QUANTIFYING THE FUNCTIONAL VALUE OF STREAM MITIGATION STRUCTURES ON RECLAIMED SURFACE MINES IN WEST VIRGINIA

J. Todd Petty, Gretchen Gingerich, James T. Anderson and Paul F. Ziemkiewicz

West Virginia University

ABSTRACT

Large scale surface mining in southern West Virginia causes extensive alteration of headwater streams, and there remains considerable uncertainty over whether or not mitigation structures on reclaimed mines are effective in recovering lost headwater functions. The objectives of this research were to: 1) quantify the functional value of on-site constructed stream channels (i.e., perimeter channels); 2) compare the values of perimeter channels to those of intact headwater streams (i.e., reference channels); and 3) develop ecological currencies that can be used to compare constructed perimeter channels to reference streams. We quantified the ecological value of 5 perimeter stream channels constructed on three large reclaimed surface mines in southern West Virginia. The age of each reclaimed channel varied in age from 3-20 years old. The value of these structures was compared to those at 5 intact headwater streams. Ecological response variables examined included: flow, geomorphology and habitat, water chemistry, riparian vegetation, organic matter (OM) decomposition, and biological communities.

Although dissolved metal concentrations remained relatively low, perimeter channels produced significantly higher levels of alkalinity, sulfate, specific conductivity, and total dissolved solids (TDS) as compared to intact headwater streams. Perimeter channels tended to be vegetated with obligate wetland species, creating a considerable difference between mined and reference channels with regard to vegetation, canopy cover, and structural habitat quality. Species richness of macroinvertebrates and amphibians were comparable between mined and reference channels. However, there was a distinct shift from sensitive, lotic taxa in reference channels to tolerant, lentic taxa in perimeter channels. Reclaimed perimeter channels had reduced OM decomposition rates most likely as a result of reduced mechanical abrasion and reduced microbial activity. Nevertheless, mined channels had significantly higher OM retention than reference channels, and consequently, perimeter channels exhibited significantly higher overall OM processing power than reference channels. Perimeter channels also exported significantly more dissolved organic carbon (DOC) than reference streams. As the time since reclamation increased from 3 to 20 years, we observed slight declines in conductivity, significant increases in total invertebrate richness, and significant declines in percent grassland amphibians.

Our results provide the most comprehensive information available on structural and functional characteristics of aquatic habitats on reclaimed surface mines. This study also provides information needed to identify which ecological characteristics decline in function, remain unchanged, or increase in function following mine reclamation. Calculations of ecological units identified a suite of characteristics that possessed higher functional value on mine sites as compared to reference streams, including: larval amphibian biomass, OM retention, OM processing, and DOC. The only characteristic that remained largely unchanged was total invertebrate biomass. Characteristics that exhibited considerable functional declines included: TDS generation, percent lotic amphibians, EPT richness, OM decomposition rate, total invertebrate richness, and WVSCI. These results provide a basis for making better informed decisions as it relates to mine permitting and improved mine reclamation and offsite mitigation procedures.

INTRODUCTION

Effectively managing the development of large-scale surface mines in southern West Virginia may be one of the most pressing environmental issues in the U.S. at this time. Large scale surface mining (Mountaintop Mining / Valley Fill, henceforth MTM/VF) generates substantial volumes of excess spoil that are generally placed in external fills. Individual fills may range from less than 50 acres to hundreds of acres and extend for thousands of feet down drainage.

A comprehensive Environmental Impact Study demonstrated that MTM/VF can cause alteration of natural watershed functions and ecological services (USEPA 2003a). Headwater wetlands and streams provide valuable habitat for numerous wildlife, invertebrate, and plant species (Balcombe et al. 2005a, b; Drohan et al. 2006). Moreover, headwaters provide a complex network of ecological services such as flood mitigation, nutrient and organic matter cycling, aquifer recharge, improving water quality, and providing timber and other merchantable products (Mitsch and Gosselink 2000, Meyer and Wallace 2001). Consequently, there is broad concern that a cumulative loss of headwater functions can cause unacceptable impacts to larger waterbodies downstream (US EPA 2003b, Zedler 2003, Palmer et al. 2010).

The importance of headwater streams and wetlands to the overall functioning of watersheds and downstream ecosystems are well understood (Allan 1995, Mitsch and Gosselink 2000, Meyer and Wallace 2001). Headwater ecosystems are essential to the storage, processing, and downstream transport of sediments, water, organic matter, and nutrients. The balanced delivery of processed materials from headwaters is the basis of productive, diverse communities in larger rivers and estuaries (Meyer and Wallace 2001).

In an effort to avoid cumulative impacts to watershed function, the Clean Water Act and related policies now dictate that compensatory stream and wetland mitigation must replace ecological functions that are impacted by development (National Research Council 1992, 2001). Mitigation programs in West Virginia are jointly administered by the US Army Corps of Engineers and by the WV Department of Environmental Protection. In general, the USACE requires that all mitigation requirements be met on the reclaimed mine site (i.e., on-site mitigation) and that mitigation actions seek to directly replace lost functions (i.e., in-kind mitigation). In contrast, the WVDEP requires that mitigation requirements be through stream channel restoration activities further downstream or within another watershed nearby (i.e., off-site mitigation).

There are two substantive problems with the current mitigation process. First, construction of functional stream channels on mine spoil is extremely difficult, and in some settings, is impossible. Precipitation tends to infiltrate quickly into the mine spoil. Consequently, mitigation stream channels often are dry for much of the year and perform few, if any, ecosystem functions characteristic of the intact headwater streams that they are designed to replace. Second, the effectiveness of off-site mitigation in recovering lost headwater functions is questionable. The functional value of stream restoration projects has not been properly studied locally or nationwide (Bernhardt et al. 2004, Hilderbrand et al. 2005, McClurg et al. 2007, Palmer et al. 2010). Moreover, comparisons of stream restoration projects on larger rivers to the functional values associated with headwater ecosystems have never been conducted.

Our own research over the last several years has focused on improving techniques for restoring and constructing streams and wetlands in the central Appalachian region (Balcombe et al. 2005 a, b, c, and d, Petty et al. 2005, Petty and Thorne 2005, McClurg et al. 2007, Merovich and Petty 2007). Through this research, we have developed assessment measures for improving wetland monitoring and functional assessment, conducting basic ecological wetland studies to feed into the mitigation assessment process, and developing a systematic framework for identifying suitable areas for building and restoring mitigation wetlands and mitigation wetland banks. Furthermore, we have developed methods for maximizing the watershed scale benefits of stream restoration (Petty and Thorne 2005, McClurg et al. 2007, Merovich and Petty 2007). Finally, we have developed a variety of functional ecological currencies for application to both small headwater streams and larger river segments (Petty and Thorne 2005, Merovich and Petty 2007, Poplar-Jeffers et al., 2009). An ecological currency is a measure of the functional value of a given length of stream in terms of its ability to provide ecosystem services within the watershed.

Despite a good understanding of how natural headwater streams and wetlands function, two important knowledge gaps remain. We have not effectively quantified the specific ecosystem functions that are lost during large scale surface mine development. Furthermore, we have not done a good job of quantifying the extent to which the functional values of mitigation streams and wetlands constructed on reclaimed mines compare to values characteristic of natural headwater systems. To our knowledge, there are no published studies of the ecological values of stream mitigation structures constructed on reclaimed surface mines in the Appalachian region. Consequently, the overriding objective of this research is to quantify the extent to which current mitigation practices are effective in recovering lost aquatic ecosystem functions on reclaimed mine sites. Our long-range goal is to maximize the effectiveness of "on-site" mitigation and the recovery of lost headwater functions on reclaimed surface mines in the Appalachian region.

Specifically, our objectives were to: 1) Quantify the functional value of on-site stream and wetland mitigation structures constructed on large surface mines; 2) Compare the ecological values of mitigation structures to those of intact headwater streams; 3) Determine if the functional value of on-site mitigation structures improves over time; and 4) Develop ecological currencies that can be used to relate the functional value of wetland and stream mitigation structures to intact headwater ecosystems.

METHODS

Study Area

We quantified physical, chemical, and biological characteristics of five stream channels constructed on large surface mines (henceforth "perimeter channels") located in the coal-rich region southwest of Charleston, WV (Figure 1). Within the region, typical post-reclamation surface mine structures are composed of a re-contoured and re-vegetated "on-bench" site located adjacent to valley fills. The chosen sites varied in drainage area and age since reclamation but all were designed so that any overland flow from reclaimed mine lands drained towards the perimeter of the site and into the perimeter channels. All sites drained towards an "on-bench" outflow notch in the berm that surrounded the reclaimed surface mine perimeter (Figure 2).

Study reaches within the sites began one retention cell above the outflow notch and continued upstream a length of 10 times the mean channel width.

These perimeter channels were constructed primarily for the purpose of slowing and retaining water to allow the settling-out of suspended sediments and some ions from the water column before discharge of water off-site. Information on estimated age since reclamation was included in the study (Table 1), however, no information on the site's history of maintenance dredging and re-contouring since reclamation was obtained. Therefore, overall age since reclamation may not represent a continuous successional trajectory. Additionally, perimeter channel sites were not necessarily designed as ecological structures.

Reference sites consisted of five intermittent streams within the region that represented the best known sites available within a reasonable proximity to perimeter channel sites. Reference sites were selected to be completely unaffected by surface mining or other known stressors (e.g. human residences or roads). We tried to avoid any influence of historic underground mining; however, this is very difficult in this region due to numerous old, small "punch-hole" mines (ultimately it turned out that one of our reference sites exhibited signs of mild acid mine drainage; UNT to White Oak Creek). Sites were selected using winter and spring 2008 water chemistry measurements as well as topographic maps and general knowledge of the area. Reach length was measured at 10 times the mean stream width with a minimum length of 50 m (Table 1).

Physical Habitat

Habitat quality assessments were performed using classification systems that included: Virginia Unified Stream Method (USACOE 2007), West Virginia Functional Channel Unit Assessment (USACOE & VADEQ 2007), Wildland Hydrology's Bank Erosion Hazard Index (Rosgen 2001), Environmental Protection Agency Rapid Bioassessment Protocol (Barbour et al. 1999), and Ohio Rapid Assessment Method (Mack 2001). These systems use simple measurements and visual assessments to assign numerical scores to each site. Categorical habitat qualifiers (e.g. excellent, good, poor) were derived from calculated scores.

Vegetation was sampled according to protocols adapted from Batzer et al. (2004), Balcombe (2005, b), and Rentch et al. (2008). Thirty meter long transects were run across the site perpendicular to the length of the stream or perimeter channel. A pin flag was used every 2 m along transects to record any intercepting vegetation. Additionally, fifteen 1x1 m plots were placed along each transect. Within each plot, percent cover was estimated using 14 categories (bare ground, cattail, exposed substrate, fern, forb, grass, moss, open water, rush, sedge, shrub, submerged vegetation, tree, and vine). All coarse woody debris within the plot was tallied and recorded. Canopy cover was estimated using a spherical densitometer. Diameter at breast height (dbh) and species was recorded for any trees > 2.5 cm dbh and within 10 m of the transect. Any additional species not captured in survey measurements were noted.

Water Chemistry, Temperature, and Discharge

Sites were visited seasonally from February 2008 to May 2009 (a full list of sample dates and parameters measured is given in Table 2). Instantaneous water chemistry measurements were taken at each visit with a Yellow Springs Instrument (YSI) 650 multi-parameter probe equipped with a 600XL sonde. The YSI was calibrated before every site visit. The probe measured temperature, pH, conductivity, and dissolved oxygen.

Seasonal water samples were taken to analyze dissolved water chemistry including metals and nutrients following protocols by Merovich et al. (2007). Grab samples were taken by completely filling a sampling bottle under the water's surface. Filtered samples were taken using a vacuum pump and mixed ester cellulose membrane 0.45 µm filter. Nitric acid (HNO₃) was added to keep all solutes in solution. Analysis was performed by the National Research Center for Coal and Energy at West Virginia University, Morgantown, WV. Alkalinity was measured in CaCO₃ equivalents and presented as mg/L. If samples were measured below the method detection limit (MDL), one half of the MDL was used in analysis. Samples measuring below MDL are presented as "MDL" in figures. Mean seasonal data was calculated using three seasons only (spring, summer, and winter) because of the absence of water during autumn sampling.

Specific limits for parameters were taken from West Virginia guidelines for warm water fisheries and comply with EPA regulations. For limits dependent on hardness, a hardness of 0-50 mg/L was used to determine limits. Total dissolved solids (TDS) was calculated by summing seasonal measurements for dissolved ions.

Hourly temperature readings were taken from June 2008 until June 2009 using HOBO U22 Water Temp Pro v2 loggers (Onset Computer Corporation, Pocasset, MA). No temperature readings were recovered from P_WO before the temperature logger was buried by bulldozer activity. Loggers for R_WO and R_MW ceased logging prior to June 2009 and resulted in incomplete data. Data from periods when loggers were inundated were used for analysis.

Discharge was calculated from width, depth, and flow measurements taken at each site visit. Flow was measured using a Flow Mate 2000 flow meter. Measurements were summarized by season for analysis.

Amphibian and Macroinvertebrate Assemblages

Amphibian assemblages were sampled seasonally in early and late spring and early and late summer (March, May, June, and July) of 2008 (Table 2). Amphibian sampling sought to maximize the diversity captured for each site as well as sample during all potential breeding seasons for probable amphibian species. Adult assemblages were estimated using Visual Encounter Surveys (VES) performed in accordance with protocols set by Crump and Scott (1994). Larval Surveys were adapted from methods by Shaffer et al. (1994). The surveys were comprised of consistent meter-long sweeps with a D-frame net in open-water and consistent meter by half-meter area searches in stream channels. Each search was done at 10 random locations over the length of the site.

Macroinvertebrate assemblages were sampled in spring and fall 2008 (Table 2). Within lotic systems, macroinvertebrates were sampled using protocols established by Merovich and Petty (2007), which are slight modifications of procedures described by the West Virginia Department of Environmental Protection's Watershed Assessment Program and the EPA's Rapid Bioassessment Protocols for wadeable streams (WVDEP 1996, Barbour et al. 1999). Within each reach, four representative riffles were sampled using a 250 µm D-frame kicknet. Within lentic systems, a D-frame net was used to take jab samples at 10 random locations along the reach. Lentic samples were taken according to protocols set by King and Richardson (2002) and Balcombe et al. (2005a). Samples were preserved with 70% ethanol and identified to the lowest possible taxonomic level. Fall 2008 samples were obtained in perimeter sites only due to low water in reference sites. Consequently, we analyzed data from spring samples only.

Stream condition was derived from macroinvertebrate samples using a modified West Virginia Stream Condition Index (WVSCI) score developed by Merriam (2009) from original Gerritsen et al. (2000) protocols. This index uses family-level community metrics to categorize stream condition as either poor, marginal, good, or excellent.

Organic Matter Processing

OM transport was measured using methods adapted from Speaker et al. (1984), Webster et al. (1994), Raikow et al. (1995), and Lamberti (1996). Cumulative OM retention was measured using artificial sticks, consisting of painted dowel rods, and instantaneous retention was measured using artificial leaves, consisting of rectangles of blue construction paper. Fifty dowel rods were placed in a riffle at the upstream end of the reach. They remained on-site and the cumulative distance they traveled was measured at four intervals over 195 days (on the day of release and then after approximately 1 month, 2 months, and 6 months). Twenty artificial leaves were also placed in a riffle at the upstream end of each reach seasonally. They were allowed to travel downstream for 30 minutes and then their distances traveled were recorded.

Retention rate was calculated for each site using the cumulative distances traveled by artificial sticks. The equation $T_d = T_o \, e^{-kd}$, where T_d is the percentage of released sticks remaining in transport at distance d, T_o is the original number of sticks released, and k is the instantaneous rate of retention, was used to calculate retention rate (Speaker et al. 1984, Raikow et al. 1995, Minter 2009).

Seasonal water samples were taken from each site in order to quantify dissolved carbon concentrations. Samples were filtered with a 0.45 µm filter and treated with nitric acid (HNO₃). Samples were analyzed using a Sievers 5310c laboratory TOC Analyzer to estimate total dissolved carbon and dissolved organic carbon concentrations.

Leaf litter packs were constructed from plastic mesh bags (10 mm mesh size) for the purpose of quantifying leaf litter decomposition rates at each study site. Bags were filled with 10 g of *Quercus palustris* (pin oak) leaves collected after abscission and air-dried for approximately two weeks to a constant mass. Bags were grouped in sets of six and anchored in riffles throughout the reach length. An additional set of litter bags was taken to the site and returned to the lab to calculate for handling loss. Litter bags were randomly sampled after 45, 75, 90, 120, 195, and

325 days on site. Bags were returned to the lab on ice and rinsed in a 250 μ m sieve. Macroinvertebrates present in litter bags were collected, preserved, and identified. Leaf litter was placed in brown paper bags and dried for approximately 48-72 hours to a constant mass. After drying, leaf litter was reduced to particulate size and subsampled. A subsample of 250 μ g was placed into pans and incinerated to determine the ash free dry mass. Methods were adapted from protocols by Benfield (1996). Decomposition rates (-k) were determined from the linear regression of the plot of the number of days of exposure versus the log-transformed percent ashfree dry mass (% AFDM). AFDM was calculated using the following formula from Benfield (1996):

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(Equation 1) % Organic Matter = (DM_{sample} - AM_{sample}) / DM_{sample} \times 100
AFDM = DM x % Organic Matter
% AFDM remaining = 100 - ((initial - final) / initial \times 100)
Where: DM = dry mass; AM = ash mass
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The slope of the line after regression provided the rate of decomposition (k). Expected decomposition rates for *Quercus* range from k=(0.0014) to k=(0.021) (Beiser et al. 1991, D'Angelo & Webster 1992).

Organic matter processing power was defined as the ability of a site to retain and process organic matter locally. Processing power of each site was calculated by multiplying the instantaneous rate of decomposition by the instantaneous rate of retention.

Statistical Analysis

Because pre-mining reference sites were intermittent streams and post-reclamation perimeter channels sites resembled wetlands, measured parameters differed in terms of how directly comparable they were. Some parameters such as water chemistry and decomposition rate were directly comparable between site types (Fig. 3). However, some parameters such as gradient and vegetation community may not be directly comparable between site types.

Ecological units (EUs) (Petty & Thorne 2005, Merovich & Petty 2007) were calculated for parameters selected as important metrics for both perimeter and reference site types (i.e., functional metrics that have important meaning, such as OM decomposition rate, regardless of whether they were sampled in a lotic or a lentic environment) (Figure 3). They were calculated by dividing the perimeter channel mean by the reference mean for each parameter. EUs with values greater than one represent ecosystem parameters with higher values on the perimeter channel sites than in reference catchments. EUs with values less than one represent ecosystem parameters with higher values in reference streams than in perimeter channels.

Prior to analyses, data were transformed using log_{10} , arcsine, and square roots of measured values where needed in order to approximate normality within the data. Correlation analysis was run on all parameters to quantify relationships between ecosystem parameters and both conductivity and time since reclamation (with respect to perimeter channels only). T-tests were used to test for statistical differences in ecosystem parameters between site types (perimeter and reference). Repeated measures ANOVAs were used to test for seasonal variation in ecosystem

parameters between perimeter and reference sites. ANCOVAs were used to test for interactive effects of site type and specific conductivity on ecosystem parameters.

Relationships among macroinvertebrate and amphibian communities and environmental variables were examined using non-metric multidimensional scaling (NMDS). NMDS is a statistical ordination, developed by Clarke (1993). This non-parametric analysis involves iterative solutions that allow species composition data to be plotted in ordination space with reduced stress (McCune & Grace 2002). This analysis enables the determination of community similarities as well as the influence of environmental variables on community composition. Additionally, Spearman rank correlations were run between NMDS scores and community metrics (Merovich & Petty 2007, McClurg et al. 2007, Merriam 2009).

All statistical analyses were conducted using the program R Project for Statistical computing version 2.8.1 (R Development Core Team 2008) unless otherwise stated. NMDS analysis was run using the package vegan (Oksanen et al. 2008). All values were considered significant at an alpha level of 0.05 unless otherwise indicated.

RESULTS

Water Temperature, Dissolved Oxygen and Discharge

We detected few significant differences in water temperature regimes between perimeter channels and reference channels (Table 3, Figure 4). Maximum, mean, and variance of daily water temperatures were relatively similar between the two stream types. Only minimum daily temperature differed between perimeter and reference channels (Table 3). Overall, dissolved oxygen concentrations and discharge did not differ significantly between perimeter and reference channels (Table 3). Perimeter channels did exhibit significantly higher spring discharge than reference channels and this contributed to an overall difference in discharge between channel types (Table 3).

Habitat Quality

Habitat quality measures were consistently higher in reference channels than in perimeter channels regardless of the assessment protocol (Table 3). EPA rapid bioassement protocol (RBP) scores averaged 78 for perimeter channels and 150 for reference sites (Table 3). Mean scores for the Virginia unified stream method (VA USM) were 4 for perimeter channels and 6 for reference sites. West Virginia functional channel unit (WV FCU) assessment scores averaged 3 for perimeter channels and 9 for reference sites. In the case of Wildland Hydrology's bank erosion hazard index (BEHI), the higher the score, the more prone a site is to erosion. BEHI scores averaged 23 for perimeter channels and 39 for reference sites (Table 3).

The Ohio Rapid Assessment Method (ORAM) version 5.0 was designed to rank overall wetland quality and to categorize natural wetlands based on amount of disturbance (Mack 2001). The higher the score, the less disturbed the wetland. Perimeter channels averaged an ORAM score of 35 and reference sites averaged 61 (Table 3).

Vegetation

Perimeter channel sites were dominated by forb (22%), grass (22%), cattail (21%), and open water (13%) (Table 3). Reference sites were dominated by bare ground (29%), forb (22%), trees (16%), and fern (11%). Mean percent canopy cover was 4% for perimeter channels and 91% for reference sites (Table 3). Perimeter channels had, on average 0.3 ± 0.6 trees and 0.1 ± 0.2 tree species per km² survey versus an average of 9.2 ± 5 trees and 2.5 ± 0.7 species per km² in reference sites. All vegetation measures were significantly different between site types except for percent open water and forb (Table 3).

Water Chemistry

In general, pH. dissolved metals and nutrients did not differ significantly between perimeter and reference channels (Table 3). Despite a lack of statistical differences in these attributes overall, we did observe several notable trends. First, low pH was characteristic of one of the reference channels (R_WO, unnamed tributary of White Oak Creek), and all reference channels possessed relatively low pH during the summer sampling period (Figure 5). Second, dissolved metal concentrations tended to vary widely among the perimeter channels and the highest metal concentrations were nearly always observed on reclaimed mines rather than reference channels. For example, elevated Al (Figure 6), Ni, Mn (Figure 7), Se (Figure 8), and Zn were observed on the White Oak mine site relative to other mines and reference channels. Furthermore, Cd, Co, Cr, Cu, and Se (Figure 8) tended to be elevated on the Stanly Branch mine site. Third, extremely high concentrations of nitrate were observed in the White Oak mine site (P_WO) in summer and autumn (Figure 9) probably as a result of fertilizer application during mine reclamation.

Several water quality characteristics did exhibit significant differences between perimeter and reference channels (Table 3), including alkalinity (Figure 10), acidity, conductivity (Figure 11), total dissolved solids (TDS), calcium, magnesium, and sulfate (Figure 12). All variables, except acidity, were significantly elevated in the perimeter sites relative to reference streams (Table 3).

A key result of our analyses was the relative consistency of dominant ions present in the perimeter channels (Figure 13). TDS in the perimeter channels was dominated by sulfate along with very consistent percent contributions of calcium and magnesium (Figure 13). One of the reference sites (R_WO) exhibited water chemistry attributes consistent with acid mine drainage, which is characterized by high sulfate concentrations and an absence of bicarbonate (Figure 13). Another reference site (R_HC) exhibited evidence of residual waste from gas drilling, which is characterized by high concentrations of sodium and chloride (Figures, 13, 14 and 15). The perimeter channel site, P_BH, also showed evidence of drilling waste (Figures 13-15).

Amphibians

Perimeter channels contained, on average, more larval amphibians than reference sites (19 vs. 8 individuals) and supported about the same number of species (2 species) (Table 4). Mean larval biomass also averaged higher in perimeter channels (1.33 g/m²) than reference sites (0.05 g/100m²) (Table 4). Perimeter channels contained, on average, less adult amphibians than reference sites (5 vs. 28 individuals) but supported a similar number of adult species (2 vs. 3

species) (Table 4). When the effects of conductivity were removed, larval amphibian richness, total number of larval amphibians, and the percent of lotic amphibians was statistically different between site types with perimeter channels supporting more larva and reference sites supporting more lotic species (Table 6). Overall density was not statistically different between perimeter channels and reference sites (Tables 3 and 4, Figure 16). However, the lack of difference was driven largely by high amphibian densities on one perimeter channel site, P_AR (Figure 16). Amphibian survey data can be found in Appendices C-E.

Perimeter channels supported primarily terrestrial and aquatic frogs that use lentic systems. Reference sites supported primarily aquatic salamanders that use lotic systems (Tables 4, 6 and 7). The species of both site types were able to use forest habitats but perimeter channels supported a statistically significant higher percentage of grassland species (Table 4). Additionally, perimeter channels supported a significantly lower percentage of lotic species and a significantly higher percentage of lentic species (Table 4).

NMDS ordination revealed clustering by site types. Amphibian community structure was primarily influenced by vegetation and water chemistry parameters including percent open water, grass, cattail, bare ground, canopy cover, fern, and number of species per km² (Figure 17). Increasing percent open water, grass and cattail indicated a perimeter- type amphibian community while increasing percent bare ground, canopy cover, fern, and species per km² indicated a reference- type community structure. Statistically significant water chemistry influences included mean specific conductivity, sulfate, magnesium, mean total dissolved solids, calcium, alkalinity, and iron (Figure 18). Increasing measures of all these parameters indicated a perimeter- type community composition.

Additionally, ANCOVA analysis revealed significant interactions between site type and site conductivity for amphibian species richness, larval richness, total number of amphibians, mean number of amphibians, number of larval amphibians, mean density, and percent forest amphibians (Table 6, Figures 19-21).

Macroinvertebrates

Perimeter channels and reference sites had similar macroinvertebrate family richness and biomass (Tables 3 and 8). Perimeter channels had a higher percentage of tolerant species (70% vs. 42%) and chironomids (58% vs. 32%) than reference sites (Table 8). Perimeter channels had a significantly lower percentage of Ephemeroptera, Plecoptera, and Trichoptera (EPT) (5%) than reference sites (48%) and a significantly lower EPT richness (1 vs. 4) (Table 8). These parameters, along with WVSCI score, were significantly different with regards to mean conductivity (Table 7). Additional ANCOVA analysis revealed significant interactions between site type and site conductivity for percent EPT, EPT richness, and total richness (Table 7, Figures 22-24).

Macroinvertebrate data were used to rank the quality of site habitat using the West Virginia Stream Condition Index (WVSCI). WVSCI ranks overall stream quality based on measures of the benthic invertebrate community. Perimeter channels had an average WVSCI score of 48 (Poor) and reference sites had an average score of 68 (Marginal). Excluding R_WO, the mean

score for reference sites was 74 (Good). Perimeter channels ranged from poor to marginal and reference sites ranged from poor to excellent (Table 8).

Both perimeter channel and reference sites were dominated by collector-gatherer functional feeding groups (Table 9). Perimeter channels were composed of 74% collector-gatherers, 8% predators, 8% omnivores (primarily planktonic species), and 2% shredders. Reference sites were composed of 57% collector-gatherers, 27% shredders, 5% predators, and 3% omnivores. Perimeter channel communities were composed of primarily lentic-inhabiting species while reference communities were primarily lotic-inhabiting species. Macroinvertebrate abundance data are reported in Appendices F-H.

NMDS analysis showed clustering of sites by site type with P_AR tending to have a community type more similar to reference sites and R_HC having a community type more similar to perimeter sites (Figure 25). P_WO and R_WO were somewhat separate from the rest of the sites and plotted out in the positive NMDS 1 and positive NMDS 2 quadrant Additionally, WVSCI score, percent tolerant taxa, percent EPT, EPT richness, percent unknown feeding group, percent shredder, and percent omnivore were correlated with community composition (Figure 25).

Organic Matter Processing and Dissolved Carbon

Retention was significantly higher in perimeter channels than in reference sites for measures of mean cumulative stick distance traveled, mean cumulative stick distance per day, number of sticks exiting the reach, and percent of sticks retained (Tables 3 and 10). No movement of artificial sticks was recorded in any perimeter channel sites except P_WO, which was the youngest perimeter channel site. Mean cumulative distance traveled by artificial sticks was 0.00 m/day in perimeter channels, excluding P_WO, and 0.06 m/day in reference sites.

Organic matter decomposition rates tended to be lower in perimeter channels than in reference channels (Tables 3 and 11, Figure 26) Perimeter channels averaged $47 \pm 2\%$ loss of organic matter after about 325 days, whereas reference sites averaged $62 \pm 19\%$ loss (Table 11). Furthermore, when the effects of conductivity were removed mean total weight of litter bags after 325 days was significantly different between site types (Table 6, Figure 27). The mean calculated decomposition rate for perimeter channels was -0.00213 \pm 0.00038 and the mean rate for reference sites was -0.00348 \pm 0.00196 (Figure 26). Processing power, which was calculated by multiplying retention rate times decomposition rate, averaged 0.013 in perimeter channels and 0.007 in reference sites (Table 11).

Decomposition rate was positively correlated with WVSCI score, percent EPT, percent unknown invertebrates, total number of adult amphibians, total number of adult amphibian species, mean percent of organic matter lost, and mean cumulative stick distance (Table 12). Percent shredders was not correlated with decomposition as expected (Table 12). Additionally, ANCOVA analysis revealed significant interactions between site type and site conductivity for decomposition rate, mean percent organic matter remaining, and mean percent of organic matter lost.

Perimeter channels averaged higher dissolved organic carbon (DOC) concentrations than reference sites (Table 13, Figure 28). Mean DOC for three seasons with water was 3.27 ± 2.09

mg/L for perimeter channels and 1.51 ± 0.64 mg/L for reference sites. Seasonal DOC was not correlated with seasonal discharge as expected (McDowell & Likens 1988c, Collier et al. 1989, Hinton et al. 1997, Meyer et al. 1998, Dawson et al. 2002, Spencer et al. 2007). Total dissolved carbon was significantly higher in perimeter channels than reference sites for all seasons (Tables 3 and 13, Figure 29).

Effect of Mine Age on Ecosystem Attributes

Overall, we observed very few significant correlations between mine age (i.e., time in years since reclamation) and various aquatic ecosystem attributions. Significant correlations were observed for percent cattail, percent fern, tree density, conductivity (Figure 30), total macroinvertebrate family richness (Figure 31), and total adult amphibian species richness.

Ecological Units

EUs were calculated for parameters deemed to be important for characterizing functional differences between perimeter and reference channels (Table 15). EUs were calculated by dividing the perimeter channel mean by the reference channel mean. Values exceeding one indicate conditions where perimeter channels may be interpreted as having a higher "function" than the reference channel. Values less than one indicate a higher functional value in reference channels. We calculated EUs assuming the overall reference mean and assuming an "idealized" reference mean, which was calculated after removing R_WO and R_HC. R_HC was the stream that showed evidence of contamination from gas drilling waste, and R_WO exhibited characteristics of a stream impacted by acid mine drainage.

To summarize, perimeter channels functioned best as habitat for larval and lentic amphibians, as highly retentive systems capable of *in situ* processing of organic matter, and as systems capable of storing high concentrations of dissolved carbon (Table 15). Perimeter channels functioned poorly as habitat for lotic amphibians and sensitive macroinvertebrate stream taxa. Perimeter channels also functioned poorly as sources of low TDS and low conductivity freshwater (Table 15).

DISCUSSION

Key Concerns

Elevated conductivity appeared to be the primary determinant of reduced biological conditions and ecosystem processes in reclaimed perimeter channels. Although not previously indicated in published literature, we observed a significant effect of site type and conductivity on OM decomposition rate. Overall analysis indicated that conductivity may be influencing decomposition rate in perimeter channels, possibly through reduced microbial activity. Simon et al. (2009) found differences in stream OM decomposition rates along a pH gradient. They determined that reduced decomposition was the result of altered microbial assemblages and reduced microbial activity under low pH conditions (Simon et al. 2009). Additional studies have linked reduced microbial biomass and respiration with reduced decomposition rates within

acidified streams (Mulholland et al. 1987, Griffith & Perry 1994, Meegan et al. 1996, Niyogi et al. 2001). Although the sites in this study did not suffer from reduced pH (except for R_WO), increased conductivity may have similar effects on microbial community structure, biomass, or respiration.

Within the central Appalachian region, the order Ephemeroptera usually account for 25-50% of the total spring macroinvertebrate community in relatively undisturbed headwater streams (Pond et al. 2008). Ephemeroptera have also been found to show the greatest response to increases in specific conductivity in waters affected by surface mining within the region (Hartman et al. 2005, Pond et al. 2008). Consistent with previous studies, the relatively undisturbed reference sites contained a significantly higher percentage of EPT than perimeter channel sites. Additionally, a significant interaction between site type and conductivity was shown for WVSCI score, percent EPT, EPT richness, and total invertebrate richness. EPT species are considered taxa indicative of good water quality and many cannot be supported in perimeter channel sites due to conversion from lotic to lentic conditions in addition to elevated TDS and specific conductivity (Pond et al. 2008). One possible mechanism for this is the relationship between elevated conductivity and interference with the osmoregulation of macroinvertebrates (Wichard 1973, McCulloch 1993).

The shift in community composition from a community supporting a large percentage of shredders to a community supporting a large percentage of collector gatherers (primarily chironomids) may have downstream implications. Shredders play an important role within the aquatic continuum by feeding on coarse particulate organic matter (CPOM) and converting it to fine particulate organic matter (FPOM) (Cummins & Klug 1979). FPOM, in turn, is exported as a food resource base for collector-gatherers (Short & Maslin 1977). If the shredder community is lost or reduced, shortcomings in trophic linkages may affect the entire aquatic ecosystem.

Elevated conductivity levels also had a significant influence on amphibian assemblage composition. Specifically, a strong interaction was shown between conductivity and site type for the numbers of amphibians (primarily larva), overall richness, amphibian density, and percent of forest-utilizing species. Most of these metrics are associated with the larval component of the amphibian population. These metrics are quantitative indicators of the shift in community compositions between site types. Differences in water chemistry such as elevated sulfate, calcium, magnesium, and alkalinity, however, did not seem to deter amphibians from using these sites overall.

The difference in number of adults versus larval amphibians at the two site types can be explained by the differences in the communities that both inhabit and use the aquatic features. Reference sites were inhabited by stream salamanders that live and breed within the stream. The difference in quantity of adults and larva at these sites may be due to the ability to more easily locate and capture the larger, adult salamanders as well as the relatively small number of eggs deposited by these salamander species (~20) (Green & Pauley 1987). The large quantity of larval versus adult amphibians in perimeter channels may be explained by the utilization of these sites by both lentic-using species as well as tree frogs inhabiting adjacent, intact forests that utilize these lentic sites for breeding. Both lentic frogs and tree frogs may lay as many as 1000-2000 eggs (Green & Pauley 1987). Frog larva are more easily captured than stream salamander

larva as they tend to congregate at pond margins and are more visible. Additionally, frog adults are harder to locate and capture than their larva.

Ecological Units

To assist with quantitative calculation of on-site shortcomings, we calculated ecological units (EUs) to represent a proportional difference in measures found at perimeter channel sites versus those found at reference sites. The differences represent aspects of the original site that have been lost and would be difficult to restore on-site because of the conversion of low TDS lotic channels to high TDS lentic channels. Overall the EUs show that perimeter channels were functionally similar to reference channels in terms of amphibian biomass and OM processing. Substantial shortcomings were present with regards to species composition shifts, losses of sensitive invertebrate taxa, and overall invertebrate taxa richness. Attempts to compensate for shortcomings captured in the ratios by re-constructing stream structure on-site are ill-advised because of the difficulties associated with constructing a lotic system "from scratch" (Palmer et al. 2009). Therefore, perimeter channels should be designed as lentic systems and EUs should be applied to sites outside the mine permit boundary on an aquatic surface area basis (Merovich & Petty 2007). For example, to compensate for a reduction in overall WVSCI score, off-site mitigation projects to enhance lotic habitat for invertebrates can be conducted at a rate of 1 meter of mitigation for every 0.71 m of perimeter channel. The application of EUs at a watershed scale may allow off-set of functional and structural losses that occur despite reclamation efforts.

Reclamation Successes

Perimeter channel and reference sites supported two very different amphibian community types, however, overall diversity and number of species supported was comparable. Perimeter channels supported a majority of generalist species, such as *Rana clamitans* (green frog) and *Notophthalmus v. viridescens* (red spotted newt), and tree frog larva such as *Hyla chrysoscelis* (Cope's gray tree frog). Reference sites supported stream salamanders, primarily *Desmognathus monticola* (seal salamander) and *Desmognathus fuscus* (northern dusky salamander).

Overall, preferences of the species themselves to use or inhabit lentic versus lotic waters were the driving factors of community composition. Primarily those species that prefer lotic habitats were found in reference sites and those species preferring lentic habitats for breeding or feeding were found in perimeter channel habitats. However, the vegetational differences between these site types may be the second most important characteristic. The amphibian community structure is highly correlated to the type and quality of vegetation present. Young, sparsely vegetated perimeter channels (such as P_WO) did not support larval amphibians despite being equally as close to intact forest as other perimeter channel sites.

In terms of number of species, both site types supported an average of two larval species and approximately two adult species. Overall diversity remained the same; however, there was an unmistakable shift in community type. Lotic and forest species were replaced by grassland-inhabiting, lentic species. Perimeter channels supported the larva of forest species such as *Hyla chrysoscelis* (Cope's gray tree frog) and *Pseudacris c. crucifer* (northern spring peeper). These sites most likely benefited from close proximity to intact forest (Hecnar & M'Closkey 1996,

Stevens et al. 2002). Constructed wetlands have been reported to be colonized by ubiquitous anurans such as gray tree frog, American toad, spring peeper, and *Rana catesbeiana* (American bullfrog) within two years of creation (Perry et al. 1996, Mierzwa 2000, Pechmann et al. 2001). All of the perimeter channels in this study were older than three years since reclamation and showed colonization by amphibians. No positive correlation between amphibian community metrics and age since reclamation was found. Negative correlations were shown between total adult amphibian species and percent of grassland amphibians and age since reclamation. This means that the number of adult amphibians, presumably grassland species, reduced over time. It is difficult to determine why this might have occurred by sampling five perimeter channels. As the perimeter channels age, there may be a significant change in an unmeasured parameter such as mean water depth or cattail density that dissuaded use by grassland adults.

The colonization by amphibians shortly after wetland establishment is consistent with patterns found in accidentally formed and constructed wetlands (Kent & Langston 2000, Pollio 2005). Although other studies found greater numbers of species in these wetlands than we found (Pollio 2005), this study found six of nine species expected to occur in grassland areas. Additionally, studies have shown the number of amphibian species found in newly created pools was positively correlated with the distance of these pools to forests (Laan & Verboom 1990). As the perimeter channels in this study were not intentionally designed as wetlands to support amphibians, considering wetland habitat during perimeter channel construction may lead to an increase in the number of species using the wetlands, as long as minimal distance to intact forest is maintained.

Perimeter channels and reference sites had similar macroinvertebrate family richness and biomass but significantly different community composition. Perimeter channels overall, however, supported a comparable number of macroinvertebrates and number of species. Perimeter channels were dominated by lentic species, such as Odonates, and were dominated by collector-gathers (primarily chironomids). Reference sites were dominated by lotic species and had a higher percentage of shredders than perimeter channel sites.

Mean decomposition rates for both site types were within ranges suggested by Beiser et al. (1991) and D'Angelo and Webster (1992). OM decomposition rates showed a significant overall effect of site type and mean conductivity. Decomposition rates were not, however, correlated with parameters known to affect decomposition such as temperature, nutrient concentration, hydrology, dissolved oxygen, percentage of shredders, or pH (Whiles & Wallace 1997, Graça et al. 2001, Swan 2004, Simon et al. 2009). However, these parameters were also not significantly different between site types with the exception of summer pH and spring discharge. Although discharge did not differ between site types, hydrology in terms of lentic versus lotic systems did. Therefore, it is likely that, the difference in aquatic system type itself affects the potential breakdown rate between perimeter channel and reference site types.

Field observations over the course of the study support the idea that the lotic component to sites contributed to their overall loss of organic matter. The position of the bags and securing rope in relation to the rebar stake, the amount of movement from the original placement, the degree of distress to the mesh bag, and the integrity of the remaining material indicate that the majority of material lost from reference site bags was lost through mechanical abrasion.

The difference in the degree of water flow between site types is evidenced by differences in organic matter retention. No movement of artificial sticks or leaves was recorded at any perimeter channel sites except P_WO (which has the highest gradient of perimeter channels). Of the perimeter channel sites, P_WO also had the highest decomposition rate. Overall distance traveled by artificial sticks and leaves was greater in reference sites than in perimeter channels.

Winter DOC levels in perimeter channels were statistically higher than in reference sites, despite a loss of original topsoil. Because of the relation between DOC and soil type and chemistry (McDowell & Likens 1988, Dawson et al. 2002, Ankers et al. 2003), it is unlikely that the DOC in perimeter channel systems originates from the soil of the reclaimed site. Perimeter channel DOC is also unlikely to originate from leaf litter inputs as found by Hongve (1999). Perimeter channels have a low percent canopy cover and are usually at or above the canopy height of adjacent intact forest.

This knowledge, along with comparable processing power rates between site types, indicates that the high DOC concentrations within perimeter channels is most likely originating from on-site inputs of detritus from aquatic macrophytes. Specifically, the high percent cover emergent vegetation and higher retention capability of perimeter channels allows organic material to be effectively cycled within the perimeter channels. Anderson and Mitsch (2006) found that the percentage of soil organic matter content within riverine wetlands increased about one percent every three years. This concurs with correlations in this study between age since reclamation and percent cattail. The high retention ability of these sites combined with increasing cattail coverage may lead to increased mean DOC concentrations with age since reclamation. This trend is seen in all but the oldest site (P_BH).

Perimeter channels overall resemble wetlands more than streams. Because wetlands of this type are uncommon within West Virginia, it may be difficult to evaluate their function as wetlands. An expected range of DOC levels, especially, may be difficult to evaluate without local reference wetlands because of the complexity of parameters that determine DOC concentration as well as the natural DOC flux inherent from environmental conditions. For instance, Mann and Wetzel (1995) found DOC levels fluctuated naturally with seasonal macrophyte growth and bacterial production within riverine wetlands. Additionally, geographic region, elevation, season, and degree of exposure may influence local DOC fluctuation.

Seasonal DOC concentration was not correlated with seasonal discharge as expected (McDowell & Likens 1988c, Collier et al. 1989, Hinton et al. 1997, Meyer et al. 1998, Dawson et al. 2002, Spencer et al. 2007). This may be due to the mining disturbance at perimeter channel sites resulting in large, reclaimed catchments with relatively low OM inputs from tree canopies. Reference site concentrations were within the range of 0.673 - 2.94 mg/L for forested watersheds within the region (Meyer & Tate 1983; Tate & Meyer 1983). Perimeter channel mean concentrations were less than annual mean concentrations of 9.8 ± 1.5 mg/L for wetland-dominated watersheds reported by Eimers et al. (2008). Both mean concentrations, however, are comparable to mean annual concentrations ranging from 7.1 - 48.2 mg/L within the ponded portion of riverine wetlands (Mann & Wetzel 1995).

Are Ecological Functions Reclaimed Locally?

The process of site reclamation is intended to offset any geomorphic or ecological losses (Bradshaw 1984, Holl 2002). Although full re-creation of the original topography of an area during reclamation is considered the geomorphic ideal, it is not always appropriate, or feasible, in steeper areas (Nicolau 2003). Ecologically, Hilderbrand et al. (2005) urge the setting of realistic restoration goals and argue that "scientifically defensible end points of functional or structural equivalence" need to be set. In the case of mountaintop removal mining in West Virginia, the site is converted from a steep, forested headwater stream to an unforested site with rolling terrain and wetland-like aquatic features. Despite this conversion, the question remains if ecological functions such as supporting biological communities and downstream energy export are adequately reclaimed.

Functional downstream export of carbon is supported by the on-site generation of DOC. Higher retention capabilities of perimeter sites allows for increased opportunity for decomposition. Overall processing power is comparable in perimeter sites and reference sites. Mechanical breakdown of OM is lost, but decomposition rates are comparable.

In terms of biotic communities, both amphibians and macroinvertebrates showed similar diversities on mined and unmined sites. However, communities shifted from lotic communities supporting sensitive taxa to lentic communities supporting generalists and tolerant taxa. Any retention of biotic communities by perimeter channels may be favorably influenced by the proximity of intact forest. These areas may encourage biotic use of perimeter sites and act as a source for colonizers.

Watershed Scale Perspective

The change in site type from forested stream to perimeter channel resulted in a shift in vegetation, amphibian, and macroinvertebrate composition that cannot be reclaimed on-site. The new system cannot support the same community types. In response to the loss of pre-existing communities, off-site mitigation that supports healthy, native biotic communities must be pursued.

Additionally, the shift in communities from lotic, sensitive taxa to lentic, generalist taxa may become problematic as the cumulative effects from stream to stream and watershed to watershed are considered over a region (Lowe & Bolger 2002, Lowe et al. 2006, Pond et al. 2008). Disturbance at a local scale may influence the ability of populations to re-colonize at a regional scale (Lowe & Bolger 2002). Although prevention of species loss at a local scale may not be possible, it can be prevented at the watershed scale. Consideration must be given to protect portions of a mined watershed to act as sites for wildlife protection and source populations for continued re-colonization (Lowe et al. 2006, Pond et al. 2008). Consideration must be given both to the extent of the watershed affected and to the life histories of at-risk species to determine regional habitat needs (Lowe et al. 2006). Ensuring the connectivity of first-order streams may be essential for ensuring the survival of some species, such as *Gyrinophilus porphyriticus* (spring salamander) (Lowe & Bolger 2002, Lowe et al. 2006).

The change in water chemistry resulting in increased alkalinity, manganese, TDS, calcium, magnesium, sulfate, and specific conductivity also cannot be reclaimed on-site. This increase in water parameters can be a compounded problem in the watershed as a whole depending on the extent to which the watershed is mined. Pond et al. (2008) found that the evidence of mining and reclamation on the water chemistry (especially specific conductivity) was greatly reduced in watersheds that contained a higher percentage of unmined tributaries. The best way to handle changes in water chemistry resulting from watershed scale disturbance may be dilution at a watershed scale, primarily through planned protection of headwater streams (Saunders et al. 2002. Lowe et al. 2006). By preserving a percentage of tributaries within the mined watershed as undisturbed sites and sources of dilute water, cumulative downstream changes in water chemistry may be avoided (Saunders et al. 2002, Merriam 2009). This may be an especially pertinent solution because mining and its effects may not be the only stressor to local watersheds within the region. Historical mining and non-residential development may compound watershed-wide stresses to ecological function (Merriam 2009).

Reference Site Conditions

Reference sites were selected for this study based on winter and spring water chemistry measures, topographic maps, and a general knowledge of the area. The reference streams selected were known to drain watersheds with no surface mining activities and no residential development. Preliminary measures of water quality indicated that all streams were in reasonably good condition for streams in this region.

Unfortunately, two of the reference sites, R_HC and R_WO, later displayed water chemistry that was less than ideal. R_WO had lower than average pH as well as higher acidity, aluminum, manganese, calcium, manganese, conductivity, and sulfate than other reference sites. The water chemistry of this site is indicative of streams impacted by acid mine drainage (Merovich et al. 2007). Consequently, this site is presumed to be influenced by historic underground mining. It is not clear why initial water quality measures did not show signs of impairment. However, we know from studies in other watersheds that water quality in streams impacted by acid mine drainage (AMD) can vary significantly from one season to the next (Merovich et al. 2007). The poor water quality conditions of R_WO likely contributed to measures of organic matter decomposition, and amphibian and macroinvertebrate diversity that were substantially lower than those of other reference sites.

Another reference site, R_HC, was impacted by some unknown, non-mining related disturbance upstream in summer 2008. During this period, a natural gas line and access road were installed and waste water from gas well drilling may have been introduced to the stream. Prior to disturbance, R_HC possessed water quality characteristics very similar to the other high quality reference sites. Following disturbance, all pore spaces within the R_HC stream bed were filled with sediment resulting in loss of habitat for amphibians and macroinvertebrates. Additionally, R_HC water chemistry measures showed an increase in nitrite, conductivity, barium, sodium, and chloride.

Although R_HC and R_WO possessed less-than-ideal biotic and abiotic conditions, these sites were included in most analyses comparing un-mined reference stream channels to reclaimed mine perimeter channels. Excluding them would have reduced our sample size from five to three and made direct comparisons between site types difficult. In addition, we believe that the range of conditions observed at reference sites is representative of streams draining watersheds that have not been surface mined (Minter 2009, Merriam 2009). Because of the topography and geology of the area, it is likely that one in five watersheds in the region will be affected by legacy impacts from underground mining and increasingly streams are being impacted by gas extraction or other non-mining related disturbance (Merriam 2009).

Additional Questions

Previous literature has emphasized the connectivity of upstream functional and ecological processes to downstream ecosystem function and value (Vannote et al. 1980, Gomi et al. 2002, Lowe et al. 2006, Meyer et al. 2007, Wipfli et al. 2007). If downstream functions and values are to be maintained, the watershed function as a whole must be considered (Lowe & Bolger 2002, Saunders et al. 2002, Lowe et al. 2006, Pond et al. 2008). The functions considered by this study were the downstream export of energy in terms of on-site OM retention and decomposition and off-site DOC export and the support of biologic community structure and diversity. Additional studies may seek to understand possible losses in function of the downstream export of particulate organic matter (POM), emphasized by Vannote et al. (1980), and gaps in ecological function originating from the physical gap between perimeter channel off-site outflow points and native stream channels.

Native channels act as a source of coarse and fine particulate organic matter (CPOM/FPOM) for downstream trophic webs (Cummins & Klug 1979, Vannote et al. 1980, Cummins et al. 1989, Wallace et al. 1997). This is generated by macroinvertebrate activity as well as mechanical breakdown of organic matter. Since perimeter channels have a different macroinvertebrates community than reference sites, and because the mechanical component caused by lotic waters is lost, there may be an additional functional loss of CPOM/FPOM export. Additional studies may investigate potential shortcomings in CPOM and FPOM production and export.

The reclaimed perimeter channels in this study consisted of retained water prevented from escaping off the mine perimeter by berms. These features drained towards a central point where an off-site flow was created by perforating the berm. The water was then allowed to drain off-site, downhill and rejoin native streams. The physical area in between the point where water exits the reclaimed site and rejoins native channels may act as an additional site of disturbance as no native channel exists and exported water creates a new channel. Future studies may seek to measure the differences in water chemistry between the uphill off-site flow point and the junction where the flow joins native channels. Specifically, what changes in DOC may occur between those two points? Additionally, does the transitional zone allow for increases or decreases in TDS, total suspended solids (TSS), and specific conductivity?

The reclaimed perimeter channels in this study resemble wetlands. However, no comparison was made between the ecological function of these sites as wetlands and the ecological function of similar wetlands within the region. Rough comparisons can be made via published literature.

Overall, DOC concentrations and decomposition rates were comparable to published values (Beiser et al. 1991, D'Angelo & Webster 1992, Mann & Wetzel 1995). The DOC concentrations and decomposition rates expected for wetlands within the study site may differ, however, because of regional factors such as elevation, growing season, geology, etc. Future studies may seek to compare perimeter site function to the function of comparable wetlands within the region.

This study also did not investigate differences in the functions of perimeter channels in terms of their overall construction and design. Comparison to native wetlands may aid in guiding design suggestions such as recommended percent of open water and water depth. At this point, variation exists from site to site in terms of how mine sites are reclaimed and how perimeter channels are designed. These differences seem to originate from both the time period of the reclamation and the company performing the reclamation. Future studies may seek to discover if increases in functional recovery can be gained simply by perimeter channel design. Specifically, can conductivity and TDS be further reduced on-site and can intentional design increase the diversity and structure of biotic communities?

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Table 1. Summary characterists of study sites. P = reclaimed mine perimeter sites and R = reference channel sites. Approximate age is the number of years since the mine was reclaimed. "na" = not applicable. DA = drainage area.

Site Names	Site Code	Approx. Age (years)	Mean Discharge (m³/s)	Calculated DA (ha)	Latitude (DD)	Longitude (DD)
White Oak	P_WO	3	0.0159	45	38.04778	-81.52139
Argus	P_AR	5	0.0013	6	37.98972	-82.25222
Stanley Branch	P_ST	10	0.0116	33	38.08306	-81.93472
Sugartree	P_SU	10	0.0130	37	38.09007	-81.95751
Big Horse	P_BH	20	0.0030	12	38.08500	-81.89750
UT Hell Creek	R_HC	na	0.0005	7	37.73044	-82.23232
UT Lukey Fork	R_LF	na	0.0028	10	38.05944	-81.95306
UT Mud Creek East	R_ME	na	0.0011	5	38.04647	-81.91148
UT Mud Creek West	R_MW	na	0.0029	11	38.06105	-81.94331
UT White Oak	R_WO	na	0.0044	16	38.05250	-81.52278

Table 2. Sampled parameters for reclaimed mine perimeter channels and reference sites with sampling dates.

Response Variables	Sample Dates
Water Temerature (°C)	June 08 - June 09 (continuous)
Discharge (m ³ /s)	February 08 - May 09 (4 seasonal samples)
Water Chemistry	February 08 – May 09 (4 seasonal samples)
Habitat Quality	June 08 (single sample)
Vegetation	June 08 (single sample)
Macroinvertebrates	May 08 (single sample)
Amphibians	March - July 08 (4 samples at 6 wk intervals)
Organic Matter Retention	March 08 - February 09 (4 seasonal samples)
Organic Matter Decomposition	May 08 - February 09 (4 seasonal samples)

Table 3. Mean (S.E) for various aquatic ecosystem attributes quantified at perimeter channel and reference channel sites. Also presented are statistics for T-tests comparing attributes between site types (d.f.=8). *p=<0.05, **p=<0.005, ***p=<0.001.

Response Variables	Perimeter	Reference	T-Stat
Temperature and DO			
Max Daily Temp (°C)	30.7 (3.2)	31.9 (2.4)	-0.29
Min Daily Temp (°C)	0.15 (0.08)	0.00 (0.00)	2.28*
Mean Daily Temp (°C)	10.5 (0.2)	9.6 (0.7)	1.12
CV for Mean Daily Temp	63.6 (3.5)	58.9 (4.8)	0.80
Dissolved Oxygen (mg/L)	8.7 (0.5)	9.9 (1.0)	-1.11
Discharge			
Annual Mean Discharge (m³/s)	0.009 (0.003)	0.002 (0.001)	2.16*
Habitat Assessment			
EPA RBP	78 (6)	150 (10)	6.22***
VA USM	4 (0)	6 (0)	-5.34***
WV FCU	3 (0)	9 (0)	-14.67***
ВЕНІ	23 (4)	39 (3)	-3.29*
ORAM	35 (6)	61 (4)	-3.59*
<u>Vegetation</u>			
Pct Bare Ground	3 (2)	29 (8)	-3.22*
Pct Cattail	21 (7)	0 (0)	3.06*
Pct Fern	0 (0)	11 (2)	-5.19***
Pct Forb	22 (8)	22 (5)	0.03
Pct Grass	22 (4)	2(1)	5.52***
Pct Open Water	13 (6)	0 (0)	2.04*
Pct Tree	1 (1)	16 (3)	-4.18**
Pct Vine	2(1)	8 (2)	-3.20*
Species per km ²	0.1 (0.1)	2.5 (0.3)	-9.01***
Trees per km ²	0.3 (0.3)	9.2 (2.2)	-6.33***
Pct Canopy Cover	4 (4)	91 (1)	-26.10***
Water Chemistry			
pH	7.4 (0.1)	6.7 (0.4)	1.56
Alkalinity (mg/L)	138 (13)	5 (1)	12.82***
Acidity (mg/L)	0 (0)	13 (5)	-4.88**
Conductivity (µS/cm)	2197 (414)	461 (326)	3.29*
Total Dissolved Solids (mg/L)	25.2 (3.0)	2.7 (0.4)	6.81***

Response Variables	Perimeter	Reference	T-Stat	
	162 (21)	21 (15)	4 O Calcula	
Ca (mg/L)	163 (31)	21 (15)	4.96**	
Cl (mg/L)	13.8 (9.7)	11.4 (10.2)	0.45	
Mg (mg/L)	154 (34)	9 (4)	6.12***	
Na (mg/L)	13.2 (6.3)	23.7 (22.2)	0.09	
$SO_4 (mg/L)$	1008 (196)	32 (20)	7.71***	
Al (mg/L)	0.1 (0.0)	0.5 (0.5)	-0.87	
Ba (mg/L)	0.016 (0.002)	0.093 (0.060)	-1.72	
Cd (mg/L)	0.007 (0.000)	0.008 (0.000)	-0.63	
Co (mg/L)	0.011 (0.002)	0.009 (0.001)	1.00	
Cr (mg/L)	0.006 (0.001)	0.009 (0.001)	-2.51*	
Cu (mg/L)	0.008 (0.001)	0.008 (0.001)	0.20	
Fe (mg/L)	0.10 (0.04)	0.06 (0.00)	1.03	
Mn (mg/L)	0.3 (0.2)	0.4 (0.3)	0.20	
Ni (mg/L)	0.034 (0.019)	0.024 (0.011)	0.48	
Se (mg/L)	0.024 (0.002)	0.030 (0.005)	-1.02	
Zn (mg/L)	0.020 (0.008)	0.042 (0.031)	-0.45	
NO_2 (mg/L)	0.04 (0.03)	3.10 (3.08)	-0.94	
NO_3 (mg/L)	11.0 (10.4)	0.8 (0.2)	0.88	
TP (mg/L)	0.08 (0.04)	0.05 (0.01)	0.66	
NH_3 (mg/L)	0.007 (0.003)	0.013 (0.002)	-1.97	
<u>Macroinvertebrates</u>				
WVSCI Score	48 (5)	68 (8)	-2.02*	
Pct Chironomid	58 (16)	32 (11)	1.42	
Pct Tolerant	70 (15)	42 (13)	1.56	
Pct EPT	5 (4)	48 (16)	-2.48*	
EPT Richness	1 (0)	4(1)	-2.67*	
Total Richness	8 (2)	7 (1)	0.53	
Total Inverts	763 (273)	213 (87)	1.81	
Pct 2 Dominant Sp	78 (10)	66 (6)	1.24	
Total Biomass (g/m ²)	31.8 (18.9)	34.5 (14.9)	-0.09	
Pct Collector- Gatherer	74 (12)	57 (9)	1.33	
Pct Filterer	2 (2)	0 (0)	1.32	
Pct Scraper	6 (4)	3 (3)	0.53	
Pct Shredder	2 (2)	27 (11)	-2.18*	
Pct Predator	8 (3)	5 (3)	0.82	
Pct Omnivore	8 (5)	3 (3)	0.78	

Response Variables	Perimeter	Reference	T-Stat
Amphibians			
Number Total Species	3 (1)	3 (0)	-0.60
Number Larval Species	2(1)	2 (0)	0.20
Number Adult Species	1 (0)	2 (0)	-2.19*
Larval Biomass (100g/m ²)	0.0133 (0.0103)	0.0005 (0.0002)	1.25
Total Density (#/m ²)	1.2 (0.7)	1.8 (0.7)	-0.64
% Grassland	58 (18)	0 (0)	2.76*
% Forest	95 (5)	100 (0)	-1.00
% Lotic	5 (3)	86 (13)	-5.11***
% Lentic	89 (8)	54 (8)	3.06*
,, 2011.0	0, (0)	0.(0)	2.00
Organic Matter Processing			
OM Decomp Rate	0.0021 (0.0002)	0.0035 (0.0009)	-1.52
Total Wt (g) (after 325 d)	6.7 (0.9)	5.3 (1.6)	0.73
% Organic (after 325 d)	85 (7)	78 (6)	0.57
Organic Mass (g) (after 325 d)	5.3 (0.1)	3.8 (0.9)	1.76
% Organic Mass Lost (after 325 d)	47 (1)	62 (9)	-1.74
Leaf Transport (m)	0.52 (0.52)	0.42 (0.15)	-0.67
Stick transport (m)	7.44 (7.44)	26.89 (4.81)	-2.94*
Stick Transport / Day	0.04 (0.04)	0.14 (0.02)	-2.94*
OM Retention Rate	-0.064 (0.014)	-0.020 (0.005)	2.07*
% Sticks Retained	87 (13)	59 (9)	2.55*
# Sticks Exiting Reach	6 (6)	21 (5)	-3.76*
Dissolved Organic Carbon (mg/L)	3.5 (0.9)	1.5 (0.3)	2.02*
Total Dissolved Carbon (mg/L)	27.0 (5.0)	3.7 (0.7)	9.70***
Processing Power * 100	0.013 (0.003)	0.007 (0.002)	1.63

Table 4. Amphibian abundance survey totals, for four sample periods, observed on reclaimed mine perimeter channels and reference sites. Frog and salamander species' preference for grassland or forest was based on information from Green and Pauley (1987). Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p<0.05 in t-tests) is indicated by **bold**.

Site Code	Total Larva	Larval Species	Total Adults	Adult Species	Total Individuals	Total Species	Pct Grassland	Pct Forest	Pct Lotic	Pct Lentic
P_WO	0	0	6	2	6	2	100	100	17	100
P_AR	63	5	17	2	80	5	89	100	0	81
P_ST	9	1	3	1	12	1	25	100	8	100
P_SU	12	4	1	1	13	4	69	77	0	62
P_BH	12	2	0	0	12	2	8	100	0	100
R_HC	3	1	8	2	11	2	0	100	100	55
R_LF	8	2	45	4	53	5	0	100	98	57
R_ME	13	2	69	2	82	3	0	100	100	44
R_MW	14	2	15	2	29	3	0	100	100	83
R_WO	1	1	2	2	3	3	0	100	33	33
Perimeter	19 ± 25	2 ± 2	5 ± 7	1 ± 1	25 ± 31	3 ± 2	58 ± 40	42 ± 10	5 ± 7	89 ± 17
Reference	8 ± 6	2 ± 1	28 ± 28	2 ± 1	36 ± 32	3 ± 1	0 ± 0	100 ± 0	86 ± 30	54 ± 18

Table 5. Larval amphibian biomass $(g/100m^2)$ for four sampling occasions observed on reclaimed mine perimeter channels and in reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Site Code	March	May	June	July	Total	Mean
P_WO	0.00	0.00	0.00	0.00	0.00	0.00
P_AR	6.91	2.91	8.22	3.37	21.41	5.35
P_ST	0.00	0.06	0.00	0.00	0.06	0.01
P_SU	0.00	0.01	0.10	0.09	0.20	0.05
P_BH	0.00	4.87	0.11	0.00	4.99	1.25
R_HC	0.00	0.06	0.00	0.00	0.06	0.02
R_LF	0.05	0.14	0.02	0.00	0.21	0.05
R_ME	0.06	0.10	0.08	0.00	0.23	0.06
R_MW	0.08	0.20	0.02	0.11	0.41	0.10
R_WO	0.00	0.00	0.05	0.00	0.05	0.01
Perimeter	1.38 ± 3.09	1.57 ± 2.23	1.69 ± 3.65	0.69 ± 1.50	5.33 ± 9.24	1.33 ± 2.31
Reference	0.04 ± 0.04	0.10 ± 0.08	0.03 ± 0.03	0.02 ± 0.05	0.19 ± 0.14	0.05 ± 0.04

Table 6. ANCOVA analysis of the effects of site type, conductivity, and their interaction on various ecological measures in reclaimed mine perimeter channels and reference sites. Degrees of freedom = 7. Statistical significance is indicated by *=<0.05, **=<0.005, ***=<0.001.

Parameters	Type	Cond	Type x Cond
Decomp Rate	-0.085	-1.020	3.417*
Mean Total Wt (g) (325 d)	3.394*	4.829**	-1.895
Mean % Organic (325 d)	-2.162	-1.972	5.035**
Mean Organic Mass (g) (325 d)	0.615	1.837	0.744
Mean % Org Mass Lost (325 d)	-0.854	-0.854	4.168**
WVSCI Score	-0.900	-2.673*	5.089**
Pct Chironomid	0.214	1.123	0.030
Pct Tolerant	0.428	1.518	-0.120
Pct EPT	-0.440	-2.420*	2.463*
EPT Richness	-0.877	-3.551**	3.820**
Total Richness	-1.789	0.133	3.318*
Total Inverts	-1.185	-0.520	1.759
Pct 2 Dominant Sp	0.241	1.035	1.056
Total Biomass (g/m²)	-0.756	-0.935	1.682
Pct Collector- Gatherer	0.175	1.010	0.762
Pct Filterer	-0.465	0.206	0.285
Pct Scraper	-1.636	-1.562	2.009
Pct Shredder	0.593	-0.566	0.601
Pct Predator	0.110	0.606	0.118
Pct Omnivore	-0.989	-0.700	1.180
Pct Unknown	0.915	-0.405	0.416
Amphibian Richness	-1.738	-2.502*	5.533***
Larval Amphibian Richness	0.027*	0.018*	4.775**
Adult Amphibian Richness	0.703	-0.432	1.228
Total No Amph	-1.331	-2.007	3.062*
Mean No Amph	-1.331	-2.007	3.062*
Total No Adult Amph	-0.054	-1.265	1.738
Mean No Adult Amph	0.102	-1.163	1.598
Total No Larval Amph	-2.465*	-2.337	3.511**
Mean No Larval Amph	-1.674	-0.739	-1.469
Total Larval Amph Biomass (100g/m²)	-1.558	-1.058	1.694
Mean Larval Amph Biomass (100g/m²)	-1.558	-1.058	1.694
Mean Amph Density (ind/m ²)	-1.326	-2.003	3.059*
Pct Grassland Amph	-1.018	0.392	0.643
Pct Forest Amph	1.429	1.061	3.041*
Pct Lotic Amph	2.472*	-0.003	0.079
Pct Lentic Amph	-0.723	1.009	1.269

Table 7. Frog and salamander species expected (Exp) to occur in grassland and forest in southwestern West Virginia, based on Green and Pauley (1987) compared to those actually observed (Obs) as (a) adults during visual encounter surveys (VES) (seen or heard), in (l) larval surveys, or (b) for both larval and VES. The preceding "p" indicates individuals encountered in perimeter sites and "r" indicates occurrence within reference sites.

		Gras	sland	Forest	
Aquatic Salamanders		Exp	Obs	Exp	Obs
Appalachian Seal Salamander	Desmognathus monticola			X	r.a
Eastern Hellbender	Cryptobranchus alleganiensis			X	
Midland Mud Salamander	Pseudotriton montanus			X	
Mudpuppy	Necturus maculosus	X		X	
Northern Dusky Salamander	Desmognathus fuscus			X	r.b
Northern Red Salamander	Pseudotriton ruber	X		X	
Northern Two-lined Salamander	Eurycea bislineata			X	r.a
Red-spotted Newt	Notophthalmus v. viridenscens	X	p.b	X	
Southern Two-lined Salamander	Eurycea cirrigera		_	X	r.l
Spring Salamander	Gyrinophilus porphyriticus			X	r.a
Terrestrial Salamanders					
Cumberland Plateau Salamander	Plethodon kentucki			X	
Four-toed Salamander	Hemidactylium scutatum			X	
Green Salamander	Aneides aeneus			X	
Jefferson Salamander	Ambystoma jeffersonianum			X	
Longtail Salamander	Eurycea longicauda	X		X	
Marbled Salamander	Ambystoma opacum			X	
Ravine Salamander	Plethodon richmondi			X	
Redback Salamander	Plethodon cinereus			X	
Slimy Salamander	Plethodon glutinosus			X	
Spotted Salamander	Ambystoma maculatum			X	
Wehrle's Salamander	Plethodon wherlei			X	
Ambystoma species	Ambystoma sp.		p.l	X	
Aquatic Frogs					
Bullfrog	Rana catesbeiana	X	p.a	X	
Greenfrog	Rana clamitans	X	p.b	X	
Pickerel frog	Rana palustris	X	p.a	X	
Northern Leopard Frog	Rana pipiens	X		X	
Terrestrial Frogs					
Eastern American Toad	Bufo americana	\boldsymbol{x}	p.l		
Eastern Spadefoot	Scaphiopus holbrookii			X	
Fowler's Toad	Bufo woodhouseii				
Gray Treefrog	Hyla chrysoscelis		p.l	X	
Mountain Chorus Frog	Pseudacris brachyphona			X	
Northern Peeper	Pseudacris c. cricifer		p.l	X	r.a
Wood Frog	Rana sylvatica			X	

Table 8. Macroinvertebrate measurements from reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p < 0.05 in t-tests) is indicated by **bold**.

Site Code	WVSCI Rating	WVSCI Score	Pct Chironomid	Pct Tolerant	Pct EPT	EPT Richness	Total Richness	Total Inverts	Pct 2 Dominant	# 1 Dominant	# 2 Dominant
P_WO	Poor	33	94	100	0	0	2	1329	98	Chironomidae	Unknown Diptera
P_AR	Marginal	64	10	40	19	1	8	86	39	Snails	Baetidae
P_ST	Marginal	56	29	29	8	1	7	191	75	Cyclopoida	Chironomidae
P_SU	Poor	44	85	87	0	2	10	1384	92	Chironomidae	Cyclopoida
P_BH	Poor	44	72	92	0	0	13	823	86	Chironomidae	Oligochaeta
R_HC	Poor	52	38	58	6	1	6	47	62	Chironomidae	Cyclopoida
R_LF	Excellent	88	0	11	89	6	7	145	57	Ameletidae	Peltoperlidae
R_ME	Good	80	20	23	74	4	7	301	69	Peltoperlidae	Chironomidae
R_MW	Good	76	34	38	52	7	10	512	57	Chironomidae	Ameletidae
R_WO	Poor	45	68	82	18	2	4	60	87	Chironomidae	Capniidae/Leuctridae
Perimeter	-	48 ± 12	58 ± 37	70 ± 33	5 ± 8	1 ± 1	8 ± 4	763 ± 611	78 ± 23	-	-
Reference	-	68 ± 19	32 ± 25	42 ± 28	48 ± 35	4 ± 3	7 ± 2	213 ± 195	66 ± 12	-	

Table 9. Percent of macroinvertebrates by feeding guild observed on reclaimed mine perimeter channels and reference sites. Guilds include collector gatherer (CG), filterer (FI), scraper (SC), shredder (SH), predator (PR), omnivore (OM), and unknown (UN). Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Site Code	CG	FI	SC	SH	PR	OM	UN
P_WO	94	0	0	0	6	0	0
P_AR	30	0	21	9	21	19	0
P_ST	70	2	2	0	4	22	0
P_SU	93	0	6	0	1	0	1
P_BH	82	8	2	0	8	0	0
R_HC	55	0	17	6	2	17	2
R_LF	61	0	0	20	14	0	6
R_ME	25	0	0	69	0	0	5
R_MW	61	0	0	22	6	0	10
R_WO	82	0	0	18	0	0	0
Perimeter	74 ± 26	2 ± 3	6 ± 9	2 ± 4	8 ± 8	8 ± 11	0 ± 0
Reference	57 ± 20	0 ± 0	3 ± 8	27 ± 24	5 ± 6	3 ± 8	5 ± 4

Table 10. Mean organic matter transport distances and retention rate for reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p < 0.05) is indicated in **bold**.

Site Code	Mean Cum Stick Distance (m)	Mean Cum Stick Dist/Day	Retention Rate	Pct Sticks Retained	Dowels Exiting Reach	Gradient
P_WO	37.2	0.2	-0.0089	36	32	6
P_AR	0.0	0.0	-0.0782	100	0	1
P_ST	0.0	0.0	-0.0782	100	0	1
P_SU	0.0	0.0	-0.0782	100	0	1
P_BH	0.0	0.0	-0.0782	100	0	1
R_HC	11.7	0.1	-0.0367	84	8	17
R_LF	30.2	0.2	-0.0204	64	18	6
R_ME	22.9	0.1	-0.0216	66	17	16
R_MW	28.6	0.1	-0.0147	52	24	8
R_WO	41.1	0.2	-0.0066	28	36	7
Perimeter	7.4 ± 16.6	0.0 ± 0.1	-0.0644 ± 0.0310	87 ± 29	6 ± 14	2 ± 2
Reference	26.9 ± 10.7	$\textbf{0.1} \pm \textbf{0.1}$	-0.0200 ± 0.0111	59 ± 21	21 ± 10	11 ± 5

Table 11. Mean total weight (g), mean organic (g) and inorganic mass (g), percent organic, percent organic mass lost, decomposition rate (k) and processing power observed on reclaimed mine perimeter channels and reference sites after \sim 325 days of exposure. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p < 0.05) is indicated in **bold**.

Site Code	Days of Exposure	Total Mass	Organic Mass	Inorganic Mass	% Organic	% Oganic Mass Lost	Decomposition Rate	Process Power *100
P_WO	199	10.2	5.6	4.6	57	45	-0.00268	0.002
P_AR	325	5.8	5.1	0.7	88	50	-0.00231	0.018
P_ST	328	6.0	5.5	0.5	92	45	-0.00167	0.013
P_SU	329	5.6	5.2	0.4	92	49	-0.00205	0.016
P_BH	325	5.7	5.4	0.3	94	47	-0.00194	0.015
R_HC	200	11.0	6.0	5.0	57	41	-0.00248	0.009
R_LF	327	1.4	1.1	0.3	82	89	-0.00666	0.014
R_ME	329	3.4	2.9	0.5	85	71	-0.00380	0.008
R_MW	328	5.1	3.8	1.4	75	63	-0.00297	0.004
R_WO	326	5.6	5.3	0.3	94	48	-0.00149	0.001
Perimeter		6.7 ± 2.0	5.3 ± 0.2	1.3 ± 1.8	85 ± 16	47 ± 2	-0.00213 ± 0.0004	0.013 ± 0.006
Reference		5.3 ± 3.6	3.8 ± 1.9	1.5 ± 2.0	78 ± 14	62 ± 19	-0.00348 ± 0.0020	0.007 ± 0.005

Table 12. Significant (p < 0.05) correlation of biological parameters with decomposition rates. Mean and standard error are given in the first two rows.

Response Variables	Perimeter Average	Reference Average	Correlation with Decomp
<u>Macroinvertebrates</u>			
WVSCI Score	48 (5)	68 (8)	0.75
Pct EPT	5 (4)	48 (16)	0.86
EPT Richness	1 (0)	4(1)	
Pct Predator	8 (3)	5 (3)	
Pct Unknown	0 (0)	5 (2)	0.83
<u>Amphibians</u>			
Total Adult Amph Sp	1 (0)	2(0)	0.78
Total No Adult Amph	5 (3)	28 (13)	0.80
Pct Lotic Amph	5 (3)	86 (13)	
Pct Lentic Amph	89 (8)	54 (8)	

Table 13. Dissolved organic carbon measures (mg/L) for reference sites and reclaimed mine perimeter channels. Reference sites did not contain water at the time of autumn sampling. Autumn samples for P_ST were contaminated. Site mean is the mean of spring, summer, and winter only. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p < 0.05) is indicated in **bold**.

Site Code	Spring	Summer	Autumn	Winter	Site Mean
P_WO	0.42	1.10	1.29	1.18	0.90
P_AR	5.71	1.53	2.99	3.20	3.48
P_ST	7.74	2.34	-	2.94	4.34
P_SU	8.99	7.49	8.01	1.72	6.07
P_BH	1.72	1.47	4.41	1.52	1.57
R_HC	0.32	0.27	-	0.67	0.42
R_LF	3.43	1.42	-	0.98	1.94
R_ME	2.61	0.86	-	0.92	1.46
R_MW	2.77	1.39	-	1.18	1.78
R_WO	3.37	1.29	-	1.12	1.93
Perimeter	4.92 ± 3.73	2.79 ± 2.65	4.18 ± 3.10	2.11 ± 0.90	3.27 ± 2.09
Reference	2.50 ± 1.27	1.05 ± 0.49	-	$\boldsymbol{0.97 \pm 0.20}$	1.51 ± 0.64

Table 14. Total dissolved carbon measures (mg/L) for reference sites and reclaimed mine perimeter channels. Reference sites did not contain water at the time of autumn sampling. Autumn samples for P_ST were contaminated. Site mean is the mean of spring, summer, and winter only. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation. Statistical significance (p < 0.05) is indicated in **bold**.

Site Code	Spring	Summer	Autumn	Winter	Site Mean
P_WO	44.4	26.3	2.0	2.1	24.3
P_AR	28.1	18.9	2.9	2.9	16.6
P_ST	26.1	33.7	-	3.0	20.9
P_SU	22.8	29.0	5.1	5.8	19.2
P_BH	13.6	18.3	3.3	3.1	11.7
R_HC	1.2	2.1	-	1.1	1.5
R_LF	5.3	3.2	-	1.4	3.3
R_ME	4.0	3.7	-	1.5	3.0
R_MW	4.1	3.1	-	1.6	2.9
R_WO	4.1	1.7	-	1.6	2.5
Perimeter	27.0 ± 5.0	25.2 ± 3.0	3.3 ± 0.6	3.4 ± 0.6	18.5 ± 2.1
Reference	3.7 ± 0.7	2.7 ± 0.4	-	1.4 ± 0.1	2.6 ± 0.3

Table 15. Ecological units (EU) ratios, EU_I ratios (calculated using ideal reference means), perimeter means, reference means, and ideal reference means for reclaimed mine perimeter channels and reference sites. Biological parameters are standardized by the area of aquatic feature sampled. EU ratios greater than 1.0 represent conditions where the values observed in the perimeter channels exceeded those observed in the reference channels.

Response Variables	Perimeter	Reference	Ideal Reference	EU Ratio	EU _I Ratio
Mean Larval Amph Biomass (100g/m²)	1.33 (1.03)	0.05 (0.02)	0.07 (0.02)	26.60	19.00
Retention Rate	-0.0644 (0.0139)	-0.0199 (0.0049)	-0.0189 (0.0021)	3.24	3.41
Mean DOC (mg/L)	3.51 (0.94)	1.51 (0.28)	1.73 (0.14)	2.32	2.03
Processing Power *100	0.013 (0.003)	0.007 (0.002)	0.009 (0.003)	1.86	1.44
Pct Lentic Amph	89 (8)	54 (8)	61 (11)	1.65	1.46
Total Invertebrate Richness	8 (2)	7 (1)	8 (1)	1.14	1.00
Total Invert Biomass (g/m²)	31.8 (18.9)	34.5 (14.9)	44.2 (22.3)	0.92	0.72
WVSCI Score	48 (5)	68 (8)	81 (3)	0.71	0.59
Decomp Rate	0.0021 (0.0002)	0.0035 (0.0009)	0.0045 (0.0011)	0.57	0.44
EPA RBP	78 (6)	150 (10)	134 (1)	0.52	0.58
EPT Richness	1 (0)	4 (1)	6 (1)	0.25	0.17
Mean Conductivity (µS/cm)	2197 (414)	461 (326)	61 (2)	0.21	0.03
Pct EPT	5 (4)	48 (16)	72 (11)	0.10	0.07
Pct Lotic Amph	5 (3)	86 (13)	99 (1)	0.06	0.05

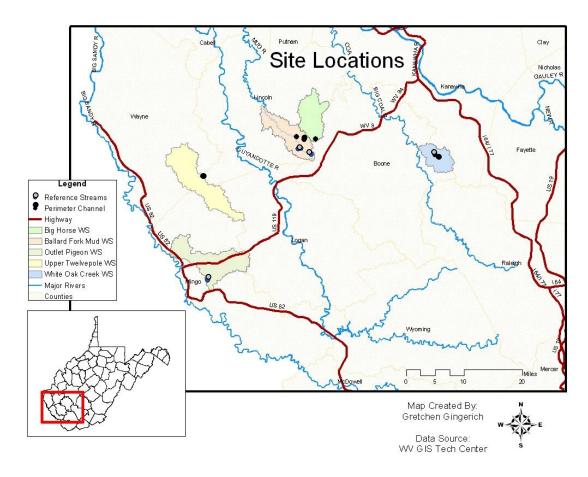


Figure 1. Site locations and HUC 12 watershed boundaries for reference sites (gray dots) and reclaimed mine perimeter channel sites (black dots).

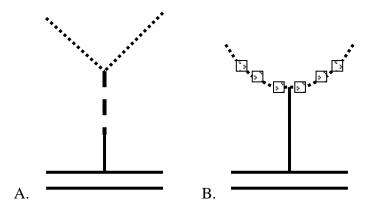


Figure 2. A) Typical native headwater catchment with two ephemeral streams (dotted line) draining into an intermittent stream (dashed line), which transitions into a perennial stream (solid line), and finally drains into a broad river. Typically there is a continuous gradient and linkage as you move from the ephemeral channels downstream to the larger river mainstem. B) Typical reclaimed mine headwater catchment consisting of a series of sediment retention cells located along the perimeter of a larger surface mine / valley fill complex. The upper headwater complex typically is low gradient with an extremely steep ephemeral or perennial channel connecting the "on-bench" perimeter channel complex with the larger river mainstem downslope.

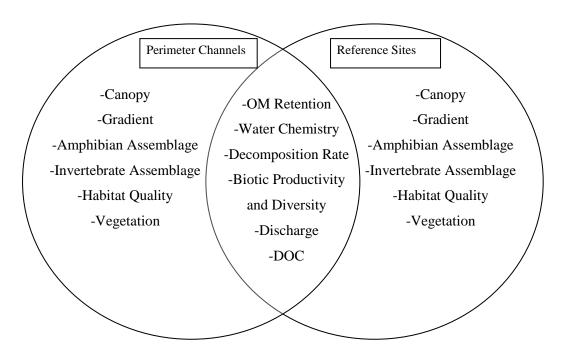


Figure 3. Several structural characteristics of aquatic systems were expected to be dramatically different between reclaimed mine perimeter channels and reference channels, such as canopy cover, gradient, biological assemblages and vegetation. However, other characteristics, such as OM processing, DOC, and biological productivity, provide a measure of ecosystem function and can be compared directly across the site types. These "functional" attributes of aquatic systems were used to construct measures of ecological units (i.e., EUs) for perimeter channels and reference channels.

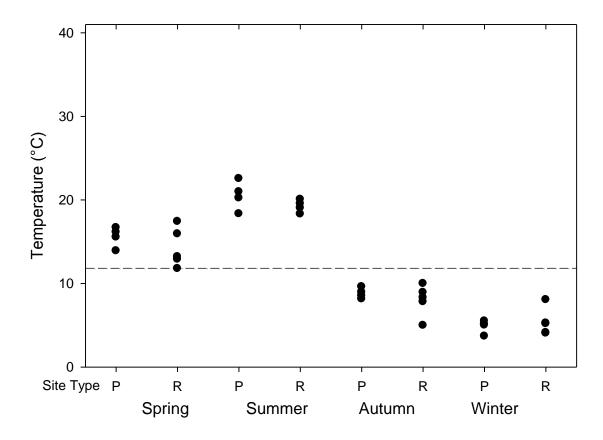


Figure 4. Mean daily temperature for reclaimed mine perimeter channels (P) and reference sites (R) during periods when streams contained water. The mean of these temperatures is $11.7\,^{\circ}$ C (dashed line).

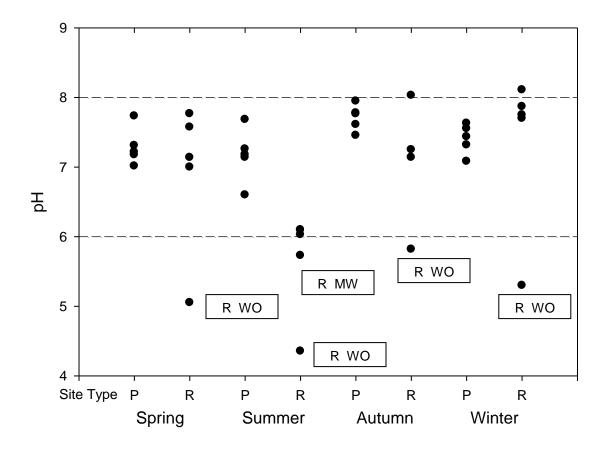


Figure 5. Seasonal pH for reclaimed mine perimeter channels and reference streams combined by site type. A range of pH 6.0-8.0 (dashed lines) is considered normal or acceptable.

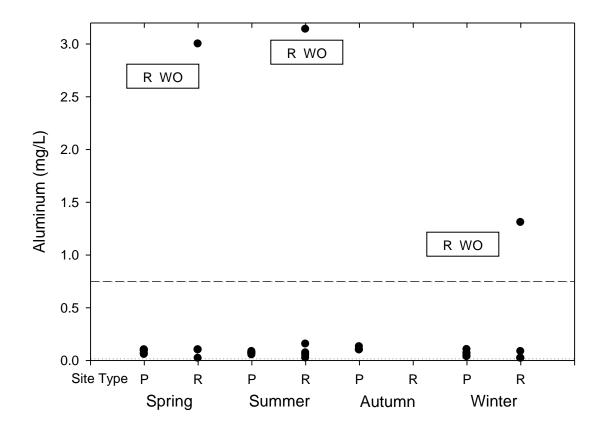


Figure 6. Seasonal aluminum (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.75 mg/L (dashed line). Method detection limits (MDL) were 0.021 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

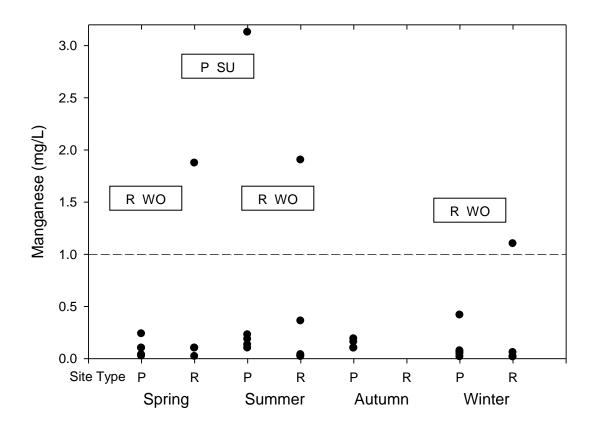


Figure 7. Seasonal manganese (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 1 mg/L (dashed line). Method detection limits (MDL) were 0.017 mg/L. Reference sites did not contain enough water for sampling in autumn.

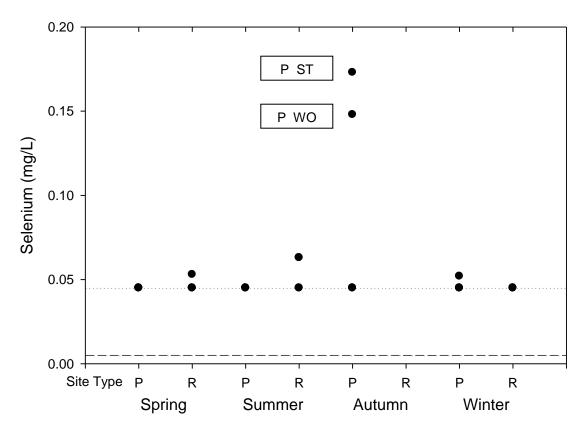


Figure 8. Seasonal selenium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 0.005 mg/L (dashed line). MDL was 0.045 mg/L (dotted line). Reference sites did not contain enough water for sampling in autumn.

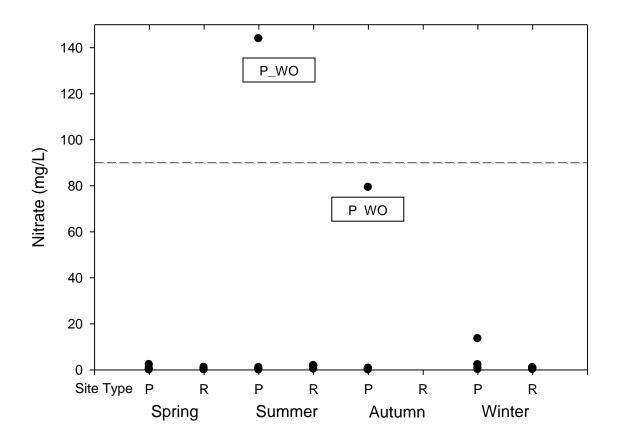


Figure 94. Seasonal nitrate (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. The WWF limit is 90 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

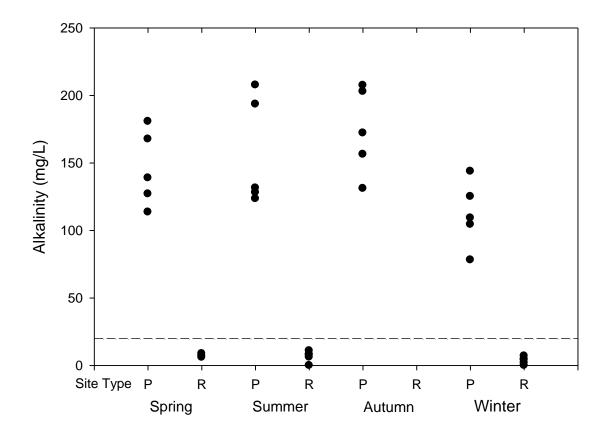


Figure 10. Seasonal alkalinity (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. An alkalinity of > 20mg/L (dashed line) is considered to have good buffering capacity. Reference sites did not contain enough water for sampling in autumn.

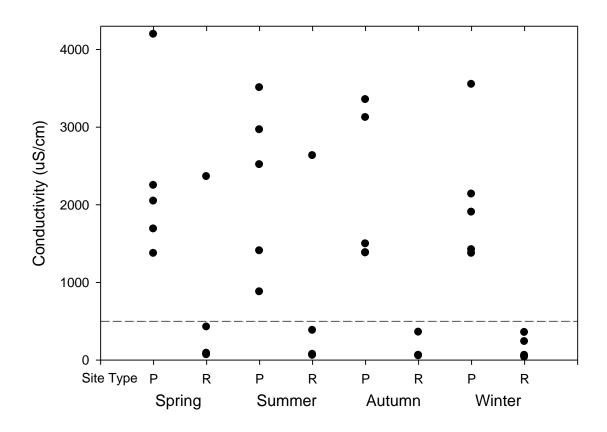


Figure 11. Seasonal conductivity (μ S/cm) for reclaimed mine perimeter channels and reference streams combined by site type. A conductivity of 500 μ S/cm (dashed line) is the EPA recommended upper limit for healthy fisheries.

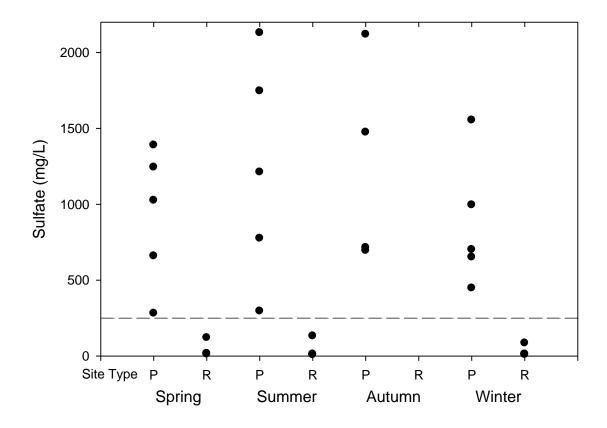


Figure 12. Seasonal sulfate (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Perimeter sites measured above 250 mg/L (dashed line). Reference sites did not contain enough water for sampling in autumn.

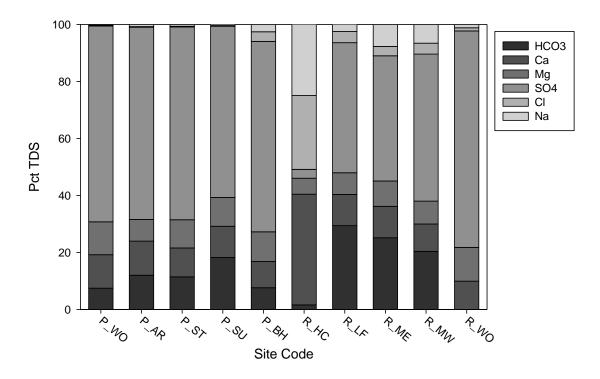


Figure 13. The percent of mean TDS that was composed of bicarbonate, calcium, magnesium, sulfate, chloride, and sodium for reclaimed surface mine perimeter channel and reference sites. Perimeter channels are presented in order of age since reclamation.

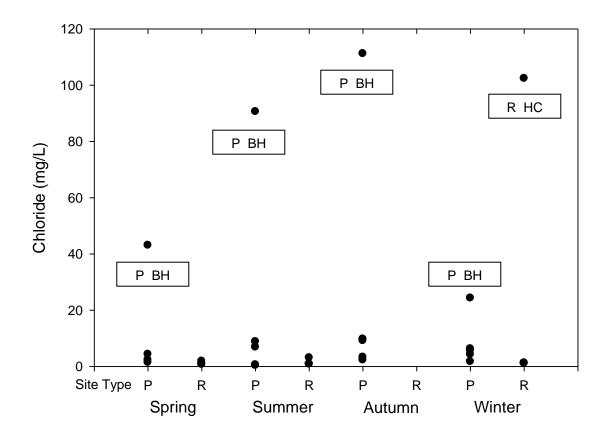


Figure 14. Seasonal chloride (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. R_HC experienced a summer measure of 1070.73 mg/L (not shown) after disturbance. Reference sites did not contain enough water for sampling in autumn.

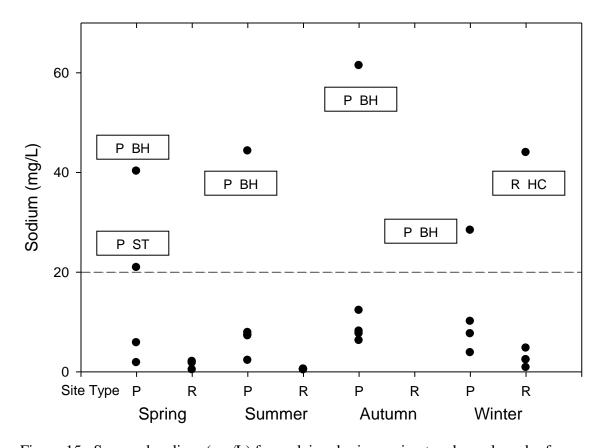


Figure 15. Seasonal sodium (mg/L) for reclaimed mine perimeter channels and reference streams combined by site type. Summer R_HC measured 293.22 mg/L (not shown). Reference sites did not contain enough water for sampling in autumn.

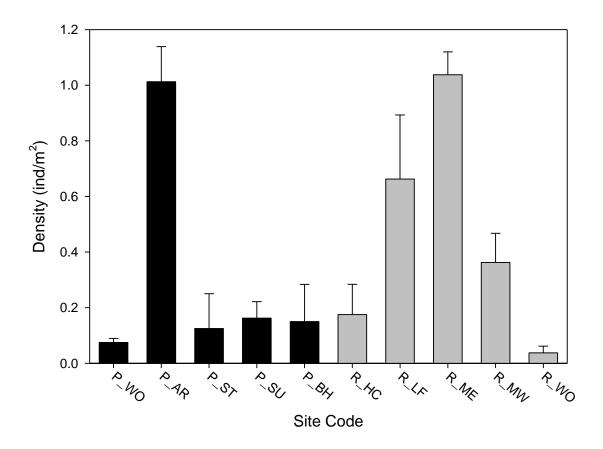


Figure 16. Mean amphibian density and standard error on reclaimed mine perimeter channels and reference sites determined from amphibian abundance surveys performed on four sample dates. Perimeter channel sites are listed in increasing age since reclamation. Perimeter channel sites are shown in black, and reference channel sites are shown in gray.

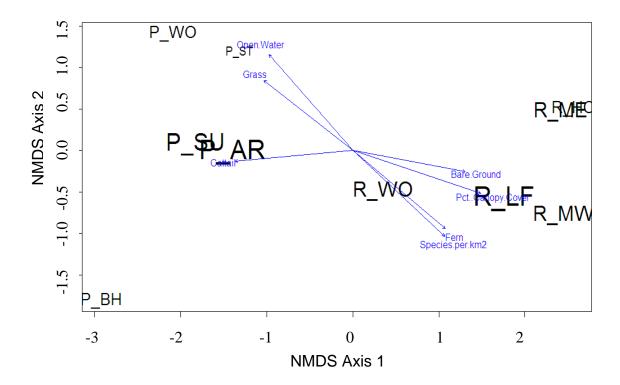


Figure 17. NMDS ordination analysis distinguishing site type by amphibian community data with overlaying significant vegetation vectors. Vegetation vectors include percent open water, grass, cattail, bare ground, percent canopy cover (Pct.Canopy.Cover), fern, and species per km². The direction of the vector indicates the direction of influence the vector has on determining community composition. The size of the character indicates the species richness of the site with larger characters indicating sites with greater richness.

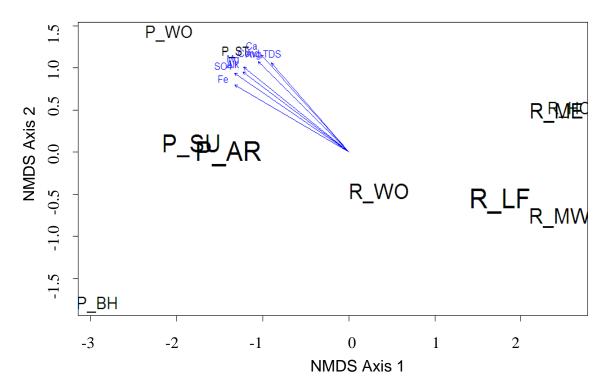


Figure 18. NMDS ordination analysis distinguishing site type by amphibian community data with overlaying significant water chemistry vectors. The direction of the vector indicates the direction of influence the vector has on determining community composition. Vector measures include mean conductivity (Cond), sulfate (SO₄), magnesium (Mg), mean total dissolved solids (Avg.TDS), calcium (Ca), alkalinity (Alk), and iron (Fe). The size of the character indicates the species richness of the site with larger characters indicating sites with greater richness.

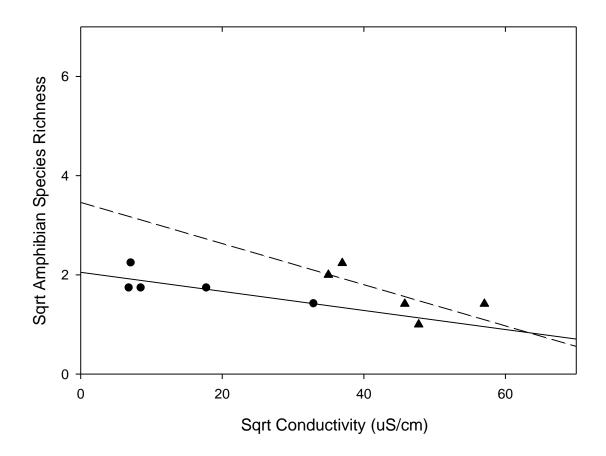


Figure 5. Mean conductivity versus amphibian species richness for four sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

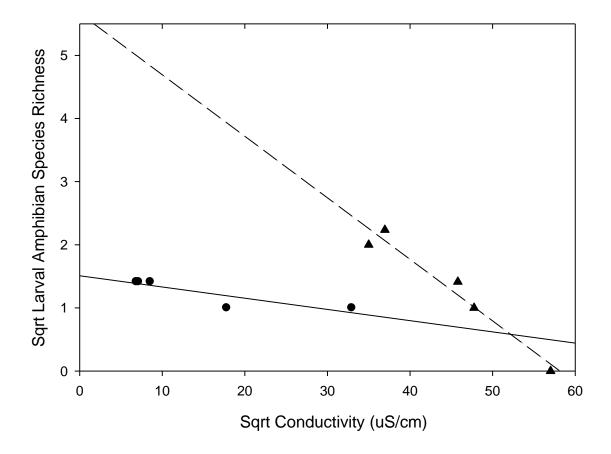


Figure 60. Mean conductivity versus the total number of larval amphibian species for four sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

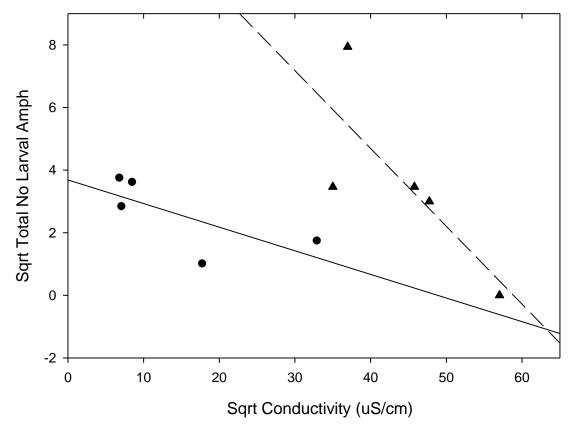


Figure 21. Mean conductivity versus the total number of larval amphibians captured during four amphibian sampling periods. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

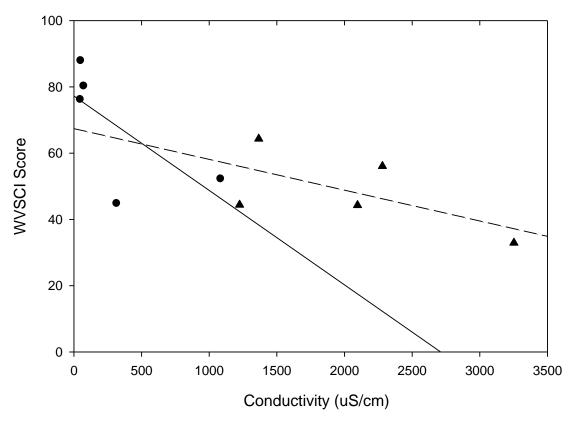


Figure 22. Mean conductivity versus WVSCI score for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

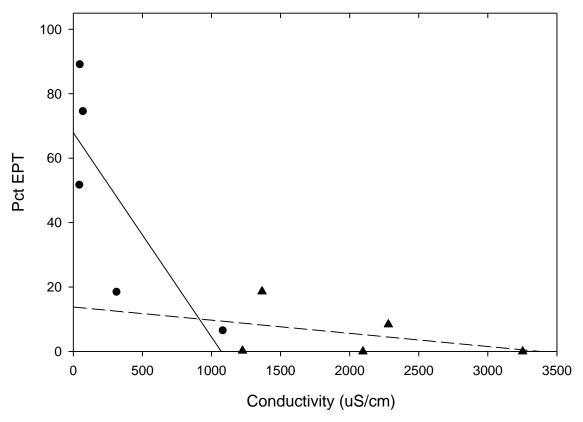


Figure 23. Mean conductivity versus percent EPT for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

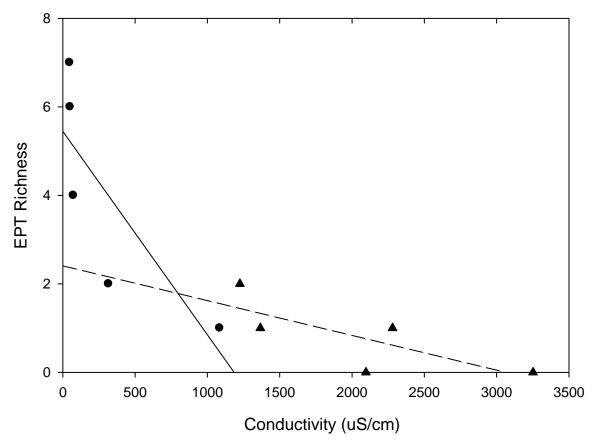


Figure 24. Mean conductivity versus EPT species richness for the spring 2008 sampling period. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

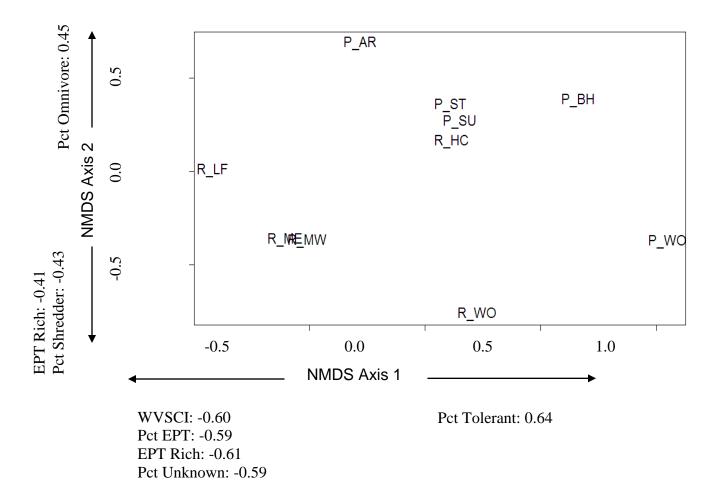


Figure 25. NMDS ordination analysis distinguishing site type by macroinvertebrate community data with Spearman rank correlations are annotated along each axis. The size of the site character indicates the species richness of the site with larger characters indicating sites with greater richness.

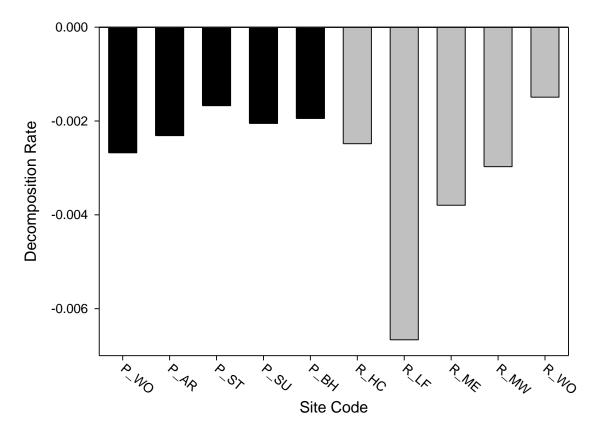


Figure 26. Decomposition rate (-k) of *Quercus palustris* (pin oak) leaf litter on reclaimed mine perimeter channels and reference streams after ~325 days of exposure. Perimeter channel sites are listed in increasing age since reclamation. Perimeter channel sites are shown in black and reference channel sites are shown in gray.

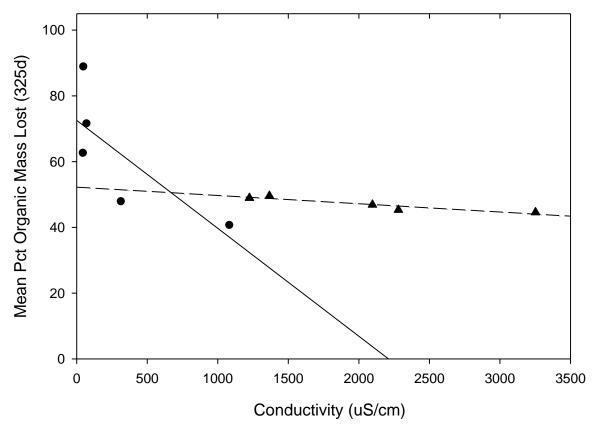


Figure 27. Mean conductivity versus mean percent organic mass lost from leaf litter packs after 325 days. Reclaimed mine perimeter channel sites are represented by black triangles and dotted regression line. Reference sites are represented by black circles and solid regression line.

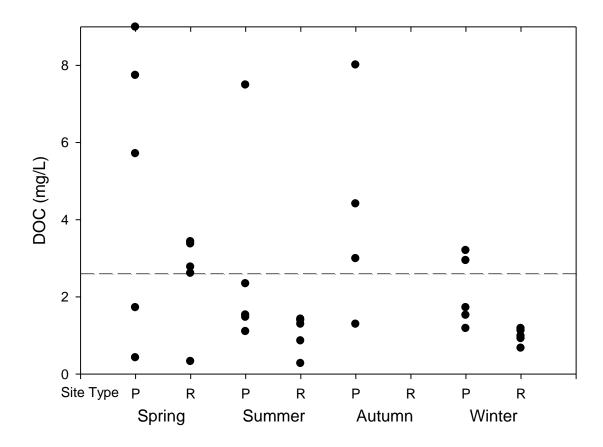


Figure 28. Seasonal DOC measurements for reclaimed mine perimeter channels and reference sites combined by site type. Mean DOC is 2.60 mg/L (dashed line). Reference sites did not contain enough water for autumn sampling.

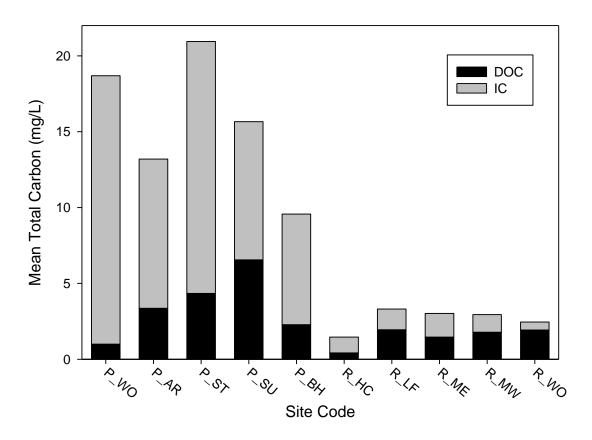


Figure 29. Mean total carbon for reclaimed mine perimeter channels and reference sites given by concentration of dissolved organic carbon (DOC) and inorganic carbon (IC). Perimeter channel sites are listed in increasing age since reclamation.

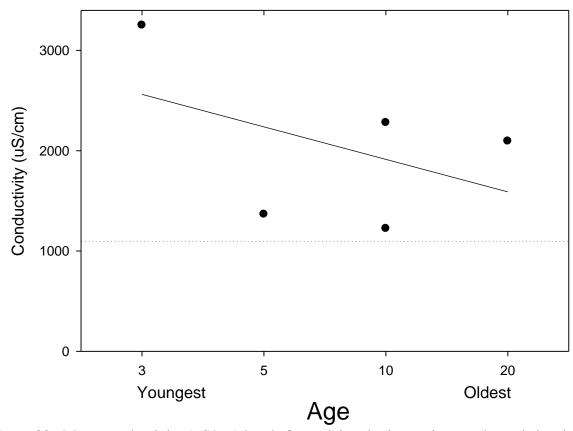
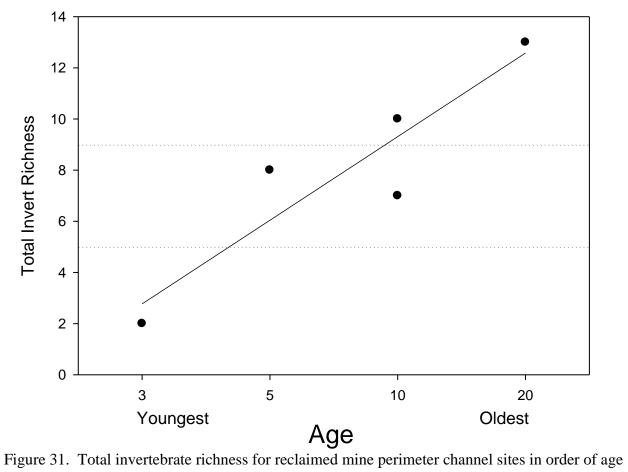


Figure 30. Mean conductivity (μ S/cm) levels for reclaimed mine perimeter channel sites in order of age since reclamation. The upper 95% confidence interval for reference sites (1101 μ S/cm) is shown (dotted line).



since reclamation. Confidence interval for reference sites (5-9) is shown (dotted line).

Appendices
Appendix A. Seasonal water chemistry measures for reference sites and reclaimed mine perimeter channels. Reference sites did not contain enough water for sampling during autumn. MDL= method detection limit. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

	Season	Acidity	Alk	Cond	Ca	Mg	SO_4	Na	Cl
		mg/L	mg/L	μS/cm	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	0	181	-	241	231	1245	-	-
P_WO	Summer	0	194	3230	316	302	2131	7.43	6.9
P_WO	Autumn	0	207	3620	403	442	2120	7.66	9.2
	Winter	0	144	2905	266	276	1555	7.64	6.3
	Spring	0	139	1382	28	66	660	5.84	2.3
P_AR	Summer	0	123	1343	176	77	776	7.21	0.6
r_AK	Autumn	0	131	1428	219	95	715	8.15	2.3
	Winter	0	104	1311	185	92	652	10.11	4.3
	Spring	0	168	-	222	206	1390	20.91	4.3
P_ST	Summer	0	208	2560	246	254	1747	7.87	8.8
P_31	Autumn	0	203	3250	404	486	2290	12.32	9.8
	Winter	0	125	1030	67	65	448	3.84	1.7
	Spring	0	114	769	40	39	282	1.83	1.4
P_SU	Summer	0	128	741	61	50	297	2.29	0.3
P_3U	Autumn	0	156	1452	161	140	695	6.27	3.3
	Winter	0	109	1937	197	194	996	-	5.7
	Spring	0	127	-	149	152	1026	40.25	43.1
D DII	Summer	0	131	2120	155	179	1212	44.31	90.6
P_BH	Autumn	0	172	2580	263	382	1475	61.43	111.2
	Winter	0	78	1588	98	120	702	28.40	24.3
	Spring	0 ± 0	146 ± 28	1076 ± 628	136 ± 99	139 ± 84	921 ± 451	17.21 ± 16.94	12.8 ± 18.4
D	Summer	0 ± 0	157 ± 40	1999 ± 982	191 ± 96	172 ± 109	1233 ± 734	13.82 ± 17.19	21.4 ± 38.8
Perimeter	Autumn	0 ± 0	174 ± 32	2466 ± 1008	290 ± 110	309 ± 179	1459 ± 752	19.17 ± 23.73	27.1 ± 47.1
	Winter	0 ± 0	112 ± 24	1754 ± 726	163 ± 80	150 ± 86	871 ± 430	12.50 ± 10.98	8.5 ± 9.0

	Season	Acidity	Alk	Cond	Ca	Mg	SO_4	Na	Cl
		mg/L	mg/L	μS/cm	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	21	-	54	123	1	-	0.42	1.9
R_HC	Summer	12	6	2740	103	56	8	293.22	-
к_пс	Autumn	-	-	-	-	-	-	-	-
	Winter	-	4	462	16	12	10	44.00	102.4
	Spring	4	7	-	3	2	13	-	0.8
R_LF	Summer	-	8	52	3	2	10	0.41	0.9
K_LF	Autumn	-	-	-	-	-	-	-	-
	Winter	4	7	50	2	2	11	0.86	1.3
	Spring	6	9	67	4	3	16	1.78	1.1
D ME	Summer	5	11	64	3	3	13	0.42	0.9
R_ME	Autumn	-	-	-	-	-	-	-	-
	Winter	7	5	89	3	3	13	4.76	1.1
	Spring	4	6	47	3	2	17	2.07	1.0
R_MW	Summer	6	8	51	2	2	11	0.54	0.9
IX_IVI VV	Autumn	-	-	-	-	-	-	-	-
	Winter	8	2	44	2	2	13	2.41	1.1
	Spring	28	-	-	15	17	121		0.7
R_WO	Summer	35	0	356	17	20	132	0.54	3.1
K_WO	Autumn	-	-	-	-	-	-	-	-
	Winter	32	0	278	12	15	86	2.41	1.2
	Spring	13 ± 11	7 ± 4	56 ± 31	30 ± 53	5 ± 7	42 ± 50	1.42 ± 1.00	1.1 ± 0.5
Reference	Summer	14 ± 14	7 ± 4	652 ± 1174	26 ± 44	17 ± 23	35 ± 54	59.03 ± 130.92	1.4 ± 1.2
Reference	Autumn	-	-	-	-	-	-	-	-
	Winter	13 ± 12	4 ± 3	185 ± 182	7 ± 6	7 ± 6	26 ± 33	10.89 ± 18.56	21.4 ± 45.3

	Season	Al	Fe	Se	Zn	Cd	Cr	Со
		mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	0.065	0.01	MDL	MDL	MDL	MDL	0.016
D WO	Summer	0.068	0.02	MDL	0.053	MDL	MDL	0.021
P_WO	Autumn	0.130	1.06	0.148	0.141	MDL	MDL	MDL
	Winter	0.104	0.11	0.052	0.097	0.012	0.013	0.013
	Spring	0.100	0.10	MDL	MDL	MDL	MDL	MDL
P_AR	Summer	0.085	0.15	MDL	MDL	MDL	MDL	MDL
P_AR	Autumn	0.100	0.54	MDL	MDL	MDL	MDL	MDL
	Winter	0.036	0.05	MDL	MDL	MDL	MDL	MDL
	Spring	0.088	0.04	MDL	MDL	MDL	MDL	MDL
р ст	Summer	0.053	0.08	MDL	MDL	MDL	MDL	MDL
P_ST	Autumn	0.100	1.11	0.173	0.071	0.057	0.066	0.054
	Winter	0.061	0.07	MDL	0.018	MDL	MDL	0.013
	Spring	0.100	0.10	MDL	MDL	MDL	MDL	MDL
P_SU	Summer	0.072	0.64	MDL	MDL	MDL	MDL	0.018
1_30	Autumn	0.100	0.69	MDL	MDL	MDL	MDL	MDL
	Winter	0.037	0.03	MDL	0.035	MDL	MDL	MDL
	Spring	0.056	0.01	MDL	MDL	MDL	MDL	0.016
P_BH	Summer	0.065	0.07	MDL	0.020	MDL	MDL	MDL
г_вп	Autumn	0.100	1.02	MDL	0.115	0.023	0.025	0.021
	Winter	0.069	0.08	MDL	0.023	MDL	MDL	0.013
	Spring	0.082 ± 0.020	0.05 ± 0.04	-	-	-	-	0.016 ± 0.009
Dominanter	Summer	0.069 ± 0.012	0.19 ± 0.25	-	0.037 ± 0.023	-	-	0.020 ± 0.011
Perimeter	Autumn	0.106 ± 0.013	0.88 ± 0.25	0.161 ± 0.088	0.109 ± 0.065	0.040 ± 0.025	0.046 ± 0.029	0.038 ± 0.024
	Winter	0.061 ± 0.028	0.07 ± 0.03	0.052 ± 0.023	0.043 ± 0.037	0.012 ± 0.005	0.013 ± 0.006	0.013 ± 0.007

	Season	Al	Fe	Se	Zn	Cd	Cr	Co
		mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	0.100	0.10	MDL	MDL	MDL	0.016	MDL
R_HC	Summer	0.155	0.12	MDL	MDL	MDL	MDL	MDL
к_пс	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
	Spring	MDL	0.01	MDL	MDL	MDL	MDL	0.018
R_LF	Summer	0.051	0.10	MDL	MDL	MDL	MDL	MDL
K_LF	Autumn	-	-	-	-	-	-	-
	Winter	0.085	0.06	MDL	0.018	0.012	0.020	0.013
	Spring	0.100	0.10	MDL	0.020	MDL	MDL	MDL
R_ME	Summer	0.072	0.09	MDL	MDL	MDL	MDL	MDL
K_ME	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
	Spring	0.100	0.10	MDL	0.020	MDL	MDL	MDL
R_MW	Summer	0.023	0.07	0.063	0.028	MDL	0.013	MDL
IX_IVI VV	Autumn	-	-	-	-	-	-	-
	Winter	MDL	0.01	MDL	MDL	MDL	MDL	MDL
	Spring	3.000	0.01	0.053	0.458	MDL	MDL	-
R_WO	Summer	3.139	0.01	0.063	0.028	MDL	MDL	MDL
K_WO	Autumn	-	-	-	-	-	-	-
	Winter	1.308	0.10	MDL	MDL	0.012	0.015	0.016
	Spring	0.825 ± 1.309	0.07 ± 0.05	0.053 ± 0.024	0.166 ± 0.201	-	0.016 ± 0.007	0.018 ± 0.008
Deference	Summer	0.069 ± 1.371	0.08 ± 0.04	0.063 ± 0.035	0.028 ± 0.002	-	0.013 ± 0.006	-
Reference	Autumn	-	-	-	-	-	-	-
	Winter	0.696 ± 0.569	0.04 ± 0.04	-	0.018 ± 0.008	0.012 ± 0.007	0.017 ± 0.010	0.014 ± 0.008

	Season	Cu	Ba	Mn	Ni	NO_2	NO_3	NH_3	TP
		mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	MDL	0.02	0.02	0.05	MDL	MDL	MDL	MDL
P_WO	Summer	MDL	0.02	0.19	0.13	0.40	143.83	0.033	0.05
r_wo	Autumn	MDL	MDL	0.19	0.06	0.10	79.27	MDL	MDL
	Winter	MDL	MDL	0.42	0.14	0.07	13.53	0.009	0.68
	Spring	MDL	0.01	0.10	MDL	MDL	2.28	MDL	MDL
P_AR	Summer	MDL	0.02	0.23	MDL	0.03	1.01	0.007	0.05
r_AK	Autumn	MDL	0.02	0.16	MDL	MDL	0.24	-	MDL
	Winter	MDL	0.02	0.05	MDL	MDL	2.25	MDL	0.07
	Spring	MDL	0.02	0.04	0.03	MDL	0.74	MDL	MDL
P_ST	Summer	MDL	0.02	0.13	MDL	0.03	-	0.039	0.05
1_51	Autumn	0.061	0.07	0.10	0.06	MDL	0.70	0.087	0.07
	Winter	MDL	0.02	0.06	0.02	MDL	0.17	MDL	MDL
	Spring	MDL	0.02	0.10	MDL	MDL	0.06	0.002	0.03
P_SU	Summer	MDL	0.03	3.13	0.04	0.03	0.02	0.009	0.05
1_30	Autumn	MDL	0.04	0.10	MDL	MDL	MDL	MDL	MDL
	Winter	MDL	0.02	0.02	MDL	MDL	-	MDL	0.06
	Spring	MDL	MDL	0.24	MDL	MDL	MDL	MDL	MDL
P_BH	Summer	MDL	0.01	0.10	MDL	0.03	0.02	0.003	0.05
г_вп	Autumn	0.027	0.02	0.10	MDL	MDL	0.03	MDL	0.05
	Winter	MDL	MDL	0.07	0.02	MDL	0.93	MDL	0.08
	Spring	-	0.02 ± 0.01	0.10 ± 0.08	0.04 ± 0.02	-	1.03 ± 0.98	0.002 ± 0.001	0.03 ± 0.01
Darimatar	Summer	-	0.02 ± 0.01	0.75 ± 1.33	0.09 ± 0.06	0.10 ± 0.17	36.22 ± 64.21	0.018 ± 0.017	0.05 ± 0.00
Perimeter	Autumn	0.044 ± 0.027	0.04 ± 0.03	0.13 ± 0.04	0.06 ± 0.03	0.10 ± 0.04	20.06 ± 35.34	0.087 ± 0.039	0.06 ± 0.03
	Winter		0.02 ± 0.01	0.12 ± 0.17	0.19 ± 0.06	0.07 ± 0.03	4.22 ± 5.75	0.009 ± 0.004	0.22 ± 0.28

	Season	Cu	Ba	Mn	Ni	NO_2	NO_3	NH_3	TP
		mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L	mg/L
	Spring	MDL	0.01	0.10	MDL	MDL	0.11	0.006	0.03
R_HC	Summer	MDL	0.89	0.36	0.03	46.23	1.85	0.056	0.05
K_IIC	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.10	0.02	MDL	MDL	0.97	MDL	MDL
	Spring	MDL	0.03	0.02	MDL	MDL	0.24	MDL	0.06
R_LF	Summer	MDL	0.03	0.03	MDL	0.06	1.61	0.012	0.05
K_LI	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.06	0.06	0.02	MDL	0.57	0.029	0.10
	Spring	MDL	0.03	0.10	MDL	MDL	1.06	0.009	0.08
R_ME	Summer	MDL	0.04	0.04	MDL	0.03	1.79	0.019	0.05
K_WIL	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.02	0.02	MDL	MDL	0.69	MDL	0.06
	Spring	MDL	0.03	0.10	MDL	MDL	MDL	0.010	MDL
R_MW	Summer	MDL	0.03	0.02	MDL	0.03	0.29	0.026	0.06
10_10100	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.01	0.02	MDL	MDL	0.12	MDL	0.08
	Spring	MDL	0.07	1.87	0.19	MDL	0.23	MDL	0.05
R_WO	Summer	MDL	0.04	1.90	MDL	0.03	1.52	0.026	0.05
K_WO	Autumn	-	-	-	-	-	-	-	-
	Winter	MDL	0.03	1.10	MDL	MDL	0.43	0.003	MDL
	Spring	-	0.03 ± 0.02	0.44 ± 0.80	0.03 ± 0.08	-	0.41 ± 0.42	0.008 ± 0.005	0.05 ± 0.03
Reference	Summer	-	0.20 ± 0.38	0.47 ± 0.81	0.02 ± 0.01	9.28 ± 20.66	1.41 ± 0.64	0.028 ± 0.017	0.05 ± 0.01
Kelelelice	Autumn	-	-	-	-	-	-	-	-
	Winter	-	0.04 ± 0.03	0.24 ± 0.48	0.02 ± 0.01	-	0.56 ± 0.32	0.016 ± 0.012	0.08 ± 0.04

Appendix B. Seasonal temperature data for reference sites and perimeter channels for periods when streams contained water. Mean and standard deviation by site type are given in the last two rows. Perimeter channel sites are listed in order of increasing age since reclamation.

Site Code	Season	Max Daily Temp (°C)	Min Daily Temp (°C)	Mean Daily Temp (°C)	CV for Mean Daily Temp
	Spring	-	-	-	-
P_WO	Summer	-	-	-	-
r_wo	Autumn	-	-	-	-
	Winter	-	-	-	-
	Spring	20.5	5.3	13.9	23.0
P_AR	Summer	22.1	15.4	18.3	5.3
r_AK	Autumn	16.6	3.9	9.0	40.9
	Winter	13.4	0.4	5.0	53.0
	Spring	26.1	6.3	16.7	23.2
P_ST	Summer	28.2	13.8	20.2	7.8
r_s1	Autumn	18.2	1.4	8.5	52.2
	Winter	19.2	0.1	5.5	72.6
	Spring	40.6	5.3	15.6	26.7
P SU	Summer	29.1	15.8	22.6	6.4
F_50	Autumn	22.1	0.0	9.6	55.4
	Winter	16.4	0.0	5.1	59.2
	Spring	34.6	2.0	16.1	27.6
P_BH	Summer	30.3	13.4	21.0	8.2
г_вп	Autumn	19.9	0.0	8.1	66.9
	Winter	22.3	0.0	3.7	112.7
	Spring	30.4 ± 15.6	4.7 ± 2.7	15.6 ± 7.0	-
Perimeter	Summer	27.4 ± 12.7	14.6 ± 6.6	20.5 ± 9.3	-
Perimeter	Autumn	19.2 ± 8.8	1.3 ± 1.7	8.8 ± 4.0	-
	Winter	17.8 ± 8.6	0.1 ± 0.2	4.8 ± 2.3	-

Site Code	Season	Max Daily Temp (°C)	Min Daily Temp (°C)	Mean Daily Temp (°C)	CV for Mean Daily Temp
	Spring	33.2	10.0	17.4	15.6
R_HC	Summer	38.9	11.7	20.1	6.5
к_пс	Autumn	18.2	4.2	10.0	31.3
	Winter	13.9	0.0	8.1	34.3
	Spring	31.4	2.7	12.9	33.0
R_LF	Summer	29.3	10.5	19.6	9.0
K_LF	Autumn	23.9	0.0	7.8	66.0
	Winter	10.7	0.0	4.1	69.0
	Spring	22.5	6.2	11.8	23.9
D ME	Summer	24.3	11.0	18.3	8.3
R_ME	Autumn	19.5	0.0	8.3	51.0
	Winter	10.6	0.0	5.3	42.4
	Spring	34.1	1.7	13.2	32.1
D MW	Summer	16.1	0.0	5.2	63.9
R_MW	Autumn	6.6	3.8	5.0	15.5
	Winter	17.4	0.0	5.2	62.2
-	Spring	28.2	13.4	15.9	6.4
D WO	Summer	32.2	12.3	19.0	10.4
R_WO	Autumn	24.9	0.0	8.9	55.4
	Winter	15.8	0.0	4.0	71.2
	Spring	29.9 ± 4.7	6.8 ± 4.9	14.3 ± 2.3	-
Defense	Summer	28.2 ± 8.6	9.1 ± 5.1	16.4 ± 6.3	-
Reference	Autumn	18.6 ± 7.3	1.6 ± 2.2	8.0 ± 1.9	-
	Winter	13.2 ± 6.5	0.0 ± 10.0	5.7 ± 2.9	

Appendix C. Adult amphibian abundance totals observed on reclaimed mine perimeter channels and reference streams for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Desmognathus fuscus	0	0	0	0	0	3	22	23	10	0	0	58
Desmognathus monticola	0	0	0	0	0	5	20	46	4	0	0	75
Desmognathus unknown	0	0	0	0	0	0	1	0	1	0	0	2
Eurycea bislineata	0	0	0	0	0	0	1	0	0	1	0	2
Gyrinophilus porphyriticus	0	0	0	0	0	0	1	0	0	0	0	1
Notophthalmus v. viridescens	5	1	0	1	0	0	0	0	0	0	7	0
Pseudacris c. crucifer	0	0	0	0	0	0	0	0	0	1	0	1
Rana catesbeiana	0	0	1	0	0	0	0	0	0	0	1	0
Rana clamitans	0	1	1	0	0	0	0	0	0	0	2	0
Rana palustris	1	0	1	0	0	0	0	0	0	0	2	0
Rana sp.	0	15	0	0	0	0	0	0	0	0	15	0
Total Individuals	6	17	3	1	0	8	45	69	15	2	27	139
Total Species	2	3	3	1	0	2	5	2	3	2	5	6

Appendix D. Larval amphibian abundance survey totals observed on reclaimed mine perimeter channels and reference streams for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Ambystoma sp.	0	1	0	0	0	0	0	0	0	0	1	0
Bufo americana	0	0	0	3	0	0	0	0	0	0	3	0
Desmognathus fuscus	0	0	0	0	0	3	5	12	2	1	0	23
Eurycea cirrigera	0	0	0	0	0	0	3	1	12	0	0	16
Hyla chrysoscelis	0	1	0	1	11	0	0	0	0	0	13	0
Notophthalmus v. viridescens	0	15	0	5	0	0	0	0	0	0	20	0
Pseudacris c. crucifer	0	7	9	3	0	0	0	0	0	0	19	0
Rana clamitans	0	39	0	0	1	0	0	0	0	0	40	0
Total Individuals	0	63	9	12	12	3	8	13	14	1	96	39
Total Species	0	5	1	4	2	1	2	2	2	1	6	2

Appendix E. Combined larval and adult amphibian abundance survey totals observed on reclaimed mine perimeter channels and reference sites for four sample periods. Totals by site type are given in the last two columns. Totals by site are given in the last two rows. Perimeter channel sites are listed in order of increasing age since reclamation.

Species	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Ambystoma sp.	0	1	0	0	0	0	0	0	0	0	1	0
Bufo americana	0	0	0	3	0	0	0	0	0	0	3	0
Desmognathus fuscus	0	0	0	0	0	6	27	35	12	1	0	81
Desmognathus monticola	0	0	0	0	0	5	20	46	4	0	0	75
Desmognathus unknown	0	0	0	0	0	0	1	0	1	0	0	2
Eurycea bislineata	0	0	0	0	0	0	1	0	0	1	0	2
Eurycea cirrigera	0	0	0	0	0	0	3	1	12	0	0	16
Gyrinophilus porphyriticus	0	0	0	0	0	0	1	0	0	0	0	1
Hyla chrysoscelis	0	1	0	1	11	0	0	0	0	0	13	0
Notophthalmus v. viridescens	5	16	0	6	0	0	0	0	0	0	27	0
Pseudacris c. crucifer	0	7	9	3	0	0	0	0	0	1	19	1
Rana catesbeiana	0	0	1	0	0	0	0	0	0	0	1	0
Rana clamitans	0	40	1	0	1	0	0	0	0	0	42	0
Rana palustris	1	0	1	0	0	0	0	0	0	0	2	0
Rana sp.	0	15	0	0	0	0	0	0	0	0	15	0
Total Individuals	6	80	12	13	12	11	53	82	29	3	123	178
Total Species	2	6	4	4	2	2	6	3	4	3	9	7

Appendix F. Macroinvertebrate abundance data given by Order (when known) for reclaimed mine perimeter channels and reference sites. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Order	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Cladocera	0	0	210	0	0	0	0	1	16	0	42 ± 94	3 ± 7
Clams	0	0	8	0	16	0	0	0	0	0	5 ± 7	0 ± 0
Coleoptera	0	8	15	0	3	0	0	0	0	0	5 ± 7	0 ± 0
Collembola	0	0	0	0	24	0	0	8	56	0	5 ± 11	13 ± 24
Cyclopoida	0	0	88	96	0	8	0	0	0	0	37 ± 50	2 ± 4
Diptera	1329	33	56	1196	705	20	16	68	184	41	664 ± 611	66 ± 69
Ephemeroptera	0	16	16	2	0	0	88	15	128	0	7 ± 8	46 ± 58
Hemiptera	0	0	2	0	0	0	0	0	16	0	0 ± 1	3 ± 7
Odonata	0	10	0	6	3	0	0	1	0	0	4 ± 4	0 ± 0
Oligochaeta	0	1	0	8	56	8	0	0	8	8	13 ± 24	5 ± 4
Plecoptera	0	0	0	1	0	3	41	208	135	11	0 ± 0	80 ± 89
Snails	0	18	8	75	16	8	0	0	0	0	23 ± 30	2 ± 4
Trichoptera	0	0	0	0	0	0	0	1	1	0	0 ± 0	0 ± 1

Appendix G. Macroinvertebrate abundance data given as a percent by Order (when known) for reclaimed mine perimeter channels and reference streams. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Order	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Cladocera	0	0	52	0	0	0	0	0	3	0	10 ± 23	1 ± 1
Clams	0	0	2	0	2	0	0	0	0	0	1 ± 1	0 ± 0
Coleoptera	0	9	4	0	0	0	0	0	0	0	3 ± 4	0 ± 0
Collembola	0	0	0	0	3	0	0	3	10	0	1 ± 1	3 ± 4
Cyclopoida	0	0	22	7	0	17	0	0	0	0	6 ± 9	3 ± 8
Diptera	100	38	14	86	86	43	11	23	34	68	65 ± 37	36 ± 22
Ephemeroptera	0	19	4	0	0	0	61	5	24	0	5 ± 8	18 ± 26
Hemiptera	0	0	0	0	0	0	0	0	3	0	0 ± 0	1 ± 1
Odonata	0	12	0	0	0	0	0	0	0	0	2 ± 5	0 ± 0
Oligochaeta	0	1	0	1	7	17	0	0	1	13	2 ± 3	6 ± 8
Plecoptera	0	0	0	0	0	6	28	69	25	18	0 ± 0	29 ± 24
Snails	0	21	2	5	2	17	0	0	0	0	6 ± 9	3 ± 8
Trichoptera	0	0	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0

Appendix H. Macroinvertebrate abundance data given by Genus (when known) for reclaimed mine perimeter channels and reference streams. Mean and standard deviation by site type are given in the last two columns. Perimeter channel sites are listed in order of increasing age since reclamation.

Class/Order	Genera	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Oligochaeta	-	0	1	0	8	56	8	0	0	8	8	13 ± 24	5 ± 4
Bivalvia (clam)	-	0	0	8	0	16	0	0	0	0	0	5 ± 7	0 ± 0
Gastropoda (snail)	-	0	18	8	75	16	8	0	0	0	0	23 ± 30	2 ± 4
Ephemeroptera	Baetis	0	3	0	0	0	0	11	0	0	0	1 ± 1	2 ± 5
Ephemeroptera	Centroptilum	0	13	0	0	0	0	0	0	0	0	3 ± 6	0 ± 0
Ephemeroptera	Acerpenna	0	1	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0
Ephemeroptera	Ephemerellidae(UNK)	0	2	16	0	0	0	0	7	0	0	4 ± 7	1 ± 3
Ephemeroptera	Ephemerella	0	0	0	0	0	0	2	0	35	0	0 ± 0	7 ± 15
Ephemeroptera	Ephemera	0	0	0	0	0	0	0	0	5	0	0 ± 0	1 ± 2
Ephemeroptera	Ameletus	0	0	0	2	0	0	67	0	102	0	0 ± 1	34 ± 48
Ephemeroptera	Mayfly(UNK)	0	0	0	0	0	0	8	8	0	0	0 ± 0	3 ± 4
Trichoptera	Hydropsyche	0	0	0	0	0	0	0	0	1	0	0 ± 0	0 ± 0
Trichoptera	Caddisfly(UNK)	0	0	0	0	0	0	0	1	0	0	0 ± 0	0 ± 0
Plecoptera	Capnia	0	0	0	0	0	0	1	0	0	0	0 ± 0	0 ± 0
Plecoptera	Leuctridae(UNK)	0	0	0	0	0	3	0	49	74	11	0 ± 0	27 ± 33
Plecoptera	Leuctra	0	0	0	0	0	0	12	0	0	0	0 ± 0	2 ± 5
Plecoptera	Capniidae/Leuctridae(UNK)	0	0	0	0	0	0	0	0	16	0	0 ± 0	3 ± 7
Plecoptera	Perlodidae(UNK)	0	0	0	0	0	0	0	0	16	0	0 ± 0	3 ± 7
Plecoptera	Isoperla	0	0	0	0	0	0	12	0	0	0	0 ± 0	2 ± 5
Plecoptera	Yugus	0	0	0	1	0	0	0	0	0	0	0 ± 0	0 ± 0
Plecoptera	Peltoperla	0	0	0	0	0	0	16	150	0	0	0 ± 0	33 ± 66
Plecoptera	Nemouridae(UNK)	0	0	0	0	0	0	0	9	3	0	0 ± 0	2 ± 4

Class/Order	Genera	P_WO	P_AR	P_ST	P_SU	P_BH	R_HC	R_LF	R_ME	R_MW	R_WO	Perimeter	Reference
Odonata	Gomphidae(UNK)	0	8	0	0	0	0	0	0	0	0	2 ± 4	0 ± 0
Odonata	Libellulidae	0	2	0	3	0	0	0	1	0	0	1 ± 1	0 ± 0
Odonata	Coenagrionidae	0	0	0	3	3	0	0	0	0	0	1 ± 2	0 ± 0
Coleoptera	Dytiscidae(UNK)	0	0	15	0	1	0	0	0	0	0	3 ± 7	0 ± 0
Coleoptera	Agabus	0	0	0	0	2	0	0	0	0	0	0 ± 1	0 ± 0
Coleoptera	Peltodytes	0	8	0	0	0	0	0	0	0	0	2 ± 4	0 ± 0
Diptera	Chironomidae	1246	0	55	1112	0	0	0	0	0	24	483 ± 638	5 ± 11
Diptera	Tipulidae(UNK)	0	0	0	0	1	1	0	0	0	0	0 ± 0	0 ± 0
Diptera	Tabanus	0	0	0	0	0	0	8	0	0	0	0 ± 0	2 ± 4
Diptera	Chrysops	0	0	0	0	1	0	0	0	0	0	0 ± 0	0 ± 0
Diptera	Simulium	0	0	0	2	49	0	0	0	0	0	10 ± 22	0 ± 0
Diptera	Ceratopogonidae(UNK)	83	8	1	0	0	0	0	0	0	0	18 ± 36	0 ± 0
Diptera	Bezzia	0	0	0	0	1	0	0	0	0	0	0 ± 0	0 ± 0
Diptera	Stratiomyidae	0	0	0	4	0	0	0	0	0	0	1 ± 2	0 ± 0
Diptera	Tanyderidae	0	0	0	0	2	0	0	0	0	0	0 ± 1	0 ± 0
Diptera	Diptera(UNK)	0	16	0	8	2	1	8	8	0	0	5 ± 7	3 ± 4
Diptera	Non-Tanypodinae	0	9	0	35	593	18	0	51	176	17	127 ± 261	52 ± 72
Diptera	Tanypodinae	0	0	0	35	56	0	0	0	0	0	18 ± 26	0 ± 0
Collembola	Sminthuridae(UNK)	0	0	0	0	0	0	0	8	0	0	0 ± 0	2 ± 4
Collembola	Sminthurides	0	0	0	0	0	0	0	0	56	0	0 ± 0	11 ± 25
Collembola	Agrenia bidenticulata	0	0	0	0	24	0	0	0	0	0	5 ± 11	0 ± 0
Cyclopoida	Cladocera	0	0	210	0	0	0	0	0	16	0	42 ± 94	3 ± 7
Hemiptera	Hemiptera(UNK)	0	0	2	0	0	0	0	0	16	0	0 ± 1	3 ± 7
Hemiptera	Mesouelia	0	0	0	0	0	0	0	0	0	0	0 ± 0	0 ± 0
Decapoda	Crayfish(UNK)	0	0	0	0	0	0	0	1	0	0	0 ± 0	0 ± 0
Coleoptera	Hydrocanthus	0	0	0	0	0	0	0	0	0	0	3 ± 0	0 ± 0
Calanoida	Copepod(UNK)	0	0	88	96	0	16	0	0	0	0	37 ± 50	3 ± 7