$	Calibrated digital probe	9.39	0.10	11.49	0.59	-3.93	0.176
Dissolved Oxygen	% saturation	Calibrated digital probe	92.7	0.6	120.6	9.5	-2.95	0.208
pН			7.76	0.34	8.79	0.02	-3.02	0.203
Discharge	L s¹	Calibrated digital probe	10.8	2.3	3.9	1.4	0.082	0.082
Substrate size: gravel	proportion	Gravelometer	0.07	n/a	0.20	n/a	n/a	n/a
cobble	proportion		0.93	n/a	0.80	n/a	n/a	n/a
Median Particle Size	mm	Gravelometer	90	n/a	90	n/a	n/a	n/a
Total Suspended Solids	$mg L^{-1}$	Filtering, Dry-ash weights	0.183	0.145	31.833	12.0	-2.64	0.046*
Inorganic Sediment	% of TSS	Filtering, Dry-ash weights	0.27	100	69.2	12.3	-0.66	0.541
Sodium (Na <sup>†</sup> )	$mgL^{-1}$	Ion chromatography	2.527	0.084	5.049	0.722	-3.97	0.029*
Potassium (K <sup>+</sup> )	$mgL^{-1}$	Ion chromatography	1.049	0.036	1.628	0.120	-4.62	0.019*
Magnesium (Mg <sup>2+</sup> )	$mgL^{-1}$	Ion chromatography	3.092	0.307	5.149	0.135	-6.18	0.003*
Calcium (Ca <sup>2+</sup> )	$mgL^{-1}$	Ion chromatography	20.670	2.052	45.594	3.041	-6.79	0.001*
Chloride (Cl <sup>*</sup> )	$mgL^{-1}$	Ion chromatography	0.784	0.054	1.268	0.081	-4.96	0.004*
Nitrate (NO3 <sup>-</sup> )	$mgL^{-1}$	Ion chromatography	3.546	0.532	12.853	0.511	-12.61	<0.001*
Sulfate (SO <sub>4</sub> 2-)	$mgL^{-1}$	Ion chromatography	6.879	0.192	19.486	1.706	-7.35	0.005*
Phosphorus	μg L <sup>-1</sup>	Spectrophotometer	7	0.1	7	0.3	-0.18	0.866

<sup>\*</sup> p-values confirm statistical significance

#### Algal Biomass and Productivity of Buffalo Creek Tributaries

Service-Learning Project for Limnology T.S. Clavelle, D.K. Gilhuly, D. J. Lavender

#### Introduction

Measuring primary production in a stream ecosystem can be a difficult task. In order to determine algal growth and any limiting nutrients, an *in situ* nutrient limitation experiment was conducted. The area that we were assigned to study was in the Buffalo Creek Watershed, and our study is combined with other groups to create an outline for the agricultural impairment of streams. Different streams will have different amounts of biomass and algal production. Algal biomass and production in streams is determined by many factors including key nutrients such as nitrogen and phosphorus, light, water velocity, substrate type, and pH. Nitrogen and phosphorus are often the most important limiting factors affecting growth and are very likely allochthonous inputs to an agricultural stream (Gerloff et al, 1957). Determining which nutrients in the stream are responsible for limiting the growth of the algae is key in determining how to manage impacts on the landscape surrounding a stream.

The two streams that were chosen for this study were Conley Run, our agriculturally impaired stream, and Stony Run, our forested control stream in this experiment. Stony Run is an unimpaired natural stream, chosen for its proximity to Conley Run without impairment by the agriculture that affects Conley. In our experiment, nutrient diffusing substrates were infused with nutrient enriched agar. The four enrichments were control, nitrogen, phosphorus and nitrogen and phosphorus. These were used to determine which nutrients, if any, were limiting algal growth in Conley Run and Stony Run, and to determine if available levels of these nutrients would provide enough to stimulate algal growth. Once we collected the substrates, we processed them for chlorophyll *a* content. We expected overall algal biomass to be higher in Conley Run because it has higher background nutrient concentrations and high light availability (Luis Rivera et al., 2007). However, we also expected added nutrients, especially P, to stimulate algal growth in both streams

#### **Materials and Methods**

Stony Run (reference stream) and Conley Run (agriculturally impaired stream) were tested for algal biomass and productivity. Nutrient diffusing artificial substrates were constructed on which to grow our algae, 72 10.2cm diameter clay pots were glued to 18 pieces of 25x25cm Plexiglas sheets; 4 pots per piece of Plexiglas. The eighteen sets of 4 clay pots were filled with 225 mL of 2% agar solution. Four types of agar were used: (1) N enrichment, 0.5 mol/L NaNO<sub>3</sub>; (2) P enrichment, 0.1 mol/ L KH<sub>2</sub>PO<sub>4</sub>; (3) N+P enrichment, 0.5 mol/L NaNO<sub>3</sub> and 0.1 mol/L KH<sub>2</sub>PO<sub>4</sub>; (4) C, control with no nutrients which is unenriched agar only. The drainage holes on the bottom of the pots were plugged with neoprene stoppers to limit leaking of our substrate. The pots were then all glued, one pot of each nutrient treatment to each Plexiglas sheet. Sheets were placed in runs to support good water flow over the pots while minimizing disturbance for growing algae. Sheets were oriented in the stream in the following fashion; controls upstream, N pots oriented perpendicular to the flow of the stream, P pots oriented on the other side of the stream, and N and P pots downstream so as to minimize contamination of our control. We collected one set of 4 pots from both streams after periods of 9, 19 and 26 days.

Algae was scraped from the substrate once it is back at the lab and then processed for assessment of chlorophyll a. Algae was filtered using vacuum filtration and diluted as necessary. These filters were placed in the freezer until extraction of the chlorophyll a using 10 ml of 90% basic acetone solution for each filter. The filters were stored in the refrigerator for a period of four hours, after which 3 ml of the resulting solution was examined using a spectrophotometric assay. Absorbance readings were taken at 750 nm and 665 nm for each filter. Chlorophyll a was then calculated from this data.

#### Results

The first pick up of pots (10/26/07) was after nine days incubation in Conley and Stony Run. The average chlorophyll a level for the control pots of Conley Run was  $0.452~(\mu g/cm^2)$  compared with only  $0.118~(\mu g/cm^2)$  in Stony Run. Furthermore, there was no significant difference in algal growth between the four treatment pots in Conley Run, and Conley run had much higher algal growth than Stony Run for all four treatments. The second pick-up (11/5/07) after nineteen days in the stream, yielded increased levels of algal growth for all four treatments in Conley Run while no net increase in growth was found in Stony Run (Figure 2). For Conley Run, the two phosphorus treatments yielded slightly lower algal growth  $(3.42~\mu g/cm^2)$  for P and  $3.17~\mu g/cm^2$  for N+P) than the nitrogen  $(4.23~\mu g/cm^2)$  and control  $(4.43~\mu g/cm^2)$ , however these differences were not significant (Figure 1).

The final pick up (11/12/07) after twenty-six days in the streams, showed the same trend as the second pick-up with increased growth for all four Conley Run pots while there was no additional growth on any of the pots from Stony Run. In Conley Run, mean chlorophyll a values were higher for all treatments from the second pick-up compared with the first, and mean chlorophyll values from 11/12/07 were higher than those of the 11/5/07, resulting in a linear growth pattern for all four treatments (Figure 1). Stony Run did not show any net algal growth over time for any treatment as was found in Conley Run (Figure 2).

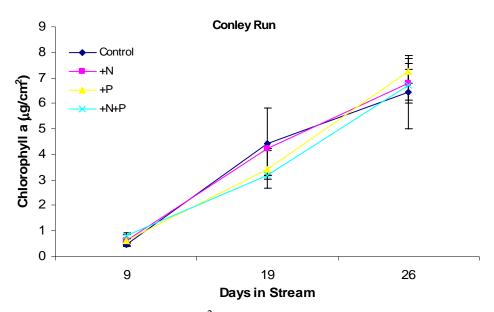


Figure 1. Mean chlorophyll a ( $\mu$ g/cm<sup>2</sup>) abundance extracted from treatment pots in Conley Run.

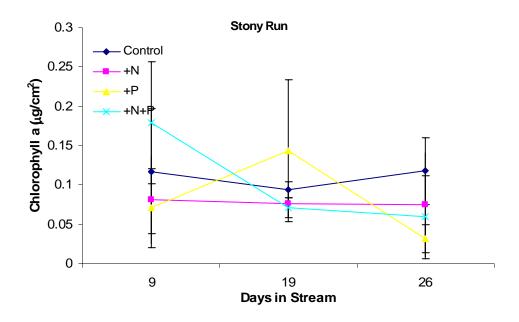


Figure 2. Mean chlorophyll a (µg/cm<sup>2</sup>) abundance extracted from treatment pots in Stony Run

#### **Discussion**

The results of algal growth as measured via chlorophyll extraction and analysis in Conley Run show that the algal growth is not limited by any nutrients. The four treatments in Conley Run (control, phosphorus, nitrogen, and phosphorus and nitrogen) did not show significantly different values for chlorophyll a in any of the three pickups. If nitrogen or phosphorus concentrations in Conley Run were limiting the growth of algae then the results would show greater productivity in the nitrogen, phosphorus, or nitrogen and phosphorus treatments. However, there was no difference between the control and the other treatments, which verifies that the control treatment pots were just as productive as the nutrient treatments. This confirmed a part of our initial hypothesis; there would be greater algal growth in the agriculturally impaired stream, due to the higher nutrient levels and PAR.

Stony Run on the other hand displayed close to no algal growth over the course of the study, whether nutrients were added to the substrate or not. Our nutrient-diffusing substrates may not have released enough nutrients to stimulate algal growth. Artificial substrates were controlled to provide the same concentrations of nutrients in both streams, so higher nutrient concentrations in Conley Run compared to Stony Run may explain the disparity between the streams. However, the amount of PAR was also higher in Conley Run compared to Stony Run, which could possibly explain the lopsided growth pattern that was observed over the course of this study (E. Ercolano et al., 2007). This seems to be the next logical reason to explain the difference in growth between the two streams.

These results are significant because with an excessive amount of nutrients comes an excessive amount of algal growth. Once the algae begin to decompose, the respiration by the decomposers can lead to depletion of dissolved oxygen. This in turn can seriously affect the

abundance and variety of aquatic life. Observed levels of dissolved oxygen were high during the day (13 ppm) while at night they would significantly drop (7-8 ppm) (Crumb et al., 2007). Most aquatic organisms require oxygen to breathe, and these streams become increasingly more difficult to live in at night when oxygen levels are extremely low. Only certain macroinvertebrates can tolerate such conditions, while most species of fish cannot. Aside from making streams unattractive and slimy with large algae blooms, an excess amount of nutrients can be toxic to aquatic life and unsafe for grazing animals.

This experiment was important for a number of reasons. First it showed that our results supported our hypothesis in that none of the nutrients we tested were limiting in Conley Run. These results implicated to us the extent of the agricultural impairment. Conley Run had very high levels of nutrients such that even upon the addition of external macronutrients in our experiment, we could not statistically show any net effects of nutrient addition. Growth was highly limited in Stony Run, our forested control stream, due possibly to limiting light and/or improper diffusion of nutrients through the substrate. Overall our experiment indicated agricultural impairment and eutrophic conditions in Conley Run but was unsuccessful in determining limiting macronutrients in either stream. When tied to the other parts of this study, we see interesting results and can draw some definite conclusions about the nature of agricultural impairment in the Buffalo Creek Watershed Area.

#### **Literature Cited**

- Bulle, Ercolano, E., Muli. "Physical characteristics of an agriculturally impaired stream" McTammany, Limnology Symposium, 2007.
- Crumb et al. "Metabolism of an Agriculturally Impaired Stream. McTammany, Limnology Symposium, 2007.
- Gerald C. Gerloff, Folke Skoog. "Nitrogen as a Limiting Factor for the Growth of Microcystis Aeruginosa in Southern Wisconsin Lakes" *Ecology*, Vol. 38, No. 4 (Oct., 1957), pp. 556-561.
- Lluis Rivera, Joan. Eugenia Marti. Daniel Von Schiller. "Effects of nutrients and light on periphyton biomass and nitrogen uptake in Mediterranean streams with contrasting land uses". *Freshwater biology.* 52, no. 5 (2007 May): p. 891-906.
- Pringle, M. Catherine. Frank J. Triska. "Effects of Nutrient Enrichment on Periphyton". *Methods in Stream Ecology.* 1996. 607-623.

#### A Comparison of Forested and Agricultural Stream Metabolism

N. A. Chacosky, T. L. Crumb, and S. L. Haupt

#### Introduction

Streams are linked through their watersheds and can be significantly affected by land use (Mulholland et al., 2005). Increases in agriculture have impacted the physical and chemical characteristics of streams (Young and Huryn, 1999). An agriculturally impaired stream is submitted to nutrient loading, increased sunlight, and low allochthonous input. These streams characteristically have higher dissolved inorganic matter and can have intense eutrophic conditions. Conversely, forested streams have decreased sunlight, increased allochthonous input, and less inorganic matter. Stream metabolism, a measure of the combined rates of primary production and respiration, are tightly linked to these characteristics. Both primary production and respiration can be greatly stimulated by nutrients and light in agricultural streams (Young and Huryn, 1999, McTammany et al., 2007), while forested streams may have limited photosynthesis but respiration supported by detritus.

In order to study the effects of agriculture on stream metabolism, we used concentrations of dissolved oxygen to calculate gross primary production (GPP), net ecosystem production (NEP), respiration (R), and the ratio of photosynthesis to respiration (P/R) in Stony Run, a forested stream, and Conley Run, an agriculturally impaired stream. A high GPP, NEP, and P/R value would indicate a highly productive system, especially if the value for NEP was positive since NEP is equal to the rate of photosynthesis minus the rate of respiration. Conversely, a negative NEP would mean that the system was consuming carbon and oxygen at a faster rate than producing it.

Both streams are tributaries to Buffalo Creek near Cowan, Pennsylvania. Agriculture is a significant contributor to the economy in central Pennsylvania; therefore it is important to examine how land use can affect stream ecology in this area. We predict that Conley Run will have higher GPP, NEP, and P/R values than Stony Run because Conley Run is agriculturally impaired.

#### Methods

Dissolved oxygen concentration was measured in Stony Run, a forested reference stream, and Conley Run, an agriculturally impaired stream. Dissolved oxygen levels were measured using a sonde probe left in a stream for an extended period. The sonde probe took measurements of dissolved oxygen and temperature at 15 minute intervals. The sondes collected data simultaneously in both streams in order to prevent environmental variability. Atmospheric pressure and time of sunrise and sunset each day were also measured by Bucknell University's weather station. Data were collected during the month of October, the 12<sup>th</sup> through the 16<sup>th</sup>, 2007.

Basically, metabolism is estimated in flowing water systems by measuring the change in dissolved oxygen concentration between 2 points in time and correcting this value for gas exchange with the atmosphere (diffusion). We calculated the absolute change in dissolved oxygen every 15 minutes from our sonde data. To calculate the direction of oxygen diffusion (into or out of the stream), we needed to know whether dissolved oxygen concentrations in the

stream were above or below saturation. We calculated saturation oxygen concentrations using stream temperature and barometric pressure measured continuously throughout the sonde deployment. Subtracting the measured dissolved oxygen concentrations and the oxygen saturation concentrations gave us the saturation deficit.

Diffusion was estimated using the energy-dissipation method (Tsivoglou and Neal 1976), which uses the relationship between water velocity, stream slope, and diffusion. The gas exchange rate was corrected for stream temperature using the equation (Elmore and West 1961),

$$k_2 = k_{2(20)} \ 1.024^{(T-20)}$$

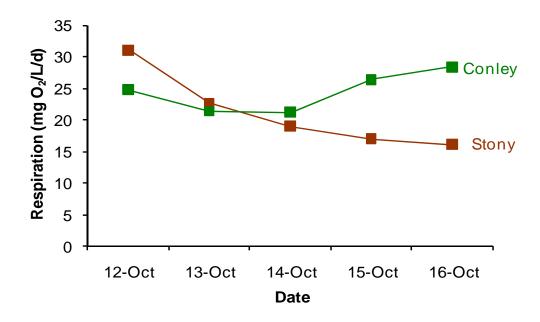
Oxygen exchange due to diffusion was calculated by multiplying the saturation deficit by the gas exchange rate. We then added the rate of diffusion to the measured change in dissolved oxygen to correct for change due to gas exchange with the atmosphere, which gives us a measure of the change in dissolved oxygen concentration in the stream due to biological processes alone.

These data were calculated for 24-hour periods from midnight to midnight to calculate daily metabolism. At night, there is no light to support photosynthesis, so all change in dissolved oxygen is due to respiration. During daytime, both respiration and photosynthesis occur, so we needed to correct for continuing oxygen consumption due to respiration. Therefore, a regression was made then to connect diffusion-corrected oxygen change from dusk and dawn to find the area above the curve of these lines, assuming respiration is constant between dawn and dusk.

R was calculated using the change in dissolved oxygen, corrected for diffusion and estimated as linear during daytime, and adjusted for 24 hours. GPP was then calculated by subtracting the amount of R from each 15-minute interval and adjusting for hours of daylight. Net ecosystem production was GPP minus R, and the P/R ratio was GPP divided by R. A paired t-test was then used for each metabolic parameter to find a significant difference from one stream to the other.

#### **Results**

Respiration rates over the five-day study in both creeks remained fairly constant with Stony Run only decreasing slightly and Conley Run only increasing slightly over time (Figure 1). There was no significant difference in respiration rates between the two streams (paired t-test, p=0.19) GPP values however were very different between the streams. Stony Run, the reference stream, had a very constant and low GPP, but Conley Run, the impaired stream, had a high GPP that initially decreased but then increased by the end of the study (Figure 2). These values were found to be significantly different (paired t-test, p<0.0001). NEP also showed major differences between the streams although both streams had negative values. Conley Run had a constant small negative value for NEP, but Stony Run started out with a large negative value, increased quickly, and then started to level off to a value still far below Conley Run by day five (Figure 3). These values were also significantly different between the two streams (paired t-test, p=0.004) In contrast, both streams showed constant values for their ratio of photosynthesis to respiration (Figure 4); however, the values for these ratios were completely opposite with Conley Run maintaining values between 0.8 and 0.9 and Stony run around 0.1. P/R values were significantly different between the two streams (paired t-test, p<0.0001).



**Figure 1.** Respiration in Conley and Stony Run from Oct. 12, 2007 to Oct. 16, 2007.

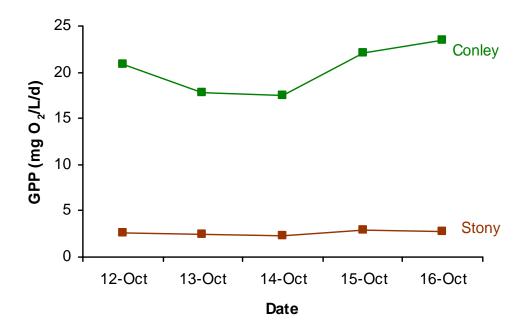


Figure 2. GPP in Conley and Stony Run from Oct. 12, 2007 to Oct. 16, 2007.

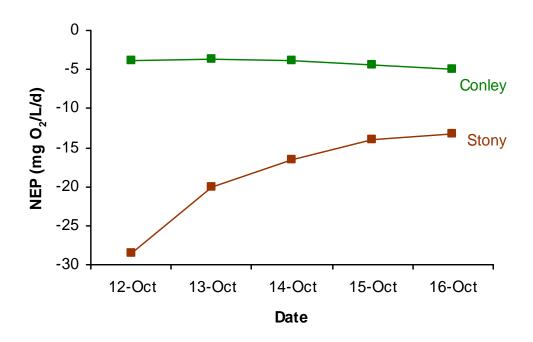


Figure 3. NEP in Conley and Stony Run from Oct. 12, 2007 to Oct. 16, 2007.

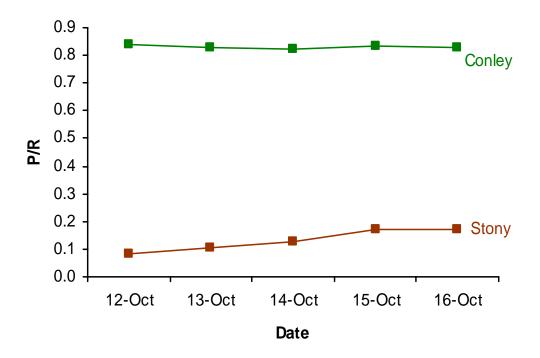


Figure 4. P/R ratio in Conley and Stony Run from Oct. 12, 2007 to Oct. 16, 2007.

#### **Discussion**

In 1980, Vannote and others presented the river continuum concept, providing a model for predicting stream characteristics. The dynamics of stream metabolism and stream energy sources follow a predictable pattern as you move from small to larger order streams. The change from headwater streams dependent on energy from terrestrial inputs to larger streams more dependent on internal energy from photosynthetic algae is thought to reflect a change in the ratio of gross primary production to community respiration. Furthermore, downstream characteristics are inextricably linked to upstream processes such as nutrient release from organic material (Vannote et al, 1980) However, changes in land use, particularly agricultural practices, can disrupt this pattern by changing the energetics of the stream.

Forested streams are shaded and receive significant amounts of organic material from trees. Light, a necessary component of photosynthesis, is reduced and primary production is low. Furthermore, breakdown of organic matter fueled by respiration, supports a community of heterotrophic organisms. Metabolism in forested streams is thus dominated by heterotrophic respiration. Our values for GPP and the P/R ratio in Stony Run are consistent with this assertion and comparable to other forested streams (McTammany et al, 2007, Young and Huron, 1999). NEP, a measure of the amount of carbon available for storage in the system, was significantly negative in Stony Run further indicating that energy inputs from outside the system are extremely important.

