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Steven W. Bailey University of Louisville

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#### COMMUNITY STRUCTURE AND DYNAMICS OF BENTHIC MACROINVERTEBRATES IN A

#### RECREATED HEADWATER STREAM SYSTEM ON A VALLEY FILL IN A RETROFITTED

#### WATERSHED LOCATED IN THE APPALACHIAN COALFIELDS OF

SOUTHEASTERN KENTUCKY (U.S.A.)

By

Steven W. Bailey B.S., University of Louisville, 2011 M.S., University of Louisville, 2015

A Dissertation Submitted to the Faculty of the College of Arts and Sciences of the University of Louisville in Partial Fulfillment of the Requirements for the Degree of

Doctor of Philosophy in Biology

Department of Biology University of Louisville Louisville, Kentucky

August 2021

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A Dissertation Approved on

May 3, 2021

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Dr. Steve Yanoviak

#### DEDICATION

This dissertation is dedicated to the folks of Central Appalachia.

I sincerely hope that this small contribution to the Guy Cove recreated headwater stream system project will help in the effort to ameliorate the environmental havoc that surface mining has inflicted upon the Appalachian Mountains.

#### ACKNOWLEDGEMENTS

This dissertation would not have been completed without support from many people. I am grateful for their generous contributions of patience, meaningful conversation, physical labor, technical skill, scholarly mentoring, and professional advice. First, I would like to thank my advisor, Dr. Jim Alexander, for his integral guidance and genuine resolve to see me succeed. Second, I would like to thank the members of my advisory committee, Dr. Steve Yanoviak, Dr. C. Andrew Day, Dr. Gary Cobbs (emeritus), and Dr. Chris Barton for their invaluable interdisciplinary contributions. Furthermore, I would like to thank Drs. Alexander, Day, and Yanoviak for being on my committee since its inception; Dr. Cobbs for his R coding expertise and orchestration of the statistical analyses; and Dr. Barton for granting me the permission and logistical support necessary to access the study sites. Chris Osborne and David Collett, from the University of Kentucky's Robinson Forest, managed the logistics of site access. Reese, Andrew, and Campbell assisted me with fieldwork and "bug picking". Richard Schultz generated the water chemistry data. Gregory J. Pond was immensely helpful during the invertebrate identification process. Dr. Margaret Carreiro (emerita) inspired me to study ecology, and Dr. Hwa-seong Jin inspired me to study stream ecology. And finally, I will be forever grateful to the following people: my parents, Brian and Judi, for instilling in me the work ethic necessary for this endeavor; my wife Rebecca for her love, support and sacrifice; and my daughter Piper for selflessly sharing her boundless and seemingly inexhaustible excitement about life – what a blessing you are!

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#### ABSTRACT

# THE COMMUNITY ECOLOGY OF BENTHIC MACROINVERTEBRATES IN A RECREATED STREAM SYSTEM ON A VALLEY FILL IN A RETROFITTED WATERSHED LOCATED IN THE APPALACHIAN COALFIELDS OF SOUTHEASTERN KENTUCKY (U.S.A.)

Steven W. Bailey

March 28, 2021

The extraction of coal from steep-gradient surface mining sites such as in the Appalachian Coalfields of the U.S. produces excess debris that is often placed in adjacent valleys resulting in the creation of valley fills. Not only are headwater streams buried in the process, but watershed functions are either destroyed outright, or become fragmented and disconnected from adjacent ecosystems resulting in adverse effects to downstream biological communities. In this dissertation, the dynamics of stream macroinvertebrate community structure, composition, diversity, and biotic integrity are assessed at a "proof of concept" stream system recreated on a retrofitted valley fill. For comparison, two reference streams were selected with contrasting degrees of environmental impact from surface mining and deforestation. Each stream was sampled monthly over the course of one year. Sixteen environmental variables were measured and sixteen biotic metrics for benthic invertebrate dynamics were calculated. From this analysis, it was apparent that the recreated stream supported a diverse and abundant benthic macroinvertebrate community more similar to an unmined stream than to a mine-impacted stream located immediately downgradient of a traditionally constructed valley fill. These results suggest

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that the retrofitted watershed, 1) improved water quality in the recreated stream system by mitigating elevated specific conductance, and 2) improved stream habitat availability and quality by restoring, at least partially, ecological functions that were lost to deforestation, mining, and valley fill creation. Overall, these results can help inform and guide stakeholders and decision-makers considering future reclamation projects at any of the hundreds of valley fills created on the surface mined lands in the Appalachian Coalfields of the Southeastern United States.

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#### INTRODUCTION

Headwaters are the springs and intermittently flowing small streams that compose the uppermost portions of a stream or river network where the surface runoff from snowmelt or precipitation is sufficiently concentrated to form distinct banks and cause scouring action that restrict the growth of algae and other plants (Dietrich and Dunne 1993; MacDonald and Coe 2007; Meyer et al. 2007; Gomi et al. 2002; Gordon et al. 2004). The U.S. EPA (2011b) considers headwater streams and watersheds in Central Appalachia, U.S.A. to be keystone components of the region's ecology because they are sources of clean, abundant water for larger streams and rivers, and they are active sites for the biogeochemical processes that support both aquatic and terrestrial ecosystems. However, periods of recurrent natural drying are also a distinctive and ecologically influential characteristic of many headwater streams (Fritz et al. 2006). Currently, there is no universally accepted definition of headwater streams; a simple definition is likely to be insufficient (Benda et al. 2005).

Headwater streams make up about 53% of the total stream miles in the United States (USEPA 2016), and account for between 70 and 80% of total stream miles in eastern coal mining states (USEPA 2003, 2005). These estimations likely underrepresent the actual total extent of headwaters because the smallest designation for a stream on a topographic map often represents a perennial channel that generally maintains year-round flow as a result of groundwater inputs (Allan and Castillo 2007; Gomi et al. 2002;

Gordon et al. 2004; Fritz et al. 2006; Hansen 2001; Meyer and Wallace 2001; Meyer etal. 2007). However, the transition between a perennial channel and the many small channels of the upper portion of a watershed that are either spring fed (intermittent channels) or that carry water only during storm events (ephemeral and intermittent channels) is indistinct, and the transition migrates up- and downslope depending on precipitation (Allan and Castillo 2007). Furthermore, most headwater streams are either too small to be accurately identified by large scale aerial and satellite imagery, or they are concealed by canopy coverage in forested watersheds (Fritz et al. 2006; Roy et al. 2009).

Although headwater streams are connected to the larger river network and strongly influence the processes and functions of downstream ecosystems, they are also easily influenced by small-scale changes to local conditions of adjacent terrestrial ecosystems because of their narrow channels and relatively small watersheds (Pringle 1997; MacDonald and Coe 2007; Meyer et al. 2007). Therefore, headwater streams are tightly coupled, or linked, with natural watershed processes such as nutrient and energy fluxes (Vannote et al. 1980; Costanza et al. 1997; Daily et al. 1997; Meyer and Wallace 2001; Gomi et al. 2002). Thus, more than any other ecosystem, the structure and function of a stream ecosystem in general, and a headwater ecosystem in particular, is determined by its interface, or linkage with adjacent ecosystems, especially the riparian zone (Cummins and Klug 1979; Gregory et al. 1991; Lamberti et al. 2010).

An ecosystem linkage is defined as any persistent or recurring process or attribute that connects different ecosystems in some manner, and are integral—even defining, components of aquatic ecosystem structure and function (Lamberti et al. 2010). Stream structure refers to the pattern or organization of physical features within a system such as

streamflow, current (velocity), channel morphology, substrate composition, vegetation, temperature, and regional species richness and local species interactions (Gordon et al. 2004; Allan and Castillo 2007; Fritz et al. 2010). Stream function refers to the processes and rates of a system, including inputs and transformations of energy and material in processes such as primary production, nutrient cycling, organic matter processing, and secondary production (Bunn and Davies 2000; Allan and Castillo 2007; Fritz et al. 2010; Lamberti et al. 2010). The types of linkages among aquatic ecosystems can be separated broadly into those that are physical, chemical, or biological in nature (Lamberti et al. 2010). Physical linkages involve the exchange of nonbiological material such as water, sediments, heat energy, and gases, whereas chemical linkages include the inter-ecosystem movements of inorganic nutrients and other dissolved ions, and biological linkages include the movement of organisms and their products such as feces and chemical signals (Lamberti et al. 2010). Because aquatic ecosystem linkages operate as interactive pathways along lateral, longitudinal, vertical and temporal dimensions (Ward 1989), understanding linkages is important not just for protecting ecosystems, but for restoring impaired ecosystems (Lamberti et al. 2010).

The lateral pathway links the stream ecosystem to the terrestrial ecosystem via the riparian zone, which is a key ecosystem for regulating aquatic-terrestrial processes (Gregory et al. 1991), such as stream microclimate, channel bank stability, nutrient filtration and sediment trapping, and a diverse distribution of vegetation that provides habitat for a variety of aquatic, semi-aquatic, and terrestrial species (Naiman and Decamps 1997). Forested headwaters are almost completely embedded within the forest and its canopy, unlike the wider channels further downstream in the river network, and

unlike headwaters in differing ecoregions such as the Great Plains and the deserts of the USA (Naiman and Decamps 1997; Omernik and Griffith 2014). Therefore, a major source of energy to the stream food web in forested headwaters is the in-fall from the canopy of invertebrates and vegetation (in the form of woody and non-woody debris, e.g., tree branches of all sizes, leaves, fruits, flowers and seeds) that contribute significant inputs of energetic resources that are subsequently transported downstream to larger streams and rivers (Cummins 1974; Hynes 1975a; Vannote et al. 1980; Cummins et al. 1983; Naiman and Decamps 1997; Meyer and Wallace 2001; Allan and Castillo 2007).

The longitudinal pathway includes the supply of water, which mostly originates in headwater streams, and the downstream transport of sediment and organic matter (Vannote et al. 1980; Cummins et al. 1983; Knighton 1998; Moore and Wondzell 2005; MacDonald and Coe 2007). Longitudinal linkages such as nutrient dynamics, insect colonization cycles, and species diversity are critical to the maintenance of ecosystem function (Newbold et al. 1981; Muller 1982; Meyer et al. 2007). For example, dissolved inorganic nutrients such as nitrogen and phosphorous 'cycle' (Newbold et al. 1981) within the ecosystem via incorporation into living tissue and subsequent remineralization by excretion or egestion and decomposition (Allan and Castillo 2007). However, since nutrients in a stream do not cycle in place, but are displaced downstream as they complete a cycle, the coupling of the concept of cycling with downstream transport is referred to as spiraling (Wallace et al. 1977; Webster and Patten 1979; Newbold et al. 1981). Insect colonization cycles involve adult females flying upstream utilizing the channel as a dispersal corridor in search of headwater habitats suitable for oviposition and larvae development (Muller 1982). Finally, the species diversity associated with

headwater streams is attributable to a diverse array of unique habitats and therefore integral to the maintenance of biological diversity in downstream ecosystems (Meyer et al. 2007).

The vertical dimension links the stream channel to the groundwaters flowing under the stream (Ward 1989). Groundwater flow exerts a strong effect upon the flow regime, a key driver of river and floodplain wetland ecosystems because the streamflow largely determines the composition of physical habitats, which in turn largely determines the composition of the biotic communities (Bunn and Arthington 2002). Additionally, the hyporheic zone, i.e., the saturated sediments in which surface water and groundwater mix, is important both as habitat for numerous aquatic organisms, and as an ecotone that is metabolically active with complex patterns of nutrient cycling that affect ecosystem metabolism (Dahm et al. 2006).

Finally, the fourth dimension is time, which gives perspective to analyzing and understanding response times following disturbances to the ecosystems and their linkages and provides informed insight into the long-term effectiveness of reclamation efforts (Hopkins et al. 2013). Unsatisfactory restorations can result from focusing on inappropriate time scales and attempting to do in a matter of years what takes decades or centuries under natural conditions (Hilderbrand et al. 2005).

Many of the ecosystem linkages associated with headwater streams support natural processes that help to sustain and fulfill human life and are therefore defined as ecosystem services (Daily et al 1997). Headwaters provide many distinct ecosystem services such as the regulation of hydrological flows; the purification, retention and storage of water—including the supply of drinking water; flood control; groundwater

recharging; soil formation and retention; crop irrigation; and habitat that either directly or indirectly supports a variety of fish and wildlife, to name only a few (Hill et al. 2014). In the continental United States, about 117 million people, over one third of the total U.S. population, get some or all of their drinking water from public drinking water systems that rely at least in part on headwater streams (USEPA 2006). In Kentucky, surface water accounts for about 95 percent of the water used in the state and provides domestic water supplies for 92 percent of the urban population and for about 50 percent of the rural population (KGS 2020).

The Central Appalachian Coalfields cover approximately 48,000 square kilometers (12 million acres) in Kentucky, West Virginia, Virginia, and Tennessee, USA (USEPA 2011b). Surface mining and reclamation are two dominant drivers of land cover/land-use change in this region and have produced significant changes in the region's topography, hydrology, vegetation, groundwater, and wildlife (Loveland et al. 2003; USEPA 2003, 2005; Townsend et al. 2009; Bernhardt and Palmer 2011). Most estimates of surface mining area and extent that were generated prior to the mid-2000s failed to present accurate, comprehensive, and spatially explicit representations of land use change because they relied on existing permit data (Geredien 2009).

The increasing availability of low-cost analytical software tools and freely downloadable high-resolution satellite imagery datasets such as those from the National Aerial Imagery Program (USDA-FSA 2021) have enabled and empowered researchers to quantitatively assess myriad social and environmental issues through a geospatial lens (Downey 2006), from analyzing relationships between vegetation and crime with regard to urban planning (e.g., Wolfe and Mennis 2012) to illuminating the inequity of

environmental burdens on certain societal groups (Weigand et al. 2019). One such use of these technologies calculated that nearly 1.2 million acres (10%) of the Central Appalachian Coalfields had been heavily mined and over 500 mountain ridges either destroyed or heavily impacted (Geredien 2009). More recently, statistics on the spatial extent of surface mining and valley fill creation have become available at temporal resolutions that are finer than decadal timescales. For example, Pericak et al. (2018) created a yearly, 30 m dataset that is freely available to the public.

Surface mining refers to the removal of the terrain surface (plant life, soil, and potentially bedrock) to access minerals underneath, such as coal, iron, and other metals (AMS 2021). There are five recognized types of surface mining, including strip mining (area and contour) and mountaintop removal (AMS 2021). These mining methods involve removing all or some portion of the top of a mountain or ridge in order to expose banded deposits of coal (referred to as coal seams) compacted within layers of sedimentary rock (USEPA 2011b). The rock and soil overlying a coal seam, which can be as far as 300 vertical meters below the surface, is referred to as overburden and is removed through blasting and excavation in order to expose the seam for mining (USEPA 2011b). The steep slopes characteristic of the Appalachian coalfields prevents the resulting debris, referred to as spoil, from being utilized to recontour the mined surface during reclamation efforts (Blackburn-Lynch 2015). Consequently, the spoil is disposed of in constructed fills located in small valleys or hollows adjacent to the mining site (USEPA 2011b). The size of valley fills varies, but the largest have volumes of over 150 million m<sup>3</sup> and can exceed three km in length (USEPA 2005) and tens to hundreds of meters deep (USEPA 2005; Bernhardt and Palmer 2011; USEPA 2011b). Once filled,

those buried sections of headwater streams are permanently removed from production of aquatic invertebrates and fishes (Hartman et al. 2005).

Furthermore, the near continuous operation of heavy equipment, particularly during the process of overburden removal and disposal, compacts the bare soils and leads to the creation of a highly impervious surface at the mined site, which destroys the natural subsurface flow, and in turn increases surface runoff (USEPA 2011b). The excess surface runoff is often diverted into ditches and sediment ponds or directed toward the valley fill where it can then infiltrate the spoil (USEPA 2011b). As surface runoff percolates through the spoil, exposure to this unweathered rock debris produces higher concentrations of total dissolved solids (TDS) in waters emerging downstream of the valley fill compared to waters in non-mined watersheds (USEPA 2011b). One component of TDS is salinity, which is the property of water that results from the combined influence of all disassociated mineral salts (USEPA 2011a). Some mineral salts are comprised of elements that are essential nutrients, e.g., sodium chloride (NaCl), however aquatic organisms are adapted to specific ranges of salinity and experience toxic effects from excess salinity (USEPA 2011a; Canedo-Arguelles et al. 2013). In the Central Appalachian region, the prominent sources of salts are mine overburden and valley fills, with highly elevated concentrations of the dissolved ions sulfate  $(SO_4^{2-})$ , calcium  $(Ca^{2+})$ , magnesium (Mg<sup>2+</sup>), and bicarbonate (HCO<sub>3</sub><sup>-</sup>) in downstream waters (Bryant et al. 2002; Hartman et al. 2005; USEPA 2011a). These ions are major components of specific conductivity, (a measure of the stream's ability to conduct an electrical current), which reflects the concentration of dissolved ions in the water (salinity) and is highly correlated with TDS (Green et al. 2000; Howard et al. 2001; Bryant et al. 2002; Bodkin et al. 2007;

Merricks et al. 2007; Pond et al. 2008). Multiple physiological functions that enable organisms to develop, grow, move, and sense their environment are dependent upon osmotic and ionic cellular mechanisms that maintain balance via selectively permeable membranes in gills or other respiratory surfaces that are in direct contact with dissolved ions in water (USEPA 2011a). Several studies of Appalachian streams have demonstrated strong negative associations between specific conductance and benthic macroinvertebrate community metrics (Green et al. 2000; Freund and Petty 2007; Pond et al. 2008; Gerritsen et al. 2010; Bernhardt et al. 2012; USEPA 2011a; Cormier et al. 2013a, b, c). In response, the U.S. EPA (2011a) has developed an aquatic life benchmark for conductivity that is intended to protect the aquatic life in streams and rivers in the Appalachian Region where mixtures of ions are dominated by salts of Ca<sup>2+</sup>, Mg<sup>2+</sup>, SO4<sup>2-</sup>, and HCO3<sup>-</sup> at a circum-neutral to alkaline pH.

In addition to burying the stream, the constructed valley fill removes the adjacent terrestrial habitats, particularly the riparian zone, and eliminates the tree canopy, thereby greatly reducing the external inputs of deciduous organic matter to waters downstream of the valley fill (Hartman et al. 2005). This disturbance to the hydrology and natural landscape of the watershed can result in long-term impacts, i.e., legacy effects at the local and regional scale (Harding et al. 1998; Frady et al. 2007). Legacy effects resulting from the loss of headwaters as a result of mining and valley fill creation include elevated specific conductivity (Pond 2008; Hopkins et al. 2013) and increased sedimentation (Pond 2008) which are known to be negatively associated with stream invertebrate abundance and diversity (USEPA 2011b) which ultimately leads to the elimination of sources of invertebrate recolonization for downstream reaches (USEPA 2011a).

The Office of Surface Mining Reclamation and Enforcement (OSMRE) estimated that approximately 1,165 km (724 mi) of headwater streams were permanently buried under valley fills in the Central Appalachian Coalfields between 1985 and 2001 (USEPA 2003, 2005, 2011b). In a cumulative impact study, the EPA reassessed the number of stream miles lost between 1992 and 2002 by including other mining activities such as blasting and backfilling in addition to valley fill creation and revised the estimate to 1,944 km (1,208 mi) of lost headwater streams during the 10-year-period (USEPA 2002, 2003, 2005). Given the extent of headwater streams lost to mountaintop mining and valley fills, there is a growing need to develop practical stream restoration techniques for mined lands in order to restore both structure and function to impacted headwater stream systems (Bernhardt and Palmer 2011; Agouridis et al. 2017).

With an estimated 1.5 million acres of Appalachian surface-mined land available for restoration, the potential for economic investment in site preparation and tree planting alone can be measured in the billions of dollars, not to mention the associated impacts to related industries (Barton et al. 2018). However, what constitutes 'restoration' is an open definition given the wide range of policy initiatives and projects stemming from diverse political, economic, and administrative practices (Baker et al. 2014). A restoration project might embrace a variety of aims and objectives, but whether or not the effort is considered a success can be linked to a complex interrelationship between rationales, underlying values, project actions, and the chosen evaluation criteria (Baker and Eckerberg 2016). While the number of rationales might vary with the number of stakeholders in any given project, the rationale for restoration at a mining site, for example, might be aimed at the restoration of past ecosystems, or merely meeting

minimum regulatory requirements. In either case, as Baker and Eckerberg (2016) point out, the underlying values of the stakeholders can influence their decisions on project implementation and the evaluation of the outcome, which are likely to be as disparate as the achievement of historical fidelity is to the achievement of the minimal standards for regulatory compliance.

In practical terms, channel engineering efforts designed to mitigate impacts from mining typically result in constructed channels that are similar in structure (width, depth, slope, sinuosity, etc.) to the ones destroyed, but not similar in ecological function (Bernhardt and Palmer 2011). This loss of ecological function occurs because the entire watershed, not just the stream, has been dramatically altered by the surface mining operations, with the resulting disruption of both hydrological and ecological linkages between the streams and their watersheds (Bernhardt and Palmer 2011). Because the reconstruction of a properly functioning ecosystem is dependent upon the first trophic level (plants and algae), it is unrealistic to assume that any semblance to historical fidelity can be achieved on mined lands with regard to species assemblages and habitats—at least in the short term (Bradshaw 1983; Baker and Eckberg 2016). Thus, the ability to successfully restore mountainous Appalachian headwater streams on mined lands is crucially linked to both the restoration of a stream's channel and also its deforested watershed, including uplands and riparian areas (Agouridis et al. 2009; Bernhardt and Palmer 2011). Watershed reforestation is necessary because the food web in these streams is reliant upon external sources of energy (Allan and Castillo 2007), with the major source being leaf litter inputs obtained both laterally and longitudinally (Ward 1989; Wallace et al. 1995; Webster et al. 1999). Since the forces that shape community

structure are those that determine which and how many species occur together, it can be expected that similar macroinvertebrate communities should occur wherever environmental circumstances are comparable (Allan and Castillo 2006).

However, the physical restoration of many forests and streams on mined lands is likely impossible because the original forest and stream are no longer there to restore (Gunn 1991). Nonetheless, even in such circumstances where forests and streams are newly created, whether naturally or through human interventions, the restoring of ecosystem functions at watershed scale is at least theoretically possible given the science of restoration ecology (Harris et al. 2006). The practice of ecological restoration is the process of assisting the recovery of an ecosystem that has been degraded, damaged, or destroyed (SERI 2004), ideally resulting in its return to an undisturbed state (Palmer and Filoso 2009). But given the current state of the science, which strives to (re-)create complex systems from perhaps oversimplified guiding principles (Hilderbrand et al. 2005), it is unrealistic to assume that the full suite of ecosystem services can be restored when restoration efforts target sites in watersheds with deforestation, mining, and valleyfill creation (Palmer and Filoso 2009). Regardless of permanence of flow, invertebrate diversity is generally lower in constructed channels on valley fills compared to forested perennial or intermittent streams, and evidence that such channels improve water quality is limited (USEPA 2011b).

Although selecting restoration goals can be politically contentious (Hobbs 2004) for many reasons, not least the opportunity for a multi-billion dollar "restoration economy" (Holl and Howarth 2000), setting goals based on rationales that focus less on historical structure at a site and more on restoring ecological functions is more likely to

ensure ecosystem service delivery in the present-day context of global environmental change (Baker et al. 2014). This type of pragmatic goal setting allows considerations to be built into target goals that extend beyond those important for just the restoration of the site itself (Harris et al. 2006), such as restoring or enhancing ecosystem services at a regional scale (e.g., high-quality downstream water resources) by mitigating the deleterious effects of mined watersheds on headwater streams at the local scale.

For example, efforts to mitigate surface runoff infiltration of a valley fill or to mitigate elevated levels of specific conductivity downstream may both show marked improvement at project completion, with subsequent gradual improvement through the early years and thereafter (Agouridis et al. 2017). But in a mined watershed that has been clear-cut of all vegetation, it may take several decades for some ecosystem functions such as external energy inputs to return depending on forestry reclamation efforts (Zipper et al. 2011) and the subsequent maturing of riparian plantings that provide the inputs of leaves and wood to the stream ecosystem (Beechie et al. 2010). Furthermore, absent the light-limiting controls of a forest canopy, stream ecosystem metabolism may also take several decades to shift from internal energy sources such as the photosynthetic activities of benthic algae, macrophytes, and phytoplankton to the external energy sources provided by a forested riparian zone (Allan and Castillo 2007).

It follows then that the decades-long shift in energy production from internal to external sources that occurs during the continuum of forest succession shapes the composition of channel substrates available both as habitat and food. This process in turn shapes the composition of invertebrate groups with the characteristic morphological and behavioral adaptations of food acquisition suited to the general resource conditions of the

stream at that point in time (Cummins and Klug 1979). Therefore, the water quality of the stream system is dependent upon the relationships between terrestrial organic matter inputs and its processing by critical functional ecological groups of aquatic invertebrates (Cummins 1974). Because of this critical role in stream ecosystem processes, aquatic invertebrates have been used extensively in evaluation criteria for water quality monitoring and impact assessment (Cairns and Pratt 1993). Some insects such as mayflies, stoneflies, and caddisflies (taxonomic orders Ephemeroptera, Plecoptera, and Trichoptera, respectively, or EPT) are significant components of headwater habitats and are widely used as indicators of stream health (Pond 2012). However, evaluation criteria should not be narrowly defined or one-off events at project completion, but rather be ongoing and adaptable over time (Baker and Eckberg 2016) in order to effectively measure the recovery of biophysical processes (primary production, nutrient transformation, groundwater recharge, contaminant removal, water infiltration and biodiversity) that are critical not just to the restoration of ecosystem services such as clean water and food production, but to the eventual regeneration of the entire array of services (Palmer and Filoso 2009).

In 2008, in a first of its kind project following nearly four years of planning, the University of Kentucky created a headwater stream system on a valley fill utilizing natural channel design (NCD) techniques (Rosgen 1998; Hey 2006) and the Forestry Reclamation Approach (FRA) for reclaiming the mined land to support vegetation (Burger et al. 2005). This project was proof-of-concept in that it sought to answer questions on "how to" retrofit an existing valley fill in an effort to restore lost stream and watershed functions (Agouridis et al. 2017; Barton et al. 2017). The project was

monitored by researchers at the University of Kentucky over a five-year postconstruction period from 2009 through 2013 with regards to geomorphology, hydrology, vegetation, water quality, and habitat (Blackburn-Lynch 2015; Agouridis et al. 2017). Some hydrologic parameters, but not all, were deemed similar to a reference stream with the most notable difference being the reduction in baseflow due to a disconnect with the groundwater table (Blackburn-Lynch 2015; Agouridis et al. 2017).

The overarching purpose of my research was to answer the question of whether lost headwater stream and watershed ecological functions were restored to the retrofitted watershed. But first, a characterization of the ecology and structure of the benthic macroinvertebrate communities in the recreated stream was necessary and is the subject of this dissertation. Over a one-year period from 2014 through 2015, I collected monthly samples of benthic macroinvertebrates, benthic substrate, water chemistry, and various other environmental attributes from the recreated stream on the retrofitted valley fill and at two contrasting reference streams: (1) a relatively pristine headwater stream in an unmined watershed, and (2) a mine-impacted headwater stream immediately downgradient of a traditional valley fill. The primary objective was to assess the recreated stream and determine whether the benthic macroinvertebrate community structure and function was more similar to the unmined reference stream or the mineimpacted reference stream. However, biological impairment downstream of valley fills is well documented in the literature (Pond et al. 2008; USEPA 2011b) and is therefore not a primary focus of my study. The secondary objective of my study was to assess the continuity of ecological structure and function at the recreated stream by comparing the benthic macroinvertebrate community from the uppermost 100 m section of the recreated

channel to the community from the next 100 m section of contiguous channel

immediately downstream. I calculated 16 benthic macroinvertebrate metrics (Table 1)

**Figure 1.** Valley fill (a) prior to watershed retrofit (2007), (b) stream channel construction utilizing natural channel design (NCD) techniques (2008), and (c) implementation of forestry reclamation approach (FRA) principles. (Images are courtesy of University of Kentucky)



(c)

to characterize the ecology and structure of the benthic macroinvertebrate communities and grouped them into the following four categories that are defined in the Methods section: (1) structure, (2) composition, (3) diversity, and (4) biotic integrity. This was the first extensive survey of benthic macroinvertebrates in both the recreated stream and the nearby mine-impacted stream located downgradient of a valley fill and will provide a baseline dataset for follow-up ecological assessments as well as future reclamation efforts at other valley fills. Furthermore, general inventories of invertebrate taxonomic groups are needed to help document and explain patterns of biodiversity in Kentucky (Pond 2000).

For the primary objective, I predicted that the benthic macroinvertebrate community at the recreated stream would more closely resemble the benthic macroinvertebrate community at the unmined stream than at the mine-impacted stream due to the retrofitted valley fill's mitigating effects on water quality. However, I predicted that the community dynamics would vary between the recreated stream and the unmined stream regarding the general direction of individual metric values (higher / lower) due to probable legacy effects from watershed-scale deforestation and mining. Specifically, the absence of both riparian vegetation and a light-limiting forest canopy at the retrofitted valley fill suggested higher primary production, sedimentation and turbidity levels at the recreated stream than at the unmined stream, and subsequently lower interstitial space available for habit within the benthic substrate. Additionally, the exposure of water to unweathered rocks and mining spoil at the retrofitted valley fill was predicted to result in higher conductivity levels at the recreated stream than at the unmined stream, rendering the environment suitable to fewer taxa, and/or different taxa

more able to tolerate the higher salinity. Therefore, regarding community structure (as defined in the Methods section), I predicted that the recreated stream would exhibit higher total density, but lower total richness than the unmined stream. Regarding community composition, I predicted that the recreated stream would exhibit a benthic macroinvertebrate community with a lower percentage of insects, higher percent abundance for the top two dominant taxa, and lower richness for mayflies, stoneflies, and caddisflies. Regarding diversity, I predicted that the recreated stream would exhibit lower values than the unmined stream for the Shannon index (*H*), Simpson's index of diversity (*1-D*), and Hill's  $N_1$  and  $N_2$ . Lastly, regarding biotic integrity, I predicted that the recreated stream for an EPT index, percent EPT abundance, and percent mayfly abundance, as well as higher values (less desirable) for a biotic index (HBI).

For the secondary objective, I predicted that the recreated stream would exhibit within-stream differences in benthic macroinvertebrate community dynamics between the furthermost upstream 100 m section of recreated channel and the next 100 m contiguous section of channel immediately downstream. As with my primary objective, the probable legacy effects of deforestation and mining led me to predict that the macroinvertebrate communities would differ between upstream and downstream sections due to varying degrees of disruption to the linkages with adjacent ecosystems, particularly the distance from the unmined upper portion of the watershed (longitudinal dimension), and also the cross-sectional distance between forest edges on either side of the recreated stream (lateral dimension). Additionally, I predicted that community dynamics would vary between the up section and the down section regarding the general direction of individual

metric values (higher / lower). Regarding community structure, I predicted that the up section would exhibit higher total density, and higher total richness than the down section. Regarding community composition, I predicted that the up section would exhibit lower percent abundance for the top two taxa and higher percent abundance for insects, as well as higher richness for mayflies, stoneflies, and caddisflies. Regarding diversity, I predicted that the up section would exhibit higher values than the down section for the Shannon index (H), Simpson's index of diversity (I-D), and Hill's  $N_I$  and  $N_2$ . Lastly, regarding biotic integrity, I predicted that the up section would exhibit higher values than the down section for an EPT index, percent EPT abundance, and percent mayfly abundance, as well as lower values (more desirable) for a biotic index (HBI). Furthermore, I predicted that the metrics for the up section of the recreated stream would more closely resemble the metrics at the unmined stream, whereas the metrics for the down section would be less similar.

For brevity and clarity, the results section of this dissertation reports metric values from the middle month of each season, i.e., April (spring), July (summer), October (autumn), and January (winter), in addition to a generalization of the macroinvertebrate response throughout the entire study period. The complete monthly results of the metric calculations and all supporting data are provided in various figures, tables, and appendices. This dissertation represents the most comprehensive quantitative survey to date of the benthic macroinvertebrate community ecology and structure at a recreated headwater stream on a "proof-of-concept" retrofitted valley fill, and contributes 1) a baseline biological dataset that will inform long-term monitoring projects at the site, 2) a comprehensive biological component to support and expand on the work of Blackburn-

Lynch (2015) and Agouridis et al. (2017), and 3) knowledge of how benthic macroinvertebrate community dynamics respond at a headwater stream system recreated on a valley fill in a watershed retrofitted using NCD techniques and FRA principles in the Central Appalachian Coalfields region of the USA.

#### METHODS

Study area—The study was conducted at the University of Kentucky's Robinson Forest (37°27'N, 83°08'W), in the Central Appalachians region of southeastern Kentucky (Figure 2). Robinson Forest is an approximately 6,000-ha teaching, research and extension experimental forest located in the rugged eastern section of the Cumberland Plateau in portions of Breathitt, Perry and Knott counties. The forest comprises eight discontinuous properties, with a 4,200-ha main block that contains some of the least disturbed watersheds in eastern Kentucky (Villines et al. 2015; Agouridis et al. 2017; University of Kentucky 2021). Although the main block has remained isolated from surface mining (Villines et al. 2015), nearly all the adjacent properties have been surface mined for coal (Williamson et al. 2015). The forest was last harvested for timber between 1890 and 1920 (Overstreet 1984) and the 90+ year-old regenerated forest is classified as mature mixed-mesophytic (Witt 2012; Villines et al. 2015; Williamson et al. 2015). The topographic, soil, and climate data for Robinson Forest can be found elsewhere (Overstreet 1984; McDowell 1985; Blackburn-Lynch 2015; Williamson et al. 2015; and Agouridis et al. 2017).

During the mid-1990s, a nearly 810-ha section of Robinson Forest was clear-cut of vegetation and mined for coal, resulting in the creation of several valley fills—two of which were created in the Wharton Branch and Guy Cove watersheds (Blackburn-Lynch 2015). However, a 9-ha section at the uppermost portion of the Guy Cove watershed was not mined and only experienced some clearing of vegetation because the coal seam ended before it reached the valley ridgetop (Agouridis et al. 2017). A small spring-fed channel



**Figure 2.** Location map of study area with state and county boundaries. Grayed portion denotes the Appalachian coalfield. The inset map shows stream site locations within Breathitt County, KY USA. The unmined watershed (darkened circle) and the two mined watersheds (darkened triangle) are separated by approximately 7 km (4 miles).

flows nearly year-round (average baseflow of 55 m<sup>3</sup> d<sup>-1</sup>) from this unmined section and adjoins with the created stream channel (Agouridis et al. 2017).

*Stream selection*—Three headwater streams located in contrasting watersheds were selected for this study, (1) a stream constructed on the top of a valley fill in a retrofitted watershed, (2) a stream immediately down-gradient of a traditionally constructed valley fill, and (3) a stream in a relatively pristine watershed. The constructed stream is an ephemeral/intermittent stream located in the Guy Cove watershed (37°24′N, 83°10′W). It was designated as "GC" and hereafter referred to as the "created stream".



**Figure 3**. The Guy Cove valley fill viewed from downstream. The original headwater stream was at the bottom of the 'V' shaped fill. The created stream was constructed on top of the valley fill and establishes a 'natural' connection between the unmined upper watershed and the lower watershed downgradient of the valley fill.

The stream located down-gradient of a valley fill is a perennial stream located in the Wharton Branch watershed (37°25'N, 83°10'W). It was designated as "WB" and hereafter referred to as the "mine-impacted stream". The relatively pristine stream is a perennial stream located in the Little Millseat watershed in the main block of Robinson Forest (37°28'N, 83°09'W). It was designated as "LM" and hereafter referred to as the "unmined stream". The unmined and mine-impacted streams were used as reference streams to which the created stream was compared. The Little Millseat watershed has a long-term history of good water quality (Blackburn-Lynch 2015; Villines et al. 2015; Williamson et al. 2015; Agouridis et al. 2017) and has been the focus of published research on stream delineation and stream permanence, as well as forest longevity as a function of mine reclamation strategies and projected climate change (Sena et al. 2020). Detailed summaries and site characteristics for all three streams and their watersheds are described elsewhere (Cherry 2006; Blackburn-Lynch 2015; Villines et al. 2015; Agouridis et al. 2017; Sena et al. 2020). A single 100 m sampling section was selected at both the unmined stream and the mine-impacted stream. At the created stream, two contiguous 100 m sampling sections were selected that composed the first 200 m of constructed channel beginning where the head of the valley fill adjoins the 9-ha unmined portion of the watershed. The upstream section was designated as "GU" and hereafter referred to as the "up section", and the downstream section was designated as "GD" and hereafter referred to as the "down section". The sampling sections at each stream were delineated prior to the start of the study and subdivided into five 20 m subsections identified as location numbers 1, 2, 3, 4, 5 (n = 5 per reach, with 1 furthest upstream) using fixed reference objects and/or temporary markers. The sampling sections and

**Figure 4.** Study Sites: (a) a stream created on a valley-fill in a retrofitted watershed, (b) a relatively pristine stream in unmined watershed, and (c) a mine-impacted stream downgradient of a traditional valley fill



delineations remained constant throughout the year-long study period, with one random sample collected monthly from within each subsection.

*Field methods*—I collected benthic macroinvertebrates using a quantitative method once per month during a twelve-month period from February 2014 through
January 2015. Quantitative data were taken from five replicate Surber samples  $(0.09 \text{ m}^2)$ area; 0.25 mm mesh) stratified along a 100 m longitudinal transect within the thalweg (i.e., deepest path of flow) of each channel to ensure the highest species richness and abundance of macroinvertebrates (Brown and Brussock 1991; Feminella 1996). Individual samples were collected by disturbing the benthic substrate with a substrateappropriate tool to a depth of ca. 10 cm for ca. 30 seconds. If present, emergent macrophytes within the bounds of the Surber frame were sheared along the surface of the water and then along the bottom of the channel, with the portion from within the water column rinsed of macroinvertebrates and then retained in order to quantify substrate density. Large rocks located within the bounds of the Surber frame were placed into a bucket, individually rinsed and examined for macroinvertebrates, and then placed back in the stream. Samples were elutriated with buckets and a 0.25 mm mesh (U.S. No. 60) sieve and preserved in bags containing 90% ethyl alcohol. An effort was made in the field to preliminarily separate macroinvertebrates from benthic substrate, and with the exception of stones, to save as much of the debris (twigs, leaves, plants, etc.) collected in the Surber sampler as possible for later sorting and quantification. In July, the created stream channel was completely dry, therefore I dug out the substrate from within the bounds of the Surber frame to an approximate depth of 10 cm and bagged the sample for rehydration in the laboratory the next day.

*Laboratory methods*—In the lab, samples were rinsed with tap water through nested 1 mm (U.S. No. 18), 0.25 mm, and 0.63  $\mu$ m (U.S. No. 230) sieves for separation of invertebrate and non-invertebrate organic matter and fine sediments. The non-invertebrate benthic matter retained by the 1 mm sieve and larger than ca. 30 mm

(approximately the size of a quarter in U.S. currency) was separated into disposable aluminum pans and categorized by substrate type as either woody debris, aquatic vegetation, or leaf litter. The invertebrates retained by the 1 mm sieve were collected under a table-top magnifying glass and the remaining non-invertebrate benthic matter (less than ca. 30 mm) was rinsed into disposable aluminum pans and categorized as course benthic organic matter (CBOM). The portion of the sample retained by the 0.25 mm sieve was subsampled with a standard plankton splitter and the invertebrates collected from under a dissecting microscope (7.5x - 50x). The remaining particulate matter was rinsed into disposable aluminum pans and categorized as fine benthic organic matter (FBOM). The 0.63 µm sieve was used in order to retain as much of the original sample as possible during the rinsing process, with the contents rinsed into disposable aluminum pans and categorized as ultra-fine benthic organic matter (UBOM). All samples in disposable aluminum pans were dried (60 °C), weighed, ashed (500 °C) and reweighed to determine the ash-free dry mass (AFDM) for estimation of biomass (Steinman et al. 2006). Only macroinvertebrates retained by the 1 mm sieve were used in this study.

All organisms were identified to the taxonomic level of genus (Merritt et al. 2008; Thorp and Covich 2010; Morse et al. 2017), with the following exceptions: (1) most dipterans from the family Chironomidae (to sub-family or tribe), (2) some ephydrid, empidid and sciarid dipterans (to family), and (3) aquatic worms (to family). A representative sample of all invertebrates was sent to a credible specialist All organisms were further classified according to functional feeding group, behavioral habit, and biotic index tolerance. All insects from the taxonomic orders Ephemeroptera, Plecoptera, and

Trichoptera (50 genera comprising 11,733 individuals), and all mollusks from the orders Basommatophora and Sphaeriida (2 genera comprising 13,627 individuals) were measured for body length to the nearest mm. Monthly length-abundance histograms for each taxon were then generated to assess whether a taxon was completing successive life stages in the stream or merely transient at the time of sampling. Annual secondary production estimates were generated from the monthly length-abundance data to assess one measure of stream system ecological function but are not presented in this dissertation.

Epilithic periphyton was sampled monthly by randomly collecting rocks from within each stream's designated 100 m sampling area. Five flat cobble-size stones were collected from the thalweg of each stream site and transported back to the lab in an ice chest and stored in a freezer until being processed. During processing, I decided to combine each stream site's monthly set of five rocks into one aggregate sample in order to conserve time. Each set of rocks was placed into a scrubbing tray inscribed with a standardized unit of measure and saved as a digital image. Then the periphyton attached to the upper surface of each stone was removed through a process of scraping, scrubbing and rinsing followed by homogenization. The resulting slurry then was split into two subsamples in order to determine AFDM and chlorophyll a. The AFDM subsamples were dried (60 °C), weighed, ashed (500 °C) and reweighed to determine AFDM (Steinman et al. 2006); AFDM was used as a measure of food resource quantity  $(mg/cm^2)$ . The subsamples of Chlorophyll *a* were analyzed by spectrophotometry (Steinman et al. 2006). The ImageJ2 digital image analysis software (Schindelin et al. 2012) was used to measure the planar area of each rock in order to convert calculations of AFDM and chlorophyll a

to area-based measures. Because the created stream exhibited dense channel vegetation, sedimentation, and turbidity that made it difficult to locate rocks, I stocked the subsections of both reaches with transplanted rocks and marked the locations so that I could quickly collect them during subsequent visits. The transplanted rocks were a random mix that had been collected from the same three streams between 2011 and 2013 as part of a separate study that analyzed epilithic periphyton.

Monthly water samples were collected from the downstream portion of each stream site throughout the study period, except for Guy Cove during July 2014 when the stream channel was dry. Collection, preservation and analytic protocols were performed in accordance with standard procedures (Greenberg et al. 1992), and chemical variables were analyzed by the Environmental Analysis Laboratory (EAL) of the University of Louisville. Water temperature in each stream site was recorded hourly from September 2014 thru January 2015 using HOBO Pendant temperature/light data loggers (model UA-002-08; ONSET Computer Corporation). The forest overstory density at each stream was measured as percent canopy coverage using a spherical densiometer (Forestry Suppliers, Inc.) and generally followed the method of Lemmon (1956). Three readings (left bank, center, right bank) were taken at each of the five subsections along the transect, for a total of 15 readings per stream. The extent of cross-sectional distance between forest edges at the created stream was measured using aerial images recorded with a drone (a small Unmanned Aerial System, sUAS) deployed and piloted by staff members of the Agricultural Communications Services Department in the College of Agriculture, Food and Environment at the University of Kentucky.

*Metric selection*—Sixteen genus-level metrics were calculated in an effort to characterize the benthic macroinvertebrate community structure, composition, diversity, and biological integrity at each stream site during each month (Table 1). Although some state agencies within the Appalachian coalfield use family-level metrics, most of which approximate the strength of genus-based metrics, recent studies on the benefits of finer taxonomic resolution indicate more accurate assessments when genus- or species-level data are used (Pond et al. 2008).

In this study, community structure was defined as the totality of organisms that existed together in samples collected from one site (Begon et al. 2006) and was characterized by invertebrate density (no. of individuals  $\cdot$  m<sup>-2</sup>), and total species richness S (no. of genera). Community composition was defined as constituent taxa of the community structure (Begon et al. 2006) and was characterized by four richness metrics and two relative abundance metrics. The genus richness of the insect orders Ephemeroptera, Plecoptera, Trichoptera, and combined Ephemeroptera-Plecoptera-Trichoptera (EPT) were calculated as numbers of genera within each sample. The relative abundances for benthic insects and the top two dominant taxa were calculated as proportions of individuals within each sample. Community diversity incorporated richness, commonness and rarity (Begon et al. 2006) and was characterized with four indices of diversity. First, two common indices of entropy (i.e., the measure of disorder in a system, where more disorder implies more diversity) known as the Shannon Index, H(Spellerberg and Fedor 2003), and the Simpson Index, D (Simpson 1949), were calculated. Second, two measurements of the 'effective number of species' (true diversity) known as the Hill's numbers  $N_1$  and  $N_2$  (Hill 1973) were derived from the

aforementioned entropy indices. The Shannon Index quantifies the uncertainty that any two species randomly selected from a sample are different, where H = 0 when only one taxon is present in the collection (total certainty) and H is at a maximum (ln *S*) when all individuals are evenly distributed among the taxa (total uncertainty). The Shannon Index, H, is calculated as:

$$H = -\sum_{i=1}^{n} p_i \ln p_i$$

where *n* is the total richness in the community, and the proportion of individuals that a taxon contributes to the total in the sample is  $p_i$  for the *i*th taxon. The Simpson Index, *D* measures the probability that two individuals randomly selected from a sample will belong to the same taxon, and ranges from 0 to 1, where the value 0 represents infinite diversity (low probability) and the value 1 represents no diversity (high probability, where *D* is calculated as:

$$D = \sum_{i=1}^{n} p_i^2$$

where *n* is the total richness in the community, and the proportion of individuals that a taxon contributes to the total in the sample is  $p_i$  for the *i*th taxon. To overcome the counter-intuitive nature of the Simpson Index, *D* is subtracted from 1 so that the index represents the probability that two individuals randomly selected from a sample will belong to different taxa and is presented as the Simpson Index of Diversity (1-*D*). Thus, the higher the values of the Shannon Index, *H*, and the Simpson Index of Diversity, 1-*D*, the greater the diversity of taxa in the sample. However, because the entropy indices are nonlinear with respect to species addition, each added species leads to a smaller

increment in diversity than the species added before it (Jost et al. 2010) resulting in samples with higher levels of richness appearing more similar to one another than they otherwise would at lower levels of richness. Thus, these indices can easily be misinterpreted, and lead to incorrect inferences regarding the similarity and differentiation of communities (Jost et al. 2010). MacArthur (1972) and Hill (1973) resolved these problems by converting the entropy indices to 'effective number of species', which has the same linear metric as species richness and represents a perfectly even community with the same diversity as the original community (Jost et al. 2010). Thus, Hill's  $N_l$  diversity is the effective number of taxa for the calculated value of the Shannon Index, H, and was derived as  $e^{H}$ , and Hill's  $N_2$  diversity is the effective number of taxa for the calculated value of the Simpson Index, D, and was derived as 1/D. Thus, if all taxa are represented in equal numbers in a sample, then  $N_1 = N_2$  = species richness S. Accordingly, the higher the value of Hill's  $N_1$  and  $N_2$ , the greater the diversity, with a maximum value that corresponds to the number of taxa in the sample. Community biotic *integrity* refers to the water quality, or health of a stream (Karr 1981; Hilsenhoff 1982), based on the premise that biological communities reflect watershed conditions because they are sensitive to changes in a wide array of environmental factors (Karr 1981). The Biotic integrity was characterized by an EPT Index, percent EPT abundance, percent Ephemeroptera abundance, and the Hilsenhoff Biotic Index, or HBI. The EPT Index (Kerans and Karr 1994; Barbour et al. 1999) was calculated based on the percentage of the sum of Ephemeroptera, Plecoptera, and Trichoptera richness to the richness of all other taxa. The % EPT metric measures the relative abundance of the generally pollutionsensitive insect orders of Ephemeroptera, Plecoptera, and Trichoptera, where increasing

values indicate increasing water quality and/or habitat conditions (KDOW 2002). The percent Ephemeroptera metric measures the relative abundance of mayflies. This metric normally declines in the presence of metals and high conductivity associated with mining (KDOW 2002). The HBI was developed to summarize the overall pollution tolerance of a benthic arthropod community with a single value (Klemm et al. 1990) and was calculated as:

$$HBI = \frac{\sum n_i T V_i}{N}$$

where  $n_i$  is the number of individuals of each taxon,  $TV_i$  is the tolerance value (ranging from 0 to 10) associated with each taxon's demonstrated sensitivity to organic pollutants (with 0 being most sensitive and 10 being most tolerant), and N is the total number of individuals in the sample (Hilsenhoff 1982, 1987). The derived HBI index value is weighted by the relative abundance of each taxonomic group and ranges from 0 to 10, where decreasing HBI values reflect a greater relative abundance of sensitive taxa (e.g., mayflies and water pennies) and therefore a lower level of organic enrichment, whereas increasing HBI values reflect greater abundance of tolerant taxa (e.g., midges and aquatic worms) potentially indicating higher levels of organic enrichment. Several states, including Kentucky, have used a modified Hilsenhoff Biotic Index (mHBI) to assess impacts other than organic enrichment and have found the mHBI to be a valuable metric (KDOW 2002). I calculated HBI values following Hilsenhoff (1982, 1987) with the exceptions of not subsampling, and not always obtaining a minimum of 100 total invertebrates in a sample. I also did not make modifications that accounted for seasonal variability (Hilsenhoff 1988) or dominant taxa (Hilsenhoff 1998). I used tolerance values  $(TV_i)$  developed by the North Carolina Division of Environmental Management

Variables (y)	Category	Description			
Invertebrate Density (no. · m <sup>-2</sup> )	Structure	Figure 5. Number of benthic invertebrates per square meter			
Total Richness (S)	Structure	Figure 6. Genus-level except aquatic worms (family) & dipteran midges (sub-family or tribe)			
% Benthic Insects	Composition	Figure 7. Percentage of insects to total invertebrates			
% Top 2 Dominant Taxa	Composition	Figure 8. Percentage of two most abundant taxa to total invertebrates			
EPT Richness	Composition	Figure 9. Count of mayfly, stonefly, and caddisfly (EPT) genera			
Ephemeroptera Richness	Composition	Figure 10. Count of mayfly genera (E)			
Plecoptera Richness	Composition	Figure 11. Count of stonefly genera (P)			
Trichoptera Richness	Composition	Figure 12. Count of caddisfly genera (T)			
Shannon Index (H)	Diversity	Figure 13. Gives more weight to # of taxa and strongly influenced by rare taxa Sensitive to small diversity changes – assesses actual state of community			
Hill's N <sub>1</sub> Diversity ( $e^{H}$ )	Diversity	Figure 14. Effective # of species in community - weighted for abundance of rare species			
Simpson Index of Diversity (1 - D)	Diversity	Figure 15. Gives more weight to more common/abundant (dominant) taxa and sample evenr Not affected by rare taxa – assesses trending direction of community			
Hill's N <sub>2</sub> Diversity (1/D)	Diversity	Figure 16. Effective # of species in community - weighted for abundance of common specie			
% EPT Index	Biotic Integrity	Figure 17. Percentage of EPT genera to total invertebrate genera count			
% EPT Abundance	Biotic Integrity	Figure 18. Percentage of mayfly, stonefly, and caddisfly abundance to total invertebrates			
% Ephemeroptera Abundance	Biotic Integrity	Figure 19. Percentage of mayfly abundance to total invertebrates			
Hilsenhoff Biotic Index (HBI)	Biotic Integrity	Figure 20. Range from $0 - 10$ ; Low scores reflect higher abundances of taxa that are sensitivorganic pollution which suggests higher water quality and lower environmental stress			

Table 1. Summar	y of metric	variables (v	) us	ed for	statistical	analyses.
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(NCDEM) (Lenat 1993) that have been regionally modified for streams of the southeastern United States and adjusted for Kentucky streams with some values developed from Kentucky Division of Water data (KDOW 2002).

*Data analysis*—All statistical tests were conducted in the R platform (ver. 4.0.2) (R Core Team 2019). The data were modeled with the generalized linear model using the normal distribution with the response variable, *y*, being any one of the biotic variables (Table 1). The explanatory variables were all nominal and were Stream type (*Styp*: 'WB',' GD',' GU',' LM'), Month Name (*Mname*: 'Jan', 'Feb', ..., 'Dec'), and Location number (*Lno*: '1','2','3','4','5'). The explanatory variables *Styp* and *Mname* were treated as fixed effect variables and *Lno* was treated as a random effect variable. In the model, *Styp* and *Mname* are crossed and *Lno* is nested in *Styp* but crossed with *Mname*. Correlation (nonindependence) among repeated measures from within the same stream was accounted for by treating sample location (*Lno*, which were replicates) as a blocked random effect variable.

The goodness-of-fit of the model residuals to the normal distribution was determined by examination of the histogram of the residuals and the assumption of homogeneity of variance was tested by examination of the plot of residuals vs the predicted value. Many of the response variables did not agree with the assumption of normality, so tests of hypotheses were done by modeling the rank transform of the original *y* values. The global null hypothesis of no *Styp:Mname* interaction was tested by determining if dropping it from the model caused a reduction in AIC (Aikaike information criterion) and the associated p-value was obtained from the likelihood ratio test. Tukey's Honest Significant Difference (HSD) test was used as a post-hoc means

separation procedure to determine significant differences among stream types for each month (alpha=0.05). If the *Styp:Mname* interaction was not significant, then it was dropped from the model and a Tukey's comparison of the levels of *Styp* was done using the model with no interaction. If the *Styp:Mname* interaction was significant, then it was not dropped from the model and a Tukey's comparison of the levels of *Styp* was done separately for each level of *Mname* using the model with interaction.

Models involving random effects were fit using the *lme* function in the *nlme* package (Pinheiro et al. 2020) of R. Models not involving random effects were fit using the *gls* function in the *nlme* package of R. Tukey's Honest Significant Difference (HSD) test was implemented with the *predictmeans* function of R in the *predictmeans* package (Dongwen et al. 2020) of R. Standard errors of estimated means were estimated by fitting the untransformed *y* value and estimating the standard error with the *predictmeans* function.

## RESULTS

### **Environmental data**

*Summary*—Average, SE, and range values for all environmental variables are shown in Table 2. Percent canopy coverage was higher at the reference streams (LM, average = 67.8 %; WB, average = 84.7 %) than at the created stream (GU, average = 0%; GD, average = 0 %). The distance between forest edges was higher at the created stream (GU, average = 27.4 m; GD, average = 105.4 m) than at the reference streams where the intact riparian forest generally followed the edges of the bankfull channel width (not measured). Water temperature was lower at the unmined reference stream (LM, average = 8.8 °C) than at the created stream (GU, average = 10.5 °C; GD, average = 10.4 °C) and highest at the mine-impacted stream (WB, average = 13.7 °C). Periphyton and chlorophyll a were lower at both reference streams (LM, average = 0.05 mgAFDM/m<sup>2</sup> and 0.05  $\mu$ g/cm<sup>2</sup>, respectively; WB, average = 0.10 mg AFDM/m<sup>2</sup> and 0.08  $\mu$ g/cm<sup>2</sup>, respectively) than at the created stream (GU, average = 0.16 mg AFDM/m<sup>2</sup> and  $0.10 \ \mu g/cm^2$ , respectively; GD, average =  $0.23 \ mg \ AFDM/m^2$  and  $0.39 \ \mu g/cm^2$ , respectively). Aquatic vegetation and  $NO_2 + NO_3$  were lower at both reference streams (LM, average =  $0.0 \text{ mg AFDM/m}^2$  and 87.0 mg/L, respectively; WB, average = 0.0 mgAFDM/m<sup>2</sup> and 87.8 mg/L, respectively) than at the created stream (GU, average = 80.8mg AFDM/m<sup>2</sup> and 191.6 mg/L, respectively; GD, average = 41.7 mg AFDM/m<sup>2</sup> and 171.2 mg/L, respectively). Leaf detritus and woody debris were higher at both reference

streams (LM, average = 28.2 and 11.8 mg AFDM/m<sup>2</sup>, respectively; WB, average = 7.5 and 4.2 mg AFDM/m<sup>2</sup>, respectively) than at the created stream (GU, average = 2.7 and 2.1 mg AFDM/m<sup>2</sup>, respectively; GD, average = 0.5 mg and 1.2 mg AFDM/m<sup>2</sup>,

respectively). Measurements of benthic organic matter (course, fine, and ultra-fine) were lower at both reference streams (LM, average = 22.4, 6.6, 1.7 mg AFDM/m<sup>2</sup>,

respectively; WB, average = 6.6, 1.6, 0.3 mg AFDM/m<sup>2</sup>, respectively) than at the created stream (GU, average = 53.7, 41.4, 14.7 mg AFDM/m<sup>2</sup>, respectively; GD, average = 59.4, 44.3, 16.9 mg AFDM/m<sup>2</sup>, respectively). Chloride was lower at the unmined stream (LM, average = 0.63 mg/L) than at the created stream (GU, average = 1.42 mg/L; GD, average = 0.89 mg/L) and at the mine-impacted stream (WB, average = 1.20 mg/L). The pH values at all 4 steam sites were within the range of circum-neutral to mildly alkaline (6.0 – 10.0 SU), with values lower at both reference streams (LM, average = 6.9 SU; WB average = 6.6 SU) than at the created stream (GU, average = 7.3 SU; GD, average = 7.4 SU). Specific conductance and sulfate were lower at the unmined reference stream (LM, average = 57  $\mu$ S/cm and 7.8 mg/L, respectively) than at the created stream (GU, average = 479  $\mu$ S/cm and 77.2 mg/L, respectively) and highest at the mine-impacted reference stream (WB, average = 2,440  $\mu$ S/cm and 1,522.9 mg/L, respectively).

#### **Benthic macroinvertebrate communities**

*Summary*—A total of 140 taxa (137 insect, 2 mollusk, 1 annelid) representing 10 orders, 57 families and 70,382 individuals were collected from the four sites in all months

	Created	d Stream	Reference Streams			
Parameter	Down Section (GD)	Up Section (GU)	Unmined (LM)	Mine-impacted (WB)		
Sp. Conductivity (µS/cm) <sup>a</sup>	$479.0 \pm 48.0 (208 - 702)$	$427.0 \pm 44.0 (223 - 651)$	$57.0 \pm 5.0 (37 - 83)$	$2440.0 \pm 170.0 (691 - 2878)$		
SO4 <sup>2-</sup> (mg/L) <sup>a</sup>	77.2 $\pm$ 6.4 (43.3 - 120.2)	$67.7  \pm  4.8 \ (39.6 - 89.2)$	$7.8 \pm 0.7 (3.1 - 12.6)$	$1522.9 \ \pm 106.3 \ (419.3 - 1896.0)$		
Cl <sup>-</sup> (mg/L) <sup>a</sup>	$0.89 ~\pm~ 0.06 ~(0.58 - 1.29)$	$1.42 \pm 0.10 (0.88 - 1.97)$	$0.63 \pm 0.09 (0.26 - 1.15)$	$1.20 \pm 0.20 (0.48 - 2.86)$		
pH (standard units) <sup>a</sup>	$7.4 \pm 0.2 (6.0 - 8.6)$	7.3 $\pm$ 0.2 (6.1 – 8.3)	$6.9 \ \pm \ 0.3 \ (5.5 - 8.4)$	$6.6 \pm 0.2 (5.4 - 7.8)$		
$NO_2 + NO_3 (mg/L)^a$	171.2 ± 31.9 (37 – 403)	191.6 ± 38.1 (35 – 434)	87.0 ± 18.1 (21 – 211)	87.8 ± 17.1 (38 – 242)		
Aquatic Veg. (mg AFDM/m <sup>2</sup> ) <sup>b c</sup>	41.7 ± 9.2 (8.4 – 124.7)	$80.8 \pm 16.2 (21.9 - 200.7)$	$0.0 \pm 0.0 (0.0 - 0.0)$	$0.0 \pm 0.0 (0.0 - 0.0)$		
Leaf Detritus (mg AFDM/m <sup>2</sup> ) <sup>b c</sup>	$0.5 \pm 0.2 (0.0 - 1.6)$	$2.7 \pm 1.3 (0.1 - 14.0)$	$28.2 \pm 10.4 (0.0 - 127.8)$	$7.5 \pm 1.9 (0.2 - 20.6)$		
Woody Debris (mg AFDM/m <sup>2</sup> ) <sup>b c</sup>	$1.2 \pm 0.5 (0.0 - 3.9)$	$2.1 \pm 1.2 (0.0 - 14.7)$	$11.8 \pm 2.2 (1.1 - 27.2)$	$4.2 \pm 1.9 (0.0 - 22.8)$		
CBOM (mg AFDM/m <sup>2</sup> ) <sup>b d</sup>	$59.4 \pm 9.1 (24.4 - 129.9)$	$53.7 \pm 5.1 (19.1 - 91.5)$	$22.4 \pm 3.8 (5.2 - 48.5)$	$6.6 \pm 2.4 (1.8 - 32.5)$		
FBOM (mg AFDM/m <sup>2</sup> ) <sup>b e</sup>	44.3 ± 7.8 (7.0 – 98.4)	41.4 ± 6.1 (14.4 – 79.9)	$6.6 \pm 1.5 (0.6 - 19.0)$	$1.6 \pm 0.3 (0.3 - 4.8)$		
UBOM (mg AFDM/m <sup>2</sup> ) <sup>b f</sup>	$16.9 \pm 4.9 (4.5 - 55.8)$	$14.7 \pm 3.7 (6.3 - 46.0)$	$1.7 \pm 0.7 (0.2 - 8.9)$	$0.3 \pm 0.1 (0.1 - 0.7)$		
Water Temperature (°C) <sup>g</sup>	$10.4 \pm 0.1 (0.3 - 21.6)$	$10.5 \pm 0.1 (0.3 - 18.2)$	$8.8 \pm 0.1 (0.5 - 18.2)$	$13.7 \pm 0.0 (8.9 - 18.0)$		
Canopy Coverage (%) <sup>h</sup>	$0.0 \pm 0.0 (0.0 - 0.0)$	$0.0 \pm 0.0 (0.0 - 0.0)$	$67.8 \pm 1.3(58.4 - 76.1)$	$84.7 \pm 0.8 (80.2 - 90.6)$		
Length between forest edges $(m)^{i}$	$105.4 \pm 3.1 (42.2 - 137.3)$	$27.4 \pm 1.1 (14.5 - 40.3)$	*	Ť		
Periphyton (mg AFDM /m <sup>2</sup> ) <sup>j</sup>	$0.23 \pm 0.02 (0.15 - 0.40)$	$0.16 \pm 0.02 (0.11 - 0.39)$	$0.05 \pm 0.00 (0.03 - 0.08)$	$0.10 \pm 0.01 (0.07 - 0.14)$		
Chlorophyll <i>a</i> ( $\mu$ g/cm <sup>2</sup> ) <sup>k</sup>	$0.39 \pm 0.12 (0.08 - 1.55)$	$0.10 \pm 0.01 (0.02 - 0.16)$	$0.05 \pm 0.01 (0.01 - 0.11)$	$0.08 \pm 0.03 (0.01 - 0.32)$		

**Table 2.** Summarized data from measurements of physicochemical parameters, benthic substrates, and land cover. Values are averages  $(\pm SE)$  with ranges. Monthly data for sample replicates at each site can be found in Appendix D.

<u>CBOM</u> = course benthic organic matter, FBOM = fine benthic organic matter, UBOM = ultra-fine benthic organic matter

<sup>a</sup>Measured once monthly (n) in each stream (n = 12 at LM & WB; n = 11 at GD & GU due to dry channel in July 2014).

<sup>b</sup>Quantified from contents retained by Surber net during benthic invertebrate sampling (n = 60 per stream [5 samples per month x 12 months]).

<sup>e</sup> Retained by a 1-mm sieve and larger than approximately 30 mm in size (half-dollar coin).

<sup>d</sup>Any non-invertebrate organic matter retained by a 1-mm sieve and smaller than approximately 30 mm in size (half-dollar coin).

<sup>e</sup> Any non-invertebrate organic matter that passes through a 1-mm sieve and is retained by a 0.250 µm sieve.

<sup>f</sup> Any non-invertebrate organic matter that passes through a 0.250 µm sieve and is retained by a 0.125 µm sieve.

<sup>g</sup>Hourly readings from mid-September 2014 to mid-January 2015 (n = 2,997 per stream) with HOBO data loggers.

<sup>h</sup>Densiometer readings (September 16<sup>th</sup>, 2016): average of three readings (left bank, center of channel, right bank) at each of five transects.

<sup>i</sup> Cross-section length (m) between forest edges on either side of stream at approx. 1.5 m intervals using aerial image (n = 60 per 100 m section).

<sup>j</sup> Epilithic algae scraped from flat cobble-size stones collected from stream (n = 5 stones per stream per month, [replicates combined, n = 12 per stream]).

<sup>k</sup>Chlorophyll *a* calculated from epilithic periphyton scraped from stones (n = 5 stones per stream per month, [replicates combined, so n = 12 per stream]).

†Cross-sectional distance between tree-lines indeterminate by aerial image due to canopy coverage and stream embeddedness within forest.

combined (Appendix A). Aggregate taxa richness in all months combined was higher at the created stream (GU, 105; GD, 85) than at the reference streams (LM, 85; WB 28). Aggregate abundance (individuals) in all months combined was higher at the created stream (GU, 32,762; GD, 28,822) than at the reference streams (LM, 8,646; WB, 153).

The numbers of individuals were not evenly distributed among the observed taxa at the created stream and the mine-impacted stream. At GU, the 20 most abundant taxa represented 19.0% of the richness and accounted for 92.7% of the individuals, and the five most abundant taxa represented 4.8% of the richness and accounted for 61.0% of the individuals. At GD, the 20 most abundant taxa represented 23.5% of the richness and accounted for 94.7% of the individuals, and the five most abundant taxa represented 5.9% of the richness and accounted for 75.7% of the individuals. At WB, the 20 most abundant taxa represented 71.4% of richness and accounted for 94.8% of the individuals, and the five most abundant taxa represented 17.9% of the richness and accounted for 70.6% of the individuals. In contrast, the general patterns of relative abundant taxa represented 23.5% of the richness and accounted for 76.4% of the individuals, and the five most abundant taxa represented 5.9% of the richness and accounted for 76.4% of the individuals, and the five most abundant taxa represented 5.9% of the richness and accounted for 76.4% of the individuals, and the five most abundant taxa represented 5.9% of the richness and accounted for 38.9% of the individuals.

## Created Stream

*Up section (GU)*—The most diverse taxon in all months combined was Diptera (true flies) with 14 families that comprised 41 genera, followed in descending order by Coleoptera (beetles) with six families and 20 genera; Trichoptera (caddisflies) with eight

families and 13 genera; Plecoptera (stoneflies) with six families and 12 genera; Odonata (dragonflies and damselflies) with six families and eight genera; Ephemeroptera (mayflies) with five families and six genera; Megaloptera (dobsonflies and alderflies) with two families and two genera; Bassomatophora (aquatic snails) with 1 genus; Sphaeriida (pea clams) with 1 genus; and Haplotaxida (aquatic annelid worms) with 1 family.

The five most abundant taxa in all months combined were the non-biting midges in the sub-family Tanypodinae (Diptera: Chironomidae) with 18.4% of the individuals, followed in descending order by the pea clam *Pisidium* with 15.4%; non-biting midges in the tribe Tanytarsini (Chironomidae: sub-family Chironominae) with 10.4%; all other non-biting midges in the sub-family Chironominae with 9.5%; and the aquatic snail *Fossaria* with 7.3%. In all months combined, there were 1,264 mayflies, 1,751 stoneflies, and 2,158 caddisflies recorded that composed an EPT richness of 31 genera and an EPT index of 29.5%. The relative abundance of Ephemeroptera was 3.9% and the aggregate EPT abundance was 15.8%.

There were 14 taxa (13.3% of richness) recorded in GU that were not found at the other sites. Coleoptera had the most taxa (seven) found only at GU, which were the beetles *Celina*, *Desmopachria*, *Haliplus*, *Cymbiodyta*, *Enochrus*, and *Helochares*, followed in descending order by Diptera (five taxa) with the true flies *Dasyhelea*, *Haemagogus*, *Eristalsis*, *Scatella*, and *Nemotelus*; Ephemeroptera (one taxon) with the mayfly *Attenella*; Trichoptera (one taxon) with the caddisfly *Oxyethira*; and Sphaeriida (one taxon) with the pea clam *Pisidium*.

*Down section (GD)*—The most diverse taxon in all months combined was Diptera (true flies) with 14 families that comprised 34 genera, followed in descending order by Coleoptera (beetles) with five families and 15 genera; Odonata with six families and 11 genera (seven dragonfly and four damselfly); Trichoptera (caddisflies) with eight families and 10 genera; Plecoptera (stoneflies) with three families and seven genera; Ephemeroptera (mayflies) with four families and four genera; Megaloptera with two families and two genera (one alderfly and one dobsonfly); Bassompatophora (aquatic snail) with one genus; and Haplotaxida (aquatic annelid worms) with one family.

The most five abundant taxa in all months combined were the non-biting midges in the tribe Tanytarsini (sub-family Chironominae) with 23.2% of the abundance, followed in descending order by the aquatic snail *Fossaria* with 21.4%; non-biting midges in the sub-family Tanypodinae with 21.3%; non-biting midges in the sub-family Orthocladiinae with 6.1%; and the caddisfly *Ironoquia* with 3.7%. In all months combined, there were 657 mayflies, 1,054 stoneflies, and 1,290 caddisflies recorded that composed an EPT richness of 21 genera and an EPT index of 24.7%. The relative abundance of Ephemeroptera was 2.3% and the aggregate EPT abundance was 10.4%.

There were 8 taxa (9.4% of richness) recorded in GD that were not found at the other sites. Odonata had the most taxa (three) found only at GD, which were the damselflies *Amphiagrion* and *Enallagma*, and the dragonfly *Pentala*, followed in descending order by Coleoptera (two taxa) with the beetles *Copelatus* and *Hydroporus*; Diptera (two taxa) with the true flies *Alluaudomyia* and *Monohelea*; and Trichoptera (one taxon) with the caddisfly *Oecetis*.

## Reference Streams

*Unmined stream (LM)*—The most diverse taxon in all months combined was Diptera (true flies) with seven families that comprised 31 genera, followed in descending order by Trichoptera (caddisflies) with 12 families and 15 genera; Plecoptera (stoneflies) with seven families and 15 genera; Ephemeroptera (mayflies) with six families and 11 genera; Coleoptera (beetles) with three families and five genera; Odonata (dragonflies) with three families and three genera; Megaloptera with two families and one genus each of alderfly and dobsonfly; Bassomatophora (aquatic snail) with one genus; Sphaeriida (pea clam) with one genus; and Haplotaxida (aquatic annelid worms) with one family.

The five most abundant taxa in all months combined were the non-biting midges in the sub-family Chironominae (other than Tanytarsini) with 14.9% of the individuals, followed in descending order by the mayfly *Paraleptophlebia* with 7.4%; non-biting midges in the sub-family Orthocladiinae with 6.8%; non-biting midges in the sub-family Tanypodinae with 5.4%; and the crane fly *Hexatoma* (Diptera: Tipulidae) with 4.4%. In all months combined, there were 1,723 mayflies, 1,166 stoneflies, and 634 caddisflies recorded that composed an EPT richness of 40 genera and an EPT index of 47.6%. The relative abundance of Ephemeroptera was 19.9% and the aggregate EPT abundance was 40.7%.

There were 24 taxa (28.2% of richness) recorded at LM that were not found at the other sites. Ephemeroptera had the most taxa (seven) found only at LM, which were the mayflies *Drunella*, *Ephemerella*, *Ephemera*, *Epeorus*, *Maccaffertium*, *Stenacron*, and *Habrophlebia*, followed in descending order by Diptera (six taxa) with the true flies *Procleon*, *Stempellina*, *Chelifera*, *Prosimulium*, *Leptotarsus*, and *Molophilus*; Trichoptera

(six taxa) with the caddisflies *Agapetus*, *Goera*, *Lepidostoma*, *Molanna*, *Dolophilodes*, and *Lype*; Plecoptera (four taxa) with the stoneflies *Haploperla*, *Sweltsa*, *Ostrocerca*, and *Remenus*; and Coleoptera (one taxon) with the beetle *Oulimnius*.

There were 13 taxa (15.3% of richness) recorded at LM that were also found at GU, but not GD. Diptera had the most taxa (four) found in common between LM and GU, which were the true flies *Dicranota, Hexatoma, Limnophila,* and *Pedicia,* followed in descending order by Plecoptera (three taxa) with the stoneflies *Peltoperla, Acroneuria,* and *Eccoptura*; Trichoptera (three taxa) with the caddisflies *Hydropsyche, Pycnopsyche,* and *Polycentropus*; Ephemeroptera (one taxon) with the mayfly *Paraleptophlebia*; Coleoptera (one taxon) with the beetle *Helichus*; and Sphaeriida (one taxon) with the pea clam *Pisidium.* There was 1 taxon (15.3% of richness) recorded at LM that was also found at GD, but not GU. The crane fly *Ormosia* (Diptera: Tipulidae) was found in common with LM and GD.

*Mine-impacted stream (WB)*—The most diverse taxon in all months combined was Diptera (true flies) with five families that comprised 13 genera, followed in descending order by Plecoptera (stoneflies) with four families and five genera; Ephemeroptera (mayflies) with three families and three genera; Trichoptera (caddisflies) with three families and three genera; Megaloptera with two families and two genera (one alderfly and one dobsonfly); Odonata (dragonflies) with one genus; and Coleoptera (beetles) with one genus.

The five most abundant taxa in all months combined were the non-biting midges in the sub-family Orthocladiinae with 28.1% of individuals, followed in descending order by non-biting midges in the sub-family Tanypodinae with 12.4%; the crane fly *Tipula* 

with 12.4%; non-biting midges in the sub-family Chironominae with 10.5%; and the stonefly *Amphinemura* with 7.2%.

In all months combined, there were four mayflies, 22 stoneflies, and 11 caddisflies recorded that composed an EPT richness of 11 genera and an EPT index of 39.3%. The relative abundance of Ephemeroptera was 2.6% and the aggregate EPT abundance was 24.2%. There was one taxon recorded at WB that was not found at the other sites: the stonefly genus *Yugus* (Plecoptera: Perlodidae). The mayfly, stonefly, and caddisfly taxa recorded in the mine-impacted stream (WB) were excluded from EPT metric calculations and a value of 0 was used for statistical analyses of EPT richness; Ephemeroptera richness; Plecoptera richness; Trichoptera richness; percent EPT index; percent EPT abundance; Percent Ephemeroptera abundance (refer to Discussion section for rationale).

### Benthic macroinvertebrate community metrics

### Annual overview

Only two of the 16 macroinvertebrate metrics in my analysis showed contrasting patterns of community dynamics between the created stream and the unmined stream (density, Figure 5; and % benthic insects, Figure 7), whereas all 16 metrics showed contrasting patterns between the created stream and the mine-impacted stream. Nonetheless, 15 of the 16 metrics showed statistically significant differences between the created stream and the unmined stream (the exception being % top two dominant, Figure 8). Four of those 15 metrics were significantly different in all twelve months of the study: % insects; % EPT index; % Ephemeroptera abundance; and HBI. Two of those 15

metrics, Ephemeroptera richness and combined EPT richness, were significantly different in eleven of the months (all but April and March, respectively). The diversity metrics H, I-D,  $N_I$  and  $N_2$  were significantly different in 8 months (but not February, March, April, or July). Density was significantly different in seven months, but not June, July, August, September, and October. Percent EPT abundance was significantly different in seven months, but not February, March, April, September, or October. Plecoptera richness was significantly different in May, June, August, September, October, and January, whereas Trichoptera richness was significantly different in March, May, June, and August. Total richness was significantly different in only three months: March, August, and September.

Regarding the intra-stream analysis at the created stream, the dynamics of the macroinvertebrate communities were similar between the up section and the down section throughout the study period. In fact, all 16 of the macroinvertebrate metrics showed a similar pattern of community dynamics between the two sections. However, three of the 16 metrics showed statistically significant differences between the two sections: % Ephemeroptera abundance in March; total richness in April; and EPT richness in March and May. Nonetheless, 12 of the 16 metrics showed that community dynamics at the up section were statistically more similar to the unmined stream than the down section was to it. Two of those 12 metrics were significant in only one month: % EPT index in March, and Plecoptera richness in May. Three of those 12 metrics were significant in two months: % Ephemeroptera abundance in February and March, Ephemeroptera richness in March and May, and Trichoptera richness in May and June. Six of those 12 metrics were significant in three months: total richness in April, August, and September; EPT richness in February, April, and November; and *H*, *1-D*, *N*<sub>1</sub> and *N*<sub>2</sub>,

in June, November, and January. Lastly, one of those 12 metrics was significant in four months: HBI in February, March, April, and October. In contrast, seven of the 16 metrics also showed that community dynamics at the down section were statistically more similar to the unmined stream than the up section was to it. However, six of those seven metrics were significant in only one month: total richness and Trichoptera richness in March; and H, I-D,  $N_I$  and  $N_2$  in October. One of those seven metrics was significant in three months: percent insects in October, November, and January.

### **Benthic macroinvertebrate community metrics (seasonal summary)**

April (spring), July (summer), October (autumn), January (winter)

#### *Community structure*

*Density*—I found statistically significant differences in macroinvertebrate density (no. individuals  $\cdot$  m<sup>-2</sup>) among stream types (Tukey's HSD; *P* < 0.05; Figure 5). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, density was higher at the open-canopied stream created on top of the valley fill and virtually nonexistent at the mine-impacted stream downgradient of the valley fill. In April (spring), density was significantly higher at the created stream (GU, GD, average = 12,555.6 and 6,451.1, respectively) than at the unmined stream (LM, average = 1,108.9) and the mine-impacted stream (WB, average = 24.4). The density at the unmined stream was significantly higher than at the mine-impacted stream, whereas the created stream's sections did not differ significantly despite the marked contrast there. In July (summer), density was higher at the created stream's up section (GU, average = 2,577.8) and at the unmined stream (LM, average = 1,586.7) and lower at the created stream's

down section (GD, average = 164.4) and at the mine-impacted stream (WB, average = 22.2). Density did not differ significantly between the created stream and the unmined stream, or between sections at the created stream. However, the unmined stream and the created stream's up section differed significantly from the mine-impacted stream, whereas the down section did not. In October (autumn), density was higher at the created stream's up section (GU, average = 3,680.0) and the unmined stream (LM, average =1,988.9), and lower at the created stream's down section (GD, average = 1,273.3) and the mine-impacted stream (WB, average = 15.6). Density did not differ significantly between the created stream and the unmined stream, or between sections at the created stream. However, the unmined stream and both sections of the created stream differed significantly with the mine-impacted stream. In January (winter), density was significantly higher at the created stream (GU, average = 8,428.9; GD, average = 14,597.8) than at the unmined stream (LM, average = 1,962.2) and the mine-impacted stream (WB, average = 15.6). The density at the unmined stream was significantly higher than at the mine-impacted stream, whereas the created stream's sections did not differ significantly with one another despite the marked contrast there. Results for the full study period are presented in Appendix B1.

*Total richness (genera)*—I found statistically significant differences in total richness among stream types (Tukey's HSD; P < 0.05; Figure 6). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, richness was lowest at the mine-impacted site downstream of the valley fill, and higher at the up section of the created stream than at the down section. In April (spring), richness was higher at the created stream's up section (GU, average = 33.4) than at the



Figure 5. Average density (#·m<sup>-2</sup>) of benthic macroinvertebrates.
(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA).
(B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 1421.78 from the average.</li>

unmined stream (LM, average = 23.8), and lower at the created stream's down section (GD, average = 21.2) and the mine-impacted stream (WB, average = 0.8). The unmined stream and both of the created stream's sections differed significantly with the mineimpacted stream, however, neither section of the created stream differed significantly with the unmined stream. At the created stream, richness at the up section was significantly higher than at the down section. In July (summer), richness was higher at the unmined stream (LM, average = 15.0) and the created stream's up section (GU, average = 13.0), and lower at the created stream's down section (GD, average = 4.6) and the mineimpacted stream (WB, average = 1.8). However, none of the stream sites differed significantly with one another. In October (autumn), richness was higher at the unmined stream (LM, average = 26.6) than at the created stream (GU, GD, average = 20.4, and 16.2, respectively), and the mine-impacted stream (WB, average = 1.8). Richness did not differ significantly between the unmined stream and either section of the created stream, or between sections at the created stream. However, the up section and the unmined stream differed significantly with the mine-impacted stream, whereas the down section did not. In January (winter), richness was higher at the unmined stream (LM, average = 26.2) than at the created stream (GU, GD, average = 24.0, and 16.2, respectively) and the mine-impacted stream (WB, average = 1.2). Richness did not differ significantly between the unmined stream and either section at the created stream, or between sections at the created stream. However, the unmined stream and the created stream's up section differed significantly with the mine-impacted stream, whereas the down section did not. Results for the full study period are presented in Appendix B1.



Figure 6. Average total richness of benthic macroinvertebrates. (A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 1.95 from the average.

# Community composition

Percent benthic insects—I found statistically significant differences in the percentages of benthic insects among stream types (Tukey's HSD; P < 0.05; Figure 7). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, the percentage of insects was higher at the unmined stream than at the open-canopied stream created on top of the valley fill. The percentage of insects at the mine-impacted stream was high because there were virtually no invertebrates collected at the site. In April (spring), percentage benthic insects was significantly higher at the mineimpacted (WB, average = 100%) and unmined streams (LM, average = 99.8%) than at the created stream (GU, GD, average = 85.3%, and 76.9%, respectively). However, the difference between the up and down sections at the created stream was not significant. In July (summer), percentage benthic insects was significantly higher at the mine-impacted (WB, average = 100%) and unmined streams (LM, average = 98.3%) than at the created stream (GU, GD, average = 29.5%, and 64.0%, respectively). %). However, the difference between the up and down sections at the created stream was not significant, despite the marked contrast. In October (autumn), percentage benthic insects was significantly higher at the mine-impacted (WB, average = 100%) and unmined streams (LM, average = 98.4%) than at the created stream (GU, GD, average = 50.0%, and 88.9%, respectively). However, only the up section at the created stream was significantly different from the unmined and mine-impacted streams. Percent benthic insects did not differ significantly between the up and down sections at the created stream, despite the marked contrast. In January (winter), percentage benthic insects was significantly higher at the mine-impacted (WB, average = 100%) and unmined streams



## Figure 7. Average percentage of benthic insects.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 7.10 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from division by zero. (LM, average = 99.3%) than at the created stream (GU, GD, average = 72.9%, and 87.5%, respectively). However, only the up section at the created stream was significantly different from the unmined and mine-impacted streams. Percent benthic insects did not differ significantly between the up and down sections at the created stream. Results for the full study period are presented in Appendix B2).

Percent top two dominant taxa (abundance)—I did not find statistically significant differences in the percentage of top two dominant taxa among stream types (Tukey's HSD; P < 0.05; Figure 8)—although stark contrasts in taxa composition were observed. In general, the percentage of the two most abundant taxa was higher at the created stream than at the unmined stream. I did not calculate values for the mineimpacted stream (WB) during any month because there was insufficient invertebrate richness and abundance. In April (spring), percent top two taxa was higher at the created stream's down section (GD, average = 44.4%) than at the up section (GU, average = 37.3%) and the unmined stream (LM, average = 36.9%). In July (summer), percent top two taxa was higher at the unmined stream (LM, average = 63.6%) than at the created stream (GU, average = 56.1%; GD, 46.6%). In October (autumn), percent top two taxa was higher at the created stream (GU, average = 49.9%; GD, average = 34.4%) than at the unmined stream (LM. average = 26.2%). In January (winter), percent top two taxa was higher at the created stream's down section (GD, average = 65.2%) than at the up section (GU, average = 39.2%) and the unmined stream (LM, average = 25.7%). Results for the full study period are presented in Appendix B2. WB was excluded because of insufficient richness and abundance.





(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 5.88 from the average.

**EPT richness (genera)**—I found statistically significant differences in EPT richness among stream types (Tukey's HSD; P < 0.05; Figure 9). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, mayfly, stonefly, and caddisfly richness was higher at the unmined stream than at the created stream, and non-existent at the mine-impacted stream downgradient of the valley fill. In April (spring), EPT richness was significantly higher at the unmined stream (LM, average = 12.6) than at the created stream (GU, average = 10.0; GD, average = 5.8) and the mine-impacted stream (WB, average = 0.0). However, only the created stream's down section differed significantly with the unmined stream. Within the created stream, the difference between the up and down sections was not significant despite the marked contrast. In July (summer), EPT richness was significantly higher at the unmined stream (LM, average = 5.0) than at the created stream (GU, average = 0.2; GD, average = 0.2) and the mine-impacted stream (WB, average = 0.0). EPT richness did not differ significantly between the created stream and the mine-impacted stream. In October (autumn), EPT richness was significantly higher at the unmined stream (LM, average = 10.2) than at the created stream (GU, average = 3.6; GD, average = 1.8) and the mineimpacted stream (WB, average = 0.0). However, differences between the up and down sections at the created stream were not significant. In January (winter), EPT richness was significantly higher at the unmined stream (LM, average = 13.2) than at the created stream (GU, average = 6.0; GD, average = 4.6), and the mine-impacted stream (WB, average = 0.0). However, differences between the up and down sections at the created stream were not significant. Results for the full study period are presented in Appendix B3.



# Figure 9. Average genus-level EPT Richness.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.79 from the average.

Ephemeroptera (E) richness (genera)—I found statistically significant differences in Ephemeroptera richness among stream types (Tukey's HSD; P < 0.05; Figure 10). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, mayfly richness was higher at the unmined stream than at the created stream, and non-existent at the mine-impacted site downstream of the valley fill. In April (spring), Ephemeroptera richness was significantly higher at the unmined stream (LM, average = 4.6) and the created stream (GU and GD, average = 1.4and 1.4, respectively) than at the mine-impacted stream (WB, average = 0.0). However, differences were not significant between the unmined and created streams, or between up and down sections within the created stream. In July (summer), Ephemeroptera richness was significantly higher at the unmined stream (LM, average = 2.8) than at the created and mine-impacted streams (GU, GD, and WB, average = 0.0, 0.0, and 0.0, respectively). In October (autumn), Ephemeroptera richness was significantly higher at the unmined stream (LM, average = 3.0) than at the created stream (GU, GD, average = 0.2 and 0.0, respectively) and the mine-impacted stream (WB, average = 0.0). However, differences were not significant between the created and mine-impacted streams, or between sections within the created stream. In January (winter), Ephemeroptera richness was significantly higher at the unmined stream (LM, average = 5.4) than at the created stream (GU, GD, average = 1.6 and 1.6, respectively) and the mine-impacted stream (WB, average = 0.0). Results for the full study period are presented in Appendix B3).

*Plecoptera (P) richness (genera)*—I found statistically significant differences in Plecoptera richness among stream types (Tukey's HSD; P < 0.05; Figure 11). Average monthly values varied between stream types and exhibited distinct patterns of



Figure 10. Average genus-level Ephemeroptera Richness.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.34 from the average. response. In general, stonefly richness was higher at the unmined stream than at the created stream, and nonexistent at the mine-impacted stream downgradient of the valley fill. In April (spring), Plecoptera richness was significantly higher at the unmined stream (LM, average = 6.0) and the created stream (GU, GD, average = 4.6 and 2.4, respectively) than at the mine-impacted stream (WB, average = 0.0). However, despite the marked contrasts, differences were not significant between the unmined stream and the created stream, or between sections at the created stream. In July (summer), Plecoptera richness was not significantly higher at the unmined stream (LM, average = (0.6) than at the created and mine-impacted streams (GU, GD, and WB, average = 0.0, 0.0, and 0.0, respectively). In October (autumn), however, Plecoptera richness was significantly higher at the unmined stream (LM, average = 4.0) than at the created and mine-impacted streams (GU, GD, and WB, average = 0.8, 0.2, and 0.0, respectively). Differences were not significant between the created and mine-impacted streams, or between sections at the created stream. In January (winter), Plecoptera richness was significantly higher at the unmined stream (LM, average = 4.8) than at the created and mine-impacted streams (GU, GD, and WB, average = 1.2, 1.4, and 0.0, respectively). Differences between sections at the created stream were not significant. Results for the full study period are presented in Appendix B4.

*Trichoptera (T) richness (genera)*—I found statistically significant differences in Trichoptera richness among stream types (Tukey's HSD; P < 0.05; Figure 12). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, caddisfly richness was higher at the unmined stream than at the created stream, particularly the down section, and nonexistent at the mine-impacted



## Figure 11. Average genus-level Plecoptera Richness.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.44 from the average.
stream downgradient of the valley fill. In April (spring), Trichoptera richness was significantly higher at the unmined (LM, average = 2.0) and created streams (GU, GD, average = 4.0 and 2.0, respectively) than at the mine-impacted stream (WB, average = 0.0). However, despite the marked contrasts, differences were not significant between the unmined and created streams, or between sections at the created stream. In July (summer), Trichoptera richness was significantly higher at the unmined stream (LM, average = 1.6) than at the mine-impacted stream (WB, average = 0.0). However, richness at the created stream (GU, GD, average = 0.2, and 0.2, respectively) did not differ significantly between up and down sections, or with either reference stream. In October (autumn), Trichoptera richness was significantly higher at the unmined (LM, average = 3.2) and created streams (GU, GD, average = 2.6, and 1.6, respectively) than at the mineimpacted stream (WB, average = 0.0). However, differences were not significant between the created stream's up and down sections, or between either section and the unmined stream. In January (winter), Trichoptera richness was significantly higher at the unmined (LM, average = 3.0) and created streams (GU, GD, average = 3.2 and 1.6, respectively) than at the mine-impacted stream (WB, average = 0.0). However, differences were not significant between the created stream's up and down sections, or between either section and the unmined stream. Results for the full study period are presented in Appendix B4.

### *Community diversity*

Shannon index (H) & Hill's  $N_1$  diversity—I found statistically significant differences for Shannon index (Figure 13) and  $N_1$  diversity (Figure 14) values among stream types (Tukey's HSD; P < 0.05). Average monthly values varied between



# Figure 12. Average genus-level Trichoptera Richness.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites that share letters in a month are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.42 from the average. stream types and exhibited distinct patterns of response. In general, diversity was higher at the unmined stream than at the created stream, and virtually nonexistent at the mineimpacted stream downgradient of the valley fill. In April (spring), Shannon and  $N_l$ diversity were significantly higher at the unmined (LM, average = 2.60 and 13.80, respectively) and created streams (GU, average = 2.40 and 11.09, respectively; GD, average = 2.03 and 8.16, respectively) than at the mine-impacted stream (WB, average = 0.08 and 0.69, respectively). However, differences were not significant between the created stream's up and down sections, or between either section and the unmined stream. In July (summer), Shannon and  $N_1$  diversity were not significantly different between the unmined stream (LM, average = 1.57 and 5.49, respectively) and the created stream (GU, average = 1.50 and 4.74, respectively; GD, average = 1.19 and 3.63, respectively) or with the mine-impacted stream (WB, average = 0.53 and 1.66, respectively). Differences were also not significant between the created stream's up and down sections. In October (autumn), Shannon and  $N_1$  diversity were significantly higher at the unmined stream (LM, average = 2.75 and 15.77, respectively) and the created stream's down section (GD, average = 2.14 and 9.07, respectively), than at the created steam's up section (GU, average = 1.83 and 7.36, respectively) and the mine-impacted stream (WB, average = 0.57 and 1.75, respectively). However, differences were not significant between the created stream's up and down sections, or either section and the mine-impacted stream. In January (winter), Shannon and  $N_I$  diversity were significantly higher at the unmined stream (LM, average = 2.72 and 15.28, respectively) and the created stream's up section (GU, average = 2.15 and 8.68, respectively) than at the created stream's down section (GD, average = 1.45 and 4.4, respectively) and mine-



# Figure 13. Average Shannon Index (H).

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.17 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from zero abundances.



# Figure 14. Average Hill's N<sub>1</sub> Diversity.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 1.11 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from zero abundances. impacted stream (WB, average = 0.27 and 0.9, respectively). However, differences were not significant between the created stream's up and down sections, even though the up section differed significantly with the mine-impacted stream whereas the down section did not. Results for the full study period are presented in Appendix B5.

Simpson index of diversity (1 – D) & Hill's N<sub>2</sub> diversity—I found statistically significant differences for Simpson index of diversity (Figure 15) and Hill's  $N_2$  diversity (Figure 16) values among stream types (Tukey's HSD; P < 0.05). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, diversity was higher at the unmined stream than at the created stream, and virtually nonexistent at the mine-impacted stream downgradient of the valley fill. In April (spring), Simpson index and  $N_2$  diversity were significantly higher at the unmined stream (LM, average = 0.88 and 8.9, respectively) and the created stream (GU, average = 0.86and 7.30, respectively; GD, average = 0.79 and 5.5, respectively) than at the mineimpacted stream (WB, 0.04 and 0.66, respectively). However, differences were not significant between the created stream's up and down sections, or either section and the unmined stream. In July (summer), diversity was not significantly different between any of the stream sites. Simpson index and  $N_2$  diversity at the unmined stream (LM, average = 0.60 and 3.48) was higher than at the mine-impacted stream (WB, average = 0.33 and 1.55, respectively) but similar to the created stream (GU, average = 0.65 and 3.38, respectively; GD, average = 0.60 and 3.22, respectively). In October (autumn), Simpson index and  $N_2$  diversity were significantly higher at the unmined stream (LM, average = 0.90 and 10.81, respectively) than at the created stream (GU, average = 0.70 and 5.00, respectively; GD, average = 0.81 and 6.38, respectively) and the mine-impacted stream



Figure 15. Average Simpson's Index (1 - D).

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.07 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from division by zero.





(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites within each month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.89 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN - not a number) from zero abundances. (WB, average = 0.34 and 1.72, respectively). However, only the created stream's up section differed significantly with the unmined stream, and neither section differed significantly with the mine-impacted stream. In January (winter), Simpson index and  $N_2$  diversity were significantly higher at the unmined stream (LM, average = 0.91 and 11.14, respectively) than at the created stream (GU, average = 0.83 and 5.87, respectively; GD, average = 0.64 and 2.97, respectively) and the mine-impacted stream (WB, average = 0.19 and 0.96, respectively). However, only the created stream's down section differed significantly with the unmined stream, and neither section differed significantly with the unmined stream, and neither section differed significantly with the mine-impacted stream. Results for the full study period are presented in Appendix B6.

### *Community biotic integrity*

*Percent EPT index*—I found statistically significant differences for percent EPT index among stream types (Tukey's HSD; P < 0.05; Figure 17). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, the percentage of mayfly, stonefly, and caddisfly genera was higher at the unmined stream than at the created steam, and zero at the mine-impacted stream downgradient of the valley fill. In April (spring), percent EPT index was significantly higher at the unmined stream (LM, average = 53.2%) than at the created stream (GU, GD, average = 30.0%, and 28.2%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between the created stream's up and down sections, however both sections differed significantly with the mine-impacted stream. In July (summer), percent EPT index was significantly higher at the unmined stream (LM, average = 33.1%) than at the created stream (GU, GD, average = 1.1%, and 3.3%, respectively) and the mine-

impacted stream (WB, average = 0.0%). Difference was not significant between the created stream's up and down sections, or between either section and the mine-impacted stream. In October (autumn), percent EPT index was significantly higher at the unmined stream (LM, average = 37.9%) than at the created stream (GU, GD, average = 17.4%, and 11.2%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between the created stream's up and down sections, however only the up section differed significantly with the mine-impacted stream. In January (winter), percent EPT index was significantly higher at the unmined stream (LM, average = 51.1%) than at the created stream (GU, GD, average = 25.1%, and 28.4, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between the created stream (GU, GD, average = 25.1%, and 28.4, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between the created stream (GU, GD, average = 25.1%, and 28.4, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significantly with the mine-impacted stream is up and down sections, however both sections differed significantly with the mine-impacted stream is up and down sections. Appendix B7.

*Percent EPT abundance*—I found statistically significant differences for percent EPT abundance among stream types (Tukey's HSD; P < 0.05; Figure 18). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, the percent abundance of mayflies, stoneflies, and caddisflies was much higher at the unmined reference stream than at the created stream, and zero at the mine-impacted stream downgradient of the valley fill. In April (spring), percent EPT abundance was significantly higher at the unmined stream (LM, average = 67.0%) and the created stream (GU, GD, average = 29.0%, and 34.4%, respectively) than at the mine-impacted stream (WB, average = 0.0%). However, despite marked contrasts, neither section at the created stream differed significantly with the unmined stream, nor with one





(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 2.59 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from division by zero.

another. In July (summer), percent EPT abundance was significantly higher at the unmined stream (LM, average = 15.0%) than at the created stream (GU, GD, average = 0.1%, and 1.7%, respectively) and the mine-impacted stream (WB, average = 0.0%). Neither section at the created stream differed significantly with the mine-impacted stream, nor with one another. In October (autumn), percent EPT abundance was significantly higher at the unmined stream (LM, average = 33.0%) and the created stream (GU, GD, average = 17.9%, and 9.9%, respectively) than at the mine-impacted stream (WB, average = 0.0%). Neither section at the created stream differed significantly with the unmined stream, nor with one another. In January (winter), percent EPT abundance was significantly higher at the unmined stream (LM, average = 56.7%) than at the created stream (GU, GD, average = 10.0%, and 7.0%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between sections at the created stream (WB, average = 0.0%). Difference was not significantly with the unmined and mine-impacted stream, however both sections differed significantly with the unmined and mine-impacted stream. Results for the full study period are presented in Appendix B7.

*Percent Ephemeroptera (E) abundance*—I found statistically significant differences for percent Ephemeroptera abundance among stream types (Tukey's HSD; P< 0.05; Figure 19). Average monthly values varied between stream types and exhibited distinct patterns of response. In general, the percent abundance of mayflies was higher at the unmined stream than at the created stream, and zero at the mine-impacted stream downgradient of the valley fill. In April (spring), percent Ephemeroptera abundance was significantly higher at the unmined stream (LM, average = 42.4%) than at the created stream (GU, GD, average = 8.2%, and 8.1%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between sections at the created



## Figure 18. Average % EPT Abundance.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 4.86 from average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from division by zero.

stream, however, both sections differed significantly with the unmined and mineimpacted streams. In July (summer), percent Ephemeroptera abundance was significantly higher at the unmined stream (LM, average = 11.2%) than at the created stream (GU, GD, average = 0.0%, and 0.0%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between sections at the created stream, or between either section and the mine-impacted stream. In October (autumn), percent Ephemeroptera abundance was significantly higher at the unmined stream (LM average = 15.6%) than at the created stream (GU, GD, average = < 0.1%, and 0.0%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between sections at the created stream, or between either section and the mine-impacted stream. In January (winter), percent Ephemeroptera abundance was significantly higher at the unmined stream (LM, average = 37.9%) than at the created stream (GU, GD, average = 6.4%, and 4.7%, respectively) and the mine-impacted stream (WB, average = 0.0%). Difference was not significant between sections at the created stream, however both sections differed significantly with the mine-impacted stream. Results for the full study period are presented in Appendix B8.

*Hilsenhoff biotic index (HBI)*—I found statistically significant differences for Hilsenhoff biotic index (HBI) among stream types (Tukey's HSD; P < 0.05; Figure 20). I did not calculate HBI values for the mine-impacted stream during any month because there was insufficient taxon richness and abundance. Average monthly values varied between stream types and exhibited distinct patterns of response. In general, the HBI was lower (higher biotic integrity) at the unmined stream than at the created stream. In April (spring), HBI was significantly lower at the unmined stream (LM, average = 3.03) than at



Figure 19. Average % Ephemeroptera Abundance.

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 2.55 from the average in LM, GU, GD, but vary in WB due to samples that generated impossible numbers (NaN = not a number) from division by zero.

the created stream's down section (GD, average = 5.61). Despite the marked contrast, difference was not significant between the unmined stream and the created stream's up section (GU, average = 5.48), or between sections at the created stream. At the unmined stream, the HBI value represented a better rating of water quality and a lower degree of environmental stress (LM, 'excellent', and 'none apparent', respectively) than at the created stream (GU, 'good', and 'some', respectively; GD, 'fair', and 'fairly significant', respectively). In July (summer), HBI was significantly lower at the unmined stream (LM, average = 5.93) than at the created stream (GU and GD, average = 7.20 and 7.09, respectively). Difference was not significant between sections at the created stream. At the unmined stream, the HBI value represented a better rating of water quality and a lower degree of environmental stress (LM, 'fair', and 'fairly significant', respectively) than at the created stream (both GU and GD, 'fairly poor', and 'significant', respectively). In October (autumn), HBI was significantly lower at the unmined stream (LM, average = 4.47) than at the created stream (GU, GD, average = 6.09 and 7.04, respectively). Difference was not significant between sections at the created stream, or between the up section and the unmined stream despite the marked contrast. At the unmined stream, the HBI value represented a better rating of water quality and a lower degree of environmental stress (LM, 'very good', and 'possible slight', respectively) than at the created stream (GU, 'fair', and 'fairly significant', respectively; GD, 'fairly poor', and 'significant', respectively). In January (winter), HBI was significantly lower at the unmined stream (LM, average = 3.68) than at the created stream (GU, GD, average = 6.42 and 6.78, respectively). Difference was not significant between sections at the created stream. At the unmined stream, the HBI value represented a better rating of water



# Figure 20. Average Hilsenhoff Biotic Index (HBI).

(A) Monthly differences across study sites at Robinson Forest, Kentucky (USA). (B) Sites in a month that share letters are not significantly different from one another (Tukey's HSD; p < 0.05). Error bars represent global SE of 0.24 from the average. quality and a lower degree of environmental stress (LM, 'very good', and 'possible slight', respectively) than at the created stream (GU, 'fair', and 'fairly significant', respectively; GD, 'fairly poor', and 'significant', respectively). Results for the full study period are presented in Appendix B8.

#### DISCUSSION

The permanent loss of headwater streams due to mountaintop mining and valley fills (MTM-VF) can have major environmental consequences for the mountain ecosystem, the nearby valleys, and the downstream water quality (US EPA 2011b). Despite Eastern Kentucky's 2016 coal production being at the lowest level since 1932 and having declined by more than 80% since peak production in 1990 (KY OEP 2017), there remains more than 2,000 km of buried headwater streams (US EPA 2011b) and an estimated 1.5 million acres of surface-mined land available for restoration in the Central Appalachian Coalfields (Barton et al. 2018). There are five primary functions that underlie the operation of a watershed (Black 1997), and the performance of the retrofitted watershed with regard to the three hydrological functions (collection, storage, and discharge) are covered extensively by Blackburn-Lynch (2015) and Agouridis et al. (2017). The other two primary functions of a watershed are ecological and serve to: 1) provide aqueous sites for chemical reactions (e.g., biogeochemical and nutrient cycling processes) to take place, and 2) define the characteristics of freshwater aquatic habitat. I am not aware of any comprehensive analyses of either of the fundamental ecological functions at the retrofitted watershed, or of the flora and fauna that constitute the biological elements of the aquatic habitats in the created channel and vernal pools. Although there is a large body of literature on stream restoration ecology in urban and agricultural streams, there is a lack of evidence on the biota and ecosystem functioning

associated with constructed channels on valley fills and the ability of created streams to mitigate the effects of stream burial (US EPA 2011b).

This study reveals important trends relating the legacy effects of mountaintop mining and valley fill creation on stream macroinvertebrate communities in a created stream. Additionally, the results of this study represent a comprehensive first step in gaining an understanding of how the creation of a headwater stream system on a valley fill utilizing NCD techniques and the FRA affects benthic macroinvertebrate colonization and community dynamics. In this study, I attempted to link multiple in-stream and watershed-scale variables originating from mining legacies to responses of macroinvertebrate communities in an effort to assess the degree to which the created stream system replaced lost headwater stream ecological function, improved water quality, and enhanced aquatic habitat. Overall, the dynamics of the macroinvertebrate communities at the created stream were more similar to the community-level dynamics at an unmined stream than to dynamics at a mine-impacted stream immediately downgradient of a traditionally constructed valley fill.

Initially, I proposed three general hypotheses for how the macroinvertebrate community dynamics might respond at the created stream on the retrofitted valley fill. First, I thought that the community dynamics at the created stream would be more similar to the dynamics at an unmined stream than at a mine-impacted stream immediately downgradient of a traditional valley fill. Second, I thought that the community dynamics at the created stream would differ between the uppermost 100 m section and a 100 m section immediately downgradient. Lastly, I thought that the community dynamics at the up section of the created stream would be more similar to the dynamics at the unmined

stream than the down section was to it. My results support the hypothesis that macroinvertebrate community dynamics at the created stream are more similar to the community dynamics at an unmined stream than at a mine-impacted stream immediately downgradient of a traditionally constructed valley fill. This observation is contradictory with many bioassessments of constructed channels at mining sites (USEPA 2011b), however the constructed channel in my study is unique in that it is one element of a retrofitted valley fill upon which an entire stream system was created using NCD techniques in conjunction with the FRA. Furthermore, my results take into consideration only the first 200 m of the more than 1400 m of created stream system at the retrofitted valley fill. My results do not support the hypothesis that the community dynamics at the created stream differ between the uppermost 100 m section and a 100 m section immediately downgradient. However, a limitation of the retrofitted valley fill is the significant reduction in baseflow due to the loss of connectivity to the groundwater table (USEPA 2011b; Blackburn-Lynch 2015; Agouridis et al. 2017). Visual observations by Blackburn-Lynch (2015) indicate that instream flow volume reductions began after approximately 450 m of created stream channel. Therefore, it is possible that macroinvertebrate community dynamics changed significantly as both length of created channel and distance from seep increased, but that the effect was not detected due to the constraints of my sampling sections. Lastly, there was at least some evidence that supported the hypothesis that community dynamics at the up section of the created stream are somewhat more similar to the unmined stream than dynamics at the down section are to it. This further suggests that there may in fact be taxonomic differences between the up section and the down section, but in the case of this hypothesis the effects may not have

been detected because of my choice of macroinvertebrate metrics, or the genus-level identification of invertebrates (instead of the higher taxonomic resolution of species).

### **Environmental variables**

There are five environmental variables associated with mining and valley fills that are commonly considered to potentially affect the ecological condition (macroinvertebrate metrics in this study) of downstream habitats: (1) ion concentration, (2) heavy metal concentration, (3) organic enrichment, (4) changes to instream habitat, and (5) changes to upstream land use/land cover (USEPA 2011b). In addition to calculating values for sixteen metrics in order to analyze the benthic macroinvertebrate communities, I also calculated values for sixteen environmental variables in order to characterize instream and watershed attributes. Although I did not subject environmental variables to statistical analyses, annual summarized values (Table 2) are discussed here to show apparent trends that could help with formulating generalizations from this study's macroinvertebrate metrics as well as to inform new or revised hypotheses.

Differences in riparian forest (edge distance) and canopy (percent coverage) are expected to influence the in-stream habitat features listed in Table 2 (e.g., temperature, periphyton, organic matter, etc.) in predictable directions (higher/lower) that in turn influence differences in macroinvertebrate assemblage structure and taxa distribution (Sponseller and Benfield 2001; Rios and Bailey 2006; Adkins et al. 2016). However, an intact riparian zone and canopy coverage may not be sufficient to protect a stream from the effects of upland disturbances such as mountaintop removal and valley fill creation (Houser et al. 2005; USEPA 2011b). Furthermore, valley fills have been implicated in higher downstream water temperatures during autumn, winter, and spring and lower

annual variation (Wiley et al. 2001; USEPA 2011b), which appears to be the case at the mine-impacted site. At the created stream, the higher temperature range of the down section in comparison to the up section correlates with the increasing distance between forest edges and the concomitant decrease in shade cast on the channel. In-stream vegetation, nitrite + nitrate, benthic organic matter, periphyton and chlorophyll a were all predictably higher at both sections of the created stream where there was zero percent canopy coverage. Woody debris and leaf matter were higher at the unmined and mineimpacted streams where the riparian forest was intact, and the percentage of canopy coverage was high. Measurements of streamflow (discharge and current velocity), turbidity (relative clarity) and stream bottom embeddedness (extent of rock burial) were not taken due to equipment and time limitations, however personal visual observations were made. At the unmined and mine-impacted streams, streamflow was evident, and turbidity was low with high relative clarity. In contrast, streamflow at the created stream was not evident (either slow-moving or stagnant), and the turbidity was high with low relative clarity. At the unmined and mine-impacted streams, rocks (gravel, cobble, and boulders) were readily observable, however the embeddedness was low at the unmined stream with abundant interstitial space, but high at the mine-impacted stream with no apparent interstitial space due to the rocks and sediment particles having been cemented together by iron precipitate (Fritz et al. 2010; USEPA 2011b). At the created stream, embeddedness was also high, however no rocks were observable or even detected during benthic sampling due to burial by organic matter. I did not observe any differences between the up section and the down section with regard to streamflow, turbidity, and embeddedness. Conductivity, sulfate, and chloride values in my study are similar to

values reported for the same streams by Agouridis et al. (2017) between 2010 and 2013. Compared to the unmined stream, sulfate and conductivity at the up section of the created stream were 8 and 7 times higher, respectively; 9 and 8 times higher at the down section of the created stream; and 195 and 42 times higher at the mine-impacted stream.

The water chemistry results are notable because (1) the samples were collected from the Central Appalachian Coalfield within Ecoregion 69, and (2) the mixture of dissolved ions from salts indicate dominant levels of sulfate (SO4<sup>2-</sup>) and low levels of chloride (Cl<sup>-</sup>) at circum-neutral to mildly alkaline pH (6.0–10.0; Table 2), both of which suggest the applicability of the U.S. EPA's (2011a) aquatic life benchmark for conductivity to my study. The aquatic life benchmark for conductivity is expected to avoid the local extirpation of 95% of the invertebrate genera from this region (USEPA 2011a). All-year data from West Virginia derived a chronic benchmark value (applicable for year-round use) of 300  $\mu$ S/cm, and an independent data set from Kentucky derived a chronic benchmark value of 282 µS/cm. In my study, I consider elevated levels of conductivity to be legacy effects (and not natural background levels; Barton 2011) because the prominent sources of salts in Ecoregion 69 are mine overburden and valley fills from large-scale surface mining (USEPA 2011a). Relative concentrations of dissolved ions dominated by salts of SO<sub>4</sub><sup>2-</sup> (among others) are believed to present some species with an insurmountable physiological challenge (USEPA 2011a). Pond and colleagues (Pond et al. 2008; 2010) found only weak relationships between mayfly metrics and habitat parameters downstream of mined Central Appalachian headwater streams in West Virginia, suggesting that mining related degradation of water quality (specific conductivity) limits aquatic life regardless of habitat quality. In my study, the

elevated conductivity levels appear to be the primary explanatory variable for the observed differences in values for benthic macroinvertebrate metrics.

In light of this evidence, mayfly, stonefly, and caddisfly (EPT) taxa collected in benthic samples at the mine-impacted site were excluded from the EPT-specific metric calculations and a numerical value of 0 was used for statistical analyses for the stream type 'WB'. The metrics affected by this decision are: EPT richness; Ephemeroptera richness; Plecoptera richness; Trichoptera richness; % EPT index; % EPT abundance; and % Ephemeroptera abundance. In all other metrics, the total aggregate account of all invertebrates collected at the mine-impacted site was used. Additional rationale for this decision took into consideration the genera collected from mine-impacted stream. For example, stonefly taxa included two genera with assigned tolerance values (TV) of 0.0 (KDOW 2002): Yugus, which was not recorded in the sampling sections of the other stream types, and Soyedina. The mayfly taxa included Ephemerella (TV 1.7). Furthermore, the monthly sampling regime revealed observable discontinuity in the larval growth stages of all taxa collected from the mine-impacted stream. Because body length was measured for all 11,733 EPT's collected during all sampling efforts combined, taxon-specific length-frequency histograms were generated that estimated the presence or absence of larval growth stage (instars) distributions in each stream's sampling section. Comparisons of site and monthly trends in taxon-specific growth patterns suggested that presence of individuals in the mine-impacted stream was transient and not a result of development through successive larval growth stages. And lastly, specific conductivity measurements (Table 2) appeared to support the transient presence of EPT individuals in the mine-impacted stream. A probable source for the macroinvertebrates collected in the

mine-impacted stream was adjacent ephemeral or intermittent hillslope channels, from which individuals either drifted or were flushed during rainfall events.

## *Macroinvertebrate metrics—Summary*

*Community Structure*—In my study, I consider invertebrate density and total richness to be representative of the overall community structure with regard to benthic macroinvertebrate populations at each stream. First, I predicted that density would be higher and total richness lower at the created stream than at the unmined stream, and higher in both cases than at the mine-impacted stream. Second, I predicted that density and total richness would be higher at the up section of the created stream than at the down section of created stream.

My results support my first prediction and suggest that the community structure at the created stream is more similar to the structure at the unmined stream than at the mineimpacted stream immediately downgradient of a traditionally constructed valley fill. Nonetheless, the created stream exhibited statistically significant differences with the unmined stream. As predicted, density was significantly higher at the created stream in late autumn, winter, and spring. However, total richness was not as I predicted, with no statistical difference between the created steam and the unmined stream in autumn, winter and summer. In fact, total richness was significantly lower in the unmined stream in spring. I attribute the higher density to the higher in-stream primary production resulting from the loss of forest canopy, and also to the relatively high degree of organic matter retention. Total richness was unexpectedly high at the created stream relative to the unmined stream in large part because of the genera richness of true flies, beetles, dragonflies, and damselflies.

However, my results do not support my second prediction that diversity and total richness would be higher at the up section of created stream than at the down section. Density was not significantly different between the two sections in any month, and total richness was significantly different in only one month (April) even though it was higher at the up section in all months.

Overall, the benthic macroinvertebrate community structure at the created stream is similar to the unmined stream with regard to total richness, but not invertebrate density. Furthermore, the community structure at the created stream is similar between the up section and the down section with regard to both density and total richness, although my results suggest that there may be important distinctions between the two sections that were not revealed by my analysis.

*Community Composition*—In my study, I consider percentage of benthic insects, percentage of the top two dominant taxa, and the individual and aggregate richness of mayfly, stonefly, and caddisfly genera to be broadly representative of the overall community composition with regard to benthic macroinvertebrate populations at each stream. First, I predicted that percent insects and the EPT richness metrics would be lower at the created stream than at the unmined stream, but higher than at the mine-impacted stream. Additionally, I predicted that the percent top two dominant taxa would be higher at the created stream than at the unmined stream. Second, I predicted that the percent insects and the EPT richness metrics of the overall created stream than at the unmined stream. Second, I predicted that the percent insects and the EPT richness metrics that the up section of the created stream than at the down section. I also predicted that the percent top two dominant taxa would be lower at the up section than at the down section.

My results support my first prediction and suggest that community composition at the created stream is more similar to the composition at the unmined stream than at the mine-impacted stream. However, the created stream exhibited statistically significant differences with the unmined stream in each composition metric except for percent top two dominant. My results show that the percentage of insects was significantly lower at the created stream than at the unmined stream. This result was due to the abundance of aquatic snails and pea clams inhabiting the created stream. Also, all of the EPT richness metrics were significantly lower at the created stream than at the unmined stream. However, both sections of the created stream exhibited high abundance of the mayfly genus Ameletus which is recognized as a constituent of the basic core taxa expected in healthy Appalachian headwaters of Kentucky (Pond 2010). It would be informative to determine 1) if the species of *Ameletus* at the created stream are the same as those found by Pond (2010), and 2) if the species of Ameletus at the created stream are more or less tolerant to environmental degradation than those found by Pond (2010). The up section exhibited four of the five stonefly genera and four of the five caddisfly genera best represented in least-disturbed eastern Kentucky streams, whereas the down section exhibited three of the stonefly genera and three of the caddisfly genera (Pond 2012). However, at the up section, one of the stonefly genera (Acroneuria) and three of the caddisfly genera (Neophylax, Rhyacophila, Wormaldia) were represented by four or fewer individuals for the entire study period. At the down section, all three of the caddisfly genera were represented by two or fewer individuals for the entire study period. These observations suggest that the presence of EPT taxa at the intermittent created stream may be a function of distance from the unmined upper watershed seep (Fritz and

Dodds 2004), but also that the created stream environment may not be suitable for colonization (e.g., due to conductivity). In contrast to the created stream, the unmined stream exhibited four of the five mayfly genera best represented in least-disturbed eastern Kentucky streams, along with all five of the best represented genera for both stonefly and caddisfly taxa. Lastly, my results do not support my prediction that percent top two dominant taxa (abundance) at the created stream would be higher than at the unmined stream.

My second prediction was also not supported by my results, suggesting that community composition does not differ significantly between the up section and the down section. None of the composition metrics were significantly different between the created stream sections in any month except for combined-EPT richness in March and May. However, underlying the metrics for percent insects and percent top two dominant taxa is the fact that a high abundance of bivalves were observed in all five subsections of the up section, but not in any of the five subsections at the down section of the created stream in any month during the study period.

Regarding the percent top two dominant metric, although my results did not indicate statistically significant differences between the created stream and the unmined stream, there was a contrast in genus composition of the top dominant taxa. At the unmined stream, insects represented the top two dominant taxa in all 12 months, and an EPT genus represented at least one of the top two taxa in nine of the months, and both of the top two taxa in four of the months. At both the up section and the down section of the created stream, insects represented the top two dominant taxa in 4 of the months, and an EPT genus represented at least one of the top two taxa in top the months, and both of

the top two taxa in none of the months. However, the EPT taxa differed between the up section and the down section, with the two instances at the up section represented by the mayfly *Ameletus* and the stonefly *Amphinemura*, and both instances at the down section represented by the caddisfly *Ironoquia*.

Overall, the benthic macroinvertebrate community composition at the created stream is significantly different than the composition at both the unmined stream and the mine-impacted stream. However, the created stream's community composition is more similar to the unmined stream's community composition than to the mine-impacted stream. At the created stream, the benthic macroinvertebrate community composition is similar between the up section and the down section.

*Community Diversity*—In my study, I consider Shannon's index (*H*), Simpson's index of diversity (1-D), and Hill's  $N_1$  and  $N_2$  to be broadly representative of the overall community diversity with regard to benthic macroinvertebrate populations at each stream. First, I predicted that community diversity at the created stream would be lower than at the unmined stream, but higher than at the mine-impacted stream. Second, I predicted that community diversity at the created stream would be higher at the up section than at the down section.

My results support my first prediction and suggest that the benthic community diversity at the created stream was more similar to the diversity at the unmined stream than at the mine-impacted stream. However, the created stream exhibited statistically significant differences with the unmined stream during late summer, autumn and the first half of winter but generally not during the spring. I attribute this pattern of diversity to two general causes, 1) low baseflow at the created stream during summer and autumn;

and 2) baseflow at the unmined stream that was sufficient to provide drought refuges during summer and autumn (Chester and Robson 2011).

However, my results do not support my second prediction that community diversity at the created stream would be higher at the up section than at the down section. None of the diversity metrics showed a significant difference in diversity between the sections in any month, although the effective number of species ( $N_1$  and  $N_2$ ) clearly trended higher at the up section during all but one month.

Overall, the benthic macroinvertebrate community diversity at the created stream is lower than at the unmined stream, but much higher than at the mine-impacted stream. At the created stream, the benthic macroinvertebrate community diversity is similar between the up section and the down section.

*Community Biotic Integrity*—In my study, I consider percent EPT index, percent EPT abundance, percent Ephemeroptera abundance, and HBI to be broadly representative of the overall community biotic integrity (stream health). First, I predicted that percent EPT index, percent EPT abundance, and percent Ephemeroptera abundance would be lower, and HBI higher at the created stream than at the unmined stream, but higher and lower, respectively, than at the mine-impacted stream. Second, I predicted that percent EPT index, percent EPT abundance, and percent Ephemeroptera abundance would be lower, respectively, than at the mine-impacted stream. Second, I predicted that percent EPT index, percent EPT abundance, and percent Ephemeroptera abundance would be

My results support my first prediction and suggest that the biotic integrity at the created stream was more similar to the biotic integrity at the unmined stream than at the mine-impacted stream. However, the created stream exhibited statistically significant differences with the unmined stream. At the created stream, percent EPT index and

percent Ephemeroptera abundance were significantly lower, and HBI significantly higher, than at the unmined stream in all months. Percent EPT abundance was significantly higher at the unmined stream except for February, March and April when the created stream exhibited high abundances of the Trichoptera genera *Ironoquia*, *Ptilotomis*, and *Chimarra*.

My results do not support my second prediction and suggest that the biotic integrity of the created stream does not differ between the up section and the down section. The only biotic integrity metric that was significantly different between the two sections was percent Ephemeroptera abundance, and in just one month (March), although percent EPT index clearly trended higher at the up section during all seasons except summer.

Overall, the benthic macroinvertebrate community biotic integrity at the created stream is lower than at the unmined stream, but much higher than at the mine-impacted stream. At the created stream, the benthic macroinvertebrate community biotic integrity is similar between the up section and the down section.

## CONCLUSION

Whether or not the retrofitted watershed is functioning both hydrologically and ecologically can be explained, at least partially, by the presence or absence of invertebrate fauna typically associated with clean, high-gradient streams in the region. Overall, my results show that despite markedly different in-channel physicochemical parameters, habitat substrate constituents, and watershed-scale site characteristics, the proof-of-concept headwater stream system created on a retrofitted valley fill improves water quality and supports a benthic macroinvertebrate community with structure, composition, diversity, and biotic integrity that more closely resembles a perennial unmined stream than a mine-impacted stream immediately down-gradient of a traditional valley fill. As a result, I conclude that the created stream system and the retrofitted valley fill (albeit in a relatively early successional stage) have at least partly restored the ecological functions of a headwater stream that were lost as a result of deforestation, mining, and valley fill creation. However, my results do not identify or quantify specific ecological functions, thereby necessitating the need for ongoing biological monitoring of the entire created stream channel, and for comprehensive research on other aspects of stream ecosystem function such as habitat; nutrient cycling; organic matter processing, retention and transport; and primary and secondary production (Pond 2000). Furthermore, it is important to note that the created stream is disconnected from the groundwater table and that my results are applicable to only the uppermost 200 m of the approximately1400

m stream system. Nonetheless, my results are suitable for generalizations that can be applied to future created streams where connectivity with an engineered groundwater table is maintained along the entire channel, for example via construction of a perched aquifer (USEPA 2011b; Blackburn-Lynch 2015). Furthermore, the results of my study

**Figure 21.** Watershed and valley fill (a) before NCD and FRA retrofit (2007), (b) nine years after retrofit (2017), and (c) eleven years after retrofit (2019). (Images are courtesy of University of Kentucky).



support the premise that the ability to successfully restore or recreate functional streams on mined lands is linked to the control of water quality (Pond 2010) through the restoration of the streams' watershed, including uplands and riparian areas, and not just the physical restoration of the stream channel itself (Agouridis et al. 2017). Finally, the results of my study are broadly applicable to the burgeoning restoration economy in the United States and specifically to the Appalachian region where addressing the environmental degradation associated with surface mining could lead to a new economic opportunity for Appalachia (Barton et al. 2018).

## REFERENCES

Agouridis CT, Barton CD, Warner RC. 2009. Recreating a headwater stream system on a head-of-hollow fill: a Kentucky case study. In: Vories KC and Caswell AH, editors. Proceedings of geomorphic reclamation and natural stream design at coal mines: A technical interactive forum. April 23–30, Bristol, VA; pp. 83–88. [accessed 2021 Jan 28]. http://citeseerx.ist.psu.edu/viewdoc/download?doi=10.1.1.732.5467&rep=rep1&t

ype=pdf.

- Agouridis C, Barton C, Warner R. 2017. Recreating a headwater stream system on a valley fill in the Appalachian coal field. In: Bolan N, Kirkham MB, and Ok YS, editors. Spoil to Soil: Mine Site Rehabilitation and Revegetation. CRC Press: Taylor and Francis, Boca Raton, FL; pp. 147–174.
- Allan JD, Castillo MM. 2007. Stream ecology: Structure and function of running waters, 2<sup>nd</sup> Ed. Chapman and Hall, New York.
- American Mine Services. 2021. Types of surface mines. Boulder Colorado. [accessed 2021 Jan 27]. https://americanmineservices.com/types-of-surface-mining/.
- American Public Health Association. 2005. Standard methods for the examination of water and wastewater, 21<sup>st</sup> Ed. Washington (DC): APHA, American Water Works Association, Water Environment Federation. p. 8–10.
- Baker S, Eckerberg K, Zachrisson A. 2014. Political science and ecological restoration. Env Polit. 23(3):509–524.
- Baker S, Eckerberg K. 2016. Ecological restoration success: a policy analysis understanding. Restor Ecol. 24(3):284–290.
- Barbour MT, Gerritsen J, Snyder BD, Stribling JB. 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish, 2<sup>nd</sup> Ed. US Environmental Protection Agency, Office of Water: Washington, DC USA. Report No.: EPA-841-B-99-002.
- Barton CD. 2011. Coal mining versus water quality: an electrifying topic. Water Resour Impact. 13(2):23–24.
- Barton C, Sena K, Angel P. 2018. Reforestation can contribute to a regenerative economy in global mining regions. In: Kingsolver A and Balasundaram S, editors. Global Mountain Regions: Conversations Toward the Future. Indiana University Press, Bloomington, Indiana; pp. 343–354.
- Barton CD, Sena K, Dolan T, Angel P, Zipper C. 2017. Restoring forests on surface coal mines in Appalachia: a regional reforestation approach with global application. In: Bolan N, Kirkham MB, and Ok YS, editors. Spoil to Soil: Mine Site Rehabilitation and Revegetation. CRC Press: Taylor and Francis, Boca Raton, FL; pp. 123–145.
- Beechie TJ, Sear DA, Olden JD, Pess GR, Buffington JM, Moir H, Pollock MM. 2010.
  Process-based principles for restoring river ecosystems. Bioscience. 60(3):209–222.

- Begon M, Townsend CR, Harper JL. 2006. Ecology: from individuals to ecosystems, 4<sup>th</sup> Ed. Blackwell Publishing, Malden, MA.
- Benda L, Hassan MA, Church M, May CL. 2005. Geomorphology of steepland headwaters: the transition from hillslopes to channels. J Am Water Resour Assoc. 41(4):835–851.
- Bernhardt ES, Palmer MA. 2011. The environmental costs of mountaintop mining valley fill operations for aquatic ecosystems of the Central Appalachians. Ann NY Acad Sci. 1223(1):39–57.
- Black PE. 1997. Watershed functions. J Am Water Resour Assoc. 33(1):1–11.
- Blackburn-Lynch WC. 2015. Development of techniques for assessing and restoring streams on surface mined lands. Theses and Dissertations--Biosystems and Agricultural Engineering. Paper 37. http://uknowledge.uky.edu/bae\_etds/37.
- Bodkin R, Kern J, McClellan P, Butt AJ, Martin C. 2007. Limiting total dissolved solids to protect aquatic life. J Soil Water Conserv. 62(3):57A–61A.
- Boehme EA, Zipper CE, Schoenholtz SH, Soucek DJ, Timpano AJ. 2016. Temporal dynamics of benthic macroinvertebrate communities and their response to elevated specific conductance in Appalachia coalfield headwater streams. Ecol Indic. 64(1):171–180.
- Brown AV, Brussock PP. 1991. Comparisons of benthic invertebrates between riffles and pools. Hydrobiologia. 220(2):99–108.

- Bryant G, McPhilliamy S, Childers H. 2002. A survey of the water quality of streams in the primary region of mountaintop/valley fill coal mining. Mountaintop mining/valley fill programmatic environmental impact statement. Region 3, US Environmental Protection Agency, Wheeling, WV.
- Bunn SE, Arthington AH. 2002. Basic principles and ecological consequences of altered flow regimes for aquatic biodiversity. Environ Manage. 30(4):492–507.
- Bunn SE, Davies PM. 2000. Biological processes in running waters and their implications for the assessment of ecological integrity. Hydrobiologia. 422(0):61– 70.
- Burger J, Graves D, Angel P, Davis V, Zipper C. 2005. The forestry reclamation approach. US Office of Surface Mining. Forestry Reclamation Advisory No. 2. [accessed 2020 Sep 21]. https://arri.osmre.gov/FRA/Advisories/FRA\_No.2.7-18-07.Revised.pdf.
- Cairns Jr. J, Pratt JR. 1993. A history of biological monitoring using benthic macroinvertebrates. In: Rosenberg DM and Resh VH, editors. Freshwater biomonitoring and benthic macroinvertebrates. Chapman/Hall, New York; pp. 10–27.
- Cañedo-Argüelles M, Kefford BJ, Piscart C, Prat N, Schäfer RB, Schulz C-J. 2013. Salinisation of rivers: an urgent ecological issue. Eviron Pollut. 173(1):157–167.
- Cherry MA. 2006. Hydrochemical characterization of ten headwater catchments in eastern Kentucky. Lexington, KY: University of Kentucky, Department of Forestry. MS Thesis.

- Chester ET, Robson BJ. 2011. Drought refuges, spatial scale and recolonisation by invertebrates in non-perennial streams. Freshwater Biol. 56(10):2094–2104.
- Costanza R, d'Arge R, de Groot R, Farber S, et al. 1997. The value of the world's ecosystem services and natural capital. Nature. 387(6630):253–260.

Cummins KW. 1974. Structure and function of stream ecosystems. Bioscience. 24(11):631–641.

- Cummins KW, Klug MT. 1979. Feeding ecology of stream invertebrates. Annu Rev Ecol Syst. 10(1):147–172.
- Cummins KW, Sedell JR, Swanson FJ, Minshall GW, et al. 1983. Organic matter budgets for stream ecosystems: problems in their evaluation. In: Barnes JR and Minshall GW, editors. Stream ecology: application and testing of general ecological theory. New York, NY, Plenum Press; pp. 299–353.
- Dahm CN, Valett HM, Baxter CV, Woessner WW. 2006. Hyporheic zones. Pp. 119-142.*In*: FR Hauer and GA Lamberti (Eds), Methods in Stream Ecology 2nd ed.Academy Press, San Diego, California.
- Daily GC, Alexander S, Ehrlich PR, et al. 1997. Ecosystem services: benefits supplied to human societies by natural ecosystems. Issues in Ecology: #2. The Ecological Society of America.
- Dietrich WE, Dunne T. 1993. The Channel Head. Pp. 175-219. *In*: Beven K, and Kirby MJ (Eds), Channel Network Hydrology. Wiley, New York.
- Dongwen L, Ganesh S, Koolaard J. 2020. predictmeans: Calculate predicted means for linear models. R package version 1.0.4. https://CRAN.Rproject.org/package=predictmeans.

- Downey L. 2006. Using geographic information systems to reconceptualize spatial relationships and ecological context. Am J Sociol. 112(2):567–612.
- Feminella JW. 1996. Comparison of benthic macroinvertebrate assemblages in small streams along a gradient of flow permanence. J North Am Benthol Soc. 15(4):651–669.
- Frady C, Johnson S, Li J. 2007. Stream macroinvertebrate community responses as legacies of forest harvest at the H.J. Andrews Experimental Forest, Oregon. For Sci. 53(2):281–293.
- Freund J, Petty JT. 2007. Response of fish and macroinvertebrate bioassessment indices to water chemistry in a mined Appalachian watershed. Environ Manage. 39():707–720.
- Fritz KM, Dodds WK. 2004. Resistance and resilience of macroinvertebrate assemblages to drying and flood in a tallgrass prairie stream system. Hydrobiologia. 527:99– 112.
- Fritz KM, Johnson BR, Walters DM. 2006. Field operations manual for assessing the hydrologic permanence and ecological condition of headwater streams. US Environmental Protection Agency, Office of Research and Development, Washington DC. EPA/600/R-06/126.
- Fritz KM, Fulton S, Johnson BR, Barton CD, Jack JD, Word DA, Burke RA. 2010. Structural and functional characteristics of natural and constructed channels draining a reclaimed mountaintop removal and valley fill coal mine. J North Am Benthol Soc. 29(2):673–689.

- Geredien R. 2009. Assessing the extent of mountaintop removal in Appalachia: an analysis using vector data. Appalachian Voices: Reports — Mountaintop Removal — Reclamation FAIL. https://www.ilovemountains.org/reclamationfail/miningextent2009/Assessing\_the\_Extent\_of\_Mountaintop\_Removal\_in\_App alachia.pdf.
- Gerritsen J, Zheng L, Burton J, Boschen C, Wilkes S, Ludwig J, Cormier S. 2010.
  Inferring Causes of Biological Impairment in the Clear Fork Watershed, West
  Virginia. US Environmental Protection Agency, Office of Research and
  Development, National Center for Environmental Assessment, Cincinnati, OH.
  EPA/600/R-08/146.
- Gomi T, Sidle RC, Richardson JS. 2002. Understanding processes and downstream linkages of headwater systems. Bioscience. 52(10):905–916.
- Gordon ND, McMahon TA, Finlayson BL, Gippel CJ, Nathan RJ. 2004. Stream hydrology: an introduction for ecologists 2<sup>nd</sup> Edition. John Wiley and Sons, Hoboken, New Jersey.
- Green J, Passmore M, Childers H. 2000. A survey of the condition of streams in the primary region of mountaintop mining/valley fill coal mining. Appendix in Mountaintop mining/valley fill programmatic environmental impact statement. US EPA, Region III Aquatic Biology Group, Wheeling, WV.
- Greenberg AE, Clesceri LS, Eaton AD. 1992. Standard methods for the examination of water and wastewater. 18<sup>th</sup> ed. American Public Health Association, Washington, DC.

- Gregory SV, Swanson FJ, McKee WA, Cummins KW. 1991. An ecosystem perspective of riparian zones. Bioscience. 41():540–551.
- Gunn AS. 1991. The restoration of species and natural environments. Environ Ethics. 13(4):291–310.
- Hansen J, Ruedy R, Sato M, Imhoff M, et al. 2001. A closer look at United States and global surface temperature change. J Geophys Res Atmos. 106(D20):23,947–23,963.
- Harding JS, Benfield EF, Bolstad PV, Helfman GS, Jones EBD. 1998. Stream biodiversity: the ghost of land use past. Proc Natl Acad Sci USA. 95(25):14843– 14847.
- Harris JA, Hobbs RJ, Higgs E, Aronson J. 2006. Ecological restoration and global climate change. Restor Ecol. 14(2):170–176.
- Hartman KJ, Kaller MD, Howell JW, Sweka JA. 2005. How much do valley fills influence headwater streams? Hydrobiologia. 532(1-3):91–102.
- Hey RD. 2006. Fluvial geomorphological methodology for natural stable channel design. J Am Water Resour Assoc. 42(2):357–374.
- Hilderbrand RH, Watts AC, Randle AM. 2005. The myths of restoration ecology. Ecol Soc. 10(1):19.
- Hill MO. 1973. Diversity and evenness: a unifying notation and its consequences. Ecology. 54(2):427–432.
- Hill RA, Hawkins CP. 2014. Using modelled stream temperatures to predict macrospatial patterns of stream invertebrate biodiversity. Freshw Biol. 59(12):2632– 2644.

Hilsenhoff WL. 1982. Using a biotic index to evaluate water quality in streams.

Technical Bulletin No. 132. Department of Natural Resources, Madison, WI.

- Hilsenhoff WL. 1987. An improved biotic index of organic stream pollution. Great Lakes Entomol. 20(1):31–40.
- Hilsenhoff WL. 1988. Seasonal correction factors for the biotic index. Great Lakes Entomol. 21(1):9–13.
- Hilsenhoff WL. 1998. A modification of the biotic index of organic stream pollution to remedy problems and permit its use throughout the year. Great Lakes Entomol. 31(1):1–12.
- Hinrichs EN. 1978. Geologic map of the Noble quadrangle, eastern Kentucky. US Geological Survey, Geologic Quadrangle Map GQ-1476.
- Hobbs RJ, Davis MA, Slobodkin LB, et al. 2004. Restoration ecology: the challenge of social values and expectations. Front Ecol Environ. 2(1):43–48.

Holl KD, Howarth RB. 2000. Paying for restoration. Restor Ecol. 8(3):260–267.

- Hopkins RL, Altier BM, Haselman D, Merry AD, White JJ. 2013. Exploring the legacy effects of surface coal mining on stream chemistry. Hydrobiologia. 713(1):87–95.
- Hynes HBN. 1975a. The stream and its valley. Verh Internat Verein Theor Angew Limnol. 19:1–15.
- Jost L. 2006. Entropy and diversity. Oikos. 113(2):363–375.
- Jost L, DeVries P, Walla T, Greeney H, Chao A, Ricotta C. 2010. Partitioning diversity for conservation analyses. Divers Distrib. 16(1):65–76.
- Karr JR. 1981. Assessment of biotic integrity using fish communities. Fisheries. 6(6):21–27.

- Kentucky Division of Water (KDOW). 2002. Methods for assessing biological integrity of surface waters in Kentucky. Kentucky Department for Environmental Protection. Frankfort, Kentucky.
- Kentucky Geological Survey. 2020. Water fact sheet. University of Kentucky, Lexington, KY. [accessed 2021 Feb 1]. https://www.uky.edu/KGS/water/index.php#.
- Kentucky Office of Energy Policy (KOEP). 2017. Kentucky coal facts, 17<sup>th</sup> Edition. Kentucky Energy and Environment Cabinet. Frankfort, Kentucky.
- Kerans B, Karr JR. 1994. A benthic index of biotic integrity (B-IBI) for rivers of the Tennessee Valley. Ecol Appl. 4():768–785.
- Klemm DJ, Lewis PA, Fulk F, Lazorchak JM. 1990. Macroinvertebrate field and laboratory methods for evaluating the biological integrity of surface waters. US Environmental Protection Agency, Office of Research and Development, Washington, DC. EPA/600/4-90/030 (NTIS PB91171363).
- Lamberti GA, Chaloner DT, Hershey AE. 2010. Linkages among aquatic ecosystems. J North Am Benthol Soc. 29(1):245–263.
- Lemmon PE. 1956. A spherical densiometer for estimating forest overstory density. For Sci. 2(4):314–320.
- Lenat, DR. 1993. A biotic index for the southeastern United States: Derivation and list of tolerance values, with criteria for assigning water quality ratings. J North Am Benthol Soc. 12(3):279–290.
- MacDonald LH, Coe D. 2007. Influence of headwater streams on downstream reaches in forested areas. For Sci. 53():148–168.

- McDowell RC. 1985. The geology of Kentucky. Professional paper 1151-H, US Geological Survey. http://pubs.usgs.gov/pp/p1151h/, accessed January 27, 2021.
- Merricks TC, Cherry DS, Zipper CE, Currie RJ, Valenti TW. 2007. Coal-mine hollow fill and settling pond influences on headwater streams in Southern West Virginia, USA. Environ Monit Assess. 129(1-3):359–378.
- Merritt RW, Cummins KW, Berg MB. 2008. An Introduction to the Aquatic Insects of North America, 4<sup>th</sup> ed. Kendall Hunt, Dubuque, IA.
- Meyer JL, Wallace JB. 2001. Lost linkages and lotic ecology: rediscovering small streams. Pp. 295–317. *In*: MC Press, NJ Huntly and S Levin (Eds.), In Proceedings, The 41<sup>st</sup> Symposium of the British Ecological Society. April 10-13, 2000, Orlando, Florida. Ecological Society of America.
- Meyer JL, Strayer DL, Wallace JB, Eggert SL, Helfman GS, Leonard NE. 2007. The contribution of headwater streams to biodiversity in river networks. J Am Water Resour Assoc. 43(1):86–103.
- Moore RD, Wondzell SM. 2005. Physical hydrology and the effects of forest harvesting in the Pacific Northwest: a review. J Am Water Resour Assoc. 41(4):763–784.
- Morse JC, McCafferty WP, Stark BP, Jacobus LM (Eds). 2017. Larvae of the southeastern USA mayfly, stonefly, and caddisfly species (Ephemeroptera, Plecoptera, and Trichoptera). Biota of South Carolina. Vol 9. Clemson University Public Service Publishing, Clemson University, Clemson, South Carolina, 482 pp.

Müller K. 1982. The colonization cycle of freshwater insects. Oecologia. 52(2):202–207.

Naiman RJ, Décamps H. 1997. The ecology of interfaces: Riparian zones. Annu Rev Ecol Syst. 28(1):621–658.

- Newbold JD, Elwood JW, O'Neill RV, VanWinkle W. 1981. Measuring nutrient spiralling in streams. Can J Fish Aquat Sci. 38(7):860–863.
- Omernik JM, Griffith GE. 2014. Ecoregions of the conterminous United States: evolution of a hierarchical spatial framework. Environ Manage. 54(6):1249–1266.
- Overstreet JC. 1984. Robinson Forest Inventory, 1980-1982. Department of Forestry, College of Agriculture, University of Kentucky, Lexington, Kentucky.
- Palmer MA, Filoso S. 2009. Restoration of ecosystem services for environmental markets. Science. 325(5940):575–576.
- Pericak AA, Thomas CJ, Kroodsma DA, Wasson MF, et al. 2018. Mapping the yearly extent of surface coal mining in Central Appalachia using Landsat and Google Earth Engine. PloS ONE. 13(7): e0197758.
- Pinheiro J, Bates D, DebRoy S, Sarkar D, R Core Team. 2020. Nlme: Linear and nonlinear mixed effects models. R package version 3.1-149. https://CRAN.Rproject.org/package=nlme.
- Pond GJ. 2000. Comparison of macroinvertebrate communities of two intermittent streams with different disturbance histories in Letcher County, Kentucky. J Ky Acad Sci. 61(1):10–22.
- Pond GJ, Passmore ME, Borsuk FA, Reynolds L, Rose CJ. 2008. Downstream effects of mountaintop coal mining: comparing biological conditions using family- and genus-level macroinvertebrate bioassessment tools. J North Am Benthol Soc. 27(3):717–737.
- Pond GJ. 2010. Patterns of Ephemeroptera taxa loss in Appalachian headwater streams (Kentucky, USA). Hydrobiologia. 641(1):185–201.

- Pond GJ. 2012. Biodiversity loss in Appalachian headwater streams (Kentucky, USA): Plecoptera and Trichoptera communities. Hydrobiologia. 679(1):97–117.
- Pringle CM. 1997. How disturbance is transmitted upstream: going against the flow. J North Am Benthol Soc. 16(2):425–438.
- R Core Team. 2019. R: A language and environment for statistical computing. Version 4.0.2 Vienna (Austria): R Foundation for Statistical Computing. https://www.Rproject.org.
- Rosgen DL. 1998. The reference reach—a blueprint for natural channel design. *In*:
  Proceedings of the Wetland Engineering and River Restoration Conference.
  March 22-27, Denver, CO. American Society of Civil Engineers.
- Roy AH, Dybas AL, Fritz KM, Lubbers HR. 2009. Urbanization affects the extent and hydrologic permanence of headwater streams in a midwestern US metropolitan area. J North Am Benthol Soc. 28(4):911–928.
- Schindelin J, Arganda-Carreras I, Frise E, et al. 2012. Fiji: an open-source platform for biological-image analysis. Nat Methods. 9(7):676–682.

Sena KL, Barton CD, Williamson TN. 2020. Water quality of precipitation and streamflow, with air temperature data, in four Kentucky, Appalachian watersheds–1971 to 2018: US Geological Survey data release, https://doi.org/10.5066/P9FPLG1O.

- SERI. 2004. The SER International primer on ecological restoration. Society for Ecological Restoration International Science & Policy Working Group. www.ser.org.
- Simpson EH. 1949. Measurement of diversity. Nature. 163(4148):388.

- Spellerberg IF, Fedor PJ. 2003. A tribute to Claude Shannon (1916-2001) and a plea for more rigorous use of species richness, species diversity and the 'Shannon-Wiener' Index. Glob Ecol Biogeogr. 12(3):177–179.
- Sponseller RA, Benfield EF. 2001. Relationships between land use, spatial scale and stream macroinvertebrate communities. Freshwater Biol. 46(10):1409–1424.
- Steinman AD, Lamberti GA, Leavitt PR. 2006. Biomass and pigments of benthic algae.
  Pp. 357–379. *In*: FR Hauer and GA Lamberti (Eds), Methods in Stream Ecology 2<sup>nd</sup> ed. Academy Press, San Diego, California.
- Thorp JH, Covich AP. 2010. Ecology and Classification of North American Freshwater Invertebrates. London: Academic Press.
- Townsend PA, Helmers DP, Kingdon CC, et al. 2009. Changes in the extent of surface mining and reclamation in the Central Appalachians detected using a 1976-2006 Landsat time series. Remote Sens Environ. 113(1):62–72.
- University of Kentucky. 2021. Robinson Forest. Department of Forestry and Natural Resources, College of Agriculture, Food and Environment. University of Kentucky, Lexington, KY. [accessed 2021 Jan 27]. https://robinsonforest.ca.uky.edu.
- United States Department of Agriculture: Farm Service Agency. 2021. National Geospatial Data Asset (NGDA): National Aerial Imagery Program (NAIP). Aerial Photography Field Office, Salt Lake City, UT. [accessed 2021 Jan 27]. https://www.fsa.usda.gov/programs-and-services/aerial-photography/imageryprograms/naip-imagery/index.

- United States Environmental Protection Agency. 2002. Landscape Scale Cumulative
  Impact Study. In: Draft programmatic environmental impact statement on
  mountaintop mining/valley fills in Appalachia 2003. Appendix I. U.S.
  Environmental Protection Agency, Region 3, Philadelphia, PA. Available online
  at http://www.epa.gov/Region3/mtntop/eis2003appendices.ht#appd.
- United States Environmental Protection Agency. 2003. Draft programmatic environmental impact statement on mountaintop mining/valley fills in Appalachia. U.S. Environmental Protection Agency, Region 3, Philadelphia, PA. Available online at http://www.epa.gov?Region3/mtntop/eis2003.htm.
- United States Environmental Protection Agency. 2005. In Final programmatic environmental impact statement (FPEIS) on mountaintop mining/valley fills in Appalachia. National Center for Environmental Assessment, US Environmental Protection Agency, Washington, DC. EPA 9-03-R-05002.
- United States Environmental Protection Agency. 2006. Wadeable Streams Assessment. Office of Water and Office of Research and Development, Washington, DC. EPA 841-B-06-002.
- United States Environmental Protection Agency. 2011a. A field-based aquatic life benchmark for conductivity in central Appalachian streams. Office of Research and Development, National Center for Environmental Assessment, Washington, DC. Report No.: EPA/600/R-10/023F.

- United States Environmental Protection Agency. 2011b. The effects of mountaintop mines and valley fills on aquatic ecosystems of the Central Appalachian Coalfields. Office of Research and Development, National Center for Environmental Assessment, Washington, DC. Report No.: EPA/600/R-09/138F.
- United States Environmental Protection Agency. 2016. National Rivers and Streams Assessment 2008-2009: A collaborative Survey. Office of Water and Office of Research and Development, Washington, DC. EPA/841/R-16/007.
- Vannote RL, Minshall GW, Cummins KW, Sedell JR, Cushing CE. 1980. The river continuum concept. Can J Fish Aquat Sci. 37(1):130–137.
- Villines JA, Agouridis CT, Warner RC, Barton CD. 2015. Using GIS to delineate headwater stream origins in the Appalachian Coalfields of Kentucky. J Am Water Resour Assoc. 51(6):1667–1687.
- Wallace JB, Webster JR, Woodall WR. 1977. The role of filter feeders in flowing waters. Arch Hydrobiol. 79:506–532.
- Wallace JB, Whiles MR, Eggert S, Cuffney TF, et al. 1995. Long-term dynamics of coarse particulate organic matter in three Appalachian mountain streams. J North Am Benthol Soc. 14(2):217–232.
- Ward JV. 1989. The four-dimensional nature of lotic ecosystems. J North Am Benthol Soc. 8(1):2–8.
- Webster JR, Patten BC. 1979. Effects of watershed perturbation on stream potassium and calcium dynamics. Ecol Monogr. 49(1):51–72.

- Webster JR, Benfield EF, Ehrman TP, Schaeffer MA, Tank JL, Hutchens JJ, D'Angelo DJ. 1999. What happens to allochthonous material that falls into streams? A synthesis of new and published information from Coweeta. Freshw Biol. 41(4):687–705.
- Weigand M, Wurm M, Dech S, Taubenböck. 2019. Remote sensing in environmental justice research. ISPRS Int J Geo-Inf. 8(20):1–28.
- Williamson TN, Agouridis CT, Barton CD, Villines JA, Lant JG. 2015. Classification of ephemeral, intermittent, and perennial stream reaches using a TOPMODEL-based approach. J Am Water Resour Assoc. 51(6):1739–1759.
- Witt EL. 2012. Evaluating streamside management zone effectiveness in forested watersheds of the Cumberland Plateau. Theses and Dissertations—Plant and Soil Sciences. 6. https://uknowledge.uky.edu/pss\_etds/6.
- Wolfe MK, Mennis J. 2012. Does vegetation encourage or suppress urban crime? Evidence from Philadelphia, PA. Landsc Urban Plan. 108(2-4):112–122.
- Zipper CE, Burger JA, Skousen JG, Angel PN, Barton CD, Davis V, Franklin JA. 2011. Restoring forests and associated ecosystem services on Appalachian coal surface mines. Environ Manage. 47(5):751–765.

### APPENDIX A

#### ANNUALIZED SUMMARY OF BENTHIC MACROINVERTEBRATES

#### ABSTRACT

The purpose of Appendix A is to help the reader compare the ecological performance at each stream site with values for density and relative abundance standardized to a twelve-month period.

		<u>C</u>	reated	Stream	<u>ן</u> (ר	<u>Re</u>	<u>Reference Streams</u> Unmined (LM)			
			& l	Up Sect	tion (GI	J)	& Mi	ine-impa	acted (	y WB)
			GI	)	G	Ű	LN	Λ	W	/B
Orde	r Family	Sub-family / Genus	D	RA	D	RA	D	RA	D	RA
Non-i	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	1141.1	21.4	443.1	7.3	1.1	0.1		
S	Sphaeriidae	Pisidium sp.			936.5	15.4	1.7	0.1		
Η	Naididae		148.7	2.8	235.6	3.9	12.4	0.8		
Insect	ts									
С	Dryopidae	Helichus sp.			13.3	0.2	8.3	0.5		
	Dytiscidae	Agabus sp.	13.7	0.3	1.5	< 0.1				
	2	Celina sp.			0.2	< 0.1				
		Copelatus sp.	0.6	< 0.1						
		Desmopachria sp.			0.2	< 0.1				
		Hydroporus sp.	0.2	< 0.1						
		Laccophilus sp.	0.7	< 0.1	0.6	< 0.1				
		Neoporus sp.	18.5	0.4	4.3	0.1				
	Elmidae	<i>Dubiraphia</i> sp.	0.4	< 0.1	1.7	< 0.1				
		Optioservus sp.	1.3	< 0.1	7.2	0.1	7.6	0.5		
		<i>Oulimnius</i> sp.					4.3	0.3		
		Stenelmis sp.	21.5	0.4	83.3	1.4	27.8	1.7		
	Haliplidae	<i>Haliplus</i> sp.			0.4	< 0.1				
		Peltodytes sp.	15.6	0.3	9.8	0.2				
	Hydrophilidae	Anacaena sp.	0.7	< 0.1	7.2	0.1				
		Berosus sp.	3.7	0.1	0.2	< 0.1				
		Cymbiodyta sp.			0.2	< 0.1				
		Enochrus sp.			0.2	< 0.1				

**Appendix A.** Summary of all macroinvertebrates collected from February 2014 – January 2015. Values are annualized average densities [ $D = (no./m^2)$ , n = 5 per month] and relative abundances [RA (%), n = 5 per month].

		$\underline{C}$	reated	Stream	<u>ו</u> (ר	Re	Reference Streams			
			Do &	wn Sec	tion (GI	<b>ר</b>	8- M	ina imp	I (LIVI)	<b>7D</b> )
					GI (OU	<u>)</u> T		<u>те-тпра</u> Л		<u>, (U)</u>
Order	Family	Sub-family / Genus		RA	D	RA		RA	 	RA
C	Hvdrophilidae	Helochares sp.	Ľ	111	0.7	< 0.1	D	IUI	Ð	101
	)	Hvdrochus sp.	0.2	< 0.1	0.4	< 0.1				
		Paracymus sp.	2.0	< 0.1	2.0	< 0.1				
		Tropisternus sp.	5.7	0.1	3.0	0.1				
	Psephenidae	<i>Ectopria</i> sp.	0.2	< 0.1	5.2	0.1	63.3	4.0	0.4	1.3
D	Ceratopogonidae	Alluaudomyia sp.	0.2	< 0.1						
	10	Atrichopogon sp.	22.8	0.4	1.5	< 0.1	0.6	< 0.1	0.2	0.7
		Bezzia/Palpomyia sp.	8.7	0.2	3.9	< 0.1	9.3	0.6	0.6	2.0
		Ceratopogon sp.	0.6	< 0.1	0.2	< 0.1	15.7	1.0		
		Culicoides sp.	15.7	0.3	13.0	0.2	2.2	0.1		
		Monohelea sp.	0.2	< 0.1						
		Dasyhelea sp.			0.2	< 0.1				
		Probezzia sp.	27.4	0.5	106.1	1.8	37.6	2.4		
		Sphaeromias sp.	41.5	0.8	32.8	0.5				
D	Chironomidae	Chironominae	181.5	3.4	574.1	9.5	239.3	14.9	3.0	10.5
		Corynoneura sp.	67.4	1.3	88.9	1.5	11.1	0.7	0.2	0.7
		Orthocladiinae	327.2	6.1	324.6	5.3	108.3	6.8	8.0	28.1
		Stempellina sp.					24.6	1.5		
		Tanypodinae	1138.3	21.3	113.9	18.4	86.7	5.4	3.5	12.4
		Tanytarsini (tribe)	1238.9	23.2	631.1	10.4	43.0	2.7	0.2	0.7
	Culicidae	Anopheles sp.	0.9	< 0.1	2.2	< 0.1			0.2	0.7
		Haemagogus sp.			0.4	< 0.1				
	Dixidae	Dixa sp.	0.2	< 0.1	2.4	< 0.1	3.9	0.2		
	Dolichopodidae	dolichopodid genus A	1.3	< 0.1	0.4	< 0.1				
	Empididae	empidid genus A			0.7	< 0.1				

	````````````````````````````````		<u>(</u> Do	Created	Stream	<u>n</u> D)	<u>R</u>	Reference Streams Unmined (LM)				
			&	Up Sec	tion (G	U)	& M	line-imp	acted (V	VB)		
			G	D	G	Ú	L	M	Ŵ	B		
Order	Family	Sub-family / Genus	D	RA	D	RA	D	RA	D	RA		
D	Empididae	<i>Chelifera</i> sp.					9.3	0.6				
		Hemerodromia sp.	11.7	0.2	12.2	0.2	0.4	< 0.1				
		<i>Neoplasta</i> sp.	0.6	< 0.1	0.9	< 0.1			0.2	0.7		
	Ephydridae	ephydrid genus A	4.4	0.1	1.1	< 0.1						
		<i>Scatella</i> sp.			0.2	< 0.1						
	Psychodidae	<i>Pericoma</i> sp.	0.2	< 0.1	1.5	< 0.1						
	Sciaridae	sciarid genus A	13.7	0.3	0.9	< 0.1						
	Sciomyzidae	Sepedon sp.	3.5	0.1	3.7	0.1						
	Simuliidae	Prosimulium sp.					20.6	1.3				
		<i>Simulium</i> sp.	50.9	1.0	48.7	0.8	0.7	0.1				
		Stegopterna sp.	37.2	0.7	44.3	0.7	4.3	0.3				
	Stratiomyidae	Caloparyphus sp.	2.0	< 0.1	12.4	0.2						
		Nemotelus sp.			0.2	< 0.1						
		Stratiomys sp.	0.7	< 0.1	12.0	0.2						
	Syrphidae	Eristalis sp.			0.2	< 0.1						
	Tabanidae	Chrysops sp.	57.0	1.1	106.9	1.8	1.3	0.1				
		<i>Tabanus</i> sp.	9.3	0.2	3.2	0.1	0.2	< 0.1				
	Tipulidae	Dicranota sp.			0.6	< 0.1	4.6	0.3	0.2	0.7		
		Helius sp.	0.9	< 0.1	3.2	0.1						
		Hexatoma sp.			0.2	< 0.1	70.9	4.4				
		Leptotarsus sp.					0.4	< 0.1				
		Limnophila sp.			2.4	< 0.1	45.0	2.8	0.4	1.3		
		<i>Limonia</i> sp.	16.9	0.3	2.4	< 0.1	0.2	< 0.1				
		Molophilus sp.					3.3	0.2	0.2	0.7		
		Ormosia sp.	1.7	< 0.1			0.4	< 0.1				

	<u>_</u>		<u>(</u>	Created	Stream	<u>n</u>	R	Reference Strea		
				wn Sec	ction (G	D)	& N	Unmine	d (LM) acted (V	WB)
			$\frac{\alpha}{G}$	D D	<u>UOII (U</u> G	U		M	W	B
Order	Family	Sub-family / Genus	$\overline{D}$	RA	D	RA	 D	RA	D	RA
D	Tipulidae	Pedicia sp.			0.2	< 0.1	0.4	< 0.1		
	-	<i>Pilaria</i> sp.	4.8	0.1	25.7	0.4	0.4	< 0.1		
		Pseudolimnophila sp.	3.3	0.1	63.9	1.1	16.7	1.0		
		<i>Tipula</i> sp.	52.0	1.0	15.6	0.3	7.8	0.5	3.5	12.4
E	Ameletidae	Ameletus sp.	89.8	1.7	224.8	3.7	18.3	1.2		
	Baetidae	Baetis sp.	30.0	0.6	6.5	0.1	8.5	0.5	0.2	0.7
		Procloeon sp.					7.0	0.4		
	Ephemerellidae	Attenella sp.			0.2	< 0.1				
		Drunella sp.					1.1	0.1		
		Ephemerella sp.					51.1	3.2	0.2	0.7
		Eurylophella sp.	0.2	< 0.1	1.7	< 0.1	17.8	1.1		
	Ephemeridae	<i>Ephemera</i> sp.					29.3	1.8		
	Heptageniidae	<i>Epeorus</i> sp.					42.0	2.6		
		Maccaffertium sp.					15.7	1.0		
		Stenacron sp.					15.7	1.0		
	Leptophlebiidae	<i>Habrophlebia</i> sp.					1.1	0.1		
		Paraleptophlebia sp.			0.2	< 0.1	118.3	7.4	0.4	1.3
	Siphlonuridae	Siphlonurus sp.	1.7	< 0.1	0.7	< 0.1				
Μ	Corydalidae	Chauliodes sp.	0.9	< 0.1	0.7	< 0.1				
		<i>Nigronia</i> sp.					5.0	0.3	0.4	1.3
	Sialidae	<i>Sialis</i> sp.	0.6	< 0.1	27.4	0.5	2.4	0.2	0.4	1.3
0	Aeshnidae	<i>Aeshna</i> sp.	0.2	< 0.1	0.4	< 0.1				
		<i>Boyeria</i> sp.	1.7	< 0.1	1.5	< 0.1	0.6	< 0.1		
	Calopterygidae	Calopteryx sp.	0.4	< 0.1	13.3	0.2				
	Coenagrionidae	Amphiagrion sp.	1.9	< 0.1						

			<u>(</u>	Created	Stream	<u>n</u>	<u>Reference Streams</u> Unmined (I M)			
				Un Sec	tion (G	D) ID	& N	line-imp	u (LM) acted ()	WR)
			<u>G</u>	<u>- D</u>	<u>G</u>	U		M	W	<u>в</u>
Order	r Family	Sub-family / Genus	D	RA	D	RA	<u>D</u>	RA	D	RA
0	Coenagrionidae	Argia sp.	22.2	0.4	30.0	0.5				
	-	Enallagma sp.	0.2	< 0.1						
	Cordulegastridae	Cordulegaster sp.	1.7	< 0.1	10.4	0.2	7.6	0.5		
	Gomphidae	Stylogomphus sp.	1.7	< 0.1	5.0	0.1	30.7	1.9	0.2	0.7
	Libellulidae	prob. Leucorrhinia sp.	14.1	0.3	2.0	< 0.1				
		<i>Leucorrhinia</i> sp.	3.2	0.1	0.7	< 0.1				
		Pentala sp.	0.2	< 0.1						
		Sympetrum sp.	14.4	0.3	2.2	< 0.1				
Р	Capniidae	<i>Allocapnia</i> sp.	54.6	1.0	39.3	0.7	47.0	2.9		
	Chloroperlidae	<i>Haploperla</i> sp.					35.2	2.2		
		Sweltsa sp.					1.1	0.1		
	Leuctridae	Leuctra sp.			11.5	0.2	69.8	4.4	0.2	0.7
	Nemouridae	Amphinemura sp.	100.0	1.9	230.6	3.8	9.8	0.6	2.0	7.2
		Ostrocerca sp.					1.1	0.1		
		Prostoia sp.	1.1	< 0.1	0.9	< 0.1				
		<i>Soyedina</i> sp.			0.7	< 0.1	5.9	0.4	1.5	5.2
	Peltoperlidae	<i>Peltoperla</i> sp.			1.9	< 0.1	0.9	0.1		
	Perlidae	Acroneuria sp.			0.2		5.7	0.4	0.2	0.7
		<i>Eccoptura</i> sp.			0.4	< 0.1	19.3	1.2		
	Perlodidae	<i>Clioperla</i> sp.	0.2	< 0.1	3.7	0.1	0.2	< 0.1		
		<i>Diploperla</i> sp.	1.3	< 0.1	4.4	0.1	2.4	0.2		
		<i>Isoperla</i> sp.	37.6	0.7	24.6	0.4	10.4	0.7		
		<i>Malirekus</i> sp.	0.4	< 0.1	6.1	0.1	6.3	0.4		
		<i>Remenus</i> sp.					0.7	0.1		
		Yugus sp.							0.2	0.7

			(	Created	Stream	<u>n</u>	R	<b>Reference Streams</b>				
			Do	own Sec	tion (G	iD)		Unmine	ed (LM)			
			&	Up Sec	tion (G	U)	& M	line-im	pacted (V	WB)		
			G	ΰD	G	ίU	L	М	W	В		
Order	· Family	Sub-family / Genus	D	RA	D	RA	D	RA	D	RA		
Т	Glossosomatidae	Agapetus sp.					11.7	0.7				
	Goeridae	Goera sp.					0.6	< 0.1				
	Hydropsychidae	Cheumatopsyche sp.	0.6	< 0.1	0.4	< 0.1	4.8	0.3				
		Diplectrona sp.	2.8	0.1	48.5	0.8	33.0	2.1	0.7	2.7		
		Hydropsyche sp.			3.2	0.1	1.7	0.1				
	Hydroptilidae	<i>Hydroptila</i> sp.	0.4	< 0.1	13.9	0.2	0.2	< 0.1				
		<i>Oxyethira</i> sp.			0.2	< 0.1						
	Lepidostomatidae	Lepidostoma sp.					2.0	0.1				
	Leptoceridae	Oecetis sp.	1.1	< 0.1								
	Limnephilidae	Ironoquia sp.	196.1	3.7	77.0	1.3						
		Pycnopsyche sp.			0.2	< 0.1	17.6	1.1	0.9	3.3		
	Molannidae	<i>Molanna</i> sp.					4.1	0.3				
	Philopotamidae	<i>Chimarra</i> sp.	1.9	< 0.1	181.7	3.0						
		Dolophilodes sp.					3.2	0.2				
		<i>Wormaldia</i> sp.	0.4	< 0.1	13.0	0.2	12.4	0.8				
	Phryganidae	Ptilostomis sp.	35.4	0.7	60.0	1.0						
	Polycentropodidae	Polycentropus sp.			0.7	< 0.1	11.7	0.7	0.4	1.3		
	Psychomyiidae	<i>Lype</i> sp.					0.2	< 0.1				
	Rhyacophilidae	<i>Rhyacophila</i> sp.	0.2	< 0.1	0.7	< 0.1	10.6	0.7				
	Thremmatidae	<i>Neophylax</i> sp.	0.2	< 0.1	0.2	< 0.1	3.9	0.2				
Total	Invertebrates			28,822		32,762		8,646		153		
Total Richness / EPT Richness					105/31		85/40		28/11			
B = Basommatophora, $C$ = Coleoptera, $D$ = Dipter				phemer	optera,	$H = Ha_1$	plotaxida,	$M = \mathbf{M}$	egalopte	ra,		
O = O	donata, $P = Plecopte$	Trichop	tera									

### APPENDIX B

## MONTHLY MACROINVERTEBRATE METRIC VALUES

### ABSTRACT

The purpose of Appendix B is to help the reader compare the ecological performance at each stream site with monthly values for 16 benthic macroinvertebrate metrics.

		•	Created	l Stream		Reference Streams				
		Down S	Section	Up Se	ection	Unm	ined	Mine-in	npacted	
		G	D	G	U		Λ	W	В	
Metric $(y)$	Month	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	
Density (no·m <sup>-2</sup> )										
	FEB 2014	4,073.3	843.9	4,431.1	605.9	1,104.4	219.7	6.7	4.4	
	MAR	3,920.0	889.1	6,786.7	1,783.7	640.0	267.4	8.9	5.4	
	APR	6,451.1	1,950.4	12,555.6	1,231.8	1,108.9	237.4	24.4	16.6	
	MAY	9,091.1	3,934.0	8,908.9	1,135.9	1,728.9	244.1	113.3	62.5	
	JUN	6,757.8	2,723.1	6,886.7	2,595.7	2,777.8	917.9	13.3	5.4	
	JUL	164.4	79.5	2,677.8	1,420.1	1,586.7	194.6	28.9	10.3	
	AUG	428.9	214.6	1,080.0	333.2	1,011.1	164.2	11.1	8.6	
	SEP	1,686.7	452.7	3,255.6	490.7	2,802.2	616.8	35.6	18.1	
	OCT	1,273.3	204.1	3,680.0	1,041.3	1,988.9	790.9	24.4	11.3	
	NOV	7,108.9	1,857.6	6,813.3	1,066.9	1,233.3	169.3	20.0	8.2	
	DEC	8,653.3	2,653.4	7,300.0	226.7	1,602.2	307.1	37.8	5.7	
	JAN 2015	14,597.8	6,385.0	8,428.9	980.2	1,962.2	630.0	15.6	5.7	
Total Richness S										
(genus)	FEB 2014	21.4	2.5	24.8	3.4	22.4	2.0	0.6	0.4	
	MAR	25.6	3.5	33.2	0.8	18.4	4.2	0.8	0.5	
	APR	21.2	2.2	33.4	0.8	23.8	2.9	0.8	0.4	
	MAY	22.8	1.6	27.2	2.5	28.0	3.2	3.0	0.4	
	JUN	18.2	3.7	24.2	2.5	23.6	1.4	0.8	0.4	
	JUL	4.6	0.7	13.0	1.8	15.0	1.8	1.8	0.6	
	AUG	5.6	0.9	12.0	0.9	23.4	1.0	1.0	0.8	
	SEP	15.0	1.8	24.4	3.4	30.8	2.4	1.0	0.4	
	OCT	16.2	2.1	20.4	2.2	26.6	3.5	1.8	0.9	
	NOV	22.2	0.7	27.0	1.6	26.4	1.6	1.6	0.6	
	DEC	19.8	1.0	25.6	0.9	28.4	1.2	2.6	0.5	
	JAN 2015	16.2	1.1	24.0	0.6	26.2	3.3	1.2	0.4	

**Appendix B1.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

••		Created Stream				Reference Streams				
	_	Down S	Section D	Up Se Gl	ection	Unm	ined 1	Mine-i V	mpacted VB	
Metric (y)	Month	x	SE		SE	$\frac{z}{\bar{x}}$	SE	$\overline{x}$	SE	
% Benthic Insects										
	FEB 2014	78.5	5.3	78.4	9.0	99.3	0.5	100	0.0	
	MAR	68.5	8.4	85.7	7.4	99.5	0.5	100	0.0	
	APR	76.9	9.1	85.3	6.0	99.8	0.2	100	0.0	
	MAY	70.0	15.8	83.3	8.9	99.9	0.1	100	0.0	
	JUN	58.9	14.0	55.4	5.0	98.8	0.4	100	0.0	
	JUL	64.0	12.0	29.5	6.1	98.3	0.9	100	0.0	
	AUG	31.7	12.1	39.7	7.5	96.1	0.9	100	0.0	
	SEP	55.4	16.3	58.8	10.2	99.6	0.3	100	0.0	
	OCT	88.9	2.4	50.0	15.1	98.4	1.2	100	0.0	
	NOV	94.7	2.1	79.3	6.1	99.3	0.5	100	0.0	
	DEC	94.8	3.3	85.0	4.6	99.4	0.2	100	0.0	
	JAN 2015	87.5	10.2	72.9	6.0	99.3	0.4	100	0.0	
% Top 2										
Dominant Taxa	FEB 2014	47.1	8.2	35.0	10.2	33.2	3.7	n/a	n/a	
(Abundance)	MAR	43.8	11.5	31.1	7.2	18.0	3.7	n/a	n/a	
、 <i>,</i>	APR	44.4	11.8	37.3	3.2	36.9	3.8	n/a	n/a	
	MAY	56.0	13.9	57.2	7.0	30.1	3.9	n/a	n/a	
	JUN	74.5	7.6	40.8	5.4	38.2	8.2	n/a	n/a	
	JUL	46.6	17.9	56.1	12.7	63.6	7.4	n/a	n/a	
	AUG	68.3	12.1	52.6	8.1	22.6	2.9	n/a	n/a	
	SEP	51.1	11.0	39.5	10.2	21.1	3.8	n/a	n/a	
	OCT	34.4	8.1	49.9	15.1	26.2	3.9	n/a	n/a	
	NOV	51.8	5.2	39.2	5.5	31.2	5.3	n/a	n/a	
	DEC	62.7	9.0	45.3	6.0	24.6	5.1	n/a	n/a	
	JAN 2015	65.2	10.9	39.2	8.6	25.7	3.2	n/a	n/a	

**Appendix B2.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

		Created Stream					Reference	e Streams	
	-	Down S	Section	Up Se	ction	Unm	ined M	Mine-in W	npacted B
Metric (v)		<u> </u>	SE	$\frac{1}{\bar{x}}$	SE	$\frac{1}{\bar{x}}$	SE	$\frac{1}{\bar{x}}$	SE
EPT Richness									
(genus)	FEB 2014	5.0	0.9	7.6	1.2	11.4	0.9	0.0	0.0
C )	MAR	5.8	0.9	11.4	1.4	8.4	1.3	0.0	0.0
	APR	5.8	0.4	10.0	0.3	12.6	1.5	0.0	0.0
	MAY	3.4	0.7	7.2	0.8	15.0	1.1	0.0	0.0
	JUN	1.2	0.7	2.2	0.4	9.8	0.9	0.0	0.0
	JUL	0.2	0.2	0.2	0.2	5.0	0.9	0.0	0.0
	AUG	0.0	0.0	0.0	0.0	8.4	0.8	0.0	0.0
	SEP	1.6	0.2	3.2	1.0	11.6	0.9	0.0	0.0
	OCT	1.8	0.4	3.6	1.1	10.2	2.2	0.0	0.0
	NOV	4.2	0.2	7.6	1.2	11.6	1.1	0.0	0.0
	DEC	4.8	0.4	7.4	0.7	13.4	0.4	0.0	0.0
	JAN 2015	4.6	0.7	6.0	1.2	13.2	1.6	0.0	0.0
Ephemeroptera									
Richness	FEB 2014	0.8	0.2	1.6	0.2	5.4	0.2	0.0	0.0
(genus)	MAR	0.6	0.2	1.6	0.2	4.2	0.6	0.0	0.0
C )	APR	1.4	0.2	1.4	0.2	4.6	0.5	0.0	0.0
	MAY	1.0	0.3	1.6	0.2	5.6	1.0	0.0	0.0
	JUN	0.4	0.2	0.4	0.2	4.6	0.7	0.0	0.0
	JUL	0.0	0.0	0.0	0.0	2.8	0.6	0.0	0.0
	AUG	0.0	0.0	0.0	0.0	3.0	0.4	0.0	0.0
	SEP	0.6	0.2	0.2	0.2	4.4	0.2	0.0	0.0
	OCT	0.0	0.0	0.2	0.2	3.3	0.7	0.0	0.0
	NOV	1.0	0.3	1.0	0.3	3.6	0.7	0.0	0.0
	DEC	1.0	0.3	1.0	0.3	5.8	0.6	0.0	0.0
	JAN 2015	1.6	0.4	1.6	0.2	5.4	0.2	0.0	0.0

**Appendix B3.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

			Created	Stream			Reference	e Streams	
	-	Down G	Section D	Up Se Gl	ection U	Unm Ll	ined M	Mine-in W	npacted B
Metric (y)	Month	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE
Plecoptera									
Richness	FEB 2014	2.0	0.4	2.2	0.7	2.8	0.6	0.0	0.0
(genus)	MAR	2.8	0.4	5.2	0.7	2.6	0.4	0.0	0.0
	APR	2.4	0.2	4.6	0.4	6.0	0.9	0.0	0.0
	MAY	0.6	0.4	2.0	0.6	5.0	0.5	0.0	0.0
	JUN	0.2	0.2	0.4	0.2	2.6	0.9	0.0	0.0
	JUL	0.0	0.0	0.0	0.0	0.6	0.6	0.0	0.0
	AUG	0.0	0.0	0.0	0.0	3.2	0.6	0.0	0.0
	SEP	0.0	0.0	0.4	0.2	4.4	0.4	0.0	0.0
	OCT	0.2	0.2	0.8	0.5	4.0	1.0	0.0	0.0
	NOV	1.2	0.2	2.0	0.8	3.8	0.4	0.0	0.0
	DEC	1.6	0.4	2.0	0.3	3.6	0.2	0.0	0.0
	JAN 2015	1.4	0.2	1.2	0.7	4.8	0.6	0.0	0.0
Trichoptera									
Richness	FEB 2014	2.2	0.4	3.8	0.6	3.2	0.4	0.0	0.0
(genus)	MAR	2.4	0.4	4.6	0.8	1.6	0.6	0.0	0.0
	APR	2.0	< 0.1	4.0	0.5	2.0	0.8	0.0	0.0
	MAY	1.8	0.4	3.6	0.2	4.4	0.5	0.0	0.0
	JUN	0.6	0.4	1.4	0.4	2.6	0.4	0.0	0.0
	JUL	0.2	0.2	0.2	0.2	1.6	0.2	0.0	0.0
	AUG	0.0	0.0	0.0	0.0	2.2	0.5	0.0	0.0
	SEP	1.0	< 0.1	2.6	0.8	2.8	0.4	0.0	0.0
	OCT	1.6	0.4	2.6	0.7	3.2	0.7	0.0	0.0
	NOV	2.0	0.3	4.6	0.7	4.2	0.6	0.0	0.0
	DEC	2.2	0.2	4.4	0.5	4.0	0.5	0.0	0.0
	JAN 2015	1.6	0.2	3.2	0.6	3.0	0.9	0.0	0.0

**Appendix B4.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

••			Created	Stream		Reference Streams				
	-	Down S	ection	Up Sec	tion	Unmi	ned	Mine-im	pacted	
	-	GE	)	GU	J	LN	1	WE	3	
Metric ( <i>y</i> )	Month	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	
Shannon Index (H)										
	FEB 2014	2.05	0.19	2.19	0.18	2.61	0.10	0.35	0.22	
	MAR	2.23	0.22	2.44	0.06	2.52	0.05	0.69	0.00	
	APR	2.03	0.20	2.40	0.05	2.60	0.11	0.13	0.10	
	MAY	1.68	0.33	1.86	0.19	2.75	0.11	0.92	0.11	
	JUN	1.40	0.22	2.06	0.04	2.33	0.22	0.23	0.18	
	JUL	1.19	0.23	1.50	0.17	1.57	0.24	0.66	0.21	
	AUG	1.09	0.23	1.77	0.09	2.75	0.01	0.69	0.44	
	SEP	1.66	0.29	2.23	0.16	2.91	0.08	0.36	0.16	
	OCT	2.14	0.19	1.83	0.30	2.75	0.07	0.95	0.26	
	NOV	1.98	0.08	2.23	0.13	2.80	0.06	0.53	0.28	
	DEC	1.73	0.13	2.15	0.06	2.82	0.06	0.84	0.24	
	JAN 2015	1.45	0.12	2.15	0.06	2.72	0.07	0.33	0.17	
Hill's										
N <sub>1</sub> Diversity ( $e^{H}$ )	FEB 2014	8.38	1.66	9.45	1.31	13.86	1.29	1.50	0.32	
	MAR	10.07	1.80	11.58	0.76	12.46	0.69	2.00	0.00	
	APR	8.16	1.30	11.09	0.55	13.80	1.35	1.15	0.12	
	MAY	6.46	1.62	6.94	1.25	15.94	1.71	2.56	0.26	
	JUN	4.46	0.99	7.85	0.31	11.11	1.79	1.33	0.26	
	JUL	3.63	0.72	4.74	0.82	5.49	1.60	2.08	0.37	
	AUG	3.27	0.68	5.94	0.54	15.70	0.22	2.50	0.95	
	SEP	6.07	1.43	9.77	1.75	18.51	1.45	1.49	0.22	
	OCT	9.07	1.60	7.36	1.93	15.77	1.06	2.92	0.81	
	NOV	7.36	0.61	9.61	1.27	16.63	1.08	1.96	0.50	
	DEC	5.84	0.69	8.67	0.49	16.90	1.14	2.55	0.51	
	JAN 2015	4.41	0.54	8.68	0.52	15.28	1.12	1.47	0.24	

**Appendix B5.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

			Created S	Stream		Reference Streams				
	_	Down S	ection	Up Sec	tion	Unmi	ned	Mine-im	pacted	
	_	GE	)	GU	ſ	LN	1	WE	3	
Metric $(y)$	Month	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
Simpson Index										
Of Diversity (1-D)	FEB 2014	0.77	0.05	0.81	0.04	0.89	0.02	0.25	0.16	
	MAR	0.81	0.05	0.85	0.01	0.89	0.01	0.50	0.00	
	APR	0.79	0.05	0.86	0.01	0.88	0.01	0.07	0.06	
	MAY	0.65	0.13	0.73	0.05	0.90	0.01	0.56	0.04	
	JUN	0.59	0.08	0.81	0.02	0.81	0.07	0.17	0.13	
	JUL	0.60	0.10	0.65	0.06	0.60	0.08	0.41	0.13	
	AUG	0.55	0.12	0.76	0.03	0.91	< 0.00	0.38	0.24	
	SEP	0.65	0.12	0.82	0.03	0.92	0.01	0.24	0.11	
	OCT	0.81	0.05	0.70	0.10	0.90	0.01	0.56	0.10	
	NOV	0.80	0.02	0.83	0.02	0.91	0.01	0.32	0.17	
	DEC	0.74	0.04	0.81	0.02	0.91	0.01	0.50	0.13	
	JAN 2015	0.64	0.05	0.83	0.01	0.91	0.01	0.24	0.12	
Hill's										
N <sub>2</sub> Diversity $(1/D)$	FEB 2014	5.41	1.33	6.20	0.91	9.33	1.15	1.50	0.32	
• 、 /	MAR	6.25	1.11	6.98	0.69	9.30	0.72	2.00	0.00	
	APR	5.57	0.87	7.30	0.34	8.95	1.03	1.09	0.07	
	MAY	4.37	1.18	4.37	0.82	10.39	0.91	2.34	0.19	
	JUN	2.78	0.51	5.32	0.43	7.24	1.47	1.33	0.26	
	JUL	3.22	0.69	3.38	0.77	3.48	1.27	1.93	0.32	
	AUG	2.67	0.49	4.47	0.59	11.30	0.34	2.50	0.95	
	SEP	4.17	1.04	6.30	1.45	13.72	1.30	1.43	0.23	
	OCT	6.38	1.29	5.00	1.44	10.81	1.17	2.87	0.83	
	NOV	5.18	0.52	6.04	0.72	11.20	1.29	1.92	0.48	
	DEC	4.10	0.53	5.54	0.60	11.67	1.44	2.52	0.52	
	JAN 2015	2.97	0.39	5.87	0.48	11.14	0.78	1.45	0.24	

**Appendix B6.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

		Created Stream				Reference Streams					
		Down Section		Up Se	ction	Unmined		Mine-impacted			
		GD		GU		LM		WB			
Metric $(y)$	Month	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE		
% EPT Index											
(genus)	FEB 2014	23.2	3.8	30.3	1.1	51.0	1.2	0.0	0.0		
	MAR	22.8	1.6	34.5	4.3	47.9	4.0	0.0	0.0		
	APR	28.2	2.4	30.0	0.8	53.2	0.9	0.0	0.0		
	MAY	14.7	2.5	26.4	1.6	54.9	3.6	0.0	0.0		
	JUN	4.6	2.9	9.5	1.7	41.4	2.1	0.0	0.0		
	JUL	3.3	3.3	1.1	1.1	33.1	3.4	0.0	0.0		
	AUG	0.0	0.0	0.0	0.0	35.8	2.9	0.0	0.0		
	SEP	10.9	1.6	12.4	2.7	37.8	1.6	0.0	0.0		
	OCT	11.2	2.2	17.4	5.3	37.9	4.9	0.0	0.0		
	NOV	19.1	1.3	28.2	4.6	43.6	2.0	0.0	0.0		
	DEC	24.2	0.9	28.9	2.5	47.4	1.3	0.0	0.0		
	JAN 2015	28.4	4.2	25.1	5.1	51.1	3.1	0.0	0.0		
% EPT Abundance											
	FEB 2014	42.2	9.8	24.6	7.9	45.2	4.4	0.0	0.0		
	MAR	21.3	5.2	34.1	9.3	46.8	6.8	0.0	0.0		
	APR	34.4	9.7	29.0	7.5	67.0	3.1	0.0	0.0		
	MAY	6.0	5.1	5.9	2.3	62.8	4.8	0.0	0.0		
	JUN	0.4	0.3	1.6	0.5	40.7	8.2	0.0	0.0		
	JUL	1.7	1.7	0.1	0.1	15.0	4.7	0.0	0.0		
	AUG	0.0	0.0	0.0	0.0	37.3	3.1	0.0	0.0		
	SEP	7.5	3.5	7.0	3.0	27.5	3.0	0.0	0.0		
	OCT	9.9	3.3	17.9	10.0	33.0	5.2	0.0	0.0		
	NOV	12.0	3.0	14.0	3.8	48.8	6.7	0.0	0.0		
	DEC	6.8	0.9	15.2	7.3	68.6	3.1	0.0	0.0		
	JAN 2015	7.0	2.5	10.0	1.4	56.7	4.8	0.0	0.0		

**Appendix B7.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

			Created	Stream		Reference Streams					
	-	Down Section		Up Se	ction	Unmined		Mine-impacted			
		GE	)	GU	J	LN	1	WB			
Metric $(y)$	Month	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE		
% Ephemeroptera											
Abundance	FEB 2014	2.9	1.7	8.8	4.6	28.7	2.7	0.0	0.0		
	MAR	1.6	0.8	12.0	2.5	27.1	5.1	0.0	0.0		
	APR	8.1	2.7	8.2	1.8	42.4	2.7	0.0	0.0		
	MAY	0.4	0.2	0.5	0.1	22.8	3.9	0.0	0.0		
	JUN	0.2	0.1	0.1	< 0.1	19.7	4.3	0.0	0.0		
	JUL	0.0	0.0	0.0	0.0	11.2	4.3	0.0	0.0		
	AUG	0.0	0.0	0.0	0.0	13.6	5.3	0.0	0.0		
	SEP	4.3	3.0	0.1	0.1	15.6	3.9	0.0	0.0		
	OCT	0.0	0.0	< 0.1	< 0.1	15.6	3.8	0.0	0.0		
	NOV	4.6	2.8	0.5	0.2	23.7	3.6	0.0	0.0		
	DEC	1.6	0.6	2.5	1.0	38.6	4.3	0.0	0.0		
	JAN 2015	4.7	2.4	6.4	2.0	37.9	5.0	0.0	0.0		
Hilsenhoff Biotic											
Index (genus)	FEB 2014	6.57	0.35	5.75	0.25	4.10	0.27	n/a	n/a		
	MAR	6.48	0.12	5.38	0.33	3.59	0.22	n/a	n/a		
	APR	5.61	0.38	5.48	0.34	3.03	0.14	n/a	n/a		
	MAY	6.65	0.29	6.51	0.12	3.01	0.17	n/a	n/a		
	JUN	6.77	0.07	6.76	0.08	4.58	0.56	n/a	n/a		
	JUL	7.09	0.25	7.20	0.19	5.93	0.35	n/a	n/a		
	AUG	7.53	0.16	6.73	0.25	4.28	0.14	n/a	n/a		
	SEP	6.89	0.09	6.56	0.25	4.79	0.12	n/a	n/a		
	OCT	7.04	0.15	6.09	0.40	4.47	0.13	n/a	n/a		
	NOV	6.60	0.10	6.42	0.15	4.18	0.31	n/a	n/a		
	DEC	6.60	0.04	6.39	0.20	3.34	0.12	n/a	n/a		
	JAN 2015	6.78	0.32	6.42	0.09	3.68	0.25	n/a	n/a		

**Appendix B8.** Metrics used in analysis. Values are averages (n = 5) and standard error (SE).

#### APPENDIX C

#### MONTHLY PERCENT COMPOSITION OF MACROINVERTEBRATE GENERA

#### ABSTRACT

The purpose of Appendix C is to help the reader compare the ecological performance at each stream site with monthly percent composition values for all benthic macroinvertebrate genera represented in the metric calculations (Appendix B).

<u>a appe</u>		actomy cite of aces. V alues		Twooted	Strace			fomore		<u> </u>		
	Eshmany 2014				tion (C		<u>Ke</u>	Unmined (LM)				
reduary 2014						D)	0- N.					
				$\frac{\text{Op Sec}}{\text{D}}$	ction (G	<u>()</u>		& Mine-imp		Jacieu (WB)		
			G	D	G	<u>U</u>		A	V	VB		
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE		
Non-i	nsects											
В	Lymnaeidae	<i>Fossaria</i> sp.	17.0	6.5	3.1	1.7						
S	Sphaeriidae	Pisidium sp.			18.4	9.7						
Н	Naididae		4.5	2.6	0.1	0.1	0.7	0.5				
Insect	ts											
С	Dryopidae	Helichus sp.			0.1	0.1						
	Dytiscidae	Agabus sp.			< 0.1	< 0.1						
	-	<i>Celina</i> sp.										
		Copelatus sp.										
		Desmopachria sp.										
		Hydroporus sp.										
		Laccophilus sp.										
		Neoporus sp.			0.2	0.1						
	Elmidae	Dubiraphia sp.										
		Optioservus sp.	0.1	0.1			0.5	0.3				
		Oulimnius sp.										
		Stenelmis sp.	0.7	0.5	0.8	0.4	0.4	0.4				
	Haliplidae	Haliplus sp.										
		Peltodytes sp.	< 0.1	< 0.1	0.2	0.2						
	Hvdrophilidae	Anacaena sp.			•	•						
	)	Berosus sp.										
		Cymbiodyta sp.										
		Enochrus sp.										
		Helochares sp										
		inclocharcs sp.										

Appendix C1. List of macroinvertebrates. Values are average % composition (n=5) and standard error (SE).

		<u>_</u>	reated	Stream	<u>n</u>	<b><u>Reference Streams</u></b>						
February 2014			Do	wn Sec	tion (G	D)	N	Non-mined (LM)				
			& Up Section (GU)			<u>U)</u>	& Mine-impacted (WB)					
			G	0	G	U		Λ	W	/B		
Order	: Family	Sub-family / Genus	$\overline{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE		
С	Hydrophilidae	Hydrochus sp.										
		Paracymus sp.										
		Tropisternus sp.										
	Psephenidae	<i>Ectopria</i> sp.			0.1	0.1	3.3	0.8				
D	Ceratopogonidae	<i>Alluaudomyia</i> sp.										
		Atrichopogon sp.	0.3	0.2	0.1	0.1						
		<i>Bezzia/Palpomyia</i> sp.	0.2	0.2								
		Ceratopogon sp.					1.2	0.7				
		Culicoides sp.	0.6	0.1	< 0.1	< 0.1						
		<i>Monohelea</i> sp.										
		<i>Dasyhelea</i> sp.										
		<i>Probezzia</i> sp.	0.5	0.3	0.3	0.2	4.4	1.3				
		Sphaeromias sp.	4.4	2.8								
D	Chironomidae	Chironominae	0.4	0.3	0.9	0.3	0.7	0.4				
		<i>Corynoneura</i> sp.	0.1	0.1	0.5	0.2						
		Orthocladiinae	4.9	1.9	7.2	0.7	21.7	4.8				
		Stempellina sp.					3.7	1.1				
		Tanypodinae	6.8	2.9	16.6	1.0	6.6	3.2				
		Tanytarsini (tribe)	1.7	0.8	15.9	6.6	2.1	0.6				
	Culicidae	Anopheles sp.										
		Haemagogus sp.										
	Dixidae	Dixa sp.										
	Dolichopodidae	dolichopodid genus A	0.1	0.1								
	Empididae	empidid genus A										
	I	Chelifera sp.					0.3	0.3				

	February 2014		<u>C</u> Do	reated wn Sec	Stream	<u>Reference Streams</u> Non-mined (LM) & Mine-impacted (WB)				
				Up Sec	tion (G					U)
			G	D	G	U	LN	Л	W	VB
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Empididae	<i>Hemerodromia</i> sp. <i>Neoplasta</i> sp.	0.3	0.2	0.6	0.4				
	Ephydridae	ephydrid genus A Scatella sp.								
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A	2.2	1.7	0.2	0.2				
	Sciomyzidae	Sepedon sp.	0.1	0.1	< 0.1	< 0.1				
	Simuliidae	Prosimulium sp.								
		Simulium sp.	0.6	0.6						
		Stegopterna sp.	3.9	2.1	1.5	0.8	5.3	3.6		
	Stratiomyidae	Caloparyphus sp.			0.2	0.1				
		Nemotelus sp.								
		Stratiomys sp.			0.1	0.1				
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	0.9	0.6	2.0	1.0				
		Tabanus sp.	2.0	1.4						
	Tipulidae	Dicranota sp.					0.7	0.4		
	-	Helius sp.	0.2	0.2	0.1	0.1				
		Hexatoma sp.					1.1	0.5		
		Leptotarsus sp.								
		Limnophila sp.			0.1	0.1	0.4	0.3		
		Limonia sp.	2.8	1.8	0.1	0.1				
		Molophilus sp.					0.3	0.3		
		Ormosia sp. Pedicia sp.	0.2	0.1						

Appendix C1. (cont.)
	February 2014		<u>C</u> Do	Created wn Sec	Stream tion (GI	<u> </u> D)	<u>Re</u> N	ference on-min	e Strea led (LN	a <u>ms</u> A)
			&	Up Sec	tion (Gl	J)	& M	ine-imp	acted	(WB)
			G	D	GU	J	LN	Λ	V	VB
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.1	0.1	0.2	0.2				
		Pseudolimnophila sp.	0.4	0.4	3.9	2.3	0.8	0.8		
		<i>Tipula</i> sp.	1.3	0.3	0.9	0.4	0.4	0.4		
Е	Ameletidae	Ameletus sp.	2.9	1.7	8.7	4.7	2.4	0.4		
	Baetidae	Baetis sp.					0.7	0.5		
		Procloeon sp.								
	Ephemerellidae	Attenella sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.					11.5	3.3		
		<i>Eurylophella</i> sp.			0.1	0.1	1.6	0.8		
	Ephemeridae	Ephemera sp.								
	Heptageniidae	<i>Epeorus</i> sp.					4.8	1.3		
		Maccaffertium sp.					1.2	0.5		
		Stenacron sp.								
	Leptophlebiidae	Habrophlebia sp.								
		Paraleptophlebia sp.					6.6	1.2		
	Siphlonuridae	Siphlonurus sp.								
Μ	Corydalidae	Chauliodes sp.								
		Nigronia sp.								
	Sialidae	Sialis sp.			0.4	0.2				
0	Aeshnidae	Aeshna sp.								
		<i>Boyeria</i> sp.								
	Calopterygidae	<i>Calopteryx</i> sp.								
	Coenagrionidae	Amphiagrion sp.	< 0.1	< 0.1						
	-	Argia sp.			0.2	0.1				

Appendix C1. (cont.)

	February 2014		<u>(</u> Do	Created own Sec	Stream	<u>n</u> D)	]	<b>Referenc</b> Non-mir	<mark>e Strear</mark> ned (LM	<u>ns</u> )
			&	Up Sec	tion (G	U)	&	Mine-imp	pacted (	WB)
			G	D	G	U		LM	W	В
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Ο	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.	< 0.1	< 0.1	0.3	0.2				
	Gomphidae	Stylogomphus sp.	< 0.1	< 0.1			0.4	4 0.3		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		<i>Leucorrhinia</i> sp.								
		Pentala sp.								
		Sympetrum sp.								
Р	Capniidae	Allocapnia sp.	6.5	2.3	5.4	3.7	4.′	7 1.3		
	Chloroperlidae	<i>Haploperla</i> sp.					4.0	0 1.4		
		Sweltsa sp.								
	Leuctridae	Leuctra sp.			0.5	0.3				
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.					1.	0 1.0		
		Prostoia sp.	0.2	0.2						
		<i>Soyedina</i> sp.							25.0	15.8
	Peltoperlidae	<i>Peltoperla</i> sp.								
	Perlidae	Acroneuria sp.					0.4	4 0.3	50.0	31.6
		Eccoptura sp.								
	Perlodidae	<i>Clioperla</i> sp.			< 0.1	< 0.1				
		<i>Diploperla</i> sp.	0.2	0.2	0.3	0.2				
		<i>Isoperla</i> sp.	1.3	0.6	0.4	0.4	0.9	9 0.4		
		Malirekus sp.			0.1	0.1				
		Remenus sp.								
		Yugus sp.							25.0	15.8
Т	Glossosomatidae	Agapetus sp.								

Appendix C1. (cont.)

	February 2014		<u>(</u>	Created	Stream	<u>1</u>	<u>Re</u>	ference	e Strean	<u>ns</u>
	reordary 2014		&	Un See	tion (GI	D)	& Mi	ne-imr	bacted (N	, VB)
			G	D	G	U		<u>/</u>	W	B
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE
Т	Goeridae	Goera sp.								
	Hydropsychidae	Cheumatopsyche sp.					0.5	0.5		
		Diplectrona sp.	0.2	0.2	1.1	0.8	2.0	0.5		
		Hydropsyche sp.			0.3	0.3				
	Hydroptilidae	<i>Hydroptila</i> sp.								
		<i>Oxyethira</i> sp.								
	Lepidostomatidae	Lepidostoma sp.								
	Leptoceridae	<i>Oecetis</i> sp.								
	Limnephilidae	Ironoquia sp.	30.1	11.4	2.5	1.8				
		Pycnopsyche sp.					1.8	0.6		
	Molannidae	<i>Molanna</i> sp.								
	Philopotamidae	<i>Chimarra</i> sp.			3.7	2.4				
		Dolophilodes sp.								
		Wormaldia sp.			< 0.1	< 0.1				
	Phryganidae	Ptilostomis sp.	0.7	0.4	1.4	0.9				
	Polycentropodidae	Polycentropus sp.					0.5	0.4		
	Psychomyiidae	<i>Lype</i> sp.								
	Rhyacophilidae	Rhyacophila sp.					0.5	0.3		
	Thremmatidae	<i>Neophylax</i> sp.	< 0.1	< 0.1						
Total	Richness / EPT Rich	ness and Abundance	41/9	1,832	47/14	1,994	37/17	497	3/3	3
Avera	ge Total Richness ar	nd SE $(n = 5)$	21.4	2.5	24.8	3.4	22.4	2.0	0.6	0.4
Avera	ge EPT Richness an	d SE $(n = 5)$	5.0	0.9	7.6	1.2	11.4	0.9	0.6	0.4
$B = \overline{B}$	asommatophora, C =	= Coleoptera, $D = \overline{\text{Diptera}}$	a, $E = \overline{E}$	phemer	optera, I	H = Hap	lotaxida, 1	M = Me	egalopte	ra,
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $T = $	<b>Frichopt</b>	tera						

Appendix C1. (cont.)

Appe	enaix C2. List of m	acroinvertebrates. Values	are avera	age % (	compos	100n (n=3)	) and star	ndard e	rror(S)	E).
	N 1 2014		<u>_</u>	<u>reated</u>	<u>  Strear</u>	<u>n</u>	<u>Re</u>	terenc	<u>e Strea</u>	<u>ms</u>
	March 2014		Do	wn Sec	ction (G	D)		Unmine	ed (LM	)
			&	Up Sec	tion (G	U)	& M	ine-imp	pacted (	(WB)
			G	D	G	U		Ν	V	VB
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-i	insects									
В	Lymnaeidae	<i>Fossaria</i> sp.	29.5	8.3	7.2	5.9	0.1	0.1		
S	Sphaeriidae	Pisidium sp.			5.7	3.5	0.1	0.1		
Н	Naididae		2.0	0.9	1.4	0.8	0.3	0.3		
Insec	ts									
С	Dryopidae	Helichus sp.					2.9	2.8		
	Dytiscidae	Agabus sp.								
		<i>Celina</i> sp.								
		Copelatus sp.								
		<i>Desmopachria</i> sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	0.1	0.1	0.1	0.1				
	Elmidae	<i>Dubiraphia</i> sp.			< 0.1	< 0.1				
		Optioservus sp.	0.2	0.2	0.9	0.7	0.3	0.3		
		Oulimnius sp.					0.6	0.6		
		Stenelmis sp.	0.6	0.4	0.6	0.4	2.7	1.5		
	Haliplidae	Haliplus sp.								
	-	Peltodytes sp.	0.1	0.1	< 0.1	< 0.1				
	Hydrophilidae	Anacaena sp.								
	•	Berosus sp.								
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.								

Appendix C2. List of macroinvertebrates. Values are average % composition (n=5) and standard error (SE).

	March 2014		<u>(</u> Do	Created own Sec	<b>Strear</b> tion (G	<u>n</u> D)	<u>Re</u> N	ference Ion-min	e Strear led (LM	<u>ns</u> )
			&	Up Sect	tion (G	U)	& M	ine-imp	bacted (V	WB)
			G	D	G	U	LI	M	W	В
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.								
		<i>Tropisternus</i> sp.								
	Psephenidae	<i>Ectopria</i> sp.			0.1	0.1	9.0	3.6		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.	5.3	4.1	0.2	0.2			25.0	15.8
		<i>Bezzia/Palpomyia</i> sp.	1.5	1.5	0.3	0.2			25.0	15.8
		Ceratopogon sp.					1.4	0.9		
		Culicoides sp.	1.2	0.6	0.1	< 0.1				
		<i>Monohelea</i> sp.								
		<i>Dasyhelea</i> sp.								
		<i>Probezzia</i> sp.	0.6	0.3	1.9	1.1	2.6	0.7		
		Sphaeromias sp.	2.9	2.5	0.1	0.1				
D	Chironomidae	Chironominae	0.6	0.2	1.7	0.9	5.0	3.4		
		<i>Corynoneura</i> sp.	0.1	0.1	1.3	0.6				
		Orthocladiinae	3.0	1.0	7.8	1.8	7.8	1.9		
		<i>Stempellina</i> sp.					0.5	0.5		
		Tanypodinae	10.3	2.0	19.3	5.6	1.4	1.1		
		Tanytarsini (tribe)	3.0	1.6	7.4	3.2				
	Culicidae	Anopheles sp.								
		Haemagogus sp.								
	Dixidae	Dixa sp.								
	Dolichopodidae	dolichopodid genus A	< 0.1	< 0.1						
	Empididae	empidid genus A								
	-	<i>Chelifera</i> sp.					0.1	0.1		

Appendix C2. (cont.)

	March 2014		<u>(</u> Do	Created	Strear	<u>n</u> D)	<u>Re</u> N	ference	e Strea ed (LN	<u>ms</u> 1)
			&	Up Sec	tion (G	U)	& M	ine-imp	acted (	WB)
			G	D	G	U		M	W	/B
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
)	Empididae	Hemerodromia sp.	1.2	0.6	0.5	0.2				
		<i>Neoplasta</i> sp.			0.1	0.1				
	Ephydridae	ephydrid genus A								
		<i>Scatella</i> sp.								
	Psychodidae	<i>Pericoma</i> sp.			0.2	0.1				
	Sciaridae	sciarid genus A	2.9	1.4	< 0.1	< 0.1				
	Sciomyzidae	Sepedon sp.	0.2	0.2						
	Simuliidae	Prosimulium sp.					8.5	7.1		
		Simulium sp.								
		<i>Stegopterna</i> sp.	4.7	1.3	2.8	0.6				
	Stratiomyidae	Caloparyphus sp.	0.1	0.1	0.2	0.1				
		Nemotelus sp.								
		Stratiomys sp.	0.1	0.1						
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	2.8	0.9	1.4	0.6				
		Tabanus sp.	0.3	0.2						
	Tipulidae	Dicranota sp.			0.2	0.1				
	-	Helius sp.			0.1	0.1				
		Hexatoma sp.					5.4	1.6		
		<i>Leptotarsus</i> sp.								
		Limnophila sp.			< 0.1	< 0.1	0.1	0.1		
		Limonia sp.	0.9	0.3	0.2	0.1				
		Molophilus sp.					0.5	0.5		
		Ormosia sp.	< 0.1	< 0.1						
		Pedicia sp.								

# Appendix C2. (cont.)

	March 2014		<u>C</u> Dov	<mark>reated</mark> wn Sec	Stream tion (GI	<u> </u> D)	<u>Re</u> N	ference	e Strean ed (LM	<u>ns</u> )
			& U	Up Sec	tion (GU	л́	& Mi	ine-imp	acted (V	WB)
			GI	)	G	Ĵ	LN	A	Ŵ	B
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.1	0.1	0.5	0.2				
		Pseudolimnophila sp.	0.5	0.3	1.6	0.4	0.1	0.1		
		<i>Tipula</i> sp.	3.3	2.1	0.5	0.2	0.1	0.1		
E	Ameletidae	Ameletus sp.	1.6	0.8	11.9	2.5	5.4	2.4		
	Baetidae	Baetis sp.								
		Procloeon sp.								
	Ephemerellidae	Attenella sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.					5.7	3.7		
		<i>Eurylophella</i> sp.			0.1	0.1	0.6	0.6		
	Ephemeridae	<i>Ephemera</i> sp.					0.6	0.6		
	Heptageniidae	<i>Epeorus</i> sp.					9.0	1.7		
		Maccaffertium sp.					0.1	0.1		
		Stenacron sp.								
	Leptophlebiidae	<i>Habrophlebia</i> sp.								
		<i>Paraleptophlebia</i> sp.					5.6	1.2	25.0	15.8
	Siphlonuridae	Siphlonurus sp.								
М	Corydalidae	Chauliodes sp.								
		Nigronia sp.					2.1	0.8		
	Sialidae	<i>Sialis</i> sp.	0.2	0.2	0.8	0.5				
0	Aeshnidae	<i>Aeshna</i> sp.								
		<i>Boyeria</i> sp.								
	Calopterygidae	<i>Calopteryx</i> sp.								
	Coenagrionidae	Amphiagrion sp.								
		<i>Argia</i> sp.	0.1	0.1	0.4	0.3				

Appendix C2. (cont.)

••	March 2014		<u>(</u> De	Created	Strear	<u>n</u> (D)	<u>Re</u> N	ference	e Strear ned (LM	<u>ns</u> )
			&	Up Sec	tion (G	U)	& Mi	ine-imp	bacted (	WB)
			G	D	G	ĴÚ	LN	M	Ŵ	B
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.	0.2	0.1	0.1	0.1				
	Gomphidae	Stylogomphus sp.					1.6	0.9		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		<i>Leucorrhinia</i> sp.								
		<i>Pentala</i> sp.								
		<i>Sympetrum</i> sp.								
Р	Capniidae	<i>Allocapnia</i> sp.	0.5	0.2	0.3	0.3	3.9	1.8		
	Chloroperlidae	<i>Haploperla</i> sp.					5.8	1.9		
		<i>Sweltsa</i> sp.					0.3	0.3		
	Leuctridae	<i>Leuctra</i> sp.			0.4	0.1				
	Nemouridae	Amphinemura sp.	1.1	0.8	6.9	3.6			25.0	15.8
		<i>Ostrocerca</i> sp.				0.4				
		Prostoia sp.	0.2	0.2	0.2	0.1				
	D 1. 1.1	Soyedina sp.			0.0	0.1				
	Peltoperlidae	<i>Peltoperla</i> sp.			0.2	0.1				
	Perlidae	<i>Acroneuria</i> sp.			< 0.1	< 0.1	0.2	0.2		
	D 1 1 1	<i>Eccoptura</i> sp.			0.1	0.1	0.3	0.3		
	Periodidae	Choperia sp.			0.1	0.1				
		<i>Diploperla</i> sp.	2.4	1.0	0.2	0.1	1.0	1.2		
		Isoperia sp.	2.4	1.0	0.6	0.4	1.2	1.2		
		<i>Malirekus</i> sp.	< 0.1	< 0.1	0.3	0.1	1.3	1.1		
		<i>Remenus</i> sp.								
т	Glassasamatidas	<i>Tugus</i> sp.								
1	Giossosomandae	Agupeius sp.								

Appendix C2. (cont.)

	March 2014		<u>C</u> Do	Created	Stream	<u>n</u> D)	<u>Re</u> N	ference	e Strean	<u>15</u> )
			&	Up Sec	tion (G	U)	& Mi	ne-im	bacted (V	, VB)
			G	D	G	Ú	LN	Λ	Ŵ	B
Order	- Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE
Т	Goeridae	<i>Goera</i> sp.								
	Hydropsychidae	Cheumatopsyche sp.			< 0.1	< 0.1				
		Diplectrona sp.	0.3	0.1	1.1	0.5	4.3	2.7		
		Hydropsyche sp.			0.2	0.2				
	Hydroptilidae	Hydroptila sp.								
		<i>Oxyethira</i> sp.								
	Lepidostomatidae	Lepidostoma sp.					0.1	0.1		
	Leptoceridae	<i>Oecetis</i> sp.								
	Limnephilidae	Ironoquia sp.	14.3	6.2	2.7	0.9				
		Pycnopsyche sp.					0.5	0.5		
	Molannidae	<i>Molanna</i> sp.								
	Philopotamidae	<i>Chimarra</i> sp.	0.2	0.2	7.6	6.2				
		Dolophilodes sp.								
		<i>Wormaldia</i> sp.			0.2	0.1				
	Phryganidae	Ptilostomis sp.	0.7	0.5	1.0	0.4				
	Polycentropodidae	Polycentropus sp.					0.8	0.8		
	Psychomyiidae	<i>Lype</i> sp.								
	Rhyacophilidae	<i>Rhyacophila</i> sp.					1.3	0.9		
	Thremmatidae	<i>Neophylax</i> sp.								
Total	Richness / EPT Rich	ness and Abundance	43/10	1,766	55/19	3,054	41/18	290	4/2	4
Avera	ge Total Richness an	nd SE $(n = 5)$	25.6	3.5	33.2	0.8	18.4	4.2	0.8	0.5
Avera	ge EPT Richness an	d SE (n = 5)	5.8	0.9	11.4	1.4	8.4	1.3	0.4	0.2
B = Ba	asommatophora, C =	= Coleoptera, $D =$ Diptera	$\mathbf{a}, E = \mathbf{E}_{\mathbf{I}}$	phemer	optera,	H = Hap	lotaxida, <i>I</i>	M = Me	egaloptei	ra,
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $T = $ T	<b>Frichopt</b>	era						

Appendix C2. (cont.)

	April 2014		<u>(</u> Do	Created	<b>Strear</b> Stion (G	<u>n</u> D)	<u>Re</u>	<b>ference</b> Unmine	e <mark>Strea</mark> d (LM	<u>ms</u> )
	-		&	Up Sec	tion (G	U)	& M	ine-imp	acted (	WB)
			G	D	G	U	LN	M	W	/B
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
Non-in	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	22.1	9.4	3.6	1.8				
S	Sphaeriidae	Pisidium sp.			9.3	5.8	0.2	0.2		
Η	Naididae	_	1.0	0.8	1.8	0.8				
Insect	S									
С	Dryopidae	Helichus sp.					1.0	0.8		
	Dytiscidae	Agabus sp.	0.1	< 0.1	< 0.1	< 0.1				
		<i>Celina</i> sp.								
		Copelatus sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.								
	Elmidae	Dubiraphia sp.	< 0.1	< 0.1						
		Optioservus sp.	0.1	0.1	0.2	0.2	0.2	0.2		
		Oulimnius sp.								
		Stenelmis sp.	1.3	1.0	3.3	1.8	1.9	1.3		
	Haliplidae	Haliplus sp.			< 0.1	< 0.1				
	1	Peltodytes sp.	0.1	0.1	< 0.1	< 0.1				
	Hydrophilidae	Anacaena sp.								
	•	Berosus sp.	< 0.1	< 0.1						
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.								

**Appendix C3.** List of macroinvertebrates. Values are % composition (n = 5) and standard error (SE).

	April 2014		<u>C</u> Do	C <mark>reated</mark> wn Sec	Stream	<u>n</u> D)	<u>Re</u> N	ference	e Strean	<u>ns</u>
			&	Up Sec	tion (G	U)	& Mi	ine-im	bacted (V	, VB)
			G	D	G	Ú	LN	A I	Ŵ	B
Orde	er Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.								
		<i>Tropisternus</i> sp.								
	Psephenidae	<i>Ectopria</i> sp.					9.4	1.3		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.	0.3	0.1	< 0.1	< 0.1				
		<i>Bezzia/Palpomyia</i> sp.			0.3	0.3	0.2	0.2		
		Ceratopogon sp.					1.0	0.6		
		Culicoides sp.	0.4	0.3	0.1	< 0.1				
		Monohelea sp.								
		<i>Dasyhelea</i> sp.								
		<i>Probezzia</i> sp.	0.7	0.3	0.7	0.4	0.3	0.2		
		Sphaeromias sp.	0.4	0.4	0.3	0.1				
D	Chironomidae	Chironominae	1.3	0.2	6.0	2.4	0.3	0.2	4.2	3.2
		Corynoneura sp.	0.1	0.1	2.1	0.7				
		Orthocladiinae	4.4	1.3	7.2	1.9	3.8	0.9		
		Stempellina sp.					0.4	0.4		
		Tanypodinae	22.3	4.8	21.9	2.0	1.6	0.8		
		Tanytarsini (tribe)	3.7	1.6	7.3	1.2	0.4	0.2		
	Culicidae	Anopheles sp.								
		Haemagogus sp.								
	Dixidae	<i>Dixa</i> sp.								
	Dolichopodidae	dolichopodid genus A			< 0.1	< 0.1				
	Empididae	empidid genus A			0.1	0.1				
	-	Chelifera sp.					0.9	0.6		

Appendix C3. (cont.)

	April 2014		<u>C</u> Do	reated wn Sec	Stream tion (G	<u>n</u> D)	<u>Re</u> N	ference on-min	e Strea ed (LN	<u>ms</u> 1)
	1		&	Up Sec	tion (G	Ú	& Mi	ine-imp	acted (	WB)
			G	D	G	Ū	LN	Л	W	/B
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Empididae	Hemerodromia sp.	0.3	0.1	0.2	0.1	0.8	0.8		
		<i>Neoplasta</i> sp.			< 0.1	< 0.1				
	Ephydridae	ephydrid genus A								
		Scatella sp.								
	Psychodidae	Pericoma sp.			< 0.1	< 0.1				
	Sciaridae	sciarid genus A	0.4	0.2						
	Sciomyzidae	Sepedon sp.								
	Simuliidae	Prosimulium sp.					1.3	0.4		
		Simulium sp.	1.1	0.7	0.3	0.1	0.5	0.5		
		Stegopterna sp.	0.1	0.1	1.7	0.7	0.5	0.3		
	Stratiomyidae	Caloparyphus sp.	0.1	0.1	< 0.1	< 0.1				
		Nemotelus sp.			< 0.1	< 0.1				
		Stratiomys sp.								
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	2.5	0.5	1.1	0.3				
		Tabanus sp.	0.2	0.2	< 0.1	< 0.1				
	Tipulidae	Dicranota sp.					0.2	0.2		
	-	Helius sp.	0.1	0.1						
		Hexatoma sp.					3.6	1.7		
		Leptotarsus sp.								
		<i>Limnophila</i> sp.			< 0.1	< 0.1	2.1	0.9		
		Limonia sp.								
		Molophilus sp.								
		Ormosia sp.	0.1	0.1						
		<i>Pedicia</i> sp.			< 0.1	< 0.1				

Appendix C3. (cont.)

	April 2014		<u>(</u> Do	C <b>reated</b> own Sec	Stream tion (GI	<u> </u> ))	<u>Re</u> N	ference	e Strea ed (LN	<u>ms</u> (1)
	1		&	Up Sec	tion (GU	Ĵ)	& M	ine-imp	acted (	(WB)
			G	D	G	Ú	LN	M	V	VB
Order	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	< 0.1	< 0.1	0.3	0.1				
		Pseudolimnophila sp.			2.0	0.9	1.3	0.8		
		<i>Tipula</i> sp.	0.6	0.3	0.5	0.2				
Е	Ameletidae	Ameletus sp.	7.9	2.6	8.1	1.8	2.8	0.7		
	Baetidae	Baetis sp.	0.2	0.1						
		Procloeon sp.								
	Ephemerellidae	<i>Attenella</i> sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.					24.7	3.4		
		<i>Eurylophella</i> sp.			0.1	0.1				
	Ephemeridae	<i>Ephemera</i> sp.					0.3	0.2		
	Heptageniidae	<i>Epeorus</i> sp.					12.3	3.3		
		Maccaffertium sp.					1.0	0.4		
		Stenacron sp.								
	Leptophlebiidae	<i>Habrophlebia</i> sp.								
		Paraleptophlebia sp.					1.4	0.6		
	Siphlonuridae	Siphlonurus sp.								
Μ	Corydalidae	Chauliodes sp.								
		Nigronia sp.					0.4	0.4		
	Sialidae	Sialis sp.			0.1	0.1				
0	Aeshnidae	<i>Aeshna</i> sp.								
		<i>Boyeria</i> sp.								
	Calopterygidae	Calopteryx sp.								
	Coenagrionidae	Amphiagrion sp.								
		<i>Argia</i> sp.	1.7	1.4	0.3	0.1				

Appendix C3. (cont.)

			(	Created	Stream	<u>n</u>	Re	ference	e Strear	<u>ns</u>
	April 2014		Do	own Sec	tion (G	D)	Ν	on-min	ed (LM	)
			&	Up Sec	tion (G	U)	& Mi	ine-imp	acted (	WB)
			G	D	G	U	LN	Л	W	В
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.			0.1	0.1				
	Gomphidae	Stylogomphus sp.			< 0.1	< 0.1	0.6	0.5		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.			< 0.1	< 0.1				
		<i>Leucorrhinia</i> sp.	0.1	< 0.1						
		Pentala sp.								
		Sympetrum sp.								
Р	Capniidae	Allocapnia sp.	< 0.1	< 0.1	0.7	0.5	0.7	0.5		
	Chloroperlidae	<i>Haploperla</i> sp.					3.0	0.6		
		Sweltsa sp.					0.7	0.5		
	Leuctridae	Leuctra sp.			0.4	0.2	0.6	0.6		
	Nemouridae	Amphinemura sp.	12.1	5.3	15.4	4.5	9.4	1.1	62.5	24.4
		Ostrocerca sp.					1.0	0.8		
		Prostoia sp.								
		<i>Soyedina</i> sp.								
	Peltoperlidae	<i>Peltoperla</i> sp.			< 0.1	< 0.1	0.2	0.2		
	Perlidae	Acroneuria sp.					1.1	0.6		
		<i>Eccoptura</i> sp.					2.5	1.1		
	Perlodidae	<i>Clioperla</i> sp.								
		<i>Diploperla</i> sp.			0.2	0.1				
		<i>Isoperla</i> sp.	7.9	5.1	1.4	0.2	0.3	0.2		
		Malirekus sp.	0.1	0.1	0.1	0.1	1.0	0.4		
		Remenus sp.								
		Yugus sp.								
Т	Glossosomatidae	Agapetus sp.								

Appendix C3. (cont.)

	April 2014		<u>(</u>	Created	Stream	<u>n</u>	<u>Re</u>	ference	e Strean	<u>ns</u>
	April 2014		D( &	Un Sec	tion (G	D) U)	• IN & Mi	ine-imr	acted (A	) VB)
		-	G	D	G	U		Л	W	B
Order	- Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
Т	Goeridae	<i>Goera</i> sp.								
	Hydropsychidae	Cheumatopsyche sp.								
		Diplectrona sp.			0.7	0.4	1.8	0.7	33.3	25.8
		Hydropsyche sp.			< 0.1	< 0.1				
	Hydroptilidae	Hydroptila sp.					0.2	0.2		
		<i>Oxyethira</i> sp.								
	Lepidostomatidae	Lepidostoma sp.								
	Leptoceridae	Oecetis sp.								
	Limnephilidae	Ironoquia sp.	5.7	1.8	0.5	0.1				
		Pycnopsyche sp.					0.1	0.1		
	Molannidae	Molanna sp.								
	Philopotamidae	<i>Chimarra</i> sp.			0.8	0.5				
	-	Dolophilodes sp.								
		Wormaldia sp.								
	Phryganidae	Ptilostomis sp.	0.4	0.1	0.5	0.1				
	Polycentropodidae	Polycentropus sp.					0.9	0.6		
	Psychomyiidae	<i>Lype</i> sp.								
	Rhyacophilidae	Rhyacophila sp.			< 0.1	< 0.1	1.2	0.7		
	Thremmatidae	<i>Neophylax</i> sp.			< 0.1	< 0.1				
Total	Richness / EPT Rich	ness	38/8	2,894	55/16	5,650	46/22	499	3/2	11
Avera	ge Total Richness ar	nd SE $(n = 5)$	21.2	2.2	33.4	0.8	23.8	2.9	0.8	0.4
Avera	ge EPT Richness and	d SE(n=5)	5.8	0.4	10.0	0.3	12.6	1.5	0.6	0.2
B = Ba	asommatophora, C =	Coleoptera, D = Diptera	E = E	phemer	optera,	H = Hap	lotaxida, 1	M = Me	galopte	ra,
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $\overline{T} = $ T	richop	tera						

Appendix C3. (cont.)

Appe	endix C4. List of m	acroinvertebrates. Values	are aver	age % (	compos	ition (n = :	5) and st $($	andard	error (	SE).
			<u>C</u>	reated	Stream	<u>n</u>	Re	ferenc	<u>e Strea</u>	<u>ims</u>
	May 2014		Do	wn Sec	ction (G	D)	١	Unmine	ed (LM	.)
			&	Up Sec	tion (G	U)	& M	ine-imp	pacted	(WB)
			G	D	G	U	LI	M	V	VB
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-	insects									
В	Lymnaeidae	<i>Fossaria</i> sp.	28.6	15.7	4.0	2.6				
S	Sphaeriidae	Pisidium sp.			11.7	8.2	0.1	0.1		
Η	Naididae		1.4	1.0	1.0	0.5	0.1	0.1		
Insec	ts									
С	Dryopidae	Helichus sp.			0.1	< 0.1				
	Dytiscidae	Agabus sp.	0.4	0.2						
		<i>Celina</i> sp.								
		Copelatus sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	0.5	0.3	0.1	0.1				
	Elmidae	<i>Dubiraphia</i> sp.			0.1	0.1				
		Optioservus sp.	0.1	0.1			0.7	0.3		
		Oulimnius sp.					0.4	0.2		
		Stenelmis sp.	0.9	0.9	1.7	0.9	0.9	0.2		
	Haliplidae	<i>Haliplus</i> sp.								
		Peltodytes sp.	2.7	1.3	0.6	0.4				
	Hydrophilidae	Anacaena sp.								
		Berosus sp.	0.2	0.2						
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.			< 0.1	< 0.1				
		Helochares sp.								

**Appendix C4.** List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

			(	Created	Stream	<u>n</u>	]	Reference	e Strean	<u>ns</u>
	May 2014		Do	own Sec	ction (G	D)		Non-mir	ned (LM)	)
			&	Up Sec	tion (G	U)	&	Mine-imp	pacted (V	VB)
			G	D	G	U		LM	W	B
Order	: Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.			< 0.1	< 0.1				
		Tropisternus sp.	0.2	0.1						
	Psephenidae	<i>Ectopria</i> sp.					2.4	4 0.9		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.								
		<i>Bezzia/Palpomyia</i> sp.	< 0.1	< 0.1	< 0.1	< 0.1	0.	1 0.1	0.6	0.6
		Ceratopogon sp.								
		Culicoides sp.	0.1	0.1						
		<i>Monohelea</i> sp.								
		<i>Dasyhelea</i> sp.								
		<i>Probezzia</i> sp.	1.8	1.0	1.6	0.6	1.2	2 0.3		
		Sphaeromias sp.	0.8	0.2	0.6	0.3				
D	Chironomidae	Chironominae	4.5	1.5	31.7	8.4	0.	1 0.1	3.8	3.8
		Corynoneura sp.	0.2	< 0.1	2.2	0.9				
		Orthocladiinae	10.5	5.4	3.4	1.1	2.	9 1.0	54.7	3.7
		Stempellina sp.					1.0	0.3		
		Tanypodinae	27.4	10.9	25.5	6.2	3.0	6 0.9	10.3	6.5
		Tanytarsini (tribe)	5.4	2.2	5.6	2.6	0.2	2 0.2		
	Culicidae	Anopheles sp.	< 0.1	< 0.1	< 0.1	< 0.1				
		Haemagogus sp.								
	Dixidae	<i>Dixa</i> sp.								
	Dolichopodidae	dolichopodid genus A								
	Empididae	empidid genus A								
		<i>Chelifera</i> sp.					3.	1 0.7		

# Appendix C4. (cont.)

	May 2014		<u>(</u>	Created	Stream	<u>n</u>	Re	eference	e Strean	<u>ns</u>
	Way 2014		DC &	Un Sec	tion (G	D) U)	& M	ine-im	pacted (N	) WB)
			G	D	G	U		M	W	B
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Empididae	<i>Hemerodromia</i> sp.	0.1	0.1	< 0.1	< 0.1	0.1	0.1	10.0	10.0
	Ephydridae	ephydrid genus A Scatella sp.							10.0	10.0
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A								
	Sciomyzidae	Sepedon sp.	0.1	0.1	0.2	0.2				
	Simuliidae	Prosimulium sp.								
		<i>Simulium</i> sp.	1.4	1.2	0.7	0.4	0.1	0.1		
		<i>Stegopterna</i> sp.								
	Stratiomyidae	Caloparyphus sp.	0.1	< 0.1						
		Nemotelus sp.								
		Stratiomys sp.			< 0.1	< 0.1				
	Syrphidae	<i>Eristalis</i> sp.								
	Tabanidae	Chrysops sp.	2.7	1.3	0.9	0.4				
		<i>Tabanus</i> sp.	< 0.1	< 0.1	< 0.1	< 0.1				
	Tipulidae	Dicranota sp.					0.7	0.3		
		<i>Helius</i> sp.								
		Hexatoma sp.					2.6	1.0		
		Leptotarsus sp.								
		<i>Limnophila</i> sp.			< 0.1	< 0.1	15.4	3.1	5.0	5.0
		<i>Limonia</i> sp.								
		Molophilus sp.								
		Ormosia sp.								
		<i>Pedicia</i> sp.								

# Appendix C4. (cont.)

	Mary 2014			Created	Stream	<u>n</u>	Re	<u>ference</u>	Stream	<u>ns</u>
	May 2014		DC	own Sec	tion (G	D)		on-min	ed (LM	) VD)
			<u>&amp;</u>	Up Sec	tion (G	<u>U)</u>	<u>&amp; M</u>	ine-imp	acted (V	<u>wB)</u>
~ 1			<u> </u>	D ar	G	U ar		A ar		B
Order	Family	Sub-family / Genus	x	SE	x	SE	x	SE	$\overline{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.1	0.1	0.2	0.1	0.2	0.2		
		<i>Pseudolimnophila</i> sp.	0.1	0.1	0.9	0.8	0.6	0.4		
		<i>Tipula</i> sp.	0.1	0.1	0.2	0.1	0.1	0.1		
E	Ameletidae	Ameletus sp.			0.4	0.1	0.2	0.2		
	Baetidae	<i>Baetis</i> sp.	0.3	0.1	< 0.1	< 0.1	4.4	0.3	6.7	6.7
		Procloeon sp.								
	Ephemerellidae	<i>Attenella</i> sp.								
		<i>Drunella</i> sp.					0.8	0.3		
		<i>Ephemerella</i> sp.					3.4	0.6	5.0	5.0
		<i>Eurylophella</i> sp.	0.1	0.1						
	Ephemeridae	<i>Ephemera</i> sp.					0.3	0.2		
	Heptageniidae	<i>Epeorus</i> sp.					1.6	0.9		
		<i>Maccaffertium</i> sp.					0.2	0.2		
		Stenacron sp.					0.4	0.3		
	Leptophlebiidae	Habrophlebia sp.								
		Paraleptophlebia sp.					11.4	3.7		
	Siphlonuridae	Siphlonurus sp.			0.1	0.1				
М	Corydalidae	<i>Chauliodes</i> sp.			< 0.1	< 0.1				
	2	Nigronia sp.					0.1	0.1		
	Sialidae	Sialis sp.								
0	Aeshnidae	Aeshna sp.	< 0.1	< 0.1	< 0.1	< 0.1				
		Boyeria sp.								
	Calopterygidae	Caloptervx sp.	0.1	0.1	0.1	0.1				
	Coenagrionidae	Amphiagrion sp.	0.3	0.1						
	0	Argia sp.	0.2	0.2	0.2	0.1				

Appendix C4. (cont.)

	May 2014		( D	Created	Stream	<u>n</u>	Re	ference	Strean	<u>ns</u>
	May 2014		DC 8-	own Sec	tion (G	D)	IN & Mi	on-mine	ed (LM)	) VD)
			$\frac{\alpha}{C}$	Up Sec		$\frac{U}{U}$		me-mp	acted (V	<u>vd)</u> D
0 1	г 1									B
Orde	r Family	Sub-family / Genus	x	SE	x	SE	X	SE	x	SE
0	Coenagrionidae	Enallagma sp.	0.1	0.1	<b>•</b> •	0.1				
	Cordulegastridae	Cordulegaster sp.	0.1	0.1	0.2	0.1				
	Gomphidae	Stylogomphus sp.					0.3	0.1		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		<i>Leucorrhinia</i> sp.	2.8	2.3						
		<i>Pentala</i> sp.								
		<i>Sympetrum</i> sp.	< 0.1	< 0.1	0.4	0.2				
Р	Capniidae	<i>Allocapnia</i> sp.								
	Chloroperlidae	<i>Haploperla</i> sp.					3.6	0.6		
		<i>Sweltsa</i> sp.								
	Leuctridae	Leuctra sp.			< 0.1	< 0.1	14.7	3.2		
	Nemouridae	Amphinemura sp.	4.4	4.4	2.2	1.1	1.2	0.5	2.0	2.0
		<i>Ostrocerca</i> sp.								
		Prostoia sp.								
		Soyedina sp.								
	Peltoperlidae	Peltoperla sp.					0.2	0.2		
	Perlidae	Acroneuria sp.					0.5	0.2		
		<i>Eccoptura</i> sp.			0.1	0.1	0.4	0.2		
	Perlodidae	Clioperla sp.								
		Diploperla sp.			< 0.1	< 0.1				
		Isoperla sp.	0.7	0.6	0.9	0.8				
		Malirekus sp.					0.8	0.3		
		Remenus sp.					0.5	0.3		
		Yugus sp.					0.0	0.0		
Т	Glossosomatidae	Aganetus sn					7.9	1.4		
-										

Appendix C4. (cont.)

	May 2014		( De	Created	Stream	<u>n</u> D)	<u>Re</u> N	ference	e Strean	<u>15</u>
	101ay 2014		&	Un Sec	tion (G	U)	& M	ine-imr	pacted (V	/ VB)
			G	D	G	U		A N	W	B
Order	- Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE
Т	Goeridae	Goera sp.								
	Hydropsychidae	<i>Cheumatopsyche</i> sp.								
		Diplectrona sp.	0.1	0.1	0.2	0.2	0.7	0.4	2.0	2.0
		Hydropsyche sp.								
	Hydroptilidae	<i>Hydroptila</i> sp.	< 0.1	< 0.1	1.4	0.6				
	•	Oxyethira sp.			< 0.1	< 0.1				
	Lepidostomatidae	Lepidostoma sp.					0.3	0.2		
	Leptoceridae	Oecetis sp.	0.2	0.1						
	Limnephilidae	Ironoquia sp.	0.2	0.1	0.3	< 0.1				
	-	Pycnopsyche sp.					0.1	0.1		
	Molannidae	Molanna sp.								
	Philopotamidae	<i>Chimarra</i> sp.			0.2	0.2				
	-	Dolophilodes sp.					0.5	0.5		
		Wormaldia sp.					6.1	2.7		
	Phryganidae	Ptilostomis sp.	0.1	< 0.1	0.1	< 0.1				
	Polycentropodidae	Polycentropus sp.					2.2	0.5		
	Psychomyiidae	<i>Lype</i> sp.								
	Rhyacophilidae	Rhyacophila sp.					0.3	0.3		
	Thremmatidae	<i>Neophylax</i> sp.								
Total	Richness / EPT Rich	ness and Abundance	44/9	4,091	49/14	4,009	49/25	778	10/4	51
Avera	ge Total Richness ar	nd SE $(n = 5)$	22.8	1.6	27.2	2.5	28.0	3.2	3.0	0.4
Avera	ge EPT Richness an	d SE $(n = 5)$	3.4	0.7	7.2	0.8	15.0	1.1	0.8	0.4
B = Ba	asommatophora, C =	= Coleoptera, $D = $ Dipter	a, $\overline{E} = E$	phemer	optera,	H = Hap	olotaxida, I	M = Me	egalopter	a,
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $T = T$	<b>Frichop</b>	tera		_				

Appendix C4. (cont.)

Appe	ndix C5. List of m	acroinvertebrates. Values	are aver	age % (	composi	ition (n =	5) and st	andard	error (S	SE).
			<u>(</u>	Created	l Strean	<u>n</u>	Re	ferenc	<u>e Strea</u>	<u>ms</u>
	June 2014		Do	own Sec	ction (G	D)	1	Unmine	ed (LM	)
			&	Up Sec	tion (G	U)	& M	ine-imp	pacted (	(WB)
			G	D	G	U		Ν	W	VB
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-i	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	39.2	14.6	22.9	7.1	0.1	0.1		
S	Sphaeriidae	Pisidium sp.			17.9	6.5	0.1	0.1		
Η	Naididae		1.9	1.1	3.7	2.5	1.1	0.3		
Insect	ts									
С	Dryopidae	Helichus sp.			0.6	0.3	0.9	0.4		
	Dytiscidae	Agabus sp.	0.1	0.1	0.1	0.1				
		<i>Celina</i> sp.								
		Copelatus sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.	< 0.1	< 0.1						
		Neoporus sp.	0.3	0.1	0.1	< 0.1				
	Elmidae	<i>Dubiraphia</i> sp.			< 0.1	< 0.1				
		Optioservus sp.	< 0.1	< 0.1	0.1	0.1	1.1	0.4		
		Oulimnius sp.					0.2	0.2		
		Stenelmis sp.	0.5	0.4	2.3	1.5	2.4	0.9		
	Haliplidae	<i>Haliplus</i> sp.			< 0.1	< 0.1				
		Peltodytes sp.	0.1	0.1	0.1	0.1				
	Hydrophilidae	Anacaena sp.	0.1	0.1	2.6	1.6				
		Berosus sp.	0.3	0.2	0.1	0.1				
		<i>Cymbiodyta</i> sp.			< 0.1	< 0.1				
		Enochrus sp.								
		Helochares sp.								

**Appendix C5.** List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

	June 2014		<u>(</u> Do	Created	Stream	<u>n</u> D)	<u>Re</u> N	e <b>ference</b> Ion-min	e Strear ed (LM	<u>ns</u> )
			&	Up Sec	tion (G	U)	& M	ine-imp	acted (	WB)
			G	D	G	U	L	М	W	В
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.	0.1	0.1						
		Tropisternus sp.	0.7	0.3	0.1	0.1				
	Psephenidae	<i>Ectopria</i> sp.	< 0.1	< 0.1			3.0	0.8		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.			< 0.1	< 0.1	0.2	0.2		
		<i>Bezzia/Palpomyia</i> sp.					0.2	0.2	16.7	12.9
		Ceratopogon sp.								
		Culicoides sp.	0.2	0.1	0.5	0.2				
		<i>Monohelea</i> sp.								
		<i>Dasyhelea</i> sp.			< 0.1	< 0.1				
		<i>Probezzia</i> sp.	0.3	0.2	0.8	0.3	2.4	0.5		
		Sphaeromias sp.	0.4	0.4	0.4	0.3				
D	Chironomidae	Chironominae	3.5	1.0	11.1	2.0	23.0	11.3	33.3	25.8
		Corynoneura sp.			0.1	0.1				
		Orthocladiinae	2.3	0.9	3.0	2.1	0.5	0.3	50.0	22.4
		<i>Stempellina</i> sp.								
		Tanypodinae	35.3	9.2	17.7	1.7	11.8	1.5		
		Tanytarsini (tribe)	7.0	3.7	9.2	3.9	3.1	0.9		
	Culicidae	Anopheles sp.	< 0.1	< 0.1	0.3	0.1				
		Haemagogus sp.								
	Dixidae	<i>Dixa</i> sp.					0.3	0.2		
	Dolichopodidae Empididae	dolichopodid genus A empidid genus A								
	r	Chelifera sp.					1.1	0.8		

# Appendix C5. (cont.)

	June 2014		<u>(</u> De	Created	<b>Strean</b> tion (G	<u>n</u> D)	<u>Re</u> N	ference	e <u>Strea</u> ed (LN	<u>ms</u> (1)
			&	Up Sec	tion (G	U)	& M	ine-imp	acted (	(WB)
			G	D	G	U	LI	M	V	VB
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Empididae	Hemerodromia sp.	0.3	0.3	0.4	0.2				
		Neoplasta sp.								
	Ephydridae	ephydrid genus A								
		<i>Scatella</i> sp.			< 0.1	< 0.1				
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A								
	Sciomyzidae	Sepedon sp.	< 0.1	< 0.1	0.3	0.2				
	Simuliidae	Prosimulium sp.								
		<i>Simulium</i> sp.	0.3	0.3	0.7	0.4				
		Stegopterna sp.								
	Stratiomyidae	Caloparyphus sp.			< 0.1	< 0.1				
		Nemotelus sp.								
		Stratiomys sp.	0.1	0.1	< 0.1	< 0.1				
	Syrphidae	<i>Eristalis</i> sp.								
	Tabanidae	Chrysops sp.	1.8	1.1	0.6	0.3	0.1	0.1		
		<i>Tabanus</i> sp.	< 0.1	< 0.1	0.1	0.1				
	Tipulidae	Dicranota sp.					0.3	0.2		
		Helius sp.								
		Hexatoma sp.					2.4	1.7		
		Leptotarsus sp.								
		<i>Limnophila</i> sp.					0.2	0.2<		
		<i>Limonia</i> sp.								
		Molophilus sp.								
		Ormosia sp.								
		Pedicia sp.								

# Appendix C5. (cont.)

	June 2014		<u>(</u> Do	C <b>reated</b> own Sec	Stream tion (G	<u>n</u> D)	<u>Re</u> N	e <mark>ference</mark> Ion-min	e Strea led (LN	<u>ms</u> (1)
			&	Up Sec	tion (G	U)	& M	ine-imp	oacted (	(WB)
			G	D	G	U	LN	M	V	VB
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.			< 0.1	< 0.1				
		Pseudolimnophila sp.			0.1	0.1	0.2	0.2		
		<i>Tipula</i> sp.	0.1	0.1						
E	Ameletidae	Ameletus sp.								
	Baetidae	<i>Baetis</i> sp.	0.2	0.1	0.1	< 0.1				
		Procloeon sp.					3.9	2.1		
	Ephemerellidae	<i>Attenella</i> sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.								
		<i>Eurylophella</i> sp.					0.6	0.3		
	Ephemeridae	Ephemera sp.					2.0	0.9		
	Heptageniidae	<i>Epeorus</i> sp.								
		<i>Maccaffertium</i> sp.								
		Stenacron sp.					7.0	3.0		
	Leptophlebiidae	Habrophlebia sp.					0.2	0.1		
		Paraleptophlebia sp.					6.0	1.5		
	Siphlonuridae	Siphlonurus sp.								
Μ	Corydalidae	Chauliodes sp.								
	•	Nigronia sp.								
	Sialidae	Sialis sp.	< 0.1	< 0.1	1.1	0.6	0.2	0.2		
0	Aeshnidae	Aeshna sp.								
		<i>Boyeria</i> sp.	< 0.1	< 0.1	0.1	< 0.1	0.1	0.1		
	Calopterygidae	<i>Calopteryx</i> sp.			< 0.1	< 0.1				
	Coenagrionidae	Amphiagrion sp.								
	-	Argia sp.	0.5	0.2	0.3	0.2				

Appendix C5. (cont.)

<u></u>				7	64		Р	<b>6</b>	C4	
	Law - 2014		<u> </u>	<u>reated</u>	Stream		<u>Ke</u>	ierence	e strea	<u>ms</u>
	June 2014		Do	wn Sec	tion (G	D)	N	on-min	ed (LIV	I)
			<u>&amp;</u>	Up Sec	tion (G	<u>U)</u>	& M:	ine-imp	acted (	<u>WB)</u>
			G	D	G	U		Λ	N	/B
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.	< 0.1	< 0.1	0.2	0.1	0.2	0.2		
	Gomphidae	Stylogomphus sp.	0.3	0.3	0.6	0.4	4.3	1.5		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		<i>Leucorrhinia</i> sp.								
		Pentala sp.								
		<i>Sympetrum</i> sp.	3.5	1.9	< 0.1	< 0.1				
Р	Capniidae	Allocapnia sp.								
	Chloroperlidae	<i>Haploperla</i> sp.					0.7	0.6		
		Sweltsa sp.								
	Leuctridae	Leuctra sp.					15.2	7.0		
	Nemouridae	Amphinemura sp.	< 0.1	< 0.1	0.2	0.1				
		Ostrocerca sp.								
		Prostoia sp.								
		<i>Soyedina</i> sp.								
	Peltoperlidae	Peltoperla sp.					0.1	0.1		
	Perlidae	Acroneuria sp.					0.4	0.2		
		<i>Eccoptura</i> sp.					1.3	0.8		
	Perlodidae	<i>Clioperla</i> sp.								
		Diploperla sp.								
		Isoperla sp.					0.1	0.1		
		Malirekus sp.								
		Remenus sp.								
		Yugus sp.								
Т	Glossosomatidae	Agapetus sp.					0.1	0.1		

Appendix C5. (cont.)

	June 2014		<u>(</u>	Created	Stream	<u>n</u>	Re	eference	e Stream	<u>15</u>
	June 2014		&	Un Sec	tion (G	U)	& M	line-imr	acted (V	VB)
			G	D	G	iU		M	W	B
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
Т	Goeridae	Goera sp.								
	Hydropsychidae	Cheumatopsyche sp.	0.1	0.1						
		Diplectrona sp.	0.1	0.1	0.7	0.3	0.3	0.3		
		Hydropsyche sp.								
	Hydroptilidae	Hydroptila sp.	< 0.1	< 0.1	0.7	0.3				
		<i>Oxyethira</i> sp.								
	Lepidostomatidae	Lepidostoma sp.					0.6	0.3		
	Leptoceridae	Oecetis sp.								
	Limnephilidae	Ironoquia sp.								
		Pycnopsyche sp.					1.1	0.5		
	Molannidae	Molanna sp.					0.3	0.3		
	Philopotamidae	<i>Chimarra</i> sp.								
		Dolophilodes sp.					0.2	0.2		
		<i>Wormaldia</i> sp.					0.3	0.3		
	Phryganidae	Ptilostomis sp.								
	Polycentropodidae	Polycentropus sp.					0.4	0.4		
	Psychomyiidae	<i>Lype</i> sp.								
	Rhyacophilidae	<i>Rhyacophila</i> sp.								
	Thremmatidae	<i>Neophylax</i> sp.								
Total 1	Richness / EPT Rich	ness and Abundance	39/5	3,041	47/4	3,099	46/20	1,263	3/0	6
Avera	ge Total Richness ar	nd SE $(n = 5)$	18.2	3.7	24.2	2.5	23.6	1.4	0.8	0.4
Avera	ge EPT Richness an	d SE(n=5)	1.2	0.7	2.2	0.4	9.8	0.9	0.0	0.0
B = Ba	asommatophora, C =	Coleoptera, $D = Diptera$	a, $\overline{E} = E$	phemer	optera,	H = Hap	lotaxida,	M = Me	egalopter	a,
O = O	donata, $P = Plecopte$	era, $S =$ Sphaeriida, $T = T$	Frichopt	tera						

Appendix C5. (cont.)

			<u>C</u>	reated	Stream	<u>1</u>	Re	ference	Strea	ms
	July 2014		Do	wn Sec	tion (Gl	D)	τ	Unmine	d (LM	)
			&	Up Sec	tion (Gl	J)	& M:	ine-imp	acted (	WB)
			Gl	D	G	U	LN	N	W	/B
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-i	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	28.5	12.1	38.5	12.0	0.2	0.2		
S	Sphaeriidae	Pisidium sp.			17.5	9.5				
Н	Naididae		7.5	7.5	14.4	8.6	1.4	0.8		
Insect	ts									
С	Dryopidae	Helichus sp.			0.3	0.2	1.1	0.5		
	Dytiscidae	Agabus sp.								
		<i>Celina</i> sp.								
		Copelatus sp.								
		<i>Desmopachria</i> sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	3.3	3.3						
	Elmidae	<i>Dubiraphia</i> sp.			0.1	0.1				
		<i>Optioservus</i> sp.			0.1	0.1				
		<i>Oulimnius</i> sp.								
		Stenelmis sp.	8.3	5.2	0.8	0.5	2.3	2.0		
	Haliplidae	Haliplus sp.								
		Peltodytes sp.								
	Hydrophilidae	Anacaena sp.			0.2	0.1				
		Berosus sp.								
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.			< 0.1	< 0.1				

Appendix C6. List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

				reated	Stream	<u>1</u>	Re	ference	e Strear	<u>ns</u>
	July 2014		Do	wn Sec	tion (G	D)	N	on-min	ed (LM	.) (1)
			&	Up Sec	tion (Gl	J)	<u>&amp; M</u>	ine-imp	acted (	WB)
			G	D	G	U		Λ	W	В
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.	3.3	3.3	0.1	0.1				
		Paracymus sp.	18.1	15.5	1.1	0.7				
		<i>Tropisternus</i> sp.	2.5	2.5	0.1	0.1				
	Psephenidae	<i>Ectopria</i> sp.					4.8	2.8		
D	Ceratopogonidae	<i>Alluaudomyia</i> sp.								
		Atrichopogon sp.								
		<i>Bezzia/Palpomyia</i> sp.					0.2	0.2		
		Ceratopogon sp.								
		Culicoides sp.			0.9	0.4	0.6	0.3		
		<i>Monohelea</i> sp.								
		<i>Dasyhelea</i> sp.								
		<i>Probezzia</i> sp.	0.5	0.5	0.3	0.3	0.5	0.3		
		Sphaeromias sp.								
D	Chironomidae	Chironominae			4.3	2.8	57.9	10.1	5.0	4.5
		Corynoneura sp.								
		Orthocladiinae					1.0	0.6	23.3	13.0
		Stempellina sp.								
		Tanypodinae			3.5	2.1	4.5	1.0		
		Tanytarsini (tribe)			0.5	0.5	2.8	0.8		
	Culicidae	Anopheles sp.								
		Haemagogus sp.								
	Dixidae	<i>Dixa</i> sp.								
	Dolichopodidae	dolichopodid genus A								
	Empididae	empidid genus A								
	-	<i>Chelifera</i> sp.								

### Appendix C6. (cont.)

	July 2014		<u>C</u> Do	reated wn Sec	Strean tion (Gl	<u>1</u> D)	Re	e <mark>ferenco</mark> Jon-min	e <mark>Strea</mark> ied (LN	<u>ms</u> 1)
	j		&	Up Sect	tion (GU	J)	& M	line-imr	bacted (	WB)
			G	D	G	Ú	L	M	V	VB
Order	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Empididae	Hemerodromia sp.			0.1	0.1				
	1	Neoplasta sp.								
	Ephydridae	ephydrid genus A								
		<i>Scatella</i> sp.								
	Psychodidae	Pericoma sp.			0.2	0.2				
	Sciaridae	sciarid genus A								
	Sciomyzidae	Sepedon sp.	2.5	2.5						
	Simuliidae	Prosimulium sp.								
		Simulium sp.								
		Stegopterna sp.								
	Stratiomyidae	Caloparyphus sp.	1.7	1.7						
		Nemotelus sp.								
		Stratiomys sp.			9.3	3.6				
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	15.0	6.7	1.9	1.2				
		Tabanus sp.	7.1	6.6	0.7	0.7				
	Tipulidae	Dicranota sp.								
		Helius sp.								
		<i>Hexatoma</i> sp.					0.3	0.2		
		Leptotarsus sp.								
		Limnophila sp.			1.7	1.3				
		<i>Limonia</i> sp.								
		Molophilus sp.								
		Ormosia sp.								
		<i>Pedicia</i> sp.								

### Appendix C6. (cont.)

	July 2014		( Do	Created own Sec	Stream	<u>l</u> D)	<u>Re</u> N & M	ference on-min	e Stream ed (LM	ns ) WD)
			<u>~~</u> G	D Sec	uon (Ot Gl	J <u>)</u> J		M	W	w <i>b)</i> В
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\overline{x}$	SE	$\frac{z}{\bar{x}}$	SE	$\overline{x}$	SE
D	Tipulidae	Pilaria sp.			1.5	1.3				
		Pseudolimnophila sp.			0.1	0.1				
		<i>Tipula</i> sp.							16.7	14.9
E	Ameletidae	Ameletus sp.								
	Baetidae	<i>Baetis</i> sp.								
		Procloeon sp.					0.2	0.2		
	Ephemerellidae	Attenella sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.								
		<i>Eurylophella</i> sp.					0.4	0.2		
	Ephemeridae	<i>Ephemera</i> sp.					5.7	2.8		
	Heptageniidae	<i>Epeorus</i> sp.								
		Maccaffertium sp.								
		Stenacron sp.					3.9	2.4		
	Leptophlebiidae	<i>Habrophlebia</i> sp.								
		<i>Paraleptophlebia</i> sp.					1.0	0.8		
	Siphlonuridae	Siphlonurus sp.								
М	Corydalidae	<i>Chauliodes</i> sp.								
	•	Nigronia sp.							31.3	21.2
	Sialidae	Sialis sp.			1.3	1.0	0.9	0.6	6.3	5.6
0	Aeshnidae	Aeshna sp.								
		<i>Boyeria</i> sp.			< 0.1	< 0.1				
	Calopterygidae	<i>Calopteryx</i> sp.								
	Coenagrionidae	Amphiagrion sp.								
	U	Argia sp.								

Appendix C6. (cont.)

	July 2014		D	Created own Sec	Stream	<u>1</u> D)	Ret No	f <mark>erence</mark> on-min	ed (LM	<u>ns</u> )
		-	&	Up Sec	tion (Gl	J)	& Mi	ne-imp	acted (	<i>N</i> B)
			(	βD	G	U		1	W.	B
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.			0.3	0.2	2.5	1.4		
	Gomphidae	<i>Stylogomphus</i> sp.			0.1	0.1	4.0	1.4		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		<i>Leucorrhinia</i> sp.								
		<i>Pentala</i> sp.								
		<i>Sympetrum</i> sp.								
Р	Capniidae	Allocapnia sp.								
	Chloroperlidae	<i>Haploperla</i> sp.								
		Sweltsa sp.								
	Leuctridae	Leuctra sp.					0.3	0.3		
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.								
		Prostoia sp.								
		Soyedina sp.							17.5	10.6
	Peltoperlidae	<i>Peltoperla</i> sp.								
	Perlidae	Acroneuria sp.					0.2	0.2		
		<i>Eccoptura</i> sp.					0.3	0.3		
	Perlodidae	<i>Clioperla</i> sp.								
		<i>Diploperla</i> sp.								
		<i>Isoperla</i> sp.								
		Malirekus sp.								
		Remenus sp.								
		Yugus sp.								
Т	Glossosomatidae	Agapetus sp.								

Appendix C6. (cont.)

			<u>C</u> 1	reated	Stream	<u>n</u>		Ref	ference	e Strear	ns
	July 2014		Dov	vn Sec	tion (G	D)		N	on-min	ed (LM	)
			& U	Jp Sec	tion (G	U)	&	: Mi	ne-imp	acted (V	WB)
			GE	)	G	U		LN	1	W	В
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$		SE	$\bar{x}$	SE
Т	Goeridae	<i>Goera</i> sp.					0	.2	0.2		
	Hydropsychidae	Cheumatopsyche sp.									
		Diplectrona sp.			0.1	0.1					
		Hydropsyche sp.									
	Hydroptilidae	<i>Hydroptila</i> sp.									
		<i>Oxyethira</i> sp.									
	Lepidostomatidae	Lepidostoma sp.									
	Leptoceridae	Oecetis sp.									
	Limnephilidae	Ironoquia sp.									
	-	Pycnopsyche sp.									
	Molannidae	<i>Molanna</i> sp.									
	Philopotamidae	<i>Chimarra</i> sp.									
	1	Dolophilodes sp.									
		Wormaldia sp.									
	Phryganidae	Ptilostomis sp.									
	Polycentropodidae	Polycentropus sp.									
	Psychomyiidae	Lype sp.									
	Rhyacophilidae	<i>Rhyacophila</i> sp.									
	Thremmatidae	Neophylax sp.									
Total	Richness / EPT Rich	ness and Abundance	13/1	74	30/1	1,205	27/	11	714	6/1	13
Avera	ge Total Richness ar	nd SE $(n = 5)$	4.6	0.7	13.0	1.8	15	.0	1.8	1.8	0.6
Avera	ge EPT Richness and	d SE $(n = 5)$	0.2	0.2	0.2	0.2	4	.8	0.7	0.4	0.2
B = Ba	asommatophora, C =	Coleoptera, D = Diptera	a, $\overline{E} = \text{Ep}$	hemer	optera,	H = Hap	olotaxic	la, A	M = Me	galopte	ra,
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $T = $	richopte	ra		_					

Appendix C6. (cont.)

Appe	endix C7. List of m	acroinvertebrates. Values	are avera	age % c	omposi	tion $(n = 3)$	5) and sta	andard	error (S	SE).
			_ <u>C</u>	<u>reated</u>	Stream	<u>1</u>	<u>Re</u>	ference	<u>e Strea</u>	<u>ms</u>
	August 2014		Do	wn Sec	tion (G	D)	l	Unmine	ed (LM	)
			&	Up Sec	tion (Gl	J)	& M	ine-imp	pacted (	(WB)
			G	D	G	U	LN	N	V	VB
Orde	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-i	insects									
В	Lymnaeidae	<i>Fossaria</i> sp.	45.6	15.7	20.1	4.5	0.3	0.3		
S	Sphaeriidae	Pisidium sp.			32.5	5.6	0.6	0.4		
Η	Naididae		22.8	7.3	7.7	7.7	3.1	1.0		
Insect	ts									
С	Dryopidae	Helichus sp.			2.2	1.9	1.6	1.0		
	Dytiscidae	Agabus sp.			0.0	0.0				
		Celina sp.			0.3	0.3				
		Copelatus sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.								
	<b>T</b> 1 1	<i>Neoporus</i> sp.								
	Elmidae	Dubiraphia sp.			0.1	0.1	0.0	0.6		
		<i>Optioservus</i> sp.			0.1	0.1	0.8	0.6		
		<i>Oulimnius</i> sp.	0.5	0.5	<b>5</b> 1	2.4	0.1	0.1		
		Stenelmis sp.	0.5	0.5	5.1	3.4	4.0	1.6		
	Halıplıdae	Haliplus sp.	1.0			<b>.</b>				
		Peltodytes sp.	1.0	1.0	0.2	0.2				
	Hydrophilidae	<i>Anacaena</i> sp.								
		Berosus sp.								
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.			0.3	0.3				

Appendix C7. List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

	August 2014		<u>C</u> Do	reated wn Sec	Stream tion (GI	<u> </u> D)	<u>Ret</u> N	<b>ference</b> on-min	ed (LM)	<u>15</u> )
			&	Up Sec	tion (Gl	J)	& Mi	ine-imp	acted (V	VB)
			GI	C	Gl	J	LN	Л	WI	3
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	<i>Hydrochus</i> sp.								
		Paracymus sp.			0.4	0.4				
		Tropisternus sp.	3.6	2.6	0.7	0.5				
	Psephenidae	<i>Ectopria</i> sp.			0.5	0.5	11.5	3.5		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.								
		<i>Bezzia/Palpomyia</i> sp.								
		Ceratopogon sp.								
		Culicoides sp.	3.8	3.8	0.1	0.1				
		<i>Monohelea</i> sp.								
		<i>Dasyhelea</i> sp.								
		<i>Probezzia</i> sp.	1.0	1.0	0.2	0.2	5.4	1.2		
		Sphaeromias sp.								
D	Chironomidae	Chironominae	0.2	0.2	2.7	1.4	4.9	1.4	12.5	7.9
		Corynoneura sp.								
		Orthocladiinae					4.2	1.6	12.5	7.9
		Stempellina sp.					0.4	0.3		
		Tanypodinae			12.0	3.4	3.1	0.5		
		Tanytarsini (tribe)	0.2	0.2	0.9	0.6	1.4	0.6		
	Culicidae	Anopheles sp.							12.5	7.9
		Haemagogus sp.								
	Dixidae	Dixa sp.					0.6	0.3		
	Dolichopodidae	dolichopodid genus A								
	Empididae	empidid genus A								
		Chelifera sp.					0.6	0.3		

Appendix C7. (cont.)

	August 2014		<u>C</u> Do & 1	reated wn Sect	Stream tion (Gl	<u> </u> D)	<u>Re</u> N & Mi	ference on-min	ed (LM)	<u>18</u> ) WB)
			GI	D	<u>IOII (OU</u> G	<u>.</u> U		ле-тпр Л	W	<u>чы)</u> В
Order	Family	Sub-family / Genus	$\frac{1}{\bar{x}}$	SE	$\overline{x}$	SE	$\frac{z}{\bar{x}}$	SE	$\overline{x}$	SE
D	Empididae	Hemerodromia sp.			0.5	0.5				
		Neoplasta sp.								
	Ephydridae	ephydrid genus A								
		<i>Scatella</i> sp.								
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A								
	Sciomyzidae	Sepedon sp.	0.2	0.2	0.3	0.3				
	Simuliidae	Prosimulium sp.								
		<i>Simulium</i> sp.								
		Stegopterna sp.								
	Stratiomyidae	Caloparyphus sp.			0.7	0.5				
		Nemotelus sp.								
		Stratiomys sp.			1.4	0.7				
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	14.3	9.4	6.3	3.8	0.9	0.9		
		<i>Tabanus</i> sp.	0.7	0.5	0.2	0.2				
	Tipulidae	Dicranota sp.					0.2	0.2	12.5	7.9
		<i>Helius</i> sp.								
		<i>Hexatoma</i> sp.					11.1	3.4		
		Leptotarsus sp.								
		<i>Limnophila</i> sp.					1.6	0.8		
		<i>Limonia</i> sp.								
		Molophilus sp.					0.1	0.1		
		Ormosia sp.	1.3	1.3						
		<i>Pedicia</i> sp.								

Appendix C7. (cont.)
	August $2014$		$\underline{C}$	reated	Stream	<u>ו</u> גר	<u>Re</u>	ference	e Stream	<u>ns</u>
	August 2014		۵۵ & ۱	Jp Sec	tion (GI	קר ת	۱۸ & Mi	ine-im	acted ()	) NB)
			GI	)	GI	J		Λ	W	B
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	2.9	2.9	1.6	1.5				
		Pseudolimnophila sp.			0.2	0.2	0.7	0.4		
		<i>Tipula</i> sp.	1.2	0.9			0.3	0.3		
E	Ameletidae	Ameletus sp.								
	Baetidae	Baetis sp.								
		Procloeon sp.								
	Ephemerellidae	<i>Attenella</i> sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.								
		<i>Eurylophella</i> sp.					0.6	0.6		
	Ephemeridae	<i>Ephemera</i> sp.					6.3	4.0		
	Heptageniidae	<i>Epeorus</i> sp.								
		Maccaffertium sp.					3.8	1.0		
		Stenacron sp.					0.3	0.3		
	Leptophlebiidae	Habrophlebia sp.								
		Paraleptophlebia sp.					2.6	0.9		
	Siphlonuridae	Siphlonurus sp.								
М	Corydalidae	<i>Chauliodes</i> sp.	1.0	1.0	0.3	0.3				
	2	Nigronia sp.					1.9	1.1		
	Sialidae	Sialis sp.			2.0	1.0	0.6	0.3	50.0	31.6
0	Aeshnidae	Aeshna sp.								
		Boveria sp.								
	Calopterygidae	Caloptervx sp.								
	Coenagrionidae	Amphiagrion sp.								
	0	Argia sp.								

Appendix C7. (cont.)

Appendix C7.	(cont.)

Appen	dix C7. (cont.)									
	August 2014		<u>(</u> De	C <mark>reated</mark> own Sec	Stream tion (Gl	<u>1</u> D)	<u>Re</u> N	eference	Strea ed (LN	<u>ms</u> 1)
			&	Up Sec	tion (Gl	J)	& M	ine-imp	acted (	WB)
			G	ΰD	G	U	LI	M	W	/B
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	Enallagma sp.								
	Cordulegastridae	Cordulegaster sp.			0.3	0.2	0.7	0.4		
	Gomphidae	Stylogomphus sp.			0.5	0.5	2.3	0.7		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.								
		Leucorrhinia sp.								
		Pentala sp.								
		Sympetrum sp.								
Р	Capniidae	Allocapnia sp.								
	Chloroperlidae	<i>Haploperla</i> sp.					1.5	0.8		
		<i>Sweltsa</i> sp.								
	Leuctridae	Leuctra sp.					8.5	3.1		
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.								
		Prostoia sp.								
		Soyedina sp.					0.1	0.1		
	Peltoperlidae	Peltoperla sp.					0.1	0.1		
	Perlidae	Acroneuria sp.					0.4	0.4		
		<i>Eccoptura</i> sp.					5.6	1.9		
	Perlodidae	<i>Clioperla</i> sp.								
		<i>Diploperla</i> sp.								
		Isoperla sp.								
		Malirekus sp.								
		Remenus sp.								
		Yugus sp.								
Г	Glossosomatidae	Agapetus sp.								
		•								

	August 2014		<u>C</u>	reated	Stream	<u>ו</u> כ	Reference Streams Non-mined (LM)				
	Tugust 2011		& I	In Sec	tion (GI	D	& Mi	ne-imr	acted (V	VB)	
		-	GI	)	GI	J		1	W	3	
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	
Т	Goeridae	<i>Goera</i> sp.					0.6	0.3			
	Hydropsychidae	<i>Cheumatopsyche</i> sp.					2.5	2.2			
	• • •	Diplectrona sp.					2.9	1.9			
		Hydropsyche sp.									
	Hydroptilidae	<i>Hydroptila</i> sp.									
		Oxyethira sp.									
	Lepidostomatidae	Lepidostoma sp.					0.7	0.3			
	Leptoceridae	Oecetis sp.									
	Limnephilidae	Ironoquia sp.									
		Pycnopsyche sp.									
	Molannidae	Molanna sp.					0.6	0.6			
	Philopotamidae	<i>Chimarra</i> sp.									
		Dolophilodes sp.									
		<i>Wormaldia</i> sp.					0.1	0.1			
	Phryganidae	Ptilostomis sp.									
	Polycentropodidae	Polycentropus sp.									
	Psychomyiidae	<i>Lype</i> sp.									
	Rhyacophilidae	<i>Rhyacophila</i> sp.									
	Thremmatidae	<i>Neophylax</i> sp.									
Total 1	Richness / EPT Rich	ness and Abundance	16/0	193	29/0	486	44/17	455	5/0	5	
Avera	ge Total Richness ar	nd SE $(n = 5)$	5.6	0.9	12.0	0.9	23.4	1.0	1.0	0.8	
Avera	Average EPT Richness and SE (n = 5) $0.0$ $0.0$ $0.0$ $8.4$ $0.8$ $0.0$ $0.0$										
$B = \overline{Ba}$	asommatophora, C =	Coleoptera, $D = \overline{\text{Diptera}}$	$E = \overline{\mathrm{Ep}}$	hemer	optera, <i>l</i>	H = Hapl	otaxida, A	I = Me	galopter	·a,	
O = O	donata, $P = Plecopte$	era, $S =$ Sphaeriida, $T =$ T	richopte	era							

Appendix C7. (cont.)

App	CHUIA CO. LIST OF H	acronivericorates. values a		$\frac{1}{1}$	omposi	uon (n – .	<i>sj</i> and st			<u>эн</u> ј.	
	G	Λ	<u> </u>	reated	Strean	<u>1</u>	<u>Keterence Streams</u>				
	September 201	4	Do	wn Sec	tion (G	D)	0.14	Unmine	a (LM	) (IVD)	
			<u>&amp;</u>	Up Sec	tion (G	<u> </u>	& M	ine-imp	bacted (	(WB)	
			G	)	G	U		M	V	VB	
Orde	er Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
Non-	insects										
В	Lymnaeidae	<i>Fossaria</i> sp.	39.6	15.8	15.7	5.9					
S	Sphaeriidae	Pisidium sp.			23.9	7.9					
Η	Naididae		5.0	2.7	2.9	1.3	0.4	0.3			
Insec	ets										
С	Dryopidae	Helichus sp.			0.6	0.5	0.3	0.2			
	Dytiscidae	Agabus sp.	0.2	0.2	0.2	0.1					
		<i>Celina</i> sp.									
		Copelatus sp.	0.4	0.3							
		Desmopachria sp.			0.1	0.1					
		Hydroporus sp.	0.2	0.2							
		Laccophilus sp.	0.5	0.4	0.2	0.2					
		Neoporus sp.	2.9	1.8	0.1	0.1					
	Elmidae	Dubiraphia sp.	0.2	0.2	0.1	0.1					
		Optioservus sp.			0.1	0.1	0.3	0.3			
		<i>Oulimnius</i> sp.					0.4	0.2			
		Stenelmis sp.	0.1	0.1	4.4	3.0	5.3	1.8			
	Haliplidae	Haliplus sp.									
	1	Peltodytes sp.			0.5	0.5					
	Hydrophilidae	Anacaena sp.									
	<b>v</b> 1	Berosus sp.	0.3	0.3							
		<i>Cymbiodyta</i> sp.									
		Enochrus sp.									
		Helochares sp.									

**Appendix C8.** List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

			<u>C</u>	reated	Stream	<u>1</u>	Ret	ference	e Strean	ns
	September 2014		Do	wn Sec	tion (Gl	D)	N	on-min	ed (LM	)
			&	Up Sec	tion (Gl	J)	& Mi	ne-imp	acted (V	NB)
			Gl	)	G	U	LN	1	WB	
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	<i>Hydrochus</i> sp.								
		Paracymus sp.	0.4	0.4						
		Tropisternus sp.			0.3	0.2				
	Psephenidae	<i>Ectopria</i> sp.			0.2	0.2	6.0	1.9		
D	Ceratopogonidae	<i>Alluaudomyia</i> sp.	0.1	0.1						
		Atrichopogon sp.	0.5	0.4			0.2	0.2		
		<i>Bezzia/Palpomyia</i> sp.					1.3	0.5		
		Ceratopogon sp.	0.2	0.2			1.4	0.6		
		Culicoides sp.	0.4	0.4	0.2	0.2	0.3	0.2		
		<i>Monohelea</i> sp.								
		Dasyhelea sp.								
		<i>Probezzia</i> sp.	0.4	0.3	0.2	0.1	3.0	0.5		
		Sphaeromias sp.								
D	Chironomidae	Chironominae	7.9	4.6	6.2	0.6	13.4	3.0	16.7	12.9
		<i>Corynoneura</i> sp.	0.1	0.1			0.2	0.1		
		Orthocladiinae	1.4	0.9	1.3	0.8	6.7	1.2	4.2	3.2
		Stempellina sp.					1.9	0.5		
		Tanypodinae	5.0	2.6	6.9	1.4	7.7	1.8		
		Tanytarsini (tribe)	11.5	7.1	2.6	1.1	4.9	0.9		
	Culicidae	Anopheles sp.	0.1	0.1						
		Haemagogus sp.			0.1	0.1				
	Dixidae	Dixa sp.					0.6	0.3		
	Dolichopodidae	dolichopodid genus A								
	Empididae	empidid genus A								
	•	Chelifera sp.								

Appendix C8. (cont.)

	September 2014		C Do & I	reated wn Sec	Stream tion (GI	<u>I</u> D)	Ret N & Mi	ference on-min	<u>e Streams</u> ed (LM) pacted (WB)		
			GI	$\mathbf{D}$	GI	J		<u>ис-ш</u>	W	B	
Order	r Family	Sub-family / Genus	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	
D	Empididae	Hemerodromia sp.			0.2	0.1					
		<i>Neoplasta</i> sp.			0.1	0.1					
	Ephydridae	ephydrid genus A			0.5	0.5					
		Scatella sp.									
	Psychodidae	Pericoma sp.									
	Sciaridae	sciarid genus A									
	Sciomyzidae	Sepedon sp.	0.8	0.5	0.1	0.1					
	Simuliidae	Prosimulium sp.									
		<i>Simulium</i> sp.	6.4	5.6	8.2	7.5					
		<i>Stegopterna</i> sp.									
	Stratiomyidae	Caloparyphus sp.	0.5	0.3	1.7	0.9					
		Nemotelus sp.									
		Stratiomys sp.									
	Syrphidae	<i>Eristalis</i> sp.									
	Tabanidae	Chrysops sp.	4.2	2.9	7.6	3.5	0.2	0.1			
		<i>Tabanus</i> sp.	0.3	0.2	0.2	0.1					
	Tipulidae	Dicranota sp.					0.2	0.1			
		<i>Helius</i> sp.			0.1	0.1					
		Hexatoma sp.			0.1	0.1	7.2	1.4			
		Leptotarsus sp.									
		<i>Limnophila</i> sp.					4.0	2.1	16.7	12.9	
		<i>Limonia</i> sp.	0.3	0.1	0.3	0.3	0.2	0.2			
		Molophilus sp.									
		<i>Ormosia</i> sp.					0.2	0.2			
		<i>Pedicia</i> sp.									

## Appendix C8. (cont.)

	September 2014		<u>C</u> Do	reated wn Sec	Stream tion (GI	<u>l</u> D)	Re N	e <mark>ference</mark> Non-min	e Strear ed (LM	<u>ns</u> )
	1		&	Up Sec	tion (GU	Ĵ)	& M	line-imp	acted (	WB)
			Gl	D	G	Ú	L	M	Ŵ	B
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.7	0.7	1.1	0.4				
		Pseudolimnophila sp.			0.1	0.1	0.7	0.4		
		<i>Tipula</i> sp.	0.4	0.4	0.1	0.1	0.4	0.2	62.5	24.4
E	Ameletidae	Ameletus sp.								
	Baetidae	Baetis sp.	1.2	0.8						
		Procloeon sp.					< 0.1	< 0.1		
	Ephemerellidae	Attenella sp.			0.1	0.1				
	-	Drunella sp.								
		<i>Ephemerella</i> sp.								
		Eurylophella sp.					1.8	0.8		
	Ephemeridae	<i>Ephemera</i> sp.					6.6	2.1		
	Heptageniidae	<i>Epeorus</i> sp.								
		Maccaffertium sp.					3.0	1.8		
		Stenacron sp.					0.1	0.1		
	Leptophlebiidae	Habrophlebia sp.								
		Paraleptophlebia sp.					4.1	1.2		
	Siphlonuridae	Siphlonurus sp.	3.2	3.2						
Μ	Corydalidae	<i>Chauliodes</i> sp.	0.7	0.4	0.1	0.1				
	•	Nigronia sp.					0.2	0.2		
	Sialidae	Sialis sp.			1.4	0.6	0.1	0.1		
0	Aeshnidae	Aeshna sp.			0.1	0.1				
		<i>Boyeria</i> sp.	0.4	0.4	0.3	0.3				
	Calopterygidae	Calopteryx sp.			2.3	0.9				
	Coenagrionidae	Amphiagrion sp.								
	-	Argia sp.			0.5	0.2				

Appendix C8. (cont.)

ADDENDIX U.S. (CONL.	App	endix	C8. (	(cont.)
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			<u>C</u>	reated	Stream		Re	ference	e Strea	<u>ms</u>
	September 2014		Do	wn Sec	tion (GI	D)	Ν	on-min	ed (LN	1)
			&	Up Sect	tion (GU	J)	& M:	ine-imp	acted (	WB)
			G	D	GU	J	LN	M	W	/B
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.			0.8	0.4	1.0	0.4		
	Gomphidae	Stylogomphus sp.			0.4	0.3	4.0	0.8		
	Libellulidae	prob. <i>Leucorrhinia</i> sp.			0.5	0.3				
		Leucorrhinia sp.			0.1	0.1				
		Pentala sp.	0.4	0.4						
		Sympetrum sp.								
Р	Capniidae	Allocapnia sp.					1.0	0.4		
	Chloroperlidae	Haploperla sp.					0.9	0.3		
	1	Sweltsa sp.								
	Leuctridae	Leuctra sp.			0.3	0.2	2.7	1.0		
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.								
		Prostoia sp.								
		Sovedina sp.					0.1	0.1		
	Peltoperlidae	Peltoperla sp.								
	Perlidae	Acroneuria sp.					0.5	0.1		
		<i>Eccontura</i> sp.					2.2	0.5		
	Perlodidae	<i>Clioperla</i> sp.								
		Diploperla sp.								
		Isoperla sp.								
		Malirekus sp.								
		Remenus sp								
		Yugus sp.								
Т	Glossosomatidae	Aganetus sn								
•	Sissessinandae									

			<u>C</u>	reated	Stream	<u>n</u>	R	eference	e Strean	<u>15</u>
	September 2014		Do	wn Sec	tion (G	D)	1	Von-min	ed (LM)	)
			&	Up Sec	tion (G	U)	& N	line-imp	pacted (V	VB)
			Gl	)	G	U	L	М	W	В
Order	- Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Т	Goeridae	<i>Goera</i> sp.								
	Hydropsychidae	Cheumatopsyche sp.								
		Diplectrona sp.			1.5	1.1	3.0	2.1		
		Hydropsyche sp.								
	Hydroptilidae	<i>Hydroptila</i> sp.								
	<b>v</b> 1	Oxyethira sp.								
	Lepidostomatidae	Lepidostoma sp.					0.1	0.1		
	Leptoceridae	<i>Oecetis</i> sp.								
	Limnephilidae	Ironoquia sp.								
	-	<i>Pycnopsyche</i> sp.								
	Molannidae	<i>Molanna</i> sp.					0.3	0.2		
	Philopotamidae	Chimarra sp.			0.6	0.6				
	Ĩ	Dolophilodes sp.					0.2	0.2		
		Wormaldia sp.			0.5	0.3	0.2	0.2		
	Phryganidae	Ptilostomis sp.	3.2	0.7	4.0	1.4				
	Polycentropodidae	Polycentropus sp.			0.1	0.1	0.6	0.3		
	Psychomyiidae	Lype sp.								
	Rhyacophilidae	<i>Rhyacophila</i> sp.			0.1	0.1	0.1	0.1		
	Thremmatidae	Neophylax sp.								
Total	Richness / EPT Rich	ness and Abundance	36/3	759	53/8	1,463	50/19	1,261	4/0	16
Avera	ge Total Richness ar	nd SE $(n = 5)$	15.0	1.8	24.4	3.4	30.8	2.4	1.0	0.4
Avera	ge EPT Richness and	d SE $(n = 5)$	1.6	0.2	3.2	1.0	11.6	0.9	0.0	0.0
B = Ba	asommatophora, C =	Coleoptera, D = Diptera	E = Ep	hemer	optera,	H = Hap	olotaxida,	M = Me	egalopter	a,
O = O	donata, P = Plecopte	era, $S = $ Sphaeriida, $\overline{T} = $ T	richopt	era		_				

Appendix C8. (cont.)

Apper	<b>ndix C9.</b> List of ma	croinvertebrates. Values a	are avera	age % c	composi	ition (n =	5) and st	andard	error (S	SE).
			<u>C</u>	reated	Stream	<u>n</u>	Re	ference	e Strea	<u>ms</u>
	October 2014		Do	wn Sec	ction (G	D)	I	Unmine	ed (LM	)
			&	Up Sec	tion (G	U)	& M	ine-imp	pacted (	(WB)
			Gl	D	G	U	LN	N	V	VB
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Non-ii	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	5.6	1.0	14.7	7.7	0.3	0.3		
S	Sphaeriidae	<i>Pisidium</i> sp.			35.2	13.3	1.3	1.3		
Н	Naididae		5.5	1.8	0.1	0.1	0.1	0.1		
Insect	S									
С	Dryopidae	Helichus sp.			0.2	0.1	0.7	0.5		
	Dytiscidae	Agabus sp.	4.6	2.7	< 0.1	< 0.1				
		<i>Celina</i> sp.								
		Copelatus sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	2.1	1.2	0.2	0.1				
	Elmidae	<i>Dubiraphia</i> sp.								
		Optioservus sp.			0.1	0.1	0.4	0.3		
		<i>Oulimnius</i> sp.					0.7	0.5		
		<i>Stenelmis</i> sp.	0.5	0.5	3.9	3.4	1.2	0.7		
	Haliplidae	<i>Haliplus</i> sp.								
		Peltodytes sp.	0.2	0.2	0.1	0.1				
	Hydrophilidae	Anacaena sp.								
		Berosus sp.	1.3	0.7						
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.								

**Appendix C9.** List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

			<u>C</u>	reated	Stream	<u>n</u>	<b>Reference Streams</b>				
	October 2014		Do	wn Sec	tion (G	D)	Ν	on-min	ed (LM	)	
			&	Up Sec	tion (G	U)	& Mi	ine-imp	pacted (WB)		
			G	D	G	U	LN	Л	W	В	
Orde	er Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
С	Hydrophilidae	Hydrochus sp.									
		Paracymus sp.									
		<i>Tropisternus</i> sp.									
	Psephenidae	<i>Ectopria</i> sp.			< 0.1	< 0.1	8.1	3.6	6.7	5.2	
D	Ceratopogonidae	Alluaudomyia sp.									
		Atrichopogon sp.	0.4	0.2							
		<i>Bezzia/Palpomyia</i> sp.					0.8	0.4			
		Ceratopogon sp.					1.3	0.6			
		Culicoides sp.					1.0	0.6			
		<i>Monohelea</i> sp.	0.1	0.1							
		<i>Dasyhelea</i> sp.									
		<i>Probezzia</i> sp.	2.0	1.1	1.4	1.4	1.9	0.7			
		Sphaeromias sp.									
D	Chironomidae	Chironominae	3.1	1.3	1.4	1.1	4.8	1.8	16.7	12.9	
		Corynoneura sp.	0.4	0.2	0.7	0.3	2.3	1.3			
		Orthocladiinae	1.3	0.8	0.6	0.2	12.5	3.1	6.7	5.2	
		Stempellina sp.					2.2	1.8			
		Tanypodinae	14.8	4.1	7.9	2.6	4.6	0.8			
		Tanytarsini (tribe)	15.0	6.9	2.4	1.7	0.7	0.4			
	Culicidae	Anopheles sp.									
		Haemagogus sp.									
	Dixidae	<i>Dixa</i> sp.			< 0.1	< 0.1	0.8	0.6			
	Dolichopodidae	dolichopodid genus A			0.1	0.1					
	Empididae	empidid genus A									
	-	Chelifera sp.									

Appendix C9. (cont.)

	October 2014		<u>C</u> Do	Created wn Sec	Stream	<u>n</u> D)	Re N	eference Non-min	e Strea led (LN	<u>ms</u> (1)
			&	Up Sec	tion (G	U)	& N	line-imp	bacted (	(WB)
			G	D	G	U	L	M	V	VB
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE
D	Empididae	<i>Hemerodromia</i> sp. <i>Neoplasta</i> sp.	0.4	0.4	0.2	0.2				
	Ephydridae	ephydrid genus A Scatella sp.	3.2	1.9						
	Psychodidae Sciaridae	<i>Pericoma</i> sp. sciarid genus A								
	Sciomyzidae Simuliidae	Sepedon sp. Prosimulium sp.	0.4	0.2	< 0.1	< 0.1				
		Simulium sp. Stegopterna sp.	3.4	3.1	1.7	1.2				
	Stratiomyidae	Caloparyphus sp. Nemotelus sp.			0.3	0.2				
	Symbidae	Stratiomys sp.			0.1	0.1				
	Tabanidae	Chrysops sp. Tabanus sp.	3.6	1.0	3.5	1.3				
	Tipulidae	<i>Dicranota</i> sp. <i>Helius</i> sp.					0.3	0.3		
		Hexatoma sp.					13.7	5.3		
		Leptotarsus sp.					0.2	0.2		
		<i>Limnophila</i> sp.					0.3	0.3		
		Limonia sp. Molophilus sp.	1.2	0.5						
		Ormosia sp. Pedicia sp.					< 0.1	< 0.1		

Appendix C9. (cont.)

	October 2014			reated	Stream	<u>n</u>	<u>Re</u>	ference	e Stream	<u>ns</u>
	000001 2014		D0	Un See	tion (C	U)	רי גיער או	ino imr	noted (LIVI	J VD)
				op sec				me-mp		ND) D
Ordor	Family	Sub family / Conus		SE SE	- U	SE	L	VI SE	<u>vv</u>	D SE
	Tanniy	Dilania an	$\frac{x}{0.2}$		<i>x</i>		X	SE	X	SE
D	Tipulidae	Fuaria sp. Dagudolimnonhila an	0.5	0.5	0.0	0.5				
		<i>Fseudolimnophila</i> sp.	10.4	10.0	0.2	0.2	26	0.0	217	174
г	A 1 (* 1	<i>Tipula</i> sp.	19.4	10.9	0.2	0.2	2.0	0.9	31./	1/.4
E	Ameletidae	Ameletus sp.								
	Baetidae	Baetis sp.					0.0	0.0		
	<b>T</b> 1 11.1	Procloeon sp.					0.3	0.3		
	Ephemerellidae	Attenella sp.								
		Drunella sp.								
		<i>Ephemerella</i> sp.								
		<i>Eurylophella</i> sp.					2.0	1.0		
	Ephemeridae	<i>Ephemera</i> sp.					2.3	1.5		
	Heptageniidae	<i>Epeorus</i> sp.								
		<i>Maccaffertium</i> sp.					2.5	1.7		
		Stenacron sp.								
	Leptophlebiidae	<i>Habrophlebia</i> sp.								
		Paraleptophlebia sp.			< 0.1	< 0.1	8.5	1.5		
	Siphlonuridae	Siphlonurus sp.								
М	Corydalidae	<i>Chauliodes</i> sp.								
	5	Nigronia sp.					0.2	0.2		
	Sialidae	Sialis sp.			1.5	0.6	0.1	0.1		
0	Aeshnidae	Aeshna sp.			-		-	-		
		<i>Boveria</i> sp.	0.6	0.6						
	Caloptervoidae	<i>Calontervx</i> sp	0.0	0.0	1.4	1.1				
	Coenagrionidae	Amphiagrion sp			1.1					
	Coonagrioniado	Argia sp.	0.4	0.4	2.4	1.2				

Appendix C9. (cont.)

	October 2014		<u>C</u> Do	Created wn Sect	<u>Strean</u> tion (Gl	<u>l</u> D)	<u>Re</u> N	ference	Stream ed (LM)	<u>15</u> )
		-	&	Up Sect	ion (Gl	J)	& M:	ine-imp	acted (V	VB)
			G	D	G	U	LN	Λ	W	В
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.	0.1	0.1						
	Cordulegastridae	Cordulegaster sp.			0.3	0.3	0.1	0.1		
	Gomphidae	Stylogomphus sp.			0.4	0.2	1.8	0.9		
	Libellulidae	prob. Leucorrhinia sp.	0.1	0.1	0.1	0.1				
		<i>Leucorrhinia</i> sp.								
		Pentala sp.								
		Sympetrum sp.								
Р	Capniidae	Allocapnia sp.	0.1	0.1			2.2	1.0		
	Chloroperlidae	<i>Haploperla</i> sp.					1.3	0.6		
		<i>Sweltsa</i> sp.								
	Leuctridae	Leuctra sp.			0.4	0.3	0.4	0.3	6.7	5.2
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.								
		Prostoia sp.								
		<i>Soyedina</i> sp.			0.3	0.2	0.1	0.1	8.3	6.5
	Peltoperlidae	<i>Peltoperla</i> sp.								
	Perlidae	Acroneuria sp.					0.4	0.3		
		<i>Eccoptura</i> sp.					2.8	1.0		
	Perlodidae	<i>Clioperla</i> sp.								
		<i>Diploperla</i> sp.					0.1	0.1		
		<i>Isoperla</i> sp.					0.8	0.6		
		<i>Malirekus</i> sp.								
		Remenus sp.								
		Yugus sp.								
Γ	Glossosomatidae	Agapetus sp.								

Appendix C9. (cont.)

October 2014			reated	Strear	<u>n</u>	<u>Re</u>	Reference Streams			
	0010001 2014		D0 &	Un Sec	tion (G	U)	* Mi	ine-imr	acted ()	) VB)
			G	D	G	iU		Λ	W	B
Order	- Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Т	Goeridae	<i>Goera</i> sp.								
	Hydropsychidae	Cheumatopsyche sp.								
		Diplectrona sp.	0.2	0.2	1.5	0.9	2.3	0.9	23.3	11.3
		Hydropsyche sp.					0.1	0.1		
	Hydroptilidae	<i>Hydroptila</i> sp.								
		<i>Oxyethira</i> sp.								
	Lepidostomatidae	Lepidostoma sp.								
	Leptoceridae	Oecetis sp.								
	Limnephilidae	Ironoquia sp.								
		Pycnopsyche sp.					3.4	1.9		
	Molannidae	Molanna sp.								
	Philopotamidae	<i>Chimarra</i> sp.	1.2	1.2	10.0	6.9				
		Dolophilodes sp.								
		Wormaldia sp.			0.7	0.7	0.4	0.3		
	Phryganidae	Ptilostomis sp.	8.2	2.8	4.7	1.2				
	Polycentropodidae	Polycentropus sp.					0.3	0.2		
	Psychomyiidae	<i>Lype</i> sp.					0.3	0.3		
	Rhyacophilidae	<i>Rhyacophila</i> sp.	0.2	0.2	0.2	0.2	2.8	1.0		
	Thremmatidae	<i>Neophylax</i> sp.								
Total	Richness / EPT Rich	ness and Abundance	32/5	573	41/8	1,656	50/20	899	7/3	11
Average Total Richness and SE $(n = 5)$				2.1	20.4	2.2	26.6	3.5	1.8	0.9
Avera	Average EPT Richness and SE $(n = 5)$			0.4	3.6	1.1	10.2	2.2	0.8	0.4
B = Ba	C = Basommatophora, C = Coleoptera, D = Dipter		Piptera, $E =$ Ephemeroptera, $H =$ Haplotaxida, $M =$ Megaloptera,							ra,
O = O	P = Odonata, P = Plecoptera, S = Sphaeriida, T =			era						

Appendix C9. (cont.)

	November 201	4	<u>(</u> Do	Created	Strean	<u>1</u> D)	<u>Re</u>	<b>ference</b> Inmine	e <b>Strea</b> d (LM)	<u>ms</u>
			&	Up Sec	tion (G	U)	& Mi	ine-imp	acted (	, WB)
			G	D	G	U	LN	Λ	W	/B
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\overline{x}$	SE
Non-i	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	1.4	1.2	1.8	0.5				
S	Sphaeriidae	Pisidium sp.			17.4	5.7				
Н	Naididae	-	3.9	2.0	1.5	0.6	0.7	0.6		
Insect	S									
С	Dryopidae	Helichus sp.			0.9	0.7	0.4	0.4		
	Dytiscidae	Agabus sp.	0.3	0.2						
		Celina sp.								
		<i>Copelatus</i> sp.								
		Desmopachria sp.								
		Hydroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	0.5	0.3	0.1	0.1				
	Elmidae	Dubiraphia sp.								
		<i>Optioservus</i> sp.			0.1	0.1	0.6	0.4		
		Oulimnius sp.								
		Stenelmis sp.	< 0.1	< 0.1	0.6	0.2	1.0	0.7		
	Haliplidae	Haliplus sp.								
	1	Peltodytes sp.								
	Hydrophilidae	Anacaena sp.								
	• 1	Berosus sp.	0.1	< 0.1						
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.								

<u>Appendix C10. List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).</u>

	November 2014		Do	reated	Strear	<u>n</u> (D)	<u>Re</u> N	ference	e Strear	<u>ns</u>
			&	Up Sec	tion (G	U)	& Mine-impacted (W			WB)
			G	D	G	U		M	W	B
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.								
		Tropisternus sp.			0.1	0.1				
	Psephenidae	<i>Ectopria</i> sp.			0.2	0.1	3.0	1.0		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.	0.7	0.4						
		<i>Bezzia/Palpomyia</i> sp.					2.2	0.7		
		Ceratopogon sp.					1.4	0.6		
		Culicoides sp.	0.5	0.2	0.5	0.2				
		Monohelea sp.								
		Dasyhelea sp.								
		Probezzia sp.	0.5	0.3	5.6	5.2	3.8	1.1		
		Sphaeromias sp.	0.1	0.1	< 0.1	< 0.1				
D	Chironomidae	Chironominae	7.9	0.9	4.7	2.2	6.5	2.2	8.3	7.5
		Corynoneura sp.	1.3	0.3	1.7	0.2	0.6	0.6		
		Orthocladiinae	14.8	2.5	4.7	2.0	4.9	1.3	6.3	5.6
		Stempellina sp.					3.6	1.5		
		Tanypodinae	31.2	3.6	21.8	6.4	5.7	0.8	33.3	21.1
		Tanytarsini (tribe)							6.3	5.6
	Culicidae	Anopheles sp.	0.1	0.1						
		Haemagogus sp.								
	Dixidae	Dixa sp.	0.1	0.1	0.4	0.2	0.7	0.3		
	Dolichopodidae	dolichopodid genus A	0.2	0.1						
	Empididae	empidid genus A <i>Chelifera</i> sp.			< 0.1	< 0.1				

Appendix C10. (cont.)

November 2014		<u>(</u> Do	Created	<b>Strean</b> Stion (G	<u>n</u> D)	Re N	e <b>ference</b> Ion-min	e Strea led (LN	<u>ms</u> 1)	
			&	Up Sec	tion (G	U)	& M	line-imp	bacted (	WB)
			G	D	G	U	L	М	W	/B
Orde	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Empididae	Hemerodromia sp.	0.1	0.1	0.4	0.3				
		Neoplasta sp.								
	Ephydridae	ephydrid genus A			< 0.1	< 0.1				
		Scatella sp.								
	Psychodidae	Pericoma sp.	0.1	0.1						
	Sciaridae	sciarid genus A			< 0.1	< 0.1				
	Sciomyzidae	Sepedon sp.	< 0.1	< 0.1	< 0.1	< 0.1				
	Simuliidae	Prosimulium sp.					0.1	0.1		
		Simulium sp.	0.5	0.1	0.2	0.1				
		Stegopterna sp.								
	Stratiomyidae	Caloparyphus sp.	< 0.1	< 0.1	0.3	0.2				
	-	Nemotelus sp.								
		Stratiomys sp.								
	Syrphidae	Eristalis sp.								
	Tabanidae	Chrysops sp.	0.3	0.2	1.9	0.7				
		Tabanus sp.	0.1	0.1			0.1	0.1		
	Tipulidae	Dicranota sp.					0.2	0.2		
	-	Helius sp.			0.2	0.1				
		Hexatoma sp.					2.6	0.9		
		Leptotarsus sp.								
		Limnophila sp.					1.6	1.2		
		Limonia sp.	0.5	0.2						
		Molophilus sp.					1.6	1.4		
		Ormosia sp.								
		Pedicia sp.					0.2	0.2		

Appendix C10. (cont.)

	November 2014		<u>(</u>	Created	Stream	<u>n</u> D)	<u>Re</u> N	ference	e <mark>Strea</mark> ed (LM	<u>ms</u>
			8.	Un Sec	tion (G	U)	& M	ine-imr	acted (	WB)
			G	<u>D</u>	G	U		Л	W	VB
Order	Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.3	0.2	0.3	0.2				
	-	Pseudolimnophila sp.	< 0.1	< 0.1	1.3	1.0	3.5	1.0		
		<i>Tipula</i> sp.	2.1	1.2	0.3	0.2	0.5	0.3		
E	Ameletidae	Ameletus sp.	1.6	1.2	0.5	0.2	0.9	0.5		
	Baetidae	Baetis sp.	3.0	2.9	< 0.1	< 0.1				
		Procloeon sp.								
	Ephemerellidae	<i>Attenella</i> sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.					0.2	0.2		
		<i>Eurylophella</i> sp.					1.0	0.5		
	Ephemeridae	Ephemera sp.					0.3	0.3		
	Heptageniidae	<i>Epeorus</i> sp.					0.3	0.3		
		Maccaffertium sp.					1.0	0.5		
		Stenacron sp.								
	Leptophlebiidae	Habrophlebia sp.					0.1	0.1		
		Paraleptophlebia sp.					20.0	4.0		
	Siphlonuridae	Siphlonurus sp.								
Μ	Corydalidae	Chauliodes sp.								
	-	Nigronia sp.								
	Sialidae	Sialis sp.			0.4	0.2				
0	Aeshnidae	Aeshna sp.								
		<i>Boyeria</i> sp.								
	Calopterygidae	<i>Calopteryx</i> sp.			< 0.1	< 0.1				
	Coenagrionidae	Amphiagrion sp.								
	-	Argia sp.			1.3	1.1				

Appendix C10. (cont.)

<u>Appendi</u>			
	November 2014		
Order	Family	Sub-family / Genus	

Appendix C10. (cont.)

	November 2014		<u>(</u> Do	Created	Strear tion (G	<u>n</u> D)	<u>Re</u> N	<u>ns</u> )		
			&	Up Sec	tion (G	U)	& M	ine-imp	oacted (V	WB)
			G	D	G	iU	LN	M	W	В
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
0	Coenagrionidae	<i>Enallagma</i> sp.								
	Cordulegastridae	Cordulegaster sp.			0.1	0.1	0.9	0.5		
	Gomphidae	Stylogomphus sp.			0.1	< 0.1	0.4	0.4		
	Libellulidae	prob. Leucorrhinia sp.								
		Leucorrhinia sp.								
		Pentala sp.								
		<i>Sympetrum</i> sp.								
Р	Capniidae	<i>Allocapnia</i> sp.	3.3	0.9	0.5	0.2	11.2	2.4		
	Chloroperlidae	<i>Haploperla</i> sp.					2.9	1.4		
		Sweltsa sp.								
	Leuctridae	Leuctra sp.			0.1	0.1	0.8	0.5		
	Nemouridae	Amphinemura sp.								
		Ostrocerca sp.								
		Prostoia sp.								
		<i>Soyedina</i> sp.			< 0.1	< 0.1	0.3	0.3	37.5	21.4
	Peltoperlidae	<i>Peltoperla</i> sp.								
	Perlidae	Acroneuria sp.					0.1	0.1		
		Eccoptura sp.					0.6	0.4		
	Perlodidae	<i>Clioperla</i> sp.								
		<i>Diploperla</i> sp.	< 0.1	< 0.1						
		<i>Isoperla</i> sp.			0.2	0.2	1.0	0.5		
		<i>Malirekus</i> sp.			0.4	0.4				
		<i>Remenus</i> sp.								
		Yugus sp.								
Т	Glossosomatidae	<i>Agapetus</i> sp.								

			<u>(</u>	Created	Stream	<u>n</u>	<b>Reference Streams</b>				
	November 2014		Do	own Sec	tion (G	D)	Ν	on-min	ed (LM)	)	
			&	Up Sec	tion (G	U)	& Mi	ne-imp	bacted (WB)		
			G	D	G	U	LN	Л	WI	3	
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
Т	Goeridae	<i>Goera</i> sp.									
	Hydropsychidae	Cheumatopsyche sp.									
		Diplectrona sp.			1.6	1.0	1.6	0.8			
		Hydropsyche sp.			< 0.1	< 0.1	1.3	0.5			
	Hydroptilidae	Hydroptila sp.									
		<i>Oxyethira</i> sp.									
	Lepidostomatidae	Lepidostoma sp.									
	Leptoceridae	Oecetis sp.									
	Limnephilidae	Ironoquia sp.	3.5	2.2	4.4	1.4					
		Pycnopsyche sp.			< 0.1	< 0.1	1.7	0.5			
	Molannidae	Molanna sp.									
	Philopotamidae	<i>Chimarra</i> sp.			2.9	2.2					
	-	Dolophilodes sp.									
		Wormaldia sp.	0.1	0.1	0.8	0.4	1.2	0.7			
	Phryganidae	Ptilostomis sp.	0.5	0.2	2.3	0.7					
	Polycentropodidae	Polycentropus sp.			< 0.1	< 0.1	1.0	0.6	8.3	7.5	
	Psychomyiidae	<i>Lype</i> sp.									
	Rhyacophilidae	Rhyacophila sp.					0.6	0.4			
	Thremmatidae	<i>Neophylax</i> sp.					0.7	0.5			
Total	Richness / EPT Rich	ness and Abundance	36/7	3,129	50/15	3,066	48/22	529	6/2	9	
Avera	ge Total Richness ar	nd SE $(n = 5)$	22.2	0.7	27.0	1.6	26.4	1.6	1.6	0.6	
Avera	Average EPT Richness and SE $(n = 5)$		4.2	0.2	7.6	1.2	11.6	1.1	0.6	0.2	
B = B	asommatophora, $\overline{C}$ =	Coleoptera, D = Diptera	E = E	phemer	optera,	H = Hap	lotaxida, <i>I</i>	M = Me	galopter	a, 🗌	
O = O	donata. $P = Plecopte$	era, $S = Sphaeriida, T = T$	richop	tera							

Appendix C10. (cont.)

	Descuber 2014		<u>(</u>	Created	Stream	<u>1</u>	Re	ference	Stream	<u>ms</u>
	December 2014	ł	DC &	Jun Sec	tion (GI	D)	6- M		u (LIVI) acted (	
			<u> </u>			<u>.</u>		me-mp ∡		
Orda	. Eamily	Sub family / Canua	<u> </u>	D SE		U SE				D
<u>Unde</u>	г гашиу	Sub-family / Genus	X	SE	X	SE	X	SE	<u>x</u>	SE
Non-l	nsects			2.5	0.4	0.1				
В	Lymnaeidae	<i>Fossaria</i> sp.	4.4	3.5	0.4	0.1	0.0	• •		
S	Sphaeriidae	Pisidium sp.	0.0	0.0	7.7	2.6	0.2	0.2		
Н	Naididae		0.8	0.3	7.0	2.8	0.4	0.2		
Insect	<sup>t</sup> S									
С	Dryopidae	Helichus sp.			0.1	0.1	0.2	0.2		
	Dytiscidae	Agabus sp.	0.2	0.1						
	•	Celina sp.								
		Copelatus sp.								
		Desmopachria sp.								
		Hvdroporus sp.								
		Laccophilus sp.								
		Neoporus sp.	0.2	0.2						
	Elmidae	Dubiranhia sp	••=	0.2						
	Liiiidae	Ontioservus sp	< 0.1	< 0.1			0.4	0.2		
		Oulimnius sp	. 0.1	. 0.1			0.1	0.2		
		Stenelmis sp.			0.5	03	0.5	03		
	Haliplidae	Halinlus sp.			0.5	0.5	0.5	0.5		
	manphuae	Paltodutas sp.	< 0.1	< 0.1						
	Undrophilidaa	Angegeng sp.	< 0.1	< 0.1						
	Trydropinnuae	Anacaena sp.								
		<i>Gerosus</i> sp.								
		<i>Cymbioayia</i> sp.								
		<i>Enochrus</i> sp.								
		<i>Helochares</i> sp.								

<u>Appendix C11. List of macroinvertebrates</u>. Values are average % composition (n = 5) and standard error (SE).

Appendix	C11.	(cont.)

		<u>(</u>	Created	Stream	<u>n</u>	<u>Reference Streams</u>					
	December 2014		Do	wn Sec	tion (G	D)	Ν	on-min	ed (LM	)	
			&	Up Sec	tion (G	U)	& M	ine-imp	acted (V	l (WB)	
			G	D	G	U	LN	Ν	W	В	
Order	· Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
С	Hydrophilidae	Hydrochus sp.									
		Paracymus sp.									
		Tropisternus sp.									
	Psephenidae	<i>Ectopria</i> sp.			0.3	0.2	1.3	0.5	5.0	5.0	
D	Ceratopogonidae	<i>Alluaudomyia</i> sp.									
		Atrichopogon sp.	< 0.1	< 0.1							
		<i>Bezzia/Palpomyia</i> sp.					0.4	0.3			
		Ceratopogon sp.	< 0.1	< 0.1	< 0.1	< 0.1	1.3	0.3			
		Culicoides sp.	0.2	0.1	0.2	0.1					
		Monohelea sp.									
		Dasyhelea sp.									
		<i>Probezzia</i> sp.	0.6	0.4	2.4	1.8	2.6	1.0			
		Sphaeromias sp.	< 0.1	< 0.1	1.2	0.3					
D	Chironomidae	Chironominae	4.7	1.5	5.4	3.6	5.1	0.9	8.0	8.0	
		Corynoneura sp.	0.6	0.2	0.7	0.4	0.3	0.3	5.0	5.0	
		Orthocladiinae	13.7	5.7	7.1	1.3	3.9	1.1	28.0	12.7	
		Stempellina sp.					1.9	0.7			
		Tanypodinae	32.9	5.0	28.6	6.7	3.8	0.9			
		Tanytarsini (tribe)	29.8	5.1	16.7	5.2	2.9	1.0			
	Culicidae	Anopheles sp.									
		Haemagogus sp.									
	Dixidae	Dixa sp.			< 0.1	< 0.1	0.2	0.2			
	Dolichopodidae	dolichopodid genus A									
	Empididae	empidid genus A									
	•	Chelifera sp.									

	December 2014		<u>(</u> Do	C <b>reated</b> own Sec	Strear	<u>n</u> D)	<u>R</u>	eferenco Non-mir	e Strean ned (LM	<u>ns</u> )
			&	Up Sec	tion (G	Ú	& N	Aine-imp	bacted (	WB)
			G	D	G	Ú	Ι	LM	Ŵ	B
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Empididae	Hemerodromia sp.	0.4	0.3						
		Neoplasta sp.	0.1	0.1						
	Ephydridae	ephydrid genus A								
		Scatella sp.								
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A								
	Sciomyzidae	Sepedon sp.			< 0.1	< 0.1				
	Simuliidae	Prosimulium sp.					0.4	0.2		
		<i>Simulium</i> sp.	0.8	0.6	0.3	0.2				
		Stegopterna sp.								
	Stratiomyidae	Caloparyphus sp.	< 0.1	< 0.1	0.2	0.2				
		Nemotelus sp.								
		Stratiomys sp.								
	Syrphidae	<i>Eristalis</i> sp.			< 0.1	< 0.1				
	Tabanidae	Chrysops sp.	0.4	0.2	3.4	1.1				
		<i>Tabanus</i> sp.	0.1	0.1	0.2	0.1				
	Tipulidae	Dicranota sp.					0.2	0.2		
		Helius sp.			< 0.1	< 0.1				
		<i>Hexatoma</i> sp.					0.6	0.2		
		Leptotarsus sp.					0.2	0.2		
		Limnophila sp.			0.1	0.1	1.5	0.8		
		<i>Limonia</i> sp.	0.5	0.2	< 0.1	< 0.1				
		Molophilus sp.							5.0	5.0
		Ormosia sp.								
		Pedicia sp.					0.2	0.2		

Appendix C11. (cont.)

December 2014		<u>C</u> Do	reated wn Sec	Stream	<u>n</u> D)	<u>Re</u> N	ference	ed (LM)	<u>15</u> )	
			&	Up Sec	tion (G	U)	& M	& Mine-impacted (WF		
			GI	)	G	Ú	LN	A	Ŵ	B
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.	0.4	0.3	1.2	0.4				
		Pseudolimnophila sp.			0.1	0.1	1.2	0.6		
		<i>Tipula</i> sp.	2.2	1.3	0.1	0.1	0.2	0.2		
E	Ameletidae	Ameletus sp.	1.4	0.7	2.1	0.8	4.4	2.4		
	Baetidae	Baetis sp.	0.2	0.2	0.4	0.4	0.7	0.3		
		Procloeon sp.								
	Ephemerellidae	Attenella sp.								
	1	Drunella sp.								
		<i>Ephemerella</i> sp.					11.8	5.1		
		Eurvlophella sp.					3.0	1.2		
	Ephemeridae	Ephemera sp.								
	Heptageniidae	<i>Epeorus</i> sp.					6.7	1.0		
	1 0	Maccaffertium sp.					0.5	0.3		
		Stenacron sp.								
	Leptophlebiidae	Habrophlebia sp.					0.2	0.2		
		Paralentonhlebia sp.					11.2	2.3	6.7	6.7
	Siphlonuridae	Siphlonurus sp.							,	
М	Corvdalidae	<i>Chauliodes</i> sp.			< 0.1	< 0.1				
	e erj	Nigronia sp.			0.11	0.1	0.2	0.2		
	Sialidae	Sialis sp.			0.4	0.2	0.2	0.2		
0	Aeshnidae	Aeshna sp.			0	0.2				
0		<i>Boveria</i> sp.								
	Calontervoidae	<i>Calontervx</i> sp								
	Coenagrionidae	Amphiagrion sp								
	Coonagrioniado	Aroja sn			0.4	03				
		· · · S · · · · · · ·			0.1	0.5				

Appendix C11. (cont.)

Appendix C	<b>11.</b> (cont.)	
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<u>3)</u> <u>SE</u> 5.0
3) <u>SE</u> 5.0
<u>SE</u> 5.0
<u>SE</u> 5.0
5.0
5.0
5.0

				Created	Stream	<u>n</u>	R	<b>Reference Streams</b>			
	December 2014		Do	own Sec	tion (G	D)		Non-min	ed (LM	)	
			&	Up Sec	tion (G	U)	& N	Mine-imp	bacted (V	WB)	
			G	D	G	U	I	LM		В	
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	
Т	Goeridae	<i>Goera</i> sp.									
	Hydropsychidae	Cheumatopsyche sp.									
		Diplectrona sp.	0.2	0.2	0.9	0.6	2.2	0.7			
		Hydropsyche sp.			< 0.1	< 0.1					
	Hydroptilidae	<i>Hydroptila</i> sp.									
		<i>Oxyethira</i> sp.									
	Lepidostomatidae	Lepidostoma sp.									
	Leptoceridae	<i>Oecetis</i> sp.									
	Limnephilidae	Ironoquia sp.	1.8	0.8	2.6	1.6					
		Pycnopsyche sp.					1.0	0.4	33.3	18.3	
	Molannidae	<i>Molanna</i> sp.									
	Philopotamidae	<i>Chimarra</i> sp.			6.5	4.7					
		Dolophilodes sp.					0.7	0.5			
		<i>Wormaldia</i> sp.	0.1	0.1	0.3	0.3	0.8	0.8			
	Phryganidae	Ptilostomis sp.	1.5	0.7	1.3	0.3					
	Polycentropodidae	Polycentropus sp.			0.1	0.1	0.3	0.2	4.0	4.0	
	Psychomyiidae	<i>Lype</i> sp.									
	Rhyacophilidae	<i>Rhyacophila</i> sp.					0.2	0.2			
	Thremmatidae	<i>Neophylax</i> sp.					2.0	0.8			
Total	Richness / EPT Rich	ness and Abundance	34/9	3,894	47/13	3,284	48/2	1 736	9/3	17	
Average Total Richness and SE $(n = 5)$		19.8	1.0	25.6	0.9	28.4	- 1.2	2.6	0.5		
Avera	Average EPT Richness and SE $(n = 5)$		4.8	0.4	7.4	0.7	13.4	0.4	1.0	0.3	
B = Basommatophora, C = Coleoptera, D = Diptera				phemer	optera,	H = Hap	lotaxida	, M = Me	egalopte	ra,	
O = O	D = Odonata, P = Plecoptera, S = Sphaeriida, T = Trichoptera										

Appendix C11. (cont.)

			<u>C</u>	reated	Stream	<u>n</u>	ŀ	Referenc	e Strea	ms
	January 2015		Do	wn Sec	tion (G	D)		Unmine	ed (LM)	)
			&	Up Sec	tion (G	U)	& ]	Mine-im <sub>l</sub>	pacted (	WB)
			G	D	G	U		LM	W	'B
Order	r Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
Non-ii	nsects									
В	Lymnaeidae	<i>Fossaria</i> sp.	0.2	0.1	0.7	0.4				
S	Sphaeriidae	Pisidium sp.			19.8	7.0				
Н	Naididae	-	12.3	10.1	6.6	2.6	0.7	7 0.4		
Insect	S									
С	Dryopidae	Helichus sp.								
	Dytiscidae	Agabus sp.	0.2	0.1						
		<i>Celina</i> sp.								
		Copelatus sp.								
		<i>Desmopachria</i> sp.								
		<i>Hydroporus</i> sp.								
		Laccophilus sp.								
		Neoporus sp.	0.4	0.3	0.1	0.1				
	Elmidae	<i>Dubiraphia</i> sp.								
		<i>Optioservus</i> sp.								
		<i>Oulimnius</i> sp.					0.2	2 0.1		
		Stenelmis sp.			0.1	0.1				
	Haliplidae	<i>Haliplus</i> sp.								
		Peltodytes sp.			< 0.1	< 0.1				
	Hydrophilidae	Anacaena sp.								
		Berosus sp.								
		<i>Cymbiodyta</i> sp.								
		Enochrus sp.								
		Helochares sp.								

**Appendix C12.** List of macroinvertebrates. Values are average % composition (n = 5) and standard error (SE).

	January 2015		<u>C</u> Do	Created wn Sec	Stream tion (GI	<u>i</u> D)	<u>Re</u> N	<b>ference</b> on-min	e Strean ed (LM	<u>ns</u> )
	-		&	Up Sec	tion (GU	J)	& Mi	ine-imp	acted (V	WB)
			G	D	G	Ĵ	LN	Λ	W	B
Order	r Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
С	Hydrophilidae	Hydrochus sp.								
		Paracymus sp.								
		Tropisternus sp.								
	Psephenidae	<i>Ectopria</i> sp.			0.2	0.2	1.3	0.4		
D	Ceratopogonidae	Alluaudomyia sp.								
		Atrichopogon sp.	< 0.1	< 0.1						
		<i>Bezzia/Palpomyia</i> sp.								
		Ceratopogon sp.	< 0.1	< 0.1			1.6	0.7		
		Culicoides sp.	0.1	0.1	0.2	0.1				
		<i>Monohelea</i> sp.								
		Dasyhelea sp.								
		<i>Probezzia</i> sp.	0.2	0.2	1.6	0.9	1.8	0.5		
		Sphaeromias sp.	0.8	0.5	2.0	0.9				
D	Chironomidae	Chironominae	3.2	0.4	6.9	4.3	10.1	2.9		
		Corynoneura sp.	3.2	1.8	3.9	2.3	0.8	0.7		
		Orthocladiinae	6.2	2.5	8.8	1.2	10.6	3.3	12.5	11.2
		Stempellina sp.					0.7	0.4		
		Tanypodinae	24.5	6.3	19.5	4.9	3.2	1.3	79.2	11.2
		Tanytarsini (tribe)	40.7	11.2	15.2	4.0	1.8	0.7		
	Culicidae	Anopheles sp.								
		Haemagogus sp.								
	Dixidae	Dixa sp.					0.1	0.1		
	Dolichopodidae	dolichopodid genus A								
	Empididae	empidid genus A <i>Chelifera</i> sp.					0.1	0.1		

Appendix C12. (cont.)

	I 2016		<u>(</u>	<u>Created</u>	Stream	<u>n</u>	<u>Re</u>	ference	e Stream	<u>ms</u>
	January 2015		Do	own Sec	ction (G	D)	N	on-min	ied (LM	l)
			&	Up Sec	tion (G	U)	<u>&amp; M</u>	ine-imp	bacted (	WB)
			G	D	G	U		M	W	/B
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE
D	Empididae	Hemerodromia sp.			< 0.1	< 0.1				
		<i>Neoplasta</i> sp.								
	Ephydridae	ephydrid genus A								
		<i>Scatella</i> sp.								
	Psychodidae	Pericoma sp.								
	Sciaridae	sciarid genus A			< 0.1	< 0.1				
	Sciomyzidae	Sepedon sp.			< 0.1	< 0.1				
	Simuliidae	Prosimulium sp.					5.8	2.5		
		<i>Simulium</i> sp.	0.4	0.3	0.6	0.6				
		Stegopterna sp.	0.4	0.2	0.7	0.6				
	Stratiomyidae	Caloparyphus sp.			0.2	0.1				
		Nemotelus sp.								
		Stratiomys sp.								
	Syrphidae	<i>Eristalis</i> sp.								
	Tabanidae	Chrysops sp.	< 0.1	< 0.1	1.0	0.3				
		<i>Tabanus</i> sp.								
	Tipulidae	Dicranota sp.					0.5	0.3		
		Helius sp.			< 0.1	< 0.1				
		Hexatoma sp.					1.4	0.6		
		Leptotarsus sp.								
		<i>Limnophila</i> sp.			< 0.1	< 0.1	0.1	0.1		
		Limonia sp.								
		Molophilus sp.								
		Ormosia sp.	0.1	0.1						
		Pedicia sp.								

Appendix C12. (cont.)

<u></u>	Ianuary 2015		<u>(</u>	Created	Stream	<u>n</u> D)	<u>Re</u> N	ference	e Strea	<u>ms</u>
	bullauty 2013		&	Up Sec	tion (G	U)	& M	ine-imr	acted (	WB)
			G	D	G	Ú	LN	A I	W	VB
Order	- Family	Sub-family / Genus	$\overline{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE
D	Tipulidae	<i>Pilaria</i> sp.			0.5	0.2				
		Pseudolimnophila sp.			0.4	0.2	1.0	0.5		
		<i>Tipula</i> sp.	< 0.1	< 0.1	0.1	< 0.1	0.1	0.1		
E	Ameletidae	Ameletus sp.	2.6	1.9	5.8	1.8	3.2	1.2		
	Baetidae	Baetis sp.	2.1	1.8	0.6	0.3				
		Procloeon sp.								
	Ephemerellidae	Attenella sp.								
		<i>Drunella</i> sp.								
		<i>Ephemerella</i> sp.					14.5	2.1		
		<i>Eurylophella</i> sp.					0.1	0.1		
	Ephemeridae	<i>Ephemera</i> sp.								
	Heptageniidae	<i>Epeorus</i> sp.					7.9	1.1		
		Maccaffertium sp.					1.0	0.3		
		Stenacron sp.								
	Leptophlebiidae	<i>Habrophlebia</i> sp.								
		Paraleptophlebia sp.					11.2	1.6		
	Siphlonuridae	Siphlonurus sp.								
Μ	Corydalidae	Chauliodes sp.								
		Nigronia sp.					0.1	0.1		
	Sialidae	Sialis sp.			0.2	0.1				
0	Aeshnidae	Aeshna sp.								
		<i>Boyeria</i> sp.								
	Calopterygidae	<i>Calopteryx</i> sp.			< 0.1	< 0.1				
	Coenagrionidae	Amphiagrion sp.								
		<i>Argia</i> sp.			0.7	0.5				

Appendix C12. (cont.)

January 2015		<u>C</u> Do	reated wn Sec	Strean	<u>1</u> D)	Reference Streams					
cultury 2010			&	Up Sec	tion (G	 ຫ	& M	& Mine-impacted (WB)			
		-	G	D	GÚ		LN	LM		WB	
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
0	Coenagrionidae	<i>Enallagma</i> sp.									
	Cordulegastridae	Cordulegaster sp.			< 0.1	< 0.1					
	Gomphidae	Stylogomphus sp.			< 0.1	< 0.1	1.2	0.6			
	Libellulidae	prob. <i>Leucorrhinia</i> sp.									
		<i>Leucorrhinia</i> sp.									
		<i>Pentala</i> sp.									
		Sympetrum sp.									
Р	Capniidae	Allocapnia sp.	0.3	0.2	0.4	0.2	4.2	0.7			
	Chloroperlidae	<i>Haploperla</i> sp.					3.1	0.9			
		<i>Sweltsa</i> sp.									
	Leuctridae	Leuctra sp.			0.1	0.1	0.2	0.2			
	Nemouridae	Amphinemura sp.	1.2	0.6	0.4	0.3					
		Ostrocerca sp.									
		Prostoia sp.									
		<i>Soyedina</i> sp.					3.3	0.9			
	Peltoperlidae	<i>Peltoperla</i> sp.					0.1	0.1			
	Perlidae	Acroneuria sp.									
		Eccoptura sp.					0.1	0.1			
	Perlodidae	<i>Clioperla</i> sp.			0.1	0.1	0.1	0.1			
		<i>Diploperla</i> sp.					0.5	0.5			
		Isoperla sp.					0.4	0.3			
		Malirekus sp.					2.6	1.1			
		Remenus sp.									
		Yugus sp.									
Т	Glossosomatidae	Agapetus sp.									

Appendix C12. (cont.)

January 2015		<u>(</u>	Created	Stream	<u>n</u> D)	<u>Re</u>	Reference Streams				
Junuary 2013			&	Un Sec	tion (G	U	& Mi	& Mine-impacted (WB)			
-				GD		GU		LM		WB	
Order	Family	Sub-family / Genus	$\bar{x}$	SE	$\bar{x}$	SE	$\overline{x}$	SE	$\bar{x}$	SE	
Т	Goeridae	Goera sp.									
	Hydropsychidae	<i>Cheumatopsyche</i> sp.					0.4	0.3			
		Diplectrona sp.			0.3	0.2	1.2	0.9			
		Hydropsyche sp.			< 0.1	< 0.1					
	Hydroptilidae	<i>Hydroptila</i> sp.									
	• •	<i>Oxyethira</i> sp.									
	Lepidostomatidae	Lepidostoma sp.									
	Leptoceridae	Oecetis sp.									
	Limnephilidae	Ironoquia sp.	0.2	0.1	0.6	0.4					
		Pycnopsyche sp.					0.1	0.1	8.3	7.5	
	Molannidae	Molanna sp.									
	Philopotamidae	<i>Chimarra</i> sp.			0.6	0.6					
	-	Dolophilodes sp.					0.7	0.4			
		Wormaldia sp.			0.3	0.3	0.4	0.3			
	Phryganidae	Ptilostomis sp.	0.2	0.1	0.8	0.1					
	Polycentropodidae	Polycentropus sp.					0.8	0.6			
	Psychomyiidae	<i>Lype</i> sp.									
	Rhyacophilidae	<i>Rhyacophila</i> sp.					0.6	0.3			
	Thremmatidae	<i>Neophylax</i> sp.					0.3	0.3			
Total Richness / EPT Richness and Abundance		25/6	6,569	44/12	3,793	45/24	883	3/1	7		
Average Total Richness and SE $(n = 5)$			16.2	1.1	24.0	0.6	26.2	3.3	1.2	0.4	
Average EPT Richness and SE $(n = 5)$			4.6	0.7	6.0	1.2	13.2	1.6	0.2	0.2	
B = Ba	asommatophora, C =	Coleoptera, D = Diptera	$\overline{\mathbf{h}}, \overline{E} = \mathbf{E}$	phemer	optera,	H = Hap	lotaxida, 1	$M = M\epsilon$	galopter	a,	
O = O	donata, $P = Plecopte$	era, $S = $ Sphaeriida, $T = $ T	richop	tera		_					

Appendix C12. (cont.)

## APPENDIX D

## MONTHLY ENVIRONMENTAL VARIABLES

## ABSTRACT

The purpose of Appendix D is to help the reader compare the environmental performance at each stream site with monthly values for physico-chemical parameters, substrate constituents (food and habitat), and proxies for primary production (photosynthesis).

		Created Stream				Reference Streams				
		Down Section		Up Section		Unmined		Mine-impacted		
Variable	Month	$\overline{x}$ SE		<u> </u>	SE	$\frac{1}{\bar{\chi}}$	SE	$\frac{\overline{x}}{\overline{x}}$	SE	
Aquatic Vegetation	1	20	~1		~1	20		20		
$(mg AFDM/m^2)$	FEB/14	18.67	7.40	21.89	6.40	0.00	0.00	0.00	0.00	
(Macrophytes)	MAR	42.18	18.60	200.72	63.92	0.00	0.00	0.00	0.00	
	APR	69.61	14.47	165.92	70.90	0.00	0.00	0.00	0.00	
	MAY	57.24	9.71	80.84	20.37	0.00	0.00	0.00	0.00	
	JUN	15.66	5.19	38.96	12.59	0.00	0.00	0.00	0.00	
	JUL	30.53	14.06	35.30	12.21	0.00	0.00	0.00	0.00	
	AUG	52.32	26.84	116.65	50.72	0.00	0.00	0.00	0.00	
	SEP	124.75	44.75	94.78	33.64	0.00	0.00	0.00	0.00	
	OCT	18.16	3.93	57.56	24.36	0.00	0.00	0.00	0.00	
	NOV	8.38	3.82	40.94	19.90	0.00	0.00	0.00	0.00	
	DEC	32.68	15.82	79.27	38.72	0.00	0.00	0.00	0.00	
	JAN/15	30.61	14.84	36.70	7.71	0.00	0.00	0.00	0.00	
Leaf Detritus										
$(mg AFDM/m^2)$	FEB/14	1.32	0.83	0.40	0.24	7.14	5.07	8.12	7.14	
( C	MAR	1.60	1.09	2.51	2.51	34.51	18.91	1.19	0.89	
	APR	0.92	0.44	0.22	0.09	27.12	5.91	10.21	1.55	
	MAY	0.00	0.00	0.25	0.22	2.89	1.53	7.76	6.40	
	JUN	0.00	0.00	0.07	0.07	0.00	0.00	0.24	0.16	
	JUL	0.00	0.00	1.57	1.57	0.68	0.32	1.68	0.87	
	AUG	0.00	0.00	0.14	0.14	1.67	0.96	2.25	0.79	
	SEP	0.40	0.40	1.17	0.69	8.90	4.26	19.30	11.27	
	OCT	0.56	0.22	0.57	0.50	127.77	100.97	4.60	1.55	
	NOV	1.39	0.47	10.05	5.92	46.68	20.19	8.59	4.66	
	DEC	0.25	0.25	14.04	8.33	45.87	21.93	20.61	4.93	
	JAN/15	0.11	0.11	1.60	0.86	35.62	18.16	5.93	2.11	

**Appendix D1.** Environmental variables. Values are averages (n = 5) and standard error (SE).

		Created Stream				Reference Streams				
		Down Section Up :		Up Sec	ction	Unmined		Mine-impacted		
		GD		GL	J	LN	1	WB		
Variable	Month	$\overline{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	
Woody Debris										
$(mg AFDM/m^2)$	FEB/14	2.17	1.25	2.04	1.57	1.58	0.96	0.48	0.39	
	MAR	3.85	3.85	1.23	0.66	9.49	5.16	1.19	0.99	
	APR	2.99	1.83	0.23	0.23	14.88	4.64	4.38	2.85	
	MAY	0.00	0.00	0.00	0.00	1.11	0.53	1.08	0.46	
	JUN	0.00	0.00	0.71	0.71	17.15	6.82	0.51	0.39	
	JUL	0.00	0.00	0.00	0.00	13.04	2.53	22.83	14.57	
	AUG	0.94	0.94	1.02	1.02	12.68	4.28	3.77	1.20	
	SEP	0.91	0.91	14.72	13.49	27.23	7.01	7.61	7.37	
	OCT	0.00	0.00	2.17	2.17	5.31	2.16	0.04	0.04	
	NOV	3.83	3.17	0.03	0.03	19.95	15.47	0.15	0.15	
	DEC	0.00	0.00	2.69	2.69	6.00	1.50	7.05	6.63	
	JAN/15	0.00	0.00	0.00	0.00	13.76	3.71	1.86	1.86	
CBOM										
$(mg AFDM/m^2)$	FEB/14	24.36	6.68	19.07	3.41	5.19	1.94	2.26	0.90	
	MAR	44.58	12.96	64.69	27.09	18.70	7.66	3.24	1.06	
	APR	36.85	6.65	53.71	7.60	37.87	11.39	3.88	1.48	
	MAY	50.35	21.22	91.53	11.61	22.33	7.62	32.49	11.82	
	JUN	31.80	10.13	42.43	8.79	48.50	23.28	4.61	1.95	
	JUL	56.94	15.72	40.85	10.25	36.51	4.87	9.29	4.73	
	AUG	103.24	42.18	49.92	23.27	13.33	1.45	6.21	1.08	
	SEP	129.86	70.07	54.44	20.29	30.40	5.31	5.77	1.29	
	OCT	82.93	39.86	63.34	43.41	17.57	4.20	3.19	0.90	
	NOV	32.79	10.76	44.43	16.43	10.95	3.21	2.69	0.72	
	DEC	52.91	11.48	69.17	7.81	9.39	1.07	3.92	1.30	
	JAN/15	66.17	10.74	50.93	17.44	18.21	8.56	1.76	0.72	

**Appendix D2.** Environmental variables. Values are averages (n = 5) and standard error (SE).
		Created Stream				Reference Streams			
	-	Down S	ection	Up See	ction	ion Unmined LM		Mine-impacted WB	
	_	GI	)	GU	J				
Variable	Month	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
FBOM									
$(mg AFDM/m^2)$	FEB/14	6.97	1.18	43.36	14.48	3.00	0.60	1.48	0.48
	MAR	19.75	5.48	22.10	12.13	0.85	0.23	0.26	0.06
	APR	52.26	14.20	14.39	2.78	2.50	0.78	0.50	0.13
	MAY	58.24	12.79	57.68	13.26	0.60	0.08	4.77	1.36
	JUN	30.82	5.00	39.21	12.56	8.21	2.84	1.43	0.30
	JUL	83.31	17.35	54.92	13.64	10.53	1.40	1.78	0.37
	AUG	98.43	20.64	31.47	8.53	5.96	0.73	1.42	0.23
	SEP	60.59	21.10	74.42	54.49	19.01	3.71	2.33	0.44
	OCT	28.67	4.28	79.93	67.71	9.24	3.15	2.35	0.93
	NOV	33.88	7.04	21.31	7.04	9.02	2.82	0.80	0.15
	DEC	31.63	4.42	26.94	9.67	5.94	1.41	1.51	0.51
	JAN/15	26.74	2.55	30.96	15.05	4.45	1.78	0.93	0.24
UBOM									
(mg AFDM/m <sup>2</sup> )	FEB/14	4.49	0.74	7.33	3.05	0.24	0.10	0.16	0.06
	MAR	8.03	2.82	10.99	5.26	0.44	0.19	0.08	0.02
	APR	10.38	1.78	6.28	1.48	0.50	0.19	0.17	0.03
	MAY	13.10	3.13	15.40	5.48	0.27	0.05	0.66	0.30
	JUN	7.50	2.05	6.44	2.54	1.32	0.63	0.16	0.05
	JUL	55.85	9.58	46.03	12.01	1.10	0.67	0.20	0.08
	AUG	47.68	10.81	35.25	10.47	0.32	0.08	0.22	0.06
	SEP	21.22	7.90	13.48	8.23	8.91	5.08	0.66	0.13
	OCT	8.40	1.17	10.10	5.88	2.96	0.55	0.63	0.24
	NOV	5.78	1.30	6.48	1.27	1.51	0.32	0.30	0.07
	DEC	8.26	1.34	7.05	1.18	1.18	0.27	0.39	0.08
	JAN/15	11.60	1.26	11.43	3.41	1.54	0.49	0.25	0.04

**Appendix D3.** Environmental variables. Values are averages (n = 5) and standard error (SE).

		Created Stream			Reference Streams				
		Down Section		Up Section		Unmined		Mine-impacted	
		GD		GU		LM		WB	
Variable	Month	Х	SE	х	SE	X	SE	X	SE
Specific									
Conductivity	FEB/14	343		348		44		2549	
(µS/cm)	MAR	702		322		41		2436	
	APR	399		402		38		2644	
	MAY	496		505		81		2728	
	JUN	592		603		83		2841	
	JUL	n/a		n/a		66		2878	
	AUG	666		612		72		2727	
	SEP	632		651		69		2696	
	OCT	406		411		55		2141	
	NOV	208		223		37		691	
	DEC	322		246		48		2346	
	JAN/15	504		378		48		2604	
SO4 <sup>2-</sup>									
(mg/L)	FEB/14	79.9		78.9		5.7		1896	
	MAR	104.9		54.6		9.1		1473	
	APR	65.2		63.1		8.2		1568	
	MAY	73.7		75.8		7.4		1578	
	JUN	80.2		83.1		7.4		1579	
	JUL	n/a		n/a		7.9		1552	
	AUG	120.2		85.3		12.6		1725	
	SEP	87.3		89.2		7.8		1655	
	OCT	67.6		66.1		6.4		1473	
	NOV	43.3		39.6		3.1		419.3	
	DEC	62.9		49.1		9.2		1596	
	JAN/15	63.7		60.3		8.8		1761	

Appendix D4. Environmental variables. Values are for a single point sample.

<b>*</b> *		Created Stream				Reference Streams			
		Down Section GD		Up Section GU		Unmined LM		Mine-impacted WB	
Variable	Month	Х	SE	X	SE	Х	SE	X	SE
pН									
-	FEB/14	7.46		7.02		5.77		5.99	
	MAR	6.37		6.92		5.49		5.38	
	APR	7.12		6.91		5.57		5.93	
	MAY	8.18		7.65		6.82		6.27	
	JUN	7.49		7.37		7.62		7.52	
	JUL	n/a		n/a		7.05		6.48	
	AUG	7.15		7.22		7.23		6.88	
	SEP	7.76		7.64		7.56		7.23	
	OCT	7.54		7.52		7.41		7.04	
	NOV	8.63		7.49		7.57		7.25	
	DEC	7.98		8.28		8.35		7.76	
	JAN/15	5.99		6.05		5.78		5.97	
Periphyton									
$(mg AFDM /m^2)$	FEB/14	0.158		0.167		0.045		0.066	
	MAR	0.182		0.154		0.043		0.085	
	APR	0.215		0.157		0.037		0.115	
	MAY	0.152		0.150		0.035		0.126	
	JUN	0.335		0.117		0.044		0.135	
	JUL	0.401		0.130		0.034		0.125	
	AUG	0.188		0.110		0.050		0.103	
	SEP	0.232		0.125		0.076		0.071	
	OCT	0.197		0.385		0.075		0.111	
	NOV	0.201		0.153		0.057		0.072	
	DEC	0.193		0.149		0.071		0.095	
	JAN/15	0.301		0.161		0.044		0.102	

Appendix D5. Environmental variables. Values are for a single point sample.

		Created Stream			Reference Streams				
	_	Down SectionUp SectionGDGU		Unmined		Mine-impacted			
				GU		LM		WB	
Variable	Month	Х	SE	Х	SE	Х	SE	X	SE
Chlorophyll a									
$(\mu g/cm^2)$	FEB/14	0.084 0.161		0.127 0.097		0.061 0.090		0.018 0.016	
	MAR								
	APR	1.550		0.138		0.036		0.017	
	MAY	0.229		0.048		0.042		0.243	
	JUN	0.198		0.064		0.017		0.324	
	JUL	0.468 0.106		0.020 0.152		0.045 0.026		0.108 0.068	
	AUG								
	SEP 0.598			0.078	0.078 0.073		0.014 0.053		
	OCT	0.381	0.381						
	NOV	0.489		0.158		0.111		0.019	
	DEC	0.341		0.141		0.039	0.039		
	JAN/15	0.136		0.153		0.046		0.142	

Appendix D6. Environmental variables. Values are for a single aggregate sample.

#### APPENDIX E

### PROMINENT BENTHIC MACROINVERTEBRATES UNDERLYING THE METRICS DURING APRIL (SPRING), JULY (SUMMER), OCTOBER (AUTUMN) AND JANUARY (WINTER)

#### ABSTRACT

The purpose of Appendix E is to help the reader understand which generic-level macroinvertebrates are most prominently factored into the value derived for each metric.

#### Benthic macroinvertebrate community prominent taxa (seasonal)

#### *Community structure*

Density—April (spring). In the up section (GU) of the created stream (GC), the true flies (Diptera) were the majority of the density with  $6,548.9 \pm 784.1$  individuals m<sup>-2</sup> and a relative abundance of  $52.2\% \pm 4.0$ , followed by the stoneflies (Plecoptera) with 2,244.4  $\pm$  756.8 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 18.3%  $\pm$  5.2, and the bivalves (Sphaeriida) with  $1,291.1 \pm 831.9$  individuals  $\cdot m^{-2}$  and a relative abundance of  $9.3\% \pm 5.8$ . Individual taxa that showed the greatest densities were the non-biting midges from the sub-family Tanypodinae with 2,733.3  $\pm$  336.2 individuals m<sup>-2</sup> and a relative abundance of  $21.9\% \pm 2.0$ , followed by the stonefly Amphinemura with  $1,897.8 \pm 653.4$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 15.4% ± 4.5, and the pea clam *Pisidium* with  $1,291.1 \pm 831.9$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $9.3\% \pm 5.8$ . In the down section (GD) of the created stream (GC), the true flies (Diptera) were the majority of the density with 2,551.1  $\pm$  789.6 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 39.2%  $\pm$  6.8, followed by the snails (Basommatophora) with  $1,688.9 \pm 822.7$  individuals  $\cdot m^{-2}$  and a relative abundance of 22.1%  $\pm$  9.4, and the stoneflies (Plecoptera) with 997.8  $\pm$  361.5 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 20.1% ± 8.7. Individual taxa that showed the greatest densities were the snail *Fossaria* with 1,689.0  $\pm$  822.7 individuals m<sup>-2</sup> and a relative abundance of  $22.1\% \pm 9.4$ , followed by the non-biting midges from the subfamily Tanypodinae with  $1,424.0 \pm 396.4$  individuals  $m^{-2}$  and a relative abundance of  $22.3\% \pm 4.8$ , and the stonefly Amphinemura with 704.0  $\pm$  326.5 individuals m<sup>-2</sup> and a relative abundance of  $12.1\% \pm 5.3$ . In the unmined stream (LM), the mayflies

(Ephemeroptera) were the majority of the density with  $460.0 \pm 100.3$  individuals·m<sup>-2</sup> and a relative abundance of  $42.4\% \pm 2.7$ , followed by the true flies (Diptera) with  $224.4 \pm 59.4$  individuals·m<sup>-2</sup> and a relative abundance of  $19.2\% \pm 3.4$ , and the stoneflies (Plecoptera) with  $222.2 \pm 42.5$  individuals·m<sup>-2</sup> and a relative abundance of  $20.5\% \pm 1.4$ . Individual taxa that showed the greatest densities were the mayfly *Ephemerella* with  $257.8 \pm 66.1$  individuals·m<sup>-2</sup> and a relative abundance of  $24.7\% \pm 3.4$ , followed by the mayfly *Epeorus* with  $140.0 \pm 45.1$  individuals·m<sup>-2</sup> and a relative abundance of  $12.3\% \pm 3.3$ , and the beetle *Ectopria* with  $106.7 \pm 26.4$  individuals·m<sup>-2</sup> and a relative abundance of  $9.4\% \pm 1.3$ .

The sampling effort at the mine-impacted stream netted a total of 21 individuals comprised of true flies from the sub-family Chironominae (Diptera: Chironomidae).

*Density*—July (summer). In the up section (GU) of the created stream (GC), the snails (Basommatophora) were the majority of the density with 975.6 ± 443.9 individuals·m<sup>-2</sup> and a relative abundance of  $38.5\% \pm 12.0$ , followed by the aquatic worms (Haplotaxida) with  $717.8 \pm 684.6$  individuals·m<sup>-2</sup> and a relative abundance of  $14.4\% \pm 8.6$ , and the true flies (Diptera) with  $655.6 \pm 407.3$  individuals·m<sup>-2</sup> and a relative abundance of  $24.9\% \pm 6.2$ . Individual taxa that showed the greatest densities were the snail *Fossaria* with  $975.6 \pm 443.9$  individuals·m<sup>-2</sup> and a relative abundance of  $38.5\% \pm 12.0$ , followed by the naidid aquatic worms with  $717.8 \pm 684.6$  individuals·m<sup>-2</sup> and a relative abundance of  $38.5\% \pm 12.0$ , followed by the naidid aquatic worms with  $717.8 \pm 684.6$  individuals·m<sup>-2</sup> and a relative abundance of  $14.4\% \pm 8.6$ , and the non-biting midges in the sub-family Chironominae with  $255.6 \pm 231.2$  individuals·m<sup>-2</sup> and a relative abundance of  $4.3\% \pm 2.8$ . In the down section (GD) of the created stream (GC), the snails (Basommatophora)

were the majority of the density with  $84.4 \pm 70.6$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $28.5\% \pm 12.1$ , followed by the beetles (Coleoptera) with  $42.2 \pm 13.3$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 35.6% ± 11.8, and the true flies (Diptera) with  $28.9 \pm 9.0$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $26.8\% \pm 10.2$ . Individual taxa that showed the greatest densities were the snail Fossaria with  $84.4 \pm 70.6$  individuals m<sup>-</sup>  $^2$  and a relative abundance of 28.5%  $\pm$  12.1, followed by the beetle *Stenelmis* with 22.2  $\pm$ 15.3 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 8.3% ± 5.2, and the deer fly *Chrysops* with  $15.6 \pm 8.3$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $15.0\% \pm 6.7$ . In the unmined stream (LM), the true flies (Diptera) were the majority of the density with  $1,115.6 \pm$ 249.5 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 67.7% ± 8.8, followed by the mayflies (Ephemeroptera) with  $157.8 \pm 51.8$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $11.2\% \pm$ 4.3, and the beetles (Coleoptera) with  $113.3 \pm 62.6$  individuals  $\cdot m^{-2}$  and a relative abundance of  $8.2\% \pm 4.7$ . Individual taxa that showed the greatest densities were the nonbiting midges in the sub-family Chironominae with 964.4  $\pm$  250.4 individuals m<sup>-2</sup> and a relative abundance of 57.9%  $\pm$  10.1, followed by the mayfly *Ephemera* with 80.0  $\pm$  35.9 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 5.7% ± 2.8, and the non-biting midges in the sub-family Tanypodinae with 71.1  $\pm$  15.9 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $4.5\% \pm 1.0$ .

The sampling effort at the mine-impacted stream netted a total of 21 individuals comprised of true flies from the sub-family Chironominae (Diptera: Chironomidae). Excluded from the mine-impacted samples were 9 individuals of the stonefly *Amphinemura* and 1 individual of the caddisfly *Diplectrona*.

**Density**—**October (autumn).** In the up section (GU) of the created stream (GC), the bivalves (Sphaeriida) were the majority of the density with  $1,791.1 \pm 1,024.7$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 35.2% ± 13.3, followed by the true flies (Diptera) with 671.1  $\pm$  154.1 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 21.3%  $\pm$  5.6, and the snails (Basommatophora) with 573.3  $\pm$  371.9 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $14.7\% \pm 7.7$ . Individual taxa that showed the greatest densities were the pea clam *Pisidium* with 1.791.1  $\pm$  1.024.7 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 35.2%  $\pm$  13.3, the snail *Fossaria* with 573.3  $\pm$  371.9 individuals m<sup>-2</sup> and a relative abundance of 14.7%  $\pm$  7.7, and the non-biting midges in the sub-family Tanypodinae with 255.6  $\pm$  61.5 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 7.9% ± 2.6. In the down section (GD) of the created stream (GC), the true flies (Diptera) were the majority of the density with  $868.9 \pm$ 122.2 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 69.2% ± 3.5, followed by the beetles (Coleoptera) with 122.2  $\pm$  54.4 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 8.7%  $\pm$  3.5, and the caddisflies (Trichoptera) with  $106.7 \pm 25.5$  individuals  $\cdot m^{-2}$  and a relative abundance of  $9.8\% \pm 3.4$ . Individual taxa that showed the greatest densities were the nonbiting midges in the sub-family Tanypodinae with  $211.1 \pm 85.1$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $14.8\% \pm 4.1$ , followed by the crane fly *Tipula* with  $211.1 \pm 125.4$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 19.4%  $\pm$  10.9, and the non-biting midges in the tribe Tanytarsini with  $195.6 \pm 98.1$  individuals  $\cdot m^{-2}$  and a relative abundance of 15.0% $\pm$  6.9. In the unmined stream (LM), the true flies (Diptera) were the majority of the density with 1,108.9  $\pm$  448.5 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 52.1%  $\pm$  5.5. followed by the mayflies (Ephemeroptera) with  $280.0 \pm 102.3$  individuals m<sup>-2</sup> and a

relative abundance of  $15.6\% \pm 3.8$ , and the caddisflies (Trichoptera) with  $266.7 \pm 185.1$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $9.4\% \pm 3.1$ . Individual taxa that showed the greatest relative abundance were the non-biting midges in the sub-family Orthocladiinae with  $315.6 \pm 211.1$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $12.5\% \pm 3.1$ , followed by the crane fly *Hexatoma* with  $251.1 \pm 122.0$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $13.7\% \pm 5.3$ , and the mayfly *Paraleptophlebia* with  $171.1 \pm 57.4$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $8.5\% \pm 1.5$ .

The sampling effort at the mine-impacted stream netted a total of 21 individuals comprised of true flies from the sub-family Chironominae (Diptera: Chironomidae). Excluded from the mine-impacted samples were 9 individuals of the stonefly *Amphinemura* and 1 individual of the caddisfly *Diplectrona*.

*Density*—January (winter). In the up section (GU) of the created stream (GC), the true flies (Diptera) were the majority of the density with 4,995.6 ± 437.7 individuals·m<sup>-2</sup> and a relative abundance of  $61.5\% \pm 6.4$ , followed by the bivalves (Sphaeriida) with 1,820.0 ± 681.5 individuals·m<sup>-2</sup> and a relative abundance of 19.8% ± 7.0, and the aquatic worms (Haplotaxida) with  $582.2 \pm 213.4$  individuals·m<sup>-2</sup> and a relative abundance of  $6.6\% \pm 2.6$ . Individual taxa that showed the greatest densities were the pea clam *Pisidium* with 1,820.0 ± 681.5 individuals·m<sup>-2</sup> and a relative abundance of 19.8% ± 7.0, followed by the non-biting midges in the sub-family Tanypodinae with 1,557.8 ± 380.4 individuals·m<sup>-2</sup> and a relative abundance of 19.5% ± 4.9, and the nonbiting midges in the tribe Tanytarsini with 1,168.9 ± 246.5 individuals·m<sup>-2</sup> and a relative abundance of 15.2% ± 4.0. In the down section (GD) of the created stream (GC), the true

flies (Diptera) were the majority of the density with  $12,951.1 \pm 6,469.5$  individuals  $\cdot m^{-2}$ and a relative abundance of  $79.9\% \pm 9.0$ , followed by the aquatic worms (Haplotaxida) with  $871.1 \pm 704.2$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $12.3\% \pm 10.1$ , and the mayflies (Ephemeroptera) with  $311.1 \pm 113.3$  individuals  $\cdot m^{-2}$  and a relative abundance of  $4.7\% \pm 2.4$ . Individual taxa that showed the greatest densities were the non-biting midges in the tribe Tanytarsini with  $8,391.1 \pm 5,157.8$  individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of  $40.7\% \pm 11.2$ , followed by the non-biting midges in the sub-family Tanypodinae with 2,691.1  $\pm$  794.1 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 24.5%  $\pm$  6.3, and the naided aquatic worms with 871.1  $\pm$  704.2 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 12.3%  $\pm$ 10.1. In the unmined stream (LM), the true flies (Diptera) were the majority of the density with 880.0  $\pm$  359.3 individuals m<sup>-2</sup> and a relative abundance of 39.8%  $\pm$  4.7, followed by the mayflies (Ephemeroptera) with 646.7  $\pm$  165.3 individuals m<sup>-2</sup> and a relative abundance of  $37.9\% \pm 5.0$ , and the stoneflies (Plecoptera) with  $280.0 \pm 84.0$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 14.4% ± 1.8. Individual taxa that showed the greatest densities were the non-biting midges in the sub-family Chironominae with 246.7  $\pm$  123.6 individuals·m<sup>-2</sup> and a relative abundance of 10.1%  $\pm$  2.9, followed by the nonbiting midges in the sub-family Orthocladiinae with  $233.3 \pm 109.1$  individuals m<sup>-2</sup> and a relative abundance of  $10.6\% \pm 3.3$ , and the mayfly *Ephemerella* with  $231.1 \pm 45.2$ individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 14.5%  $\pm$  2.1.

The sampling effort at the mine-impacted stream netted a total of 21 individuals comprised of true flies from the sub-family Chironominae (Diptera: Chironomidae). Taxa

excluded from the mine-impacted samples were 9 individuals of the stonefly *Amphinemura* and 1 individual of the caddisfly *Diplectrona*.

*Total richness*—April (spring). In the up section (GU) of the created stream (GC), richness was greatest among true flies with  $16.6 \pm 0.4$  taxa and a relative richness of  $49.9\% \pm 2.3$ , followed by stoneflies with  $4.6 \pm 0.4$  taxa ( $13.9\% \pm 1.5$ ), and caddisflies with  $4.6 \pm 0.4$  taxa ( $11.9\% \pm 1.4$ ). In the down section (GD) of the created stream (GC), richness was greatest among true flies with  $11.2 \pm 1.3$  taxa and a relative richness of  $52.5\% \pm 3.3$ , followed by stoneflies with  $2.4 \pm 0.2$  taxa ( $11.6\% \pm 1.1$ ), and caddisflies with  $2.0 \pm < 0.1$  taxa ( $9.9\% \pm 1.2$ ). In the unmined stream (LM), richness was greatest among true flies with  $8.0 \pm 1.3$  taxa and a relative richness of  $32.6\% \pm 2.7$ , followed by stoneflies with  $6.0 \pm 0.9$  taxa ( $25.3\% \pm 2.9$ ), and mayflies with  $4.6 \pm 0.5$  taxa ( $19.9\% \pm 2.0$ ).

The sampling effort at the mine-impacted (WB) stream netted a single taxon of true flies from the sub-family Chironominae (Diptera: Chironomidae). Taxa excluded from the mine-impacted (WB) replicate samples were a genus of spring stonefly (*Amphinemura*) and a genus of netspinning caddisfly (*Diplectrona*).

*Total richness*—July (summer). In the up section (GU) of the created stream (GC), richness was greatest among true flies (Diptera) with  $6.4 \pm 0.5$  taxa and a relative richness of  $51.5\% \pm 4.6$ , followed by the beetles (Coleoptera) with  $2.6 \pm 0.8$  taxa ( $18.4\% \pm 4.0$ ), and the snails (Basommatophora) and bivalves (Sphaeriida) with  $1.0 \pm 0.0$  taxon ( $8.4\% \pm 1.3$ ) each. In the down section (GD) of the created stream (GC), richness was greatest among the beetles (Coleoptera) with  $1.6 \pm 0.2$  taxa and a relative richness of  $36.7\% \pm 4.9$ , followed by the true flies (Diptera) with  $1.6 \pm 0.4$  taxa ( $30.7\% \pm 7.8$ ), and

the snails (Basommatophora) with  $1.0 \pm 0.0$  taxon (25.3% ± 6.2). In the unmined stream (LM), richness was greatest among true flies (Diptera) with  $5.2 \pm 0.7$  taxa and a relative richness of  $34.8\% \pm 2.9$ , followed by the mayflies (Ephemeroptera) with  $2.8 \pm 0.6$  taxa ( $19.0\% \pm 4.0$ ), and the beetles (Coleoptera) with  $1.8 \pm 0.6$  taxa ( $12.1\% \pm 3.7$ ).

The sampling effort at the mine-impacted (WB) stream netted a single taxon of true flies from the sub-family Chironominae (Diptera: Chironomidae). Taxa excluded from the mine-impacted (WB) replicate samples were a genus of spring stonefly (*Amphinemura*) and a genus of netspinning caddisfly (*Diplectrona*).

*Total richness*—October (autumn). In the up section (GU) of the created stream (GC), richness was greatest among the true flies (Diptera) with  $8.6 \pm 0.9$  taxa and a relative richness of  $43.0\% \pm 3.6$ , followed by the beetles (Coleoptera) with  $2.8 \pm 1.1$  taxa (12.8% ± 4.3), and the caddisflies (Trichoptera) with  $2.6 \pm 0.7$  taxa (13.0% ± 3.6). In the down section (GD) of the created stream (GC), richness was greatest among the true flies (Diptera) with  $9.4 \pm 1.0$  taxa and a relative richness of  $59.4\% \pm 5.0$ , followed by the beetles (Coleoptera) with  $2.4 \pm 0.6$  taxa (14.6% ± 3.4), and the caddisflies (Trichoptera) with  $1.6 \pm 0.4$  taxa (10.4% ± 2.6). In the unmined stream (LM), richness was greatest among the true flies (Diptera) with  $11.4 \pm 1.2$  taxa and a relative richness of  $43.8\% \pm 3.8$ , followed by the stoneflies (Plecoptera) with  $4.0 \pm 1.0$  taxa (14.5% ± 2.1), and the caddisflies (Trichoptera) with  $3.2 \pm 0.7$  taxa (11.7% ± 1.3).

The sampling effort at the mine-impacted (WB) stream netted a single taxon of true flies from the sub-family Chironominae (Diptera: Chironomidae). Taxa excluded from the mine-impacted (WB) replicate samples were a genus of spring stonefly (*Amphinemura*) and a genus of netspinning caddisfly (*Diplectrona*).

*Total richness*—January (winter). In the up section (GU) of the created stream (GC), richness was greatest among the true flies (Diptera) with  $12.4 \pm 0.9$  taxa and a relative richness of  $51.5\% \pm 2.8$ , followed by the caddisflies (Trichoptera) with  $3.2 \pm 0.6$  taxa ( $13.4\% \pm 2.4$ ), and the mayflies (Ephemeroptera) with  $1.6 \pm 0.2$  taxa ( $6.8\% \pm 1.1$ ). In the down section (GD) of the created stream (GC), richness was greatest among the true flies (Diptera) with  $9.0 \pm 0.7$  taxa and a relative richness of  $55.5\% \pm 2.0$ , followed by the mayflies (Ephemeroptera) with  $1.6 \pm 0.2$  taxa ( $10.1\% \pm 2.7$ ), and the caddisflies (Trichoptera) with  $1.6 \pm 0.2$  taxa ( $9.8\% \pm 1.3$ ). In the unmined stream (LM), richness was greatest among the true flies (Diptera) with  $1.6 \pm 0.2$  taxa ( $9.8\% \pm 1.3$ ). In the unmined stream (LM), richness of  $38.5\% \pm 1.4$ , followed by the mayflies (Ephemeroptera) with  $10.2 \pm 1.5$  taxa and a relative richness of  $38.5\% \pm 1.4$ , followed by the mayflies (Ephemeroptera) with  $4.8 \pm 0.6$  taxa ( $18.7\% \pm 1.6$ ).

The sampling effort at the mine-impacted (WB) stream netted a single taxon of true flies from the sub-family Chironominae (Diptera: Chironomidae). Taxa excluded from the mine-impacted (WB) replicate samples were a genus of spring stonefly (*Amphinemura*) and a genus of netspinning caddisfly (*Diplectrona*).

#### Community composition

*Percent benthic insects*—April (spring). In the up section (GU) of the created stream (GC), the non-insect taxa were the pea clam *Pisidium* with 1,2911.1 ± 831.9 individuals·m<sup>-2</sup> and a relative abundance of 9.3% ± 5.8, the snail *Fossaria* with 424.4 ± 210.9 individuals·m<sup>-2</sup> ( $3.6\% \pm 1.8$ ), and the naidid aquatic worms with 240.0 ± 119.6 individuals·m<sup>-2</sup> ( $1.8\% \pm 0.8$ ). In the down section (GD) of the created stream (GC), the non-insect taxa was *Fossaria* with 1,688.9 ± 822.7 individuals·m<sup>-2</sup> ( $22.1\% \pm 9.4$ ), and the naidids with 40.0 ± 26.7 individuals  $\cdot$  m<sup>-2</sup> (1.0% ± 0.8). In the unmined stream (LM), the non-insect taxa was *Pisidium* with 2.2 ± 2.2 individuals  $\cdot$  m<sup>-2</sup> (0.2% ± 0.2).

*Percent benthic insects*—July (summer). In the up section (GU) of the created stream (GC), the non-insect taxa were the snail *Fossaria* with 975.6 ± 443.9 individuals·m<sup>-2</sup> and a relative abundance of  $38.5\% \pm 12.0$ , the pea clam *Pisidium* with  $240.0 \pm 138.9$  individuals·m<sup>-2</sup> ( $17.5\% \pm 9.5$ ), and the naidid aquatic worms with  $717.8 \pm$ 684.6 individuals·m<sup>-2</sup> ( $14.4\% \pm 8.6$ ). In the down section (GD) of the created stream (GC), the non-insect taxa was *Fossaria* with  $84.4 \pm 70.6$  individuals·m<sup>-2</sup> ( $28.5\% \pm 12.1$ ), and naidids with  $6.7 \pm 6.7$  individuals·m<sup>-2</sup> ( $7.5\% \pm 7.5$ ). In the unmined stream (LM), the non-insect taxa were naidids with  $22.2 \pm 11.7$  individuals·m<sup>-2</sup> ( $1.4\% \pm 0.8$ ) and *Fossaria* with  $4.4 \pm 2.7$  individuals·m<sup>-2</sup> ( $0.2\% \pm 0.2$ ).

*Percent benthic insects*—October (autumn). In the up section (GU) of the created stream (GC), the non-insect taxa were the pea clam *Pisidium* with 1,791.1  $\pm$  1,024.7 individuals·m<sup>-2</sup> and a relative abundance of 35.2%  $\pm$  13.3, the snail *Fossaria* with 573.3  $\pm$  371.9 individuals·m<sup>-2</sup> (14.7%  $\pm$  7.7), and the naidid aquatic worm (Naididae) with 2.2  $\pm$  2.2 individuals·m<sup>-2</sup> (0.1%  $\pm$  0.1). In the down section (GD) of the created stream (GC), the non-insect taxa was *Fossaria* with 71.1  $\pm$  13.0 individuals·m<sup>-2</sup> (5.6%  $\pm$  1.0), and naidids with 80.0  $\pm$  35.7 individuals·m<sup>-2</sup> (5.5%  $\pm$  1.8). In the unmined stream (LM), the non-insect taxa was *Pisidium* with 2.2  $\pm$  2.2 individuals·m<sup>-2</sup> (1.3%  $\pm$  1.3), followed by *Fossaria* with 4.4  $\pm$  4.4 individuals·m<sup>-2</sup> (0.3%  $\pm$  0.3), and naidids with 4.4  $\pm$  4.4 individuals·m<sup>-2</sup> (0.1%  $\pm$  0.1).

*Percent benthic insects*—January (winter). In the up section (GU) of the created stream (GC), the non-insect taxa were the pea clam *Pisidium* with 1,820.0 ± 681.5 individuals·m<sup>-2</sup> and a relative abundance of 19.8% ± 7.0, the naidid aquatic worms with  $582.2 \pm 213.4$  individuals·m<sup>-2</sup> (6.6% ± 2.6), and the snail *Fossaria* with  $64.4 \pm 43.9$  individuals·m<sup>-2</sup> (0.7% ± 0.4). In the down section (GD) of the created stream (GC), the non-insect taxa were the naidids with  $871.1 \pm 704.2$  individuals·m<sup>-2</sup> (12.3% ± 10.1), and *Fossaria* with  $17.8 \pm 7.5$  individuals·m<sup>-2</sup> (0.2% ± 0.1). In the unmined stream (LM), the non-insect taxa were the naidids with  $8.9 \pm 4.2$  individuals·m<sup>-2</sup> (0.7% ± 0.4).

**Percent top two dominant taxa (abundance)**—April (spring). In the up section (GU) of the created stream (GC), the two dominant taxa were the non-biting midges from the sub-family Tanypodinae with  $21.9\% \pm 2.0$  of the abundance, and the stonefly *Amphinemura* with  $15.4\% \pm 4.5$ , whereas in the down section (GD), it was the Tanypod midges with  $22.3\% \pm 4.8$  and the snail *Fossaria* with  $22.1\% \pm 9.4$ . In contrast, the two dominant taxa in the unmined stream (LM) were the mayfly *Ephemerella* with  $24.7\% \pm$ 3.4 of the abundance and the mayfly *Epeorus* with  $12.3\% \pm 3.3$ .

**Percent top two dominant taxa (abundance)**—July (summer). In the up section (GU) of the created stream (GC), the two dominant taxa were the snail *Fossaria* with  $38.5\% \pm 12.0$  of the abundance, and the pea clam *Pisidium* with  $17.5\% \pm 9.5$ , whereas in the down section (GD), it was *Fossaria* with  $46.6\% \pm 17.9$  and the beetle *Paracymus* with  $18.1\% \pm 15.5$ . In contrast, the two dominant taxa in the unmined stream (LM) were the non-biting midges from the sub-family Chironominae with  $57.9\% \pm 10.1$  of the abundance and the mayfly *Ephemera* with  $5.7\% \pm 2.8$ .

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**Percent top two dominant taxa (abundance)**—October (autumn). In the up section (GU) of the created stream (GC), the two dominant taxa were the pea clam *Pisidium* with  $35.2\% \pm 2.0$  of the abundance, and the snail *Fossaria* with  $14.7\% \pm 4.5$ , whereas in the down section (GD), it was the crane fly *Tipula* with  $19.4\% \pm 4.8$  and the non-biting midges from the tribe Tanytarsini with  $15.0\% \pm 6.9$  of the abundance. The two dominant taxa in the unmined stream (LM) were the crane fly *Hexatoma* with  $13.7\% \pm 5.3$  of the abundance and the non-biting midges in the sub-family Orthocladiinae with  $12.5\% \pm 3.1$ .

**Percent top two dominant taxa (abundance)**—January (winter). In the up section (GU) of the created stream (GC), the two dominant taxa were the pea clam *Pisidium* with 19.8%  $\pm$  7.0 of the abundance, and the non-biting midges in the sub-family Tanypodinae with 19.5%  $\pm$  4.9, whereas in the down section (GD), it was the non-biting midges from the tribe Tanytarsini with 40.7%  $\pm$  11.2 of the abundance and the non-biting midges from the sub-family Tanypodinae with 24.5%  $\pm$  6.3. In contrast, the two dominant taxa in the unmined stream (LM) were the mayfly *Ephemerella* with 14.5%  $\pm$ 2.1 of the abundance and the mayfly *Paraleptophlebia* with 11.2%  $\pm$  1.6.

*EPT richness (genera)*—April (spring). In the up section (GU) of the created stream (GC), the average EPT richness was  $10.0 \pm 0.3$  genera, and the aggregate EPT richness (sum of replicates) was 16 genera, of which two genera were the mayflies *Ameletus* and *Eurylophella*, seven genera were the stoneflies *Allocapnia, Leuctra, Amphinemura, Peltoperla, Diploperla, Isoperla,* and *Malirekus*, and seven genera were the caddisflies *Diplectrona, Hydropsyche, Ironoquia, Chimarra, Ptilostomis, Rhyacophila*, and *Neophylax*. In the down section (GD) of the created stream (GC), the

average EPT richness was  $5.8 \pm 0.4$  genera, and the aggregate EPT richness was eight genera, of which two genera were the mayflies *Ameletus* and *Baetis*, four genera were the stoneflies *Allocapnia*, *Amphinemura*, *Isoperla*, and *Malirekus*, and two genera were the caddisflies *Ironoquia* and *Ptilostomis*. In the unmined stream (LM), the average EPT richness was  $12.6 \pm 1.5$  genera, and the aggregate EPT richness was 22 genera, of which six genera were the mayflies *Ameletus*, *Ephemerella*, *Ephemera*, *Epeorus*, *Maccaffertium*, and *Paraleptophlebia*, eleven genera were the stoneflies *Allocapnia*, *Haploperla*, *Sweltsa*, *Leuctra*, *Amphinemura*, *Ostrocerca*, *Peltoperla*, *Acroneuria*, *Eccoptura*, *Isoperla*, and *Malirekus*, and five genera were the caddisflies *Diplectrona*, *Hydroptila*, *Pycnopsyche*, *Polycentropus*, and *Rhyacophila*.

*EPT richness (genera)*—July (summer). In the up section (GU) of the created stream (GC), the average EPT richness was  $0.2 \pm 0.2$  genera, and the aggregate EPT richness (sum of replicates) was one genus of the caddisfly *Diplectrona*. In the down section (GD) of the created stream (GC), the average EPT richness was  $0.2 \pm 0.2$  genera, and the aggregate EPT richness was one genus of the caddisfly *Ironoquia*. In the unmined stream (LM), the average EPT richness was  $5.0 \pm 0.9$  genera, and the aggregate EPT richness was eleven genera, of which five genera were the mayflies *Procloeon, Eurylophella, Ephemera, Stenacron,* and *Paraleptophlebia*, three genera were the stoneflies *Leuctra, Acroneuria,* and *Eccoptura,* and three genera were the caddisflies *Goera, Pycnopsyche* and *Molanna*.

*EPT richness (genera)*—October (autumn). In the up section (GU) of the created stream (GC), the average EPT richness was  $3.6 \pm 1.1$  genera, and the aggregate EPT richness (sum of replicates) was eight genera, of which one genus was the mayfly

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*Paraleptophlebia*, two genera were the stoneflies *Leuctra* and *Soyedina*, and five genera were the caddisflies *Diplectrona*, *Chimarra*, *Wormaldia*, *Ptilostomis*, and *Rhyacophila*. In the down section (GD) of the created stream (GC), the average EPT richness was  $1.8 \pm 0.4$ , and the aggregate EPT richness was five genera, of which one genus was the stonefly *Allocapnia*, and four genera were the caddisflies *Diplectrona*, *Chimarra*, *Ptilostomis*, and *Rhyacophila*. In the unmined stream (LM), EPT richness was  $10.2 \pm 2.2$  genera, and the aggregate EPT richness was 20 genera, of which five genera were the mayflies *Procloeon*, *Eurylophella*, *Ephemera*, *Maccaffertium*, and *Paraleptophlebia*, eight genera were the stoneflies *Allocapnia*, Haploperla, Leuctra, Soyedina, Acroneuria, Eccoptura, Diploperla, and *Isoperla*, and seven genera were the caddisflies *Diplectrona*, *Lype*, *Rhyacophila*.

*EPT richness (genera)*—January (winter). In the up section (GU) of the created stream (GC), the average EPT richness was  $6.0 \pm 1.2$  genera, and the aggregate EPT richness (sum of replicates) was 12 genera, of which two genera were the mayflies *Ameletus* and *Baetis*, four genera were the stoneflies *Allocapnia, Leuctra, Amphinemura,* and *Clioperla*, and six genera were the caddisflies *Diplectrona, Hydropsyche, Ironoquia, Chimarra, Wormaldia,* and *Ptilostomis.* In the down section (GD) of the created stream (GC), the average EPT richness was  $4.6 \pm 0.7$  genera, and the aggregate EPT richness was six genera, of which two genera were the mayflies *Ameletus* and *Baetis*, two genera were the stoneflies *Allocapnia* and *Baetis*, two genera were the stoneflies *Allocapnia* and *Baetis*. In the unmined stream (LM), the average EPT richness was  $13.2 \pm 1.6$  genera, and the aggregate EPT richness was 24 genera, of which six genera were the mayflies *Ameletus, Ephemerella, Eurylophella, Epeorus, Maccaffertium*, and

Paraleptophlebia, ten genera were the stoneflies Allocapnia, Haploperla, Leuctra, Soyedina, Peltoperla, Eccoptura, Clioperla, Diploperla, Isoperla, and Malirekus, and eight genera were the caddisflies Cheumatopsyche, Diplectrona, Pycnopsyche, Dolophilodes, Wormaldia, Polycentropus, Rhyacophila, and Neophylax.

*Ephemeroptera (E) richness (genera)*—April (spring). In the up section (GU) of the created stream (GC), the average Ephemeroptera richness was  $1.4 \pm 0.2$  genera with a relative richness of  $4.2\% \pm 0.8$ , and an aggregate richness (sum of replicates) of two genera that comprised *Ameletus* and *Eurylophella*. In the down section (GD) of the created stream (GC), the average Ephemeroptera richness was  $1.4 \pm 0.2$  genera with a relative richness of  $6.7\% \pm 0.9$ , and an aggregate richness of two genera that comprised *Ameletus* and *Baetis*. In the unmined stream (LM), the average Ephemeroptera richness was  $4.6 \pm 0.5$  genera with a relative richness of  $19.9\% \pm 2.0$ , and an aggregate richness of six genera that comprised *Ameletus, Ephemerella, Ephemera, Epeorus, Maccaffertium, Paraleptophlebia*.

*Ephemeroptera (E) richness (genera)*—July (summer). In the unmined stream (LM), the average Ephemeroptera richness was  $2.8 \pm 0.6$  genera with a relative richness of  $19.0\% \pm 4.0$ , and an aggregate richness (sum of replicates) of five genera that comprised *Ephemera, Stenacron, Paraleptophlebia, Eurylophella*, and *Procloeon*. There were no Ephemeroptera genera collected in the created stream (GC).

*Ephemeroptera (E) richness (genera)*—October (autumn). In the up section (GU) of the created stream (GC), the average Ephemeroptera richness was  $0.2 \pm 0.2$  genera with a relative richness of  $0.8\% \pm 0.8$ , and an aggregate richness (sum of replicates) of one genus that was *Paraleptophlebia*. There were no Ephemeroptera genera

collected in the down section (GD) of the created stream (GC). In the unmined stream (LM), the average Ephemeroptera richness was  $3.0 \pm 0.7$  with a relative richness of  $11.7\% \pm 2.8$ , and an aggregate richness of five genera that comprised *Procloeon*, *Eurylophella*, *Ephemera*, *Maccaffertium*, and *Paraleptophlebia*.

*Ephemeroptera (E) richness (genera)*—January (winter). In the up section (GU) of the created stream (GC), the average Ephemeroptera richness was  $1.6 \pm 0.2$  genera with a relative richness of  $6.8\% \pm 1.1$ , and an aggregate richness (sum of replicates) of two genera that comprised *Ameletus* and *Baetis*. In the down section (GD) of the created stream (GC), the average Ephemeroptera richness was  $1.6 \pm 0.4$  genera with a relative richness of  $10.1\% \pm 2.7$ , and an aggregate richness of two genera that comprised *Ameletus* and *Baetis*. In the average Ephemeroptera richness of two genera that comprised *Ameletus* and stream (LM), the average Ephemeroptera richness was  $5.4 \pm 0.2$  genera with a relative richness of  $21.5\% \pm 1.9$ , and an aggregate richness of six genera that comprised *Ameletus, Ephemerella, Eurylophella, Epeorus, Maccaffertium*, and *Paraleptophlebia*.

*Plecoptera (P) richness (genera)*—April (spring). In the up section (GU) of the created stream (GC), the average Plecoptera richness was  $4.6 \pm 0.4$  genera with a relative richness of  $13.9\% \pm 1.5$ , and an aggregate richness (sum of replicates) of seven genera that comprised *Allocapnia, Leuctra, Amphinemura, Peltoperla, Diploperla, Isoperla,* and *Malirekus*. In the down section (GD) of the created steam (GC), the average Plecoptera richness was  $2.4 \pm 0.2$  genera with a relative richness of  $11.6\% \pm 1.1$ , and an aggregate richness of four genera that comprised *Allocapnia, Amphinemura, Amphinemura, Isoperla,* and *Malirekus*. In the unmined stream (LM), the average Plecoptera richness was  $6.0 \pm 0.9$  genera with a relative richness of  $25.3\% \pm 2.9$ , and an aggregate richness of eleven

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genera that comprised *Allocapnia*, *Haploperla*, *Sweltsa*, *Leuctra*, *Amphinemura*, *Ostrocerca*, *Peltoperla*, *Acroneuria*, *Eccoptura*, *Isoperla*, and *Malirekus*.

*Plecoptera (P) richness (genera)*—July (summer). In the unmined stream (LM), the average Plecoptera richness was  $0.6 \pm 0.6$  genera with a relative richness of  $2.9\% \pm 2.9$ , and an aggregate richness (sum of replicates) of three genera that comprised *Leuctra*, *Acroneuria*, and *Eccoptura*. There were no Plecoptera genera collected in the created stream (GC).

*Plecoptera (P) richness (genera)*—October (autumn). In the up section (GU) of the created stream (GC), the average Plecoptera richness was  $0.8 \pm 0.5$  genera with a relative richness of  $3.6\% \pm 2.31$ , and an aggregate richness (sum of replicates) of one genus that was *Soyedina*. In the down section (GD) of the created stream (GC), the average Plecoptera richness was  $0.2 \pm 0.2$  genera with a relative richness of  $0.8\% \pm 0.8$ , and an aggregate richness of one genus that was *Allocapnia*. In the unmined stream (LM), the average Plecoptera richness was  $4.0 \pm 1.0$  genera with a relative richness of  $14.5\% \pm$ 2.1, and an aggregate richness of eight genera that comprised *Allocapnia, Haploperla, Leuctra, Soyedina, Acroneuria, Eccoptura, Diploperla*, and *Isoperla*.

*Plecoptera (P) richness (genera)*—January (winter). In the up section (GU) of the created stream (GC), the average Plecoptera richness was  $1.2 \pm 0.7$  genera with a relative richness of  $5.0\% \pm 3.1$ , and an aggregate richness (sum of replicates) of four genera that comprised *Allocapnia, Leuctra, Amphinemura,* and *Clioperla*. In the down section (GD) of the created stream (GC), the average Plecoptera richness was  $1.4 \pm 0.2$ genera with a relative richness of  $8.5\% \pm 1.2$ , and an aggregate richness of two genera that comprised *Allocapnia* and *Amphinemura*. In the unmined stream (LM), the average Plecoptera richness was  $4.8 \pm 0.6$  genera with a relative richness of  $18.7\% \pm 1.6$ , and an aggregate richness of ten genera that comprised *Allocapnia, Haploperla, Leuctra, Soyedina, Peltoperla, Eccoptura, Clioperla, Diploperla, Isoperla,* and *Malirekus*.

*Trichoptera (T) richness (genera)*—April (spring). In the up section (GU) of the created stream (GC), the average Trichoptera richness was  $4.0 \pm 0.5$  genera with a relative richness of  $11.9\% \pm 1.4$ , and an aggregate richness (sum of replicates) of seven genera that comprised *Diplectrona, Hydropsyche, Ironoquia, Chimarra, Ptilostomis, Rhyacophila,* and *Neophylax*. In the down section (GD) of the created stream (GC), the average Trichoptera richness was  $2.0 \pm < 0.1$  genera with a relative richness of  $9.9\% \pm 1.2$ , and an aggregate richness of two genera that comprised *Ironoquia* and *Ptilostomis*. In the unmined stream (LM), the average Trichoptera richness was  $2.0 \pm 0.8$  genera with a relative richness of  $7.9\% \pm 2.7$ , and an aggregate richness of five genera that comprised *Diplectrona, Hydropsyche, Polycentropus,* and *Rhyacophila.* 

*Trichoptera (T) richness*—July (summer). In the up section (GU) of the created stream (GC), the average Trichoptera richness was  $0.2 \pm 0.2$  genera with a relative richness of  $1.1\% \pm 1.1$ , and an aggregate richness (sum of replicates) of one genus that was *Diplectrona*. In the down section (GD) of the created stream (GC), the average Trichoptera richness was  $0.2 \pm 0.2$  genera with a relative richness of  $3.3\% \pm 3.3$ , and an aggregate richness of one genus that was *Ironoquia*. In the unmined stream (LM), the average Trichoptera richness was  $1.6 \pm 0.2$  genera with a relative richness of  $11.2\% \pm 1.8$ , and an aggregate richness of three genera that comprised *Goera, Pycnopsyche*, and *Molanna*.

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*Trichoptera (T) richness*—October (autumn). In the up section (GU) of the created stream (GC), the average Trichoptera richness was  $2.6 \pm 0.7$  genera with a relative richness of  $13.0\% \pm 3.6$ , and an aggregate richness (sum of replicates) of five genera that comprised *Diplectrona, Chimarra, Wormaldia, Ptilostomis,* and *Rhyacophila*. In the down section (GD) of the created stream (GC), the average Trichoptera richness was  $1.6 \pm 0.4$  genera with a relative richness of  $10.4\% \pm 2.6$ , and an aggregate richness of four genera that comprised *Diplectrona, Chimarra, Ptilostomis,* and *Rhyacophila*. In the unmined stream (LM), the average Trichoptera richness was  $3.2 \pm 0.7$  genera with a relative richness of seven genera that comprised *Diplectrona, Chimarra, Ptilostomis,* and *Rhyacophila*. In the unmined stream (LM), the average Trichoptera richness of seven genera that comprised *Diplectrona, Hydropsyche, Pycnopsyche, Wormaldia, Polycentropus, Lype,* and *Rhyacophila*.

*Trichoptera (T) richness*—January (winter). In the up section (GU) of the created stream (GC), the average Trichoptera richness was  $3.2 \pm 0.6$  genera with a relative richness of  $13.4\% \pm 2.4$ , and an aggregate richness (sum of replicates) of six genera that comprised *Diplectrona, Hydropsyche, Ironoquia, Chimarra, Wormaldia,* and *Ptilostomis*. In the down section (GD) of the created stream (GC), the average Trichoptera richness was  $1.6 \pm 0.2$  genera with a relative richness of  $9.8\% \pm 1.3$ , and an aggregate richness of two genera that comprised *Ironoquia* and *Ptilostomis*. In the unmined stream (LM), the average Trichoptera richness was  $3.0 \pm 0.9$  genera with a relative richness of  $10.8\% \pm 2.1$ , and an aggregate richness of eight genera that comprised *Cheumatopsyche, Diplectrona, Pycnopsyche, Dolophilodes, Wormaldia, Polycentropus, Rhyacophila, Neophylax*.

#### *Community diversity*

Shannon index (H) & Hill's  $N_1$  diversity—April (spring). In the up section, the Shannon index was  $2.40 \pm 0.05$  with an  $N_1$  diversity of  $11.09 \pm 0.55$  effective species. The aggregate richness was 55 taxa, 16 of which were found in only one of the five sitesamples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (8) *Atrichopogon, Bezzia/Palpomyia*, Dolichopodidae, *Neoplasta, Caloparyphus, Nemotelus, Limnophila, Pedicia*, the beetles (3) *Agabus, Haliplus*, and *Peltodytes*, the dragonflies (2) *Stylogomphus*, and prob. *Leucorrhinia*, the caddisflies (2) *Hydropsyche* and *Neophylax*) and the stonefly (1) *Peltoperla*.

In the down section, the Shannon index was  $2.03 \pm 0.20$  with an N<sub>1</sub> diversity of  $8.16 \pm 1.30$  effective species. The aggregate richness was 38 taxa, 11 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (7) *Sphaeromias, Stegopterna, Caloparyphus, Tabanus, Helius, Ormosia,* and *Pilaria*, the beetles (3) *Dubiraphia, Optioservus,* and *Berosus,* and the stoneflies (2) *Allocapnia* and *Malirekus.* 

In the unmined stream, the Shannon index was  $2.60 \pm 0.11$  with an N<sub>1</sub> diversity of  $13.80 \pm 1.35$  effective species. The aggregate richness was 46 taxa, 12 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (5) *Bezzia/Palpomyia, Stempellina, Hemerodromia, Simulium,* and *Dicranota*, the stoneflies (2) *Leuctra* and *Peltoperla*, the caddisflies (2) *Hydroptila* and *Pycnopsyche*, the beetle (1) *Optioservus,* the dobsonsfly (1) *Nigronia*, and the pea clam (1) *Pisidium*.

Shannon index (H) & Hill's  $N_1$  diversity—July (summer). In the up section, the Shannon index was  $1.50 \pm 0.17$  with an  $N_1$  diversity of  $4.74 \pm 0.82$  effective species. The aggregate richness was 30 taxa, 13 of which were found in only one of the five sitesamples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the beetles (5) *Dubiraphia, Optioservus, Helochares, Hydrochus,* and *Tropisternus*, the true flies (5) *Probezzia,* Tanytarsini, *Hemerodromia, Tabanus,* and *Pseudolimnophila,* the dragonflies (2) *Boyeria* and *Stylogomphus,* and the caddisfly (1) *Diplectrona.* 

In the down section, the Shannon index was  $1.19 \pm 0.23$  with an N<sub>1</sub> diversity of  $3.63 \pm 0.72$  effective species. The aggregate richness was 13 taxa, 8 of which were found in only one of the five site-samples (distribution rarity) and 1<sup>†</sup> of those 8 taxa also had less than 1% of the average total of individuals (abundance rarity): the beetles (3) *Neoporus, Hydrochus,* and *Tropisternus,* the true flies (3) *Probezzia<sup>†</sup>, Sepedon,* and *Caloparyphus,* the caddisfly (1) *Ironoquia,* and the naidid (1) aquatic worm.

In the unmined stream, the Shannon index was  $1.57 \pm 0.24$  with an N<sub>1</sub> diversity of  $5.49 \pm 1.60$  effective species. The aggregate richness was 27 taxa, 6 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the stoneflies (3) *Leuctra, Acroneuria* and *Eccoptura*, the true fly (1) *Bezzia/Palpomyia*, the mayfly (1) *Procloeon*, and the caddisfly (1) *Goera*.

Shannon index (H) & Hill's  $N_1$  diversity—October (autumn). In the up section, the Shannon index was  $1.83 \pm 0.30$  with an  $N_1$  diversity of  $7.36 \pm 1.93$  effective species. The aggregate richness was 41 taxa, 14 of which were found in only one of the five sitesamples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (6) *Dixa*, Dolichopodidae, *Hemerodromia*, *Sepedon*, *Stratiomys*, and *Tipula*, the beetles (3) *Agabus*, *Peltodytes*, and *Ectopria*, the caddisflies (2) *Wormaldia* and *Rhyacophila*, the mayfly (1) *Paraleptophlebia*, the dragonfly (1) prob. *Leucorrhinia*, and the aquatic worm (1) Naididae.

In the down section, the Shannon index was  $2.14 \pm 0.19$  with an N<sub>1</sub> diversity of  $9.07 \pm 1.60$  effective species. The aggregate richness was 32 taxa, 13 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (3) *Monohelea, Hemerodromia,* and *Pilaria,* the caddisflies (3) *Diplectrona, Chimarra,* and *Rhyacophila,* the beetles (2) *Stenelmis* and *Peltodytes,* the damselflies (2) *Argia* and *Enallagma,* the dragonflies (2) *Boyeria* and prob. *Leucorrhinia,* and the stonefly (1) *Allocapnia.* 

In the unmined stream, the Shannon index was  $2.75 \pm 0.07$  with an N<sub>1</sub> diversity of  $15.77 \pm 1.06$  effective species. The aggregate richness was 50 taxa, 15 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (4) *Dicranota, Leptotarsus, Limnophila,* and *Ormosia,* the stoneflies (2) *Soyedina* and *Diploperla,* the caddisflies (2) *Hydropsyche* and *Lype,* the mayfly (1) *Procloeon,* the dobsonsfly (1) *Nigronia,* the alderfly (1) *Sialis,* the dragonfly (1) *Cordulegaster,* the snail (1) *Fossaria,* the pea clam (1) *Pisidium,* and the aquatic worm (1) Naididae.

Shannon index (H) & Hill's  $N_1$  diversity—January (winter). In the up section, the Shannon index was  $2.15 \pm 0.06$  with an  $N_1$  diversity of  $8.68 \pm 0.52$  effective species. The aggregate richness was 44 taxa, 16 of which were found in only one of the five sitesamples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (6) *Hemerodromia*, Sciaridae, *Sepedon, Simulium*, *Helius*, and *Limnophila*, the beetles (3) *Neoporus*, *Peltodytes*, and *Ectopria*, the dragonflies (2) *Cordulegaster* and *Stylogomphus*, the stoneflies (2) *Leuctra* and *Clioperla*, the caddisflies (2) *Hydropsyche* and *Chimarra*, and the damselfly (1) *Calopteryx*.

In the down section, the Shannon index was  $1.45 \pm 0.12$  with an N<sub>1</sub> diversity of  $4.41 \pm 0.54$  effective species. The aggregate richness was 25 taxa, 5 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the true flies (5) *Atrichopogon, Chrysops, Ormosia,* and *Tipula*.

In the unmined stream, the Shannon index was  $2.72 \pm 0.07$  with an N<sub>1</sub> diversity of  $15.28 \pm 1.12$  effective species. The aggregate richness was 45 taxa, 11 of which were found in only one of the five site-samples (distribution rarity) and also had less than 1% of the average total of individuals (abundance rarity): the stoneflies (5) *Leuctra*, *Peltoperla*, *Eccoptura*, *Clioperla*, and *Diploperla*, the true flies (3) *Chelifera*, *Limnophila*, and *Tipula*, the caddisflies (2) *Pycnopsyche* and *Neophylax*, and the dobsonsfly (1) *Nigronia*.

Simpson index of diversity (1 - D) & Hill's N<sub>2</sub> diversity—April (spring). In the up section, the Simpson index of diversity was  $0.86 \pm 0.01$  with an N<sub>2</sub> diversity of  $7.30 \pm$ 0.34 effective species. The aggregate richness was 55 taxa, 17 of which were found in all five of the site-samples (distribution evenness) and 6<sup>†</sup> of those 17 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (10)

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Sphaeromias, Chironominae<sup>†</sup>, Corynoneura, Orthocladiinae<sup>†</sup>, Tanypodinae<sup>†</sup>,

Tanytarsini<sup>†</sup>, *Stegopterna, Chrysops, Pseudolimnophila,* and *Tipula*, the stoneflies (2) *Amphinemura*<sup>†</sup> and *Isoperla*, the mayfly (1) *Ameletus*<sup>†</sup>, the snail (1) *Fossaria*, the beetle (1) *Stenelmis*, the aquatic worm (1) Naididae, and the caddisfly (1) *Ironoquia*.

In the down section, the Simpson index of diversity was  $0.79 \pm 0.05$  with an N<sub>2</sub> diversity of  $5.57 \pm 0.87$  effective species. The aggregate richness was 38 taxa, 11 of which were found in all five of the site-samples (distribution evenness) and 6<sup>†</sup> of those 11 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (5) Chironominae, Orthocladiinae, Tanypodinae<sup>†</sup>, Tanytarsini, and *Chrysops*, the stoneflies (2) *Amphinemura*<sup>†</sup> and *Isoperla*<sup>†</sup>, the caddisflies (2) *Ironoquia*<sup>†</sup> and *Ptilostomis*, the mayfly (1) *Ameletus*<sup>†</sup>, and the snail (1) *Fossaria*<sup>†</sup>.

In the unmined stream, the Simpson index of diversity was  $0.88 \pm 0.01$  with an N<sub>2</sub> diversity of  $8.95 \pm 1.03$  effective species. The aggregate richness was 46 taxa, 7 of which were found in all five of the site-samples (distribution evenness) and 4<sup>†</sup> of those 7 taxa also had 5% or more of the mean total of individuals (abundance commonness): the mayflies (3) *Ameletus, Ephemerella*<sup>†</sup>, and *Epeorus*<sup>†</sup>, the stoneflies (2) *Haploperla* and *Amphinemura*<sup>†</sup>, the beetle (1) *Ectopria*<sup>†</sup>, and the true fly (1) Orthocladiinae.

Simpson index of diversity (1 - D) & Hill's  $N_2$  diversity—July (summer). In the up section, the Simpson index of diversity was  $0.65 \pm 0.06$  with an  $N_2$  diversity of  $3.38 \pm 0.77$  effective species. The aggregate richness was 30 taxa, 3 of which were found in all five of the site-samples (distribution evenness) and also had 5% or more of the average total of individuals (abundance commonness): the snail (1) *Fossaria*, the pea clam (1) *Pisidium*, and the true fly (1) *Stratiomys*.

In the down section, the Simpson index of diversity was  $0.60 \pm 0.10$  with an N<sub>2</sub> diversity of  $3.22 \pm 0.69$  effective species. The aggregate richness was 13 taxa, 1 of which was found in all five of the site-samples (distribution evenness) and also had 5% or more of the average total of individuals (abundance commonness): the snail (1) *Fossaria*.

In the unmined stream, the Simpson index of diversity was  $0.60 \pm 0.08$  with an N<sub>2</sub> diversity of  $3.48 \pm 1.27$  effective species. The aggregate richness was 27 taxa, 5 of which were found in all five of the site-samples (distribution evenness) and 2<sup>†</sup> of those 5 taxa also had 5% or more of the mean total of individuals (abundance commonness): the true flies (3) Chironominae<sup>†</sup>, Tanypodinae, and Tanytarsini, the mayfly (1) *Ephemera*<sup>†</sup>, and the beetle (1) *Ectopria*.

Simpson index of diversity (1 - D) & Hill's N<sub>2</sub> diversity—October (autumn). In the up section, the Simpson index of diversity was  $0.70 \pm 0.10$  with an N<sub>2</sub> diversity of  $5.00 \pm 1.44$  effective species. The aggregate richness was 41 taxa, 5 of which were found in all five of the site-samples (distribution evenness) and 3<sup>†</sup> of those 5 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (2) Tanypodinae<sup>†</sup> and *Chrysops*, the caddisfly (1) *Ptilostomis*, the snail (1) *Fossaria*<sup>†</sup>, and the pea clam (1) *Pisidium*<sup>†</sup>.

In the down section, the Simpson index of diversity was  $0.81 \pm 0.05$  with an N<sub>2</sub> diversity of  $6.38 \pm 1.29$  effective species. The aggregate richness was 32 taxa, 6 of which were found in all five of the site-samples (distribution evenness) and 5<sup>†</sup> of those 6 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (4) Tanypodinae<sup>†</sup>, Tanytarsini<sup>†</sup>, *Chrysops*, and *Tipula*<sup>†</sup>, the caddisfly (1) *Ptilostomis*<sup>†</sup>, and the snail (1) *Fossaria*<sup>†</sup>.

In the unmined stream, the Simpson index of diversity was  $0.90 \pm 0.01$  with an N<sub>2</sub> diversity of  $10.81 \pm 1.17$  effective species. The aggregate richness was 50 taxa, 6 of which were found in all five of the site-samples (distribution evenness) and 4<sup>†</sup> of those 6 taxa also had 5% or more of the mean total of individuals (abundance commonness): the true flies (3) Orthocladiinae<sup>†</sup>, Tanypodinae, and *Hexatoma*<sup>†</sup>, the beetle (1) *Ectopria*<sup>†</sup>, the mayfly (1) *Paraleptophlebia*<sup>†</sup>, the stonefly (1) *Eccoptura*.

Simpson index of diversity (1 - D) & Hill's N<sub>2</sub> diversity—January (winter). In the up section, the Simpson index of diversity was  $0.83 \pm 0.01$  with an N<sub>2</sub> diversity of  $5.87 \pm 0.48$  effective species. The aggregate richness was 44 taxa, 9 of which were found in all five of the site-samples (distribution evenness) and 6<sup>†</sup> of those 9 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (5) *Probezzia, Corynoneura,* Orthocladiinae<sup>†</sup>, Tanypodinae<sup>†</sup>, and Tanytarsini<sup>†</sup>, the mayfly (1) *Ameletus*<sup>†</sup>, the aquatic worms (1) Naididae<sup>†</sup>, the pea clam (1) *Pisidium*<sup>†</sup>, and the caddisfly (1) *Ptilostomis.* 

In the down section, the Simpson index of diversity was  $0.64 \pm 0.05$  with an N<sub>2</sub> diversity of  $2.97 \pm 0.39$  effective species. The aggregate richness was 25 taxa, 6 of which were found in all five of the site-samples (distribution evenness) and 4<sup>†</sup> of those 6 taxa also had 5% or more of the average total of individuals (abundance commonness): the true flies (5) Chironominae, *Corynoneura*, Orthocladiinae<sup>†</sup>, Tanypodinae<sup>†</sup>, and Tanytarsini<sup>†</sup>, and the caddisfly (1) *Ptilostomis*<sup>†</sup>.

In the unmined stream, the Simpson index of diversity was  $0.91 \pm 0.01$  with an N<sub>2</sub> diversity of  $11.14 \pm 0.78$  effective species. The aggregate richness was 45 taxa, 10 of which were found in all five of the site-samples (distribution evenness) and 4<sup>†</sup> of those 10

taxa also had 5% or more of the mean total of individuals (abundance commonness): the mayflies (5) *Ameletus, Ephemerella*<sup>†</sup>, *Epeorus, Maccaffertium,* and *Paraleptophlebia*<sup>†</sup>, the true flies (3) *Probezzia,* Chironominae<sup>†</sup>, and *Prosimulium*<sup>†</sup>, and the stoneflies (2) *Allocapnia* and *Haploperla*.

#### *Community biotic integrity*

**Percent EPT index**—April (spring). In the up section, the percent EPT index comprised mayflies with  $4.2\% \pm 0.8$  of genera, stoneflies with  $13.9\% \pm 1.5$ , and caddisflies with  $11.9\% \pm 1.4$ . In the down section, the percent EPT index comprised mayflies with  $6.7\% \pm 0.9$  of genera, stoneflies with  $11.6\% \pm 3.3$ , and caddisflies with  $9.9\% \pm 1.2$ . In the unmined stream, the percent EPT index comprised mayflies with  $19.9\% \pm 2.0$  of genera, stoneflies with  $25.3\% \pm 2.9$ , and caddisflies with  $7.9\% \pm 2.7$ .

**Percent EPT index**—July (summer). In the up section, the percent EPT index values comprised mayflies with  $1.1\% \pm 1.1$  of genera, stoneflies with  $0.0\% \pm 0.0$ , and caddisflies with  $0.0\% \pm 0.0$ . In the down section, the percent EPT index values comprised mayflies with  $3.3\% \pm 3.3$  of genera, stoneflies with  $0.0\% \pm 0.0$ , and caddisflies with  $0.0\% \pm 0.0$ . In the unmined stream, the percent EPT index value comprised mayflies with  $19.0\% \pm 4.0$  total genera, stoneflies with  $2.9\% \pm 2.9$ , and caddisflies with  $11.2\% \pm 1.8$ .

**Percent EPT index**—October (autumn). In the up section, the percent EPT index comprised mayflies with  $0.8\% \pm 0.8$  of genera, stoneflies with  $3.6\% \pm 2.3$ , and caddisflies with  $13.0\% \pm 3.6$ . In the down section, the percent EPT index comprised stoneflies with  $0.8\% \pm 0.8$  of genera, and caddisflies with  $10.4\% \pm 2.6$ . In the unmined

stream, the percent EPT index comprised mayflies with  $11.7\% \pm 2.8$  of genera, stoneflies with  $14.5\% \pm 2.1$ , and caddisflies with  $11.7\% \pm 1.3$ .

*Percent EPT index*—January (winter). In the up section, the percent EPT index comprised mayflies with  $6.8\% \pm 1.1$  of total genera, stoneflies with  $5.0\% \pm 3.1$ , and caddisflies with  $13.4\% \pm 2.4$ . In the down section, the percent EPT index comprised mayflies with  $10.1\% \pm 2.7$  of total genera, stoneflies with  $8.5\% \pm 1.2$ , and caddisflies with  $9.8\% \pm 1.3$ . In the unmined stream, the percent EPT index comprised mayflies with  $21.5\% \pm 1.9$  of total genera, stoneflies with  $18.7\% \pm 1.6$ , and caddisflies with  $10.8\% \pm 2.1$ .

*EPT density and percent abundance*—April (spring). In the up section, EPT density was  $3,575.6 \pm 1,095.5$  individuals·m<sup>-2</sup> and comprised mayflies with  $1,002.2 \pm 243.0$  individuals·m<sup>-2</sup> and a relative abundance of  $8.2\% \pm 1.8$ , stoneflies with  $2,244.4 \pm 756.8$  individuals·m<sup>-2</sup> and a relative abundance of  $18.3\% \pm 5.2$ , and caddisflies with  $328.9 \pm 116.1$  individuals·m<sup>-2</sup> and a relative abundance of  $2.6\% \pm 0.7$ .

In the down section, EPT density was  $1,871.1 \pm 525.3$  individuals  $\cdot m^{-2}$  and comprised mayflies with  $573.3 \pm 282.6$  individuals  $\cdot m^{-2}$  and a relative abundance of 8.1% $\pm 2.7$ , stoneflies with 997.8  $\pm 361.5$  individuals  $\cdot m^{-2}$  and a relative abundance of  $20.1\% \pm$ 8.7, and caddisflies  $300.0 \pm 62.4$  individuals  $\cdot m^{-2}$  and a relative abundance of  $6.1\% \pm 1$ .

In the unmined stream, EPT density was  $733.3 \pm 157.8$  individuals·m<sup>-2</sup> and comprised mayflies with 460.0 ± 100.3 individuals·m<sup>-2</sup> and a relative abundance of  $42.2\% \pm 2.7$ , stoneflies with  $222.2 \pm 42.5$  individuals·m<sup>-2</sup> and a relative abundance of

 $20.5\% \pm 1.4$ , and caddisflies with  $51.1 \pm 25.7$  individuals  $\cdot m^{-2}$  and a relative abundance of  $4.1\% \pm 1.4$ .

# *EPT density and percent abundance*—July (summer). In the up section, EPT density was $2.2 \pm 2.2$ individuals·m<sup>-2</sup> and comprised caddisflies with $2.2 \pm 2.2$ individuals·m<sup>-2</sup> and a relative abundance of $0.1\% \pm 0.1$ .

In the down section, EPT density was  $2.2 \pm 2.2$  individuals  $\cdot m^{-2}$  and comprised caddisflies with  $2.2 \pm 2.2$  individuals  $\cdot m^{-2}$  and a relative abundance of  $1.7\% \pm 1.7$ .

In the unmined stream, EPT density was  $217.8 \pm 54.3$  individuals·m<sup>-2</sup> and comprised mayflies with  $157.8 \pm 51.8$  individuals·m<sup>-2</sup> and a relative abundance of 11.2% $\pm 4.3$ , stoneflies with  $11.1 \pm 11.1$  individuals·m<sup>-2</sup> and a relative abundance of  $0.8\% \pm 0.8$ , and caddisflies with  $48.9 \pm 13.4$  individuals·m<sup>-2</sup> and a relative abundance of  $2.9\% \pm 0.6$ .

## *EPT density and percent abundance*—October (autumn). In the up section, EPT density was $346.7 \pm 133.1$ individuals·m<sup>-2</sup> and comprised mayflies with $2.2 \pm 2.2$ individuals·m<sup>-2</sup> and a relative abundance of $< 0.1\% \pm < 0.1$ , stoneflies with $13.3 \pm 8.2$ individuals·m<sup>-2</sup> and a relative abundance of $0.8\% \pm 0.5$ , and caddisflies with $333.1 \pm 125.8$ individuals·m<sup>-2</sup> and a relative abundance of $17.0\% \pm 9.5$ .

In the down section, EPT density was  $108.9 \pm 25.7$  individuals·m<sup>-2</sup> and comprised stoneflies with  $2.2 \pm 2.2$  individuals·m<sup>-2</sup> and a relative abundance of  $0.1\% \pm 0.1$ , and caddisflies with  $106.7 \pm 25.5$  individuals·m<sup>-2</sup> and a relative abundance of  $9.8\% \pm 3.4$ .

In the unmined stream, EPT density was 677.8  $\pm$  332.3 individuals·m<sup>-2</sup> and comprised mayflies with 280.0  $\pm$  102.3 individuals·m<sup>-2</sup> and a relative abundance of 15.6%  $\pm$  3.8, stoneflies with 131.1  $\pm$  49.6 individuals·m<sup>-2</sup> and a relative abundance of

 $8.0\% \pm 1.6$ , and caddisflies with 266.7  $\pm$  185.1 individuals  $\cdot$  m<sup>-2</sup> and a relative abundance of 9.4%  $\pm$  3.1.

*EPT density and percent abundance*—January (winter). In the up section, EPT density was  $840.0 \pm 164.6$  individuals·m<sup>-2</sup> and comprised mayflies with  $562.2 \pm 207.0$  individuals·m<sup>-2</sup> and a relative abundance of  $6.4\% \pm 2.0$ , stoneflies with  $75.6 \pm 56.8$  individuals·m<sup>-2</sup> and a relative abundance of  $1.0\% \pm 0.7$ , and caddisflies with  $202.2 \pm 105.8$  individuals·m<sup>-2</sup> and a relative abundance of  $2.6\% \pm 1.3$ .

In the down section, EPT density was  $711.1 \pm 199.6$  individuals·m<sup>-2</sup> and comprised mayflies with  $311.1 \pm 113.3$  individuals·m<sup>-2</sup> and a relative abundance of  $4.7\% \pm 2.4$ , stoneflies with  $304.4 \pm 171.7$  individuals·m<sup>-2</sup> and a relative abundance of  $1.5\% \pm 0.5$ , and caddisflies with  $95.6 \pm 33.1$  individuals·m<sup>-2</sup> and a relative abundance of  $0.8\% \pm 0.2$ .

In the unmined stream, EPT density was  $1,022.2 \pm 282.6$  individuals·m<sup>-2</sup> and comprised mayflies with 646.7 ± 165.3 individuals·m<sup>-2</sup> and a relative abundance of  $37.9\% \pm 5.0$ , stoneflies with  $280.0 \pm 84.0$  individuals·m<sup>-2</sup> and a relative abundance of  $14.4\% \pm 1.8$ , and caddisflies with  $95.6 \pm 44.9$  individuals·m<sup>-2</sup> and a relative abundance of  $4.4\% \pm 1.2$ .

**Percent Ephemeroptera (E) abundance**—April (spring). In the up section, mayfly density was 1,002.2 ± 243.0 individuals  $\cdot$ m<sup>-2</sup> with a relative abundance of 8.2% ± 1.8 and comprised *Ameletus* with 995.6 ± 244.2 individuals  $\cdot$ m<sup>-2</sup> (8.1% ± 1.8) and *Eurylophella* with 6.7 ± 4.4 individuals  $\cdot$ m<sup>-2</sup> (0.1% ± 0.1). In the down section, mayfly density was  $573.3 \pm 282.6$  individuals·m<sup>-2</sup> with a relative abundance of  $8.1\% \pm 2.7$  and comprised *Ameletus* with  $564.0 \pm 281.6$  individuals·m<sup>-2</sup> ( $7.9\% \pm 2.6$ ) and *Baetis* with  $9.0 \pm 6.5$  individuals·m<sup>-2</sup> ( $0.2\% \pm 0.1$ ).

In the unmined stream, mayfly density was  $460.0 \pm 110.3$  individuals·m<sup>-2</sup> with a relative abundance of  $42.4\% \pm 2.7$  and comprised *Ephemerella* with  $258.8 \pm 66.2$  individuals·m<sup>-2</sup> ( $24.7\% \pm 3.4$ ), *Epeorus* with  $140.0 \pm 45.1$  individuals·m<sup>-2</sup> ( $12.3\% \pm 3.3$ ), *Ameletus* with  $26.7 \pm 7.5$  individuals·m<sup>-2</sup> ( $2.8\% \pm 0.7$ ), *Paraleptophlebia* with  $20.0 \pm 10.2$  individuals·m<sup>-2</sup> ( $1.4\% \pm 0.6$ ), *Maccaffertium* with  $11.1 \pm 5.0$  individuals·m<sup>-2</sup> ( $1.0\% \pm 0.4$ ), and *Ephemera* with  $4.4 \pm 2.7$  individuals·m<sup>-2</sup> ( $0.3\% \pm 0.3$ ).

*Percent Ephemeroptera (E) abundance*—July (summer). In the unmined stream, mayfly density was  $157.8 \pm 51.8$  individuals·m<sup>-2</sup> with a relative abundance of  $11.2\% \pm 4.3$  and comprised *Ephemera* with  $80.0 \pm 35.9$  individuals·m<sup>-2</sup> ( $5.7\% \pm 2.8$ ), *Stenacron* with  $53.3 \pm 29.3$  individuals·m<sup>-2</sup> ( $3.9\% \pm 2.4$ ), *Paraleptophlebia* with  $13.3 \pm$ 10.8 individuals·m<sup>-2</sup> ( $1.0\% \pm 0.8$ ), *Eurylophella* with  $6.7 \pm 4.4$  individuals·m<sup>-2</sup> ( $0.4\% \pm$ 0.2), and *Procloeon* with  $4.4 \pm 4.4$  individuals·m<sup>-2</sup> ( $0.2\% \pm 0.2$ ).

*Percent Ephemeroptera (E) abundance*—October (autumn). In the up section, mayfly density was  $2.2 \pm 2.2$  individuals·m<sup>-2</sup> with a relative abundance of  $< 0.1\% \pm < 0.1$ and comprised *Paraleptophlebia*.

In the down section, mayfly density was  $0.0 \pm 0.0$  individuals m<sup>-2</sup>.

In the unmined stream, mayfly density was  $280.0 \pm 102.3$  individuals·m<sup>-2</sup> with a relative abundance of  $15.6\% \pm 3.8$  and comprised *Paraleptophlebia* with  $171.1 \pm 57.4$  individuals·m<sup>-2</sup> ( $8.5\% \pm 1.5$ ), *Maccaffertium* with  $31.1 \pm 14.7$  individuals·m<sup>-2</sup> ( $2.5\% \pm 1.5$ )
1.7), *Ephemera* with 20.0  $\pm$  12.4 individuals·m<sup>-2</sup> (2.3%  $\pm$  1.5), *Eurylophella* with 55.6  $\pm$  50.1 individuals·m<sup>-2</sup> (2.0%  $\pm$  1.0), and *Procloeon* with 2.2  $\pm$  2.2 individuals·m<sup>-2</sup> (0.3%  $\pm$  0.3).

**Percent Ephemeroptera (E) abundance**—**January (winter).** In the up section, mayfly density was  $562.2 \pm 207.0$  individuals·m<sup>-2</sup> with a relative abundance of  $6.4\% \pm 2.0$  and comprised *Ameletus* with  $517.8 \pm 188.8$  individuals·m<sup>-2</sup> ( $5.8\% \pm 1.8$ ) and *Baetis* with  $44.4 \pm 27.9$  individuals·m<sup>-2</sup> ( $0.6\% \pm 0.3$ ).

In the down section, mayfly density was  $311.1 \pm 113.3$  individuals·m<sup>-2</sup> with a relative abundance of  $4.7\% \pm 2.4$  and comprised *Ameletus* with  $175.6 \pm 80.4$  individuals·m<sup>-2</sup> ( $2.6\% \pm 1.9$ ) and *Baetis* with  $135.6 \pm 101.4$  individuals·m<sup>-2</sup> ( $2.1\% \pm 1.8$ ).

In the unmined stream, mayfly density was  $646.7 \pm 165.3$  individuals·m<sup>-2</sup> with a relative abundance of  $37.9\% \pm 5.0$ , and comprised *Ephemerella* with  $231.1 \pm 45.2$  individuals·m<sup>-2</sup> ( $14.5\% \pm 2.1$ ), *Paraleptophlebia* with  $208.9 \pm 71.8$  individuals·m<sup>-2</sup> ( $11.2\% \pm 1.6$ ), *Epeorus* with  $128.9 \pm 27.6$  individuals·m<sup>-2</sup> ( $7.9\% \pm 1.1$ ), *Ameletus* with  $60.0 \pm 32.7$  individuals·m<sup>-2</sup> ( $3.2\% \pm 1.2$ ), *Maccaffertium* with  $13.3 \pm 2.2$  individuals·m<sup>-2</sup> ( $1.0\% \pm 0.3$ ), and *Eurylophella* with  $4.4 \pm 2.7$  individuals·m<sup>-2</sup> ( $0.1\% \pm 0.1$ ).

*Hilsenhoff biotic index (HBI)*—April (spring). In the up section, the HBI was  $5.48 \pm 0.34$ , which represented a water quality rating of 'good' and a degree of environmental stress categorized as 'some'. Macroinvertebrate taxa sensitive to environmental stress, e.g., mayflies and water pennies (tolerance values of 0.0–4.5) were  $29.9\% \pm 8.3$  of the abundance, whereas stress-tolerant taxa, e.g., midges and aquatic worms (values of 6.6–10) were  $30.2\% \pm 3.7$  of the abundance. Macroinvertebrate taxa

with intermediate tolerance (4.6–6.5), e.g., pea clams, dragonflies and damselflies, were  $39.8\% \pm 5.2$  of the abundance.

In the down section, the HBI was  $5.61 \pm 0.38$ , which represented a water quality rating of 'fair' and a degree of environmental stress categorized as 'fairly significant'. Macroinvertebrate taxa sensitive to environmental stress were  $28.3\% \pm 8.9$  of the abundance, whereas stress-tolerant taxa were  $45.4\% \pm 7.4$ , and taxa with intermediate tolerance were  $26.2\% \pm 4.0$ .

In the unmined stream, the HBI was  $3.03 \pm 0.14$ , which represented a water quality rating of 'excellent' and a degree of environmental stress categorized as 'none apparent'. Macroinvertebrate taxa sensitive to environmental stress were  $80.9\% \pm 2.1$  of the abundance, whereas stress-tolerant taxa were  $7.8\% \pm 1.4$ , and taxa with intermediate tolerance were  $10.6\% \pm 2.1$ .

*Hilsenhoff biotic index (HBI)*—July (summer). In the up section, the HBI value was  $7.20 \pm 0.19$ , which represented a water quality rating of 'fairly poor' and a degree of environmental stress categorized as 'significant'. Macroinvertebrate taxa sensitive to environmental stress, e.g., mayflies and water pennies (tolerance values of 0.0–4.5) were  $0.5\% \pm 0.4$  of the abundance, whereas stress-tolerant taxa, e.g., midges and aquatic worms (values of 6.6–10) were  $73.8\% \pm 7.8$  of the abundance. Macroinvertebrate taxa with intermediate tolerance scores (4.6–6.5), e.g., pea clams, dragonflies and damselflies, were  $25.7\% \pm 7.8$  of the abundance.

In the down section, the HBI was  $7.09 \pm 0.25$ , which represented a water quality rating of 'fairly poor' and a degree of environmental stress categorized as 'significant'. Macroinvertebrate taxa sensitive to environmental stress were  $0.0\% \pm 0.0$  of the

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abundance, whereas stress-tolerant taxa were  $82.6\% \pm 5.7$ , and taxa with intermediate tolerance were  $14.1\% \pm 4.1$ .

In the unmined stream, the HBI was  $5.93 \pm 0.35$ , which represented a water quality rating of 'fair' and a degree of environmental stress categorized as 'fairly significant'. Macroinvertebrate taxa sensitive to environmental stress were  $20.8\% \pm 6.5$ of the abundance, whereas stress-tolerant taxa were  $65.5\% \pm 10.1$ , and taxa with intermediate tolerance were  $13.7\% \pm 3.9$ .

*Hilsenhoff biotic