Conley Run, similar to Stony run in terms of size and order, did not show similar metabolism dynamics with significantly higher values of GPP, NEP, and P/R ratio. Land use surrounding Conley Run is about 60% agriculture, with our study site running directly through a cow pasture containing no trees. GPP and P/R were found to be slightly higher (McTammany et al, 2007) and slightly lower (Young and Huron, 1999) when compared to similar studies on agricultural streams. This may reflect the amount and intensity of agricultural practices in these areas. The increased agriculture at Conley Run resulted in a reduction in the amount of allochthonous inputs and increases in the amount of light reaching the stream. Furthermore, data on chemical characteristics showed a significantly larger amount of nitrate and total suspended solids in this stream. Lack of tall vegetation could increase the amount of runoff and the input of animal waste, detritus, and soils into the stream (Young and Huryn, 1999). The increase of nutrients coupled with increases in the amount of light can lead to higher primary productivity (McTammany et al, 2007). This is further supported by data showing that there was significantly more algal growth in Conley Run.

The only variable not found to be significantly different between the two streams was respiration. Conley Run does not receive the same amount of allochthonous inputs, but this may be compensated for by the increases in algae that are also undergoing respiration. Higher productivity but similar  $\underline{R}$  rates led to a P/R ratio closer to one and a significantly less NEP value. However, respiration exceeds photosynthesis even in Conley Run, indicating that allochthonous inputs are still important for energy exchange within this system. P/R ratios in other agriculturally impaired streams can reach values greater than one (Young and Huron, 1999). This may indicate that currently Conley Run is not as impaired as other streams but with values around 0.8 it is heading toward a switch to autotrophic metabolism.

In conclusion our results support that agricultural land use is significantly impacting the metabolism of Conley Run. Because streams are linked in their watershed, this could have far

reaching impacts downstream in Buffalo Creek. Continued monitoring of these stream systems is necessary to assess the impacts of agriculture in this watershed.

#### **Literature Cited**

- Bott, Thomas L. 1996. Methods in Stream Ecology. Academic Press Inc., 533-556.
- Elmore, H.L. & West, W.F. (1961). Effects of water temperature on stream reaeration. Journal of the Sanitary Engineering Division ASCE 87, 59-71.
- McTammany, M. E., Benfield, E.F. & Webster, J.R. (2007). Recovery of stream ecosystem metabolism from historical agriculture. *Journal of the North American Benthological Society*, 26, 532-545.
- Mulholland, P. J., Houser, J.N. & Maloney, K.O. (2005). Stream diurnal dissolved oxygen profiles as indicators of in-stream metabolism and disturbance effects: Fort Benning as a Case Study. *Ecological Indicators* 5, 243-252.
- Tsiyoglou, H.C. & Neal, L.A. (1976). Tracer measurement of reaeration III. Predicting the reaeration capacity of inland streams. *Journal of the Water Pollution Control Federation*, 489, 2669-2689.
- Vannote, R. L., Minshall, G.W., Cummins, K.W., Sedell, J.R. & Cushing, C.E. (1980). The river continuum concept. *Can. J. Fish. Aquat. Sci.*, 37, 130-137.
- Young, R. G., and A. D. Huryn. 1999. "Effects of land use on stream metabolism and organic matter turnover." *Ecological applications*, 9(4), 1359-1376.

#### **Agricultural Impact on Nutrient Uptake in Streams**

Ryan Sepsy, Alison Schaffer, and Jessica Glenn

#### Introduction

Nutrients are essential to the growth and reproduction of aquatic organisms in streams. Phosphorus, in particular, is usually a limiting factor for plant growth (Phosphorus Information Sheet 2007). Nutrients are cycled in stream ecosystems through a process called *spiraling*, where nutrients move from an abiotic dissolved form to a biotic form and back to an abiotic form as they move downstream (Webster and Ehrman 1996). The distance that a nutrient molecule travels downstream while completing a cycle is referred to as *spiraling length*, which is determined by uptake length and turnover length. Uptake length refers to the distance a nutrient travels while dissolved in the water column before it is removed by through biotic processes.

Uptake length is affected by physical properties of streams, such as water temperature and velocity (D'Angelo et al. 1991). Higher temperatures tend to increase microbial activity, which results in higher uptake rates, reducing uptake length. Increased velocity correlates with an increase in uptake length because aquatic organisms have less time to utilize the available flowing nutrients. Our objective was to measure and compare uptakes lengths between an agricultural stream (Conley Run) and a naturally forested stream (Stony Run) in the Buffalo Creek Watershed in order to determine the effect of agriculture on this process. Studies have shown that agricultural systems have many negative impacts on streams due to increased pollution levels, resulting in eutrophication (Dodds 2006). In general, streams in agricultural areas have nutrient inputs that exceed the demands from the biota of the ecosystem. These increased nutrient concentrations lead to nutrient saturation within the stream, thus increasing uptake length. (Bernot et al. 2006)

Two streams (Conley Run and Stony Run) were surveyed to determine the effects of agriculture and livestock on nutrient uptake lengths of a stream. We predict that uptake length will be longer in Conley Run because of higher background nutrient concentration due to land use (cow pasture/agricultural field). In contrast, we predict Stony Run will have a shorter uptake length due to undisturbed land use and low background nutrient concentration.

#### Methods

Tracer experiments have proved to be the most successful methods for determining uptake length in streams (Mulholland et al. 2002). For each stream, a concentrated chloride and phosphorus solutions was added to the stream at constant rate using a battery-operated peristaltic pump. Once the stream reached saturation (plateau) based on constant conductivity levels, water samples were taken. Starting 10 meters downstream from where the solution was added three water samples were taken every ten meters for a total of 50 meters. The water samples were then analyzed using ion chromatography and spectrophotometry to determine chloride and phosphorus concentrations for each sample. Chloride was used as a conservative tracer, meaning its concentrations would only change through our sampling reach due to dilution from inflowing groundwater. To calculate uptake lengths by measuring decreasing phosphorus concentration in our study reach, we needed to account for dilution. Therefore, we calculated ln(P:Cl) and plotted these values against distance downstream. The inverse of the regression slope was uptake length (S<sub>w</sub>). Since uptake length is susceptible to stream size and nutrient concentration, S<sub>w</sub> was used to

calculate vertical uptake velocity of phosphorus ( $v_f$ , in mm/min) and total phosphorus uptake per unit area of stream bottom (U, in mg P/m<sup>2</sup>/min).

#### **Results**

Background nutrient concentrations, chlorophyll a concentrations, stream velocity, and discharge were measured in each stream before the nutrient solution was released. Background phosphorus concentrations were equivalent for both streams at 0.007 ppm. Despite similar background concentrations, the average chlorophyll a concentrations were different for the two streams—6.443  $\mu$ g/cm² for Conley Run and 0.118  $\mu$ g/cm² for Stony Run. Stream velocity and discharge for Conley Run was 0.111 m/s and 6.6 L/s respectively, and 0.176 m/s and 29.7 L/s respectively for Stony Run (Table 1).

The ratio of phosphorus to chloride was used to determine phosphorus uptake while eliminating the effect of dilution on phosphorus concentrations downstream (Figures 1 and 2). Because relative amounts of P and Cl enrichment were different between the 2 streams, we converted values to percent maximum in each stream and plotted them together. Uptake length for Conley Run was longer than uptake length for Stony Run (Figure 3). S<sub>w</sub> for Conley run was approximately 370.37 meters and 125 meters for Stony Run (t-value: 3.96, p-value: 0.008).

Uptake length was then used to determine uptake velocity (the rate at which a phosphorus molecule is transferred to organic form:  $v_f$ ) and phosphorus flux (the quantity of phosphorus that moves from the water column to the substrate in a given period of time: U).  $v_f$  and U were both higher in Stony Run ( $v_f$ : 1.397 mm/min; U: 0.0112 mg/m²/min) as compared to Conley Run ( $v_f$ : 1.037 mm/min; U: 0.0083 mg/m²/min) (Table 2).

#### **Discussion**

We predicted that uptake length would be longer in Conley Run as compared to Stony Run and our results supported this hypothesis. Stony Run had a higher uptake velocity as well as higher phosphorus flux as compared to Conley Run. This means that phosphorus is taken up at a faster rate in Stony Run resulting in a shorter uptake length. The opposite was observed in Conley Run; uptake velocity was slower resulting in a longer uptake length.

Many factors influence uptake length, including stream flow, the presence of organisms and detritus, organic matter source, and nutrient concentrations. Conley Run is located in an agricultural field, whereas Stony Run is in an undisturbed forested area. Although background phosphorus concentrations were similar in both streams, algal biomass was higher in Conley Run, suggesting that nutrients are not as limiting in Conley Run as compared to Stony Run. Therefore, due to the abundance of nutrients in Conley Run, our added nutrients were not readily taken up by the biota within the stream. It seems likely that algal demand for nutrients is satisfied from sources already in the stream. In contrast, nutrients are more limiting in Stony Run; therefore the added nutrients were taken up at a faster rate over a shorter distance. While algal biomass was quite low in Stony Run, other microbes may stimulate high uptake of dissolved nutrients. For example, allochthonous detritus from leaf fall gets colonized by any number of bacteria and fungi, which are known to use nutrients from the water column and from their organic substrates (Mulholland et al. 1985). These data in combination with previous studies (Bernot et al. 2006) provide plausible explanations for why Conley Run has a longer uptake length than Stony Run. Previous studies of agricultural vs. forested streams have studied

uptake lengths for nitrogen, but the values of this study are very comparable to those of other studies. The Bernot et al. study (2006) had  $v_f$  values within the same range (0.0-4.8 mm/min) suggesting our results are comparable to those measured in other studies.

Slower water velocities typically result in shorter uptake lengths (D'Angelo et al. 1991). Despite velocity being higher in Stony Run than in Conley Run, Conley Run had a longer uptake length suggesting velocity does not have the same weight as other variables that have been shown to effect uptake length. Additionally, this also shows that organisms in Conley Run are not acting as a buffer and taking up extra nutrients. If organisms were taking up additional nutrients, uptake length would be shorter in Conley Run, which was not the case.

These findings are significant because they show the effects of agriculture on streams. Increased uptake lengths are evidence that there are excessive nutrients in the water column. Even more importantly, area-specific uptake (U) was lower in Conley Run, indicating less demand of nutrients by benthic organisms in our agricultural stream. The nutrient demand of the biota is lower than the nutrient concentrations in the stream, resulting in lower nutrient retention in this type of ecosystem. Therefore, excess nutrients are flushed further downstream eventually reaching larger bodies of water (including the Susquehanna River, and ultimately the Chesapeake Bay) leading to pollution and eutrophication of these downstream ecosystems. Physical changes to stream channels may also cause reduced uptake of nutrients in agricultural streams. For example, agricultural streams tend to be narrow and deep (Sweenet et al. 2004), which means nutrients in the water column are less likely to reach benthic organisms where most biological uptake occurs.

Appropriate best management practices should be implemented to improve nutrient processing rates in agricultural streams. Outside the stream channel, steps should be taken to reduce nutrient supply to the streams so that biological demand does not get saturated by high concentrations. Riparian vegetation and cattle exclusion could help. Stream channels themselves could be made wider and shallower and include structures, like large woody debris, to improve retention of materials. These types of retentive structures can be hotspots of biological activity and greatly stimulate nutrient retention and processing in streams (Valett et al. 2002).

#### **Literature Cited**

- Bernot, M. J., J. L. Tank, T. V. Royer, and M. B. David. "Nutrient uptake in streams draining agricultural catchments of the midwestern United States" *Freshwater Biology*. 51 (2006): 499-509.
- D'Angelo, D. J., J. R. Webster, E. F. Benfield. "Mechanisms of stream phosphorus retention: an experimental study." *Journal of the North American Benthological Society.* 10 (1991): 225-237.
- Dodds, W. K. "Eutrophication and trophic state in rivers and streams." *Limnology and Oceanography*. 51 (2006): 671- 680.
- Mulholland, P. J., J. W. Elwood, J. D. Newbold, L. A. Ferrena, and J. R. Webster. 1985. Phosphorus spiralling in a woodland stream: seasonal variations. Ecology 66:1012-1023.

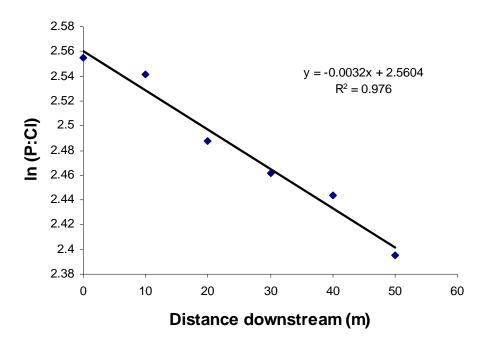
- Mulholland, P. J., J. L. Tank, J. R. Webster, W. B. Bowden, W. K. Dodds, S. V. Gregory, N. B. Grimm, S. K. Hamilton, S. L. Johnson, E. Marti, W. H. McDowell, J. L. Merriam, J. L. Meyer, B. J. Peterson, H. M. Valett, and W. M. Wollheim. "Can nutrient uptake in streams be determined by nutrient addition experiments? Results from an Interbiome comparison study." *The North American Benthological Society*. 21 (2002): 544-560.
- "Phosphorus Information Sheet" *Creek Connections*. Available from http://creekconnections.allegheny.edu/classroomresources/Chemistry/PhosphorousSheet. html. Accessed on 27 Sep. 2007.
- Sweeney, B.W., T.L. Bott, J.K. Jackson, L.A. Kaplan, J.D. Newbold, L.J. Standley, W.C. Hession, R.J. Horwitz. 2004. Riparian deforestation, stream narrowing, and loss of stream ecosystem services. Proceedings of the National Academy of Sciences 101(39):14132-14137.
- Valett, H. M., C. L. Crenshaw, and P. F. Wagner. 2002. Stream nutrient uptake, forest succession, and biogeochemical theory. Ecology 83: 2888-2901.
- Webster, J. R., and T. P. Ehrman. "Solute dynamics," in *Methods in Steam Ecology*. F. R. Hauer and G. A. Lamberti, editors. Academic Press, Inc., San Diego, California (1996). 145-160.

**Table 1** Background concentrations, chlorophyll a concentrations, velocity, and discharge for Conley Run (31 Oct 2007) and Stony Run (1 Nov 2007)

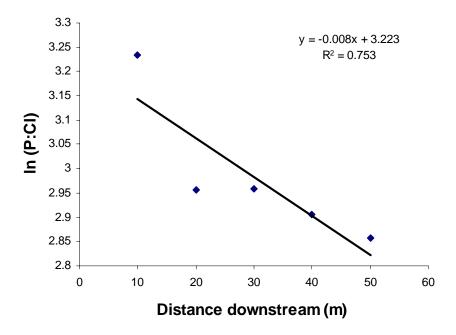
Stream	Background Concentrations (ppm)	Chlorophyll a Concentrations (µg/cm²)	Stream Velocity (m/s)	Discharge (L/s) 31 Oct 2007
<b>Conley Run</b>	0.007	6.443	0.111	6.6
Stony Run	0.007	0.118	0.176	29.7

**Table 2** Uptake length, uptake velocity, and phosphorus flux for Conley Run (31 Oct 2007) and Stony Run (1 Nov 2007)

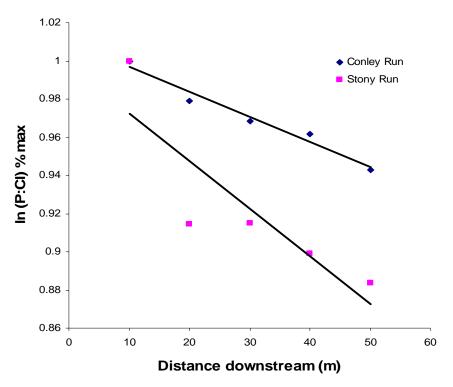
Stream	Uptake Length S <sub>w</sub> (m)	Uptake Velocity v <sub>f</sub> (mm/min)	Phosphorus Flux U (mg/m²/min)
Conley Run	370.37	1.037	0.0083
Stony Run	124.0	1.397	0.0112



**Figure 1** The natural log of the ratio of phosphorus to chloride versus distance downstream for Conley Run on 31 Oct 2007



**Figure 2** The natural log of the ratio of phosphorus to chloride versus distance downstream for Stony Run on 1 Nov 2007.



**Figure 3** A comparison of phosphorus uptake in Conley Run and Stony Run as the ratio of phosphorus to chloride decreases downstream. The steeper slope for Stony Run indicates a shorter uptake length.

## Density and diversity of benthic macroinvertebrates in an agriculturally impaired stream. K.L. Gehlhaus, B.P. Mulligan

#### Introduction

The density and diversity of macroinvertebrates in freshwater streams are directly affected by the location and physical characteristics of the stream. Agricultural land is a major source of nonpoint source pollution which increases the amount of fertilizers, pesticides, and animal wastes that become deposited into a stream (Roy et al. 2003). The diversity of macroinvertebrates in such streams will be quite different in comparison to a stream that flows through a deciduous forest. In this case, a forested riparian with a dense overhanging canopy will have a larger number of macroinvertebrates that are able to feed on leaf litter (Hauer and Resh, 1996). Furthermore if a stream's banks are subject to degradation by movement of cows, increasing the amount of sloughing, the input of fine sediments would then negatively impact particularly sensitive macroinvertebrates (Ephemeroptera, Plecoptera, and Trichoptera), and increase the amount of tolerant species (Resh et al. 1996). Our objective is to quantify and qualify two stream habitats by observing the density and diversity of macroinvertebrates in order to determine if the agricultural stream may be declared as impaired. We predict that the density and diversity of macroinvertebrates, especially those particular families less tolerant to pollution, will decrease in the agricultural stream, Conley Run, as a result of the difference in water quality, in comparison the forested reference stream, Stony Run.

#### Methods

Study region

The agricultural and deciduous forest sites in Buffalo Creek watershed, Conley Run and Stony Run, respectively, were visited on October 17<sup>th</sup>, 2007, to obtain samples of the benthic macroinvertebrates at each site. Conley Run is situated in a free grazing cow pasture with adjacent agricultural fields and is subject to herds of cow grazing and passing through its waters. The reference stream, Stony Run, is located in a thickly forested area and is surrounded by an abundant amount of deciduous native vegetation.

#### Macroinvertebrate sampling and metrics

A number of different sampling devices were used in order to representatively sample each site. For quantitative sampling, the Surber sampler was utilized to obtain the most accurate and representative sample of the invertebrate in a specific sample (Wetzel and Likens, 2000). We selected five different riffles located at different locations along the stream and collected one Surber sample per riffle. For collection of qualitative data, samples were collected using both a kick net and D-frame nets in both pools and riffles to compare the different environments within each stream. The samples were placed in 80 % ethanol to preserve specimens.

In lab, macroinvertebrates from the samples were identified by their order and family, using a dichotomous key. In addition, the density of macroinvertebrates was calculated by dividing the total number of organisms by the area sampled (0.5 m<sup>2</sup>). Taxonomic information was used to calculate several biotic indices. A family-level modified Hilsenhoff biotic index

(FBI) was calculated for each stream using tolerance scores and relative abundances of the different organisms (Resh et al. 1996). The FBI score can be used to compare relative quality of the water as a result of agricultural impact, in reference to the tolerance of the macroinvertebrates that are present and unique to each stream. Other measurements were taken, such as the percentage of pollution intolerant organisms (% EPT), in order to come to a conclusion about the state of the stream; an absence of such organisms would indicate a stream that is impaired due to pollution. The Jaccard Similarity Index was used in order to determine whether or not Conley Run had species of macroinvertebrates unique to its water, versus those found in Stony Run. The macroinvertebrates were also separated into functional feeding groups, in order to determine the sources of organic matter (algae and detritus) in the two streams.

#### **Results**

The comparison of macroinvertebrates from Conley Run to those of the reference stream, Stony Run, produced significant finds in terms of the diversity of benthic macroinvertebrates, especially those subject pollution intolerance. Overall, 367 organisms were collected from both streams combined, and more specifically 205 were collected from Conley Run, while 162 were collected from Stony Run. The density of organisms in Conley Run was 410 organisms/m², while Stony Run's density was 324 organisms/m².

The most common taxon in Conley Run was the genus *Gammarus*, of the order Amphipoda, with 29% of organisms collected (Figure 1) and 118 individuals/m² (Table 1). Flat worms (class Turbellaria) were also abundant (16% of macroinvertebrates collected, density 64 organisms/m²). Stony Run had slightly different dominating taxa, with the order Trichoptera, family Philopotamidae taking precedence with 96 organisms/m². The second most abundant taxon in Stony Run, also of the Trichoptera order, was Hydropsychidae, with a density of 44 organisms/m². These families combined made up 43% of the total macroinvertebrate density in Stony Run (Table 1).

Tolerance values, on a scale from 0-10 (0 being the lowest, 10 the highest) were assigned to each taxon for both streams in order to calculate the family biotic index (FBI). By multiplying the number of each specific taxon by its unique tolerance value, a weighted average of tolerance scores was calculated, otherwise known as the stream's FBI. Stony Run had a FBI of 4.30, while Conley Run produced an FBI of 5.23. The two prominent types of macroinvertebrates in Conley Run, Amphipoda and Turbellaria (flatworms), had tolerance scores of 4 for each.

Each stream was equally rich in taxa, with 20 different families each. Although each site contained a similar number of organisms and taxa, abundance of specific orders and families were quite different between streams, particularly with respect to pollution-sensitive orders. Conley Run produced a percentage of EPT (Ephemeroptera, Plecoptera, and Trichoptera) of only 2.0%. In comparison to our reference stream, this is quite low, as Stony Run produced a % EPT of about 61%. Furthermore, Conley Run had a sufficiently higher proportion of non-insects in comparison to Stony Run, with an abundant 72%. These include the following orders: Mollusca, Annelida, Nematodes, Turbellaria, and Crustacea. Quite contrastingly, Stony Run had a much higher number of insect orders with only 12% being non-insects. The Jaccard Similarity Index was used in order to determine the similarity of macroinvertebrate communities between the two streams directly. By dividing the number of taxa found in both streams (11) into the total number of taxa found (29), yields a Jaccard Similarity Index of 37.9% similar. Of the species unique to each stream, Conley Run species had a higher average tolerance level than Stony Run

species. Nematodes and Turbellaria produced an FBI of 4.4, while Stony Run had a lower FBI of 1.0 for its unique organisms, Plecoptera and Megaloptera.

Although the streams contained a variety of different macroinvertebrate taxa, comparing the types of functional feeding groups generated only a slight difference. Both Conley Run and Stony Run were abundant in macroinvertebrates that fed by collecting. However, Conley Run had a higher abundance of collector-gatherers (Figure 2b), whereas Stony had a majority of its organisms in the group labeled as filterers (Figure 2a). The number of predators found in each stream was relatively similar. Conley Run had more shredders, as a result of the high number of Amphipoda, more specifically, *Gammarus*, found in this stream (Figure 1b).

#### **Discussion**

Many conclusions can be drawn from the data obtained about benthic macroinvertebrate populations from Conley Run and Stony Run. The results show that the densities were more similar than predicted, however the data still shows a difference between the streams as a result of the unique species found there, and their abilities to tolerate pollution. One of the major difference between the streams was the percentage of pollution-sensitive organism as measured by the % EPT (Ephemeroptera, Plecoptera, Trichoptera). EPT organisms indicate health of streams as a result of these groups' intolerance to pollution, with Plecoptera (stoneflies) being the most sensitive order to agricultural pollution. As a biotic index, % EPT shows a distinct difference between two different systems. In Conley Run, the percentage was less than 5%, while Stony Run showed a very high percentage of EPT with 61%. The acute difference between the two streams indicates a large difference in the amount of pollution present in each. Conley Run's location in an agricultural field, surrounded by a herd of dairy cows, may be the direct cause for the low proportion of pollution-sensitive organisms. The almost complete absence of these pollution-sensitive orders, and complete absence of stoneflies, indicates that Conley Run has a higher level of pollution. Specifically, the absence of stoneflies suggests that organic pollution from dairy cows may be a problem, along with sedimentation and warm stream temperatures from active agriculture in Conley Run watershed.

The weighted tolerance levels or family biota index (FBI) of the different streams did not vary as greatly as the EPT ratio, but still represented a distinct difference between the two. The FBI for Conley Run was 5.23, while the FBI for Stony Run was 4.30. Higher scores indicate that the types of organisms found in a stream are generally more tolerant of pollution. Therefore, the organisms in Conley Run have higher pollution tolerance in general than those present in Stony Run. A range of values from 5.01 to 5.75 demonstrates "fair" water quality (Resh et al. 1996), which is where Conley Run's FBI falls. The reference stream falls within the range of 4.26 to 5.00, which shows "good" water quality (Resh et al. 1996). This set of data further supports the notion that Conley Run has poorer water quality in comparison to Stony Run.

As observed in Figure 1, there was a large difference in the taxa present in each of the streams. The richness of taxa is interestingly similar, as qualitatively there were 20 families identified from each stream. There was however a large difference in number of non-insect orders found in Conley Run in comparison to Stony Run. Conley has a great deal of non-insects, 72% which was quite higher than Stony Run's 12%; most notably, Nematodes and Turbellaria, unique to Conley Run, constituted a large portion of the organisms found in this stream. Each of these orders has high tolerance values in reference to pollution: 6 and 4 respectively. The existence of these organisms in Conley Run, and their absence from Stony Run is significant

evidence of the stream's impairment. In comparison Stony Run has a great deal of insect orders, while showing a high percentage of Trichoptera and Ephmeroptera, which are generally pollution-sensitive organisms. The species unique to Stony Run on the other hand, had tolerance values much lower than those of Conley Run. Plecoptera and Megaloptera were found in Stony Run, and by producing a weighted tolerance score of 1.0, signify that Stony Run's waters are much less polluted than that of Conley Run. The difference between the two streams' organism breakdown further corroborates the higher pollution concentration in Conley Run.

The similarity of the species found in the two streams can be quantified by the Jaccard Similarity Index. The two streams were 37.9% similar, which may be due to the streams being located very close to each other. Since they would share similar taxa because of closeness to each other, the difference in the tolerance averages of the unique taxa found in each of the streams would be a more significant measure of the impairment of Conley Run. The weighted tolerance level of the unique species found in Conley Run was 4.4, while the average for Stony Run was 1.0. The extreme difference in the two streams' averages based on the tolerance levels assigned to the species supports our hypothesis of Conley Run's impairment.

There were obvious changes in the macroinvertebrate assemblage structure, which is directly related to factors indicating variation in physical habitat, particularly bed sediment (Roy et al. 2003). Conley Run had a dense, algae infested, muddy benthic area, perfect for burrowing organisms, such as oligochate worms. Substrate in Stony Run on the other hand was comprised of larger rocks and much leaf litter, which will ultimately change the types of macroinvertebrate found. Many of the macroinvertebrates need large particles and associated interstitial space for protection from predators and high flows (Roy et al. 2003). This describes Stony Run and is a reason for there existing such a diverse number of insects, mainly the silk spinning order, Trichoptera, more commonly known as caddisflies. Conley Run, on the other hand, had a much lower discharge in comparison to Stony Run, and would therefore allow for more species that are not adapted for faster moving waters to thrive. The result of the cows plowing through the stream and leaving waste behind may play a role in decreasing the water flow, and change the availability of nutrients for various feeding groups.

Functional feeding groups are another indicator of the physicality of the two streams. The breakdown of the functional feeding groups of both groups is relatively similar except for the concentration of shredders. It would be expected that Stony Run would have a higher percentage of shredder groups, because of the higher amounts of leaf litter, and detritus located on the stream bottom. However, the data shows that Conley Run had a higher percentage of shredders present in its system. As mentioned, shredders generally gather their food through shredding organic matter, such as detritus found in the benthic area of a stream. They may also be herbivorous shredders, such as the order Amphipoda, in which the organism would shred the various types of algae found in the stream. Conley Run had a significantly higher amount of algae compared to Stony Run as a result of an increase in primary production (Figure 3), allowing for a prime habitat for these herbivorous shredders, which would explain the high percentage of this functional feeding group as well as the abundance of Amphipoda found in this stream. Meanwhile, Stony Run produced a higher number of filter feeders, while Conley Run had very few. Due to this feeding group's sensitive filter feeding structures, they are able to live in habitats with low inorganic material, as a result of this material's ability to damage these sensitive structures. Conley Run had a higher amount of total suspended solids, and therefore a higher amount of inorganic material, caused by the surrounding agricultural land and waste from

the cow herd. Therefore, the lack of filter feeders in waters demonstrates the poor quality of the water by being unable to support such groups of feeders.

In conclusion, through the various analyses of the macroinvertebrates collected it can be determined that Conley Run, in comparison to the reference stream, Stony Run, is significantly different in terms of the water quality present. Conley Run, being deemed roughly 62% agricultural, versus Stony Run which is then 68% forested would be expected to have very different taxa of macroinvertebrates, which is supported by the data found here. The presence of the cow herd along with the surrounding agricultural fields has had a negative effect on Conley Run's water quality. Biological methods may be used in order to attempt to treat this unhealthy stream. By simply planting vegetation along the stream bank, you may increase the amount of detritus and improve the benthic area of the stream. This could produce a ripple effect, by allowing for a more suitable environment for various types of insects. More drastically, the cow herd may be removed to allow the stream to repair itself. Regardless, measures need to be taken for the stream to become more ecologically sound.

#### **Literature Cited**

- Hauer, F.R., and V.H. Resh. 1996. Benthic Macroinvertebrates. Pages 339-369 in Methods in Stream Ecology. F. R. Hauer and G. A. Lamberti, editors. Academic Press, Inc., San Diego, California, USA.
- Resh, V.H., M.J. Myers, and M.J. Hannaford. 1996. Macroinvertebrates as Biotic Indicators of Environmental Quality. Pages 647-667 in Methods in Stream Ecology. F. R. Hauer and G. A. Lamberti, editors. Academic Press, Inc., San Diego, California, USA.
- Roy, A.H., A.D. Rosemond, M.J. Paul, D.S. Leigh, and J.B. Wallace. 2003. Stream macroinvertebrate response to catchment urbanization. Freshwater Biology 48:329-346.
- Wetzel, R.G., and G.E. Likens. 2000. Benthic Fauna of Streams. Pages 209-215 in Limnological Analyses. Springer-Verlag New York, Inc. New York, New York, USA. Index

Table 1. Density of organisms collected quantitatively in Conley Run on October 17<sup>th</sup>, 2007

Order/Family:	Density (organisms/m²)
Insects:	
Ephemeroptera/Caenidae	2
Ephemeroptera/Ephemerellidae	2
Odonata/Lestidae	2
Odonata/Libellulidae	4
Trichoptera/Philopotamidae	4
Coleoptera/Dryopidae	2
Coleoptera/Dytisicidae	2
Coleoptera/Elmidae	50
Coleoptera/Hydrophilidae	2
Coleoptera/Psephenidae	14
Diptera/Ceratopogonidae	12
Diptera/Chironomidae	18
Diptera/Simuliidae	2
Non-Insects:	
Turbellaria/Platyhelminthes	64
Nematodes/Nematoda	18
Annelida/Oligochaeta	26
Mollusca/Corbiculidae	24
Crustacea/Amphipoda	118
Crustacea/Isopoda	42
Crustacea/Decapoda	2
Total	410

Table 2. Density of organisms collected quantitatively in Stony Run on October 17<sup>th</sup>, 2007.

Order/Family:	Density (organisms/m²)
Insects:	
Ephemeroptera/Ephemerellidae	14
Ephemeroptera/Heptageniidae	12
Ephemeroptera/Isonychiidae	26
Plecoptera/Chloroperlidae	2
Plecoptera/Perlidae	2
Plecoptera/Peltoperlidae	2
Megaloptera/Corydalidae	2
Trichoptera/Hydropsychidae	44
Trichoptera/Philopotamidae	96
Coleoptera/Elmidae	30
Coleoptera/Psephenidae	12
Diptera/Chironomidae	34
Diptera/Simuliidae	2
Diptera/Tipulidae	8
Non-Insects:	
Annelida/Oligochaeta	14
Mollusca/Corbiculidae	2
Crustacea/Amphipoda	16
Crustacea/Isopoda	2
Crustacea/Decapoda	4
Total	324

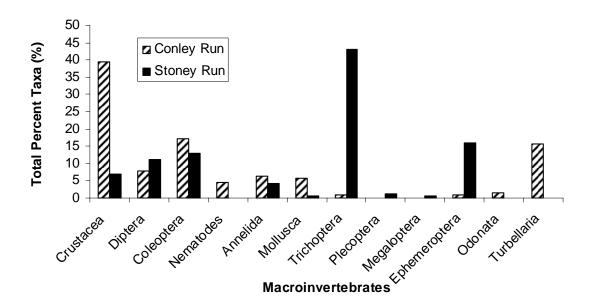


Figure 1. Percentages of the different orders of macroinvertebrates found in Conley Run and Stony Run on October  $17^{th}$ , 2007.

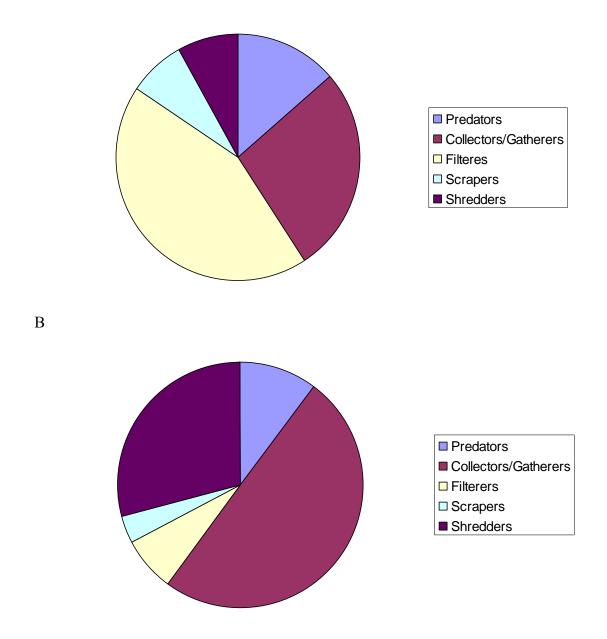


Figure 2. Functional feeding groups found in Stony Run (A) and Conley Run (B) on October  $17^{\text{th}}$ , 2007.

# Stream leaf breakdown rates as affected by vicinal land-use: a comparative analysis of forested and agricultural streams

W.S. Choi and H. Kho

#### Introduction

The breakdown of allochthonous matter is a significant process providing energy for various aquatic organisms. A consistent source of this detritus comes from senescent leaves from vegetation in the riparian zones adjacent to streams (Hagen, Webster and Benfield, 2006). Leaves that fall into streams are transported downstream until they are caught up by structures in the streambed to form leaf-packs. Leaves in leaf-packs are then processed by a variety of biological, physical, and chemical factors. The leaf-packs, over time, will chemically leach soluble compounds, become aerobically conditioned by micro-organisms, and fragment into coarse particulate matter (CPOM) or fine particulate matter (FPOM) by macro-organisms, otherwise known as "leaf shredders". Thus, the ecological integrity of a stream, which is considered to be condition of a stream relative to its historic ecosystem state, can be determined by analysis of the discrepancy in the rate that leaf-packs will breakdown due to anthropogenic disturbances (Hagen, Webster and Benfield, 2006).

The anthropogenic effects from the vicinal environment, such as in areas of agricultural or residential development, will affect the dynamics between ecological processes and physiochemical characteristics in a stream. Shredders are an integral aspect of stream ecosystems as macro-invertebrate abundance controls the rate of leaf breakdown. Therefore, the effects of agriculture, specifically the allochthonous input of leaves, may reduce shredder biomass and productivity (Hagen, Webster and Benfield, 2006).

In addition, physical and chemical changes to agricultural streams may affect decomposition rates of leaf material (Sponseller and Benfield, 2001). Higher temperatures and nutrient concentrations may cause faster microbial degradation rates on leaf detritus. However, increased sediment loads may bury leaves and create anoxic conditions on leaf surfaces, which inhibit microbial degradation. Furthermore, warmer stream temperatures may reduce dissolved oxygen and exclude leaf-shredding detritivorous insect larvae, which would tend to decrease organic matter processing rates.

Our objective was to study the extent of agricultural pressure on the ecological processes of streams through a comparative study of Stony Run, a natural-state reference stream, and Conley Run, an agriculturally affected stream. We predicted that the anthropogenic disturbances will decrease leaf decomposition rates in Conley Run; the increase of sedimentation, high nutrient content, and lack of allochthonous input will negatively affect the abundance of leaf shredding macro-invertebrates, which is a good indicator of the leaf breakdown rates. Our approach in this study was to quantify the rate of degeneration by sampling pre-weighed leaf-packs in the two streams over a period of six weeks.

#### Methods

This study of leaf breakdown rates was conducted at two sites, Stony Run and Conley Run from October 3 to November 14, 2007. Leaf breakdown rate was measured using the leaf-bag method. Three sites in each stream with similar depth, flow, and water velocity were chosen. Three leaf-bags (4-mm nylon mesh) were placed at each site in both streams for a total of 9 leaf

bags in each stream. All leaf-bags were filled with 7.0 g of senescent sugar maple (*Acer saccharum*) leaves, which had been dried to constant mass. Sugar maple leaves were used in this experiment because they have relatively fast decomposition rates (Webster and Benfield 1986). Leaf-bags were anchored to the stream bottom by a stake driven into the streambed. There were also six control leaf-bags that were brought along to the sites to account for the disturbance that would be sustained by the experimental leaf-bags during transport. Three leaf-bags were recovered bi-weekly from the two streams over six weeks during the course of the study; these leaf-bags from the sites were rinsed over a 1-mm screen mesh, dried in paper bags, and finally weighed.

Once the initial and final weights of the leaf-packs were recorded, all samples and controls were ground into fine particles and approximately 0.2 g was measured into small prefired tin cups. These sub-samples were then placed into an oven at 550 °C to combust the organic matter and be left with the ash weight of the sample, from which the ash-free dry mass (AFDM) is calculated by the difference between the sub-sample and ash weight. The total amount of organic matter in the leaf-packs was found by determining the proportion of organic matter (%AFDM) in the sub-sample (AFDM / (sub-sample of the total dry mass). The proportion of organic matter in the sub-sample (%AFDM) was then multiplied by total dry weight of each leaf-pack samples. The rate of leaf-pack decomposition rate was graphed by plotting the natural logarithm of the leaf-pack's ash-free dry mass over time in days. A linear regression-line was added and the slope of the linear regression-line will be the rate of leaf break-down.

#### **Results**

As expected, the total ash-free dry mass of Stony Run's leaf-packs steadily decreased over time (Figure 1). There are, however, occasional discrepancies in the decreasing trend of the AFDM; this is most likely due to an accumulation of detritus, sediment, microbes and macroorganisms. Linear regression showed that the rate of decomposition of leaf-packs in Stony Run was 0.0206 / d with an  $R^2$ -value of 0.7991.

Similar to Stony Run, the total ash-free dry mass of Conley Run's leaf-packs showed a decreasing pattern over time (Figure 2). Again, there are small discrepancies in the decreasing trend of the AFDM, which is because of the accumulation of detritus, sediment, microbes and macro-organisms over time. Regression data from Conley Run showed that leaf-pack breakdown rate was 0.0211 / d with an R<sup>2</sup>-value of 0.965.

ANCOVA statistical analysis was used to compare the regression slopes for Conley Run and Stony Run. Based on this analysis, leaf breakdown rates were not significantly different.

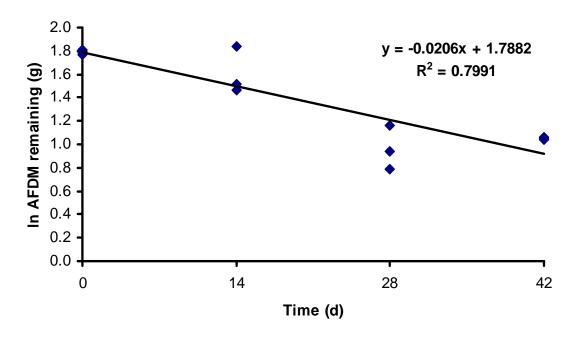


Figure 1. The natural logarithm of the AFDM (g) remaining in the leaf-packs plotted over time in days in Stony Run (10/3/07 - 11/14/07)

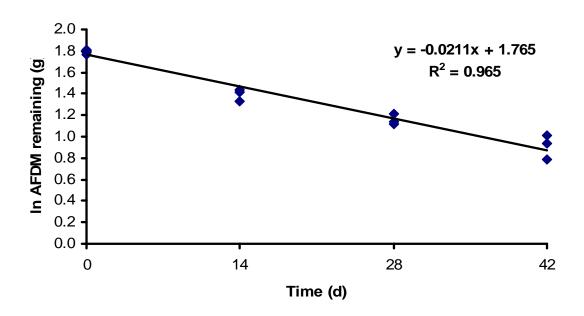


Figure 2. The natural logarithm of the AFDM (g) remaining in the leaf-packs plotted over time in days in Conley Run (10/3/07 - 11/14/07)

#### **Discussion**

Our results show that when compared through leaf breakdown data, the differences due to anthropogenic influences, such as farming or other land developing, was not significant between Conley Run, an agriculturally impaired stream, and Stony Run, the "natural" forested stream. In fact, the breakdown rates from the leaf-bags from either stream were essentially identical, 0.0206 /d and 0.0211 /d for Conley and Stony Run respectively. These breakdown rates were relatively fast, even for sugar maple leaves which average approximately 0.005 /d (Webster and Benfield 1986).

Several factors may be responsible for similar breakdown rates in these 2 streams despite drastically different water quality and biological characteristics. Stony Run had high numbers of detritivorous insect larvae (e.g., peltoperlid stoneflies) among its benthic macroinvertebrate assemblage, but Conley Run had large numbers of facultative shredders (e.g., amphipods). These shredders could be functionally redundant in leaf breakdown processes between the 2 streams. Since Stony Run has large inputs of leaf material during autumn, it supports obligate shredder populations. Shredders in Conley Run likely feed primarily on thick algal mats on the substrate but may switch to consume leaf material when available. In addition, higher temperatures and nutrient concentrations were measured in Conley Run compared to Stony Run, which may have supported larger populations of microbes (bacteria and fungi) to decompose leaves in Conley Run and compensate for lack of specific leaf-shredding invertebrates. Finally, mechanical fragmentation may have been high in Conley Run due to abrasion by transported sediment and greater fluctuations in velocity during storms. We attempted to control for physical differences between streams by placing them in locations with similar depth and velocity, but our packs were deployed during base flow and would not account for changes during storm flow.

Future studies of leaf decomposition in agricultural streams should distinguish between the roles of microbial decomposition, physical fragmentation, and shredder leaf processing to elucidate the mechanisms primarily responsible for patterns of leaf breakdown. Only with this understanding can we hope to restore ecological conditions in agriculturally impaired streams. One factor is certain, leaf detritus typically supports stream ecosystems in regions dominated by temperate deciduous forests. The drastic vegetation changes caused by agriculture throughout central Pennsylvania have caused extreme changes to the organisms in streams draining these altered landscapes. As a result, we recommend surrounding the stream with woody riparian vegetation to begin transforming Conley Run back into a natural detritus-driven ecosystem.

### **Literature Cited**

Benfield, E.F. 1996. Leaf breakdown in stream ecosystems. Pages 579-589 in F.R. Hauer and G.A. Lamberti (eds). Methods in stream ecology. Academic Press, San Diego, CA.

Hagen, E.M., J.R. Webster, and E.F. Benfield. 2006. Are leaf breakdown rates a useful measure of stream integrity along an agricultural land use gradient? Journal of the North Ameican Benthological Society 25(2):330-343.

- Sponseller R.A., and E.F. Benfield. 2001. Influences of land use on leaf breakdown in southern Appalachian headwater streams: a multiple-scaled analysis. Journal of North American Benthological Society 20(1): 44-59.
- Webster, J. R. and E. F. Benfield. 1986. Vascular Plant Breakdown in Freshwater System. Annual Review of Ecology and Systematics 17:567-594.
- Webster, J.R., E.F. Benfield, J.J. Hutchens, J.L. Tank, S.W. Golladay, and J.C. Adams. 2001. Do leaf breakdown rates actually measure leaf disappearance from streams? International Review of Hydrobiology 86:417-427.

# Professional commentary by Matthew E. McTammany, Ph.D.

My students worked extremely hard on these projects, and I hope you agree that they collected very useful and high-quality data. I revised their original submissions to include some omitted references, to explain data more accurately or completely, and to clarify certain aspects of their methods and experimental designs. Their reports appear in the preceding pages, but I wanted to provide additional interpretation of their data to summarize, highlight, and clarify some of the class's major findings from this past semester. My commentary includes the following: 1) comments on each project report, 2) additional interpretation of raw data, and 3) recommendations for management and restoration. Let me first say that my students collected all the data I will discuss, but I used some of their raw data, which may appear new. My students used some of these data for more complicated analyses and calculations, but raw data sometimes show patterns more clearly than calculated metrics can. All data from these projects are archived and available upon request.

# **Comments on Individual Projects**

Physical and chemical characteristics

This group was charged with providing general physical and chemical data to the rest of the class to support other projects. Samples were collected 2-3 times throughout the semester and therefore only represent discrete points in time at single locations in each stream. Adequate sampling of water chemistry and suspended solids would include at least monthly measurements and potentially sampling through a storm period. In the second section of my commentary, I will show more detailed data collected over 24 hours that demonstrate important and worrisome patterns of water quality in Conley Run.

Despite these limitations, Conley Run obviously has dramatically altered water quality relative to our reference stream, which should have similar water quality due to consistent size and geology between the two streams. In particular, nitrogen concentrations (as NO<sub>3</sub>) were 3x higher in Conley Run than in Stony Run. However, Stony Run also had high NO<sub>3</sub> concentrations relative to other forested streams in central PA. Streams in Buffalo Creek might receive elevated NO<sub>3</sub> deposition with precipitation. Stony Run also might receive N inputs from agricultural sources in the watershed, which are present in the watershed but not as extensive or in the immediate sampling vicinity as agriculture at Conley Run.

Neither stream had elevated phosphorus (P) concentrations, which seems non-intuitive for an extensively agricultural stream. Unfortunately, P presents unique challenges because it gets assimilated rapidly by organisms due to high demand and can adsorb onto particles in streams. Total loads of P exported from agricultural watersheds are often underestimated because water quality monitoring programs often only measure dissolved inorganic P (like our study), but most exports occur as organic or inorganic particles (e.g., dead algae, clay surfaces). Since Conley Run had higher sediment transport (TSS) and algal production than Stony Run, it seems reasonable that P export is likewise higher.

Substrate size analysis did not demonstrate dramatic differences between these two streams, but I do not feel this adequately represents the differences in streambed conditions and the obvious degradation of the stream channel in Conley Run. Conley Run was choked by heavy accumulations of silt and sand and had heavily eroded stream banks due to cows with unrestricted access to the channel. Small riffles were relatively clear of silt in Conley Run, but

runs and pools had silt several inches deep. Substrate in Stony Run was exposed cobbles and gravel, even in pools, and did not obviously suffer from heavy sediment loading. While some of this difference could be due to geological histories of the two streams, it seems that current agriculture activities in Conley Run are the primary cause of extreme sedimentation in this channel. My students used a gravelometer, which does not enable processing of particles smaller than 2 mm. In addition, my students misunderstood the particle selection procedure and grabbed the first "solid" particle they felt under their toes, which happened to be the rocks underneath the fine silt and sand on top. We were pressed for time when they conducted the substrate measurements, so I did not correct their error in the field. A more thorough and correct substrate assessment, using either gravelometry or more precise method (freeze-coring and sieving), would easily demonstrate the dramatic substrate degradation in Conley Run.

# Algal productivity

Productivity estimates were derived from artificial substrates deployed in the field to test for nutrient limitation of algal growth. Overall, algal productivity was >10x higher in Conley Run than in Stony Run, which resulted in extremely high algal biomass. Unfortunately, the nutrient supplements did not produce larger amounts of algae than non-enriched substrates, in either stream. This result was predicted for Conley Run, but algal growth in Stony Run also did not appear to be nutrient limited. I can think of 2 possible explanations for this pattern: 1) something other than nutrients is limiting algal growth in Stony Run or 2) our nutrient-diffusing substrates were not functioning properly. Light may limit algal growth and was much lower in Stony Run than in Conley Run. Light typically does not limit photosynthetic rates until light availability gets extremely low, but several studies have attributed a lack of nutrient enrichment to extreme differences in light availability between open-canopy agricultural and closed-canopy forested sites (Von Schiller et al. 2007). However, we cannot rule out that our nutrient-diffusing substrates were properly releasing nutrients to be available to the algae growing on their surfaces. These substrates could have lost all their nutrient salts before we measured algal biomass (on day 9) or might have been sealed against releasing nutrients through the pots. Agar did appear to dissolve over time, so it appears that diffusion from the substrates was occurring.

#### Stream metabolism

The extreme difference we observed in algal biomass on natural stream substrate and our artificial substrates suggested that we were likely to see major differences in ecosystem-scale primary production and respiration. Indeed, Conley Run had extremely high primary production (rate of increase of oxygen concentration as a result of photosynthesis) and respiration (rate of decrease of oxygen concentration) as a result of high algal biomass. Respiration was also high in Stony Run due to accumulations of decaying leaves. Stimulation of ecosystem metabolism is one indication of eutrophication, or the excessive productivity of algae. Eutrophication sounds like a positive pattern in that the production of algae in the stream is enhanced, but it results in serious problems in many aquatic ecosystems when the high productivity of algae exceeds the demands of consumers in the system. When this occurs, excess production can remain in the stream and be slowly decomposed, which results in depletion of oxygen, often creating anoxia. I will go into more detail about the evidence of eutrophication below.

This group also observed that 90% of respiration in Conley Run is supported by carbon fixation by algae, whereas Stony Run algae supported <10% of respiration. The transition from a stream based on detritus (like Stony Run) to a stream based on algae (like Conley Run) causes

major shifts in other microbial processes (e.g., decomposition), structure of biological communities, and dynamics of food webs and carbon processing.

# Nutrient dynamics

Nutrients (chemicals necessary to support biological organisms), especially nitrogen, were in relatively high supply in Conley Run as dissolved inorganic chemicals. These nutrients were therefore readily available to microorganisms (algae, bacteria, and fungi). However, nutrient concentrations were relatively low in Stony Run, which suggests that microorganisms may be limited by availability of nutrients. Dissolved phosphorus concentrations were not elevated in either stream, but phosphorus was likely adsorbed onto fine sediments and therefore more available in Conley Run. By adding phosphorus to each stream at a constant rate, we were able to quantify the demand for this nutrient. All uptake measurements indicate that Stony Run is more retentive of phosphorus (shorter travel distance of inorganic P, faster uptake rate of P, larger mass of P taken up per area of stream bottom) than Conley Run.

Interestingly, abiotic processes (like adsorption to particles) may contribute to measurements of phosphorus uptake. With this in mind, Conley Run, with more fine particles onto which P could adsorb, still was less retentive of P. Conley Run appears to exist in a "nutrient saturated" state, so nutrients added either experimentally or naturally through human activities are not taken up and processed by organisms in the stream without subsequently releasing nutrients already contained in sediments or biomass. On the other hand, Stony Run has the capacity to remove or retain additional nutrients and effectively buffer downstream ecosystems from nutrients added upstream. If nutrients are added to Conley Run, they would swiftly and without alteration of form or concentration, reach downstream ecosystems, including Rapid Run and probably Buffalo Creek. Other agricultural streams in Buffalo Creek watershed are also likely saturated in this manner, which seriously degrades the capacity to reduce nutrient exports until their concentrations are reduced in miles of stream or their demands are increased through improved ecosystem health.

#### Benthic macroinvertebrates

Benthic macroinvertebrates indicate a variety of stream characteristics and can be used as bioindicators of ecosystem health. As expected, Stony Run had higher diversity and abundance of pollution-sensitive organisms than Conley Run. Benthic macroinvertebrates in Conley Run also typified a sediment-impacted stream because many of the taxa collected are burrowers and thrive in streams with fine silt substrates. Even the mayflies collected in Conley Run are burrowers or sprawlers, neither of which requires clean, rocky habitats. The high abundance of non-insects and complete lack of stoneflies suggests that oxygen concentrations in Conley Run may be too low for certain macroinvertebrates. It is difficult to overlook the high densities of flatworms, isopods, and annelids, all of which indicate unhealthy conditions most likely caused by organic matter loading (cow feces, in this case). On the bright side, Conley Run supported large populations of riffle beetles (Coleoptera, Elmidae), which tend to prefer water high in dissolved oxygen. In addition, Conley Run supported large numbers of amphipods and some other shredder species. While it surprised me that the % shredders was similar between the streams, the presence of shredders in such large numbers suggests that Conley Run is fed by active springs and therefore likely remains cool, even in the height of summer. Of course, these shredders are likely feeding on algae and not leaf detritus, since there were very few leaves to contribute leaves and wood to Conley Run where we studied it. If organic matter loads from

unrestricted cow access and algal productivity from high light and nutrients could be reduced in Conley Run, the stream should be able to return to a healthier fauna, particularly if trees are planted along the stream to provide a supply of detritus to benthic organisms.

### Leaf breakdown

While leaf breakdown rates were not different between the two streams, the mechanism driving the similarity is likely quite different. Stony Run had relatively high breakdown rates due to high densities of leaf-shredding detritivorous stoneflies. Surprisingly, despite a relative lack of leaf detritus in the stream, Conley Run also had large numbers of shredders. However, these were primarily amphipods, which were likely thriving on high biomass of algae. Since amphipods are not obligate detritivores like shredding stoneflies, they can switch from algae to leaf detritus, depending on what organic matter source is most available. When we introduced leaves to Conley Run, it is possible that amphipods colonized the leaf packs and consumed the detritus, thereby causing the similarity in breakdown rates of leaves. I doubt this was the case, however, because algae are a much more nutritious source of carbon (lower C:N and C:P than leaves) and would therefore be preferred to detritus, if available. A more likely cause of similar breakdown rates is stimulation of microbe populations (bacteria and fungi) by higher nutrient availability and temperatures in Conley Run, which compensated for the lack of stonefly detritivores. We could distinguish between these competing explanations quite simply if we had sorted macroinvertebrates and counted shredders from leaf packs when we collected them. Unfortunately, time was at a premium during the semester, and we were not able to complete this aspect of the leaf breakdown study.

# **Additional Interpretations**

In my students' work to synthesize large amounts of data for their reports and descriptions of broad differences between Conley Run and Stony Run, much of the raw data must be used to determine average conditions for comparison or to calculate more complex variables. For example, measuring stream metabolism (primary production via photosynthesis and respiration) requires measuring dissolved oxygen concentrations repeatedly over short time periods for 24 hours (from midnight to midnight). However, the raw dissolved oxygen data used to calculate metabolism parameters are rarely reported and even less frequently analyzed. Unfortunately, these requirements of simpler comparisons may cause us to overlook interesting and often important variability in measured characteristics.

Oxygen concentrations fluctuate widely from nighttime to daytime because the processes controlling oxygen concentrations shift. At night, only gas exchange and respiration occur, but daytime includes these processes and photosynthesis by algae and plants. Due to the lack of photosynthesis at night, oxygen can get depleted by respiration and possibly not replaced through gas exchange, if respiration is very high or the stream is slow-flowing. Most measurements of water quality in streams do not include this type of daily fluctuation (as evidenced by the first group's dissolved oxygen measurements), and comparisons among sites (or even knowledge of a single site) depend on what time of day streams are sampled, relative to other streams.

Fortunately, we have several days of 24-hour measurements from both streams, taken on the same dates, to compare daily fluctuations in major water quality parameters, which can aid in our understanding of ecological processes and challenges in these 2 streams. Changes in dissolved oxygen are by far the most significant of these daily patterns. Oxygen concentrations are determined in surface water through a combination of 3 processes: photosynthesis,

respiration, and gas exchange or diffusion. The direction of oxygen exchange between the water and atmosphere is based on oxygen concentration relative to "saturation" concentration. Saturation concentrations are higher in cold water and low elevations (higher atmospheric pressure and oxygen content). High diffusion rates are associated with fast-flowing streams or shallow streams where the surface area of the stream is high relative to the volume of water. Conley Run is a shallow stream, but this does not result in high diffusion because it flows slowly. Instead, the shallow water and low diffusion rate in Conley Run enables biological processes of photosynthesis and respiration to have a very large effect on dissolved oxygen concentrations. Since most biological processes in small streams are associated with benthic environments (stream bottom), deeper water columns can effectively dilute the production or consumption of oxygen through photosynthesis and respiration.

In Conley Run, the shallow, slow-flowing condition of the stream results in dramatic daily fluctuation of dissolved oxygen (Figure 7.1). First, you might observe that dissolved oxygen concentrations were extremely high in Conley Run compared to both saturation and Stony Run in mid-afternoon. These supersaturated dissolved oxygen concentrations (>140% saturation) indicate that rates of photosynthesis were much higher than respiration (at that time of day) and far exceeded the diffusion rate of oxygen from the stream. Conversely, dissolved oxygen concentrations in Conley Run at nighttime were far below both saturation and Stony Run. Oxygen depletion of this magnitude (72% saturation) results from respiration occurring at such a high rate that gas exchange cannot replace the oxygen being consumed. In this case, Conley Run's low velocity reduced not only its diffusion rate but also its ability to transport materials, including dead algae. As a result, algal biomass can accumulate and decompose in the stream reach instead of being exported to downstream ecosystems with large water volumes and filter-feeders to consume the dead algae. The combination of high primary production and lack of transport capacity contributes to reveal the negative effects of eutrophication in Conley Run, namely nighttime oxygen deficits.

While other streams have shown daily fluctuations in oxygen like Conley Run, flowing water is thought to make streams resistant to eutrophication, even if algal biomass is extremely high, by enabling diffusion of oxygen when concentrations are below saturation and by transporting dead algae. Unfortunately, diffusion and water velocity in Conley Run do not seem adequate to support such high algal production without also creating lower dissolved oxygen. Of course, fluctuations themselves do not create cause for alarm, but the amount of the oxygen saturation deficit (how far below saturation the concentration is) can create problems for aquatic organisms requiring high amounts of dissolved oxygen (stoneflies, cold-water fish species). During our study, dissolved oxygen never went below 7.45 mg/L, which is high enough for even the most oxygen-sensitive, cold-water fish. However, our study was conducted during October, when cold water would cause higher dissolved oxygen concentrations than during summer when the water may warm considerably. While nighttime temperatures were similar, daily temperature fluctuation was much greater in Conley Run than in Stony Run (Figure 7.2), with Conley Run reaching almost 18°C compared to Stony Run's high of 12°C. Many trout species become physiologically stressed at 24°C and have lethal limits in the 25-30°C range. Despite being spring-fed, Conley Run may reach these temperatures during summer. Furthermore, higher warmer temperatures may lead to hypoxia and associated fish stress in Conley Run. temperature increases to 25°C and saturation deficit in Conley Run reaches 60% (quite likely, as higher temperatures cause higher respiration), dissolved oxygen would be 4.8 mg/L, which is below 5.0 mg/L, the concentration known to cause stress in most trout species. Several other

aquatic organisms (e.g., stoneflies, most caddisflies) are intolerant of low dissolved oxygen, which may explain their absence from Conley Run in our macroinvertebrate surveys.

The dramatic fluctuations in carbon dioxide concentration caused by extremely high productivity during daytime and high respiration at night can be observed in the fluctuations in pH of Conley Run (Figure 7.3). Carbon dioxide combines with water to form carbonic acid, which results in lower pH. However, carbon dioxide is consumed during photosynthesis, which raises pH in streams. Similar to dissolved oxygen, observations of pH to compare streams should account for the possibility of diel fluctuations like we observed in Conley Run. Initially, I thought Conley Run might flow through more limestone and have a limestone spring source, which could help explain its higher pH. However, at nighttime, pH was similar in Stony Run and Conley Run, which suggests that geologies were similar between the two streams and that pH in Conley Run was being influenced by photosynthesis during daytime.

### **Recommendations for Management and Restoration**

Conley Run appears to suffer from 2 primary insults related to agriculture. First, cows have unrestricted access to the stream, which contributes large fecal loads and disturbs stream banks and substrate. The results of cow access are high organic matter contamination (including fecal coliform bacteria), elevated nutrient load, collapsing and unstable stream banks, and shifting, silt substrates. These conditions are not typical of cold, spring-fed stream systems and cause greatly altered macroinvertebrate and fish communities. Secondly, agriculture along Conley Run creates conditions for eutrophication by adding nutrients (whether from fertilizers or cows) and lack of shading by woody vegetation. The combination of these properties results in extremely high algal biomass, which far exceeds consumptive capacities of macroinvertebrates and fish in the stream. Coupled with elevated temperatures and organic matter loading from cow feces, high algal biomass will lead to very high respiration and subsequent oxygen depletion.

For the particular site on Conley Run where my class worked, I strongly recommend fencing to exclude cows from all (or at least most) of the stream channel. Vegetation in the fenced area could be allowed to grow naturally, or native tree seedlings could be planted to promote healthy riparian vegetation. While guidelines for appropriate "buffer" widths are subject to interpretation, the relatively flat terrain suggests that a relatively narrow buffer of 30 feet might be enough for substantial benefit to the stream. However, the unstable stream banks caused by decades of abuse by cows may make increasing the setback of fencing a prudent decision.

Beyond proper riparian management on the reach of Conley Run we studied, I strongly recommend working with farmers throughout the Conley Run valley to improve conditions along their stream reaches. Nutrient management on all farmland, riparian management to minimize open-canopy sections, and protecting source-water and springs should be considered by everyone with property in Conley Run's watershed. Because it is a spring-fed stream, it should remain cool during summer and provide a refuge for cold-water fish from Buffalo Creek and even possibly Rapid Run. Shading the stream for its entire length would greatly help Conley Run maintain low temperatures for aquatic organisms. Reducing nutrient inputs, along with shading and cow exclusion, should reduce the build up of algae and promote healthy microbial communities. Increasing detritus inputs from woody riparian vegetation will enable obligate detritivores to recolonize the stream. Ultimately, small changes in agricultural practices through land management should accumulate to improve conditions for all organisms in Conley Run. Once healthy, Conley Run will be able to retain the limited amount of nutrients reaching the

stream, which will reduce exports to downstream ecosystems and therefore improve conditions in Buffalo Creek, Susquehanna River, and Chesapeake Bay.

#### **Literature Cited**

Von Schiller, D., E. Martí, J. L. Riera, F. Sabater. 2007. Effects of nutrients and light on periphyton biomass and nitrogen uptake in Mediterranean streams with contrasting land uses. Freshwater Biology 52: 891-906.

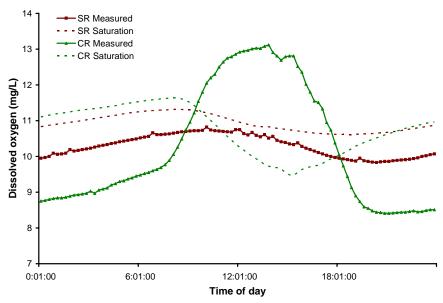


Figure 7.1. Dissolved oxygen concentrations and saturation concentrations (based on stream temperature and barometric pressure) measured on 14 October 2007 in Stony Run (SR) and Conley Run (CR).

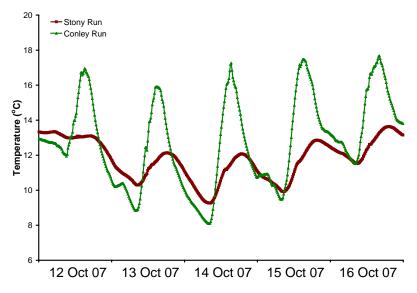


Figure 7.2. Temperature measured from 12-16 October 2007 in Stony Run and Conley Run.

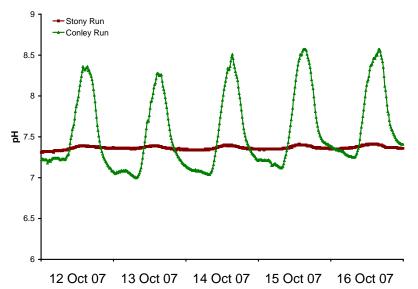


Figure 7.3. pH measured from 12-16 October 2007 in Stony Run and Conley Run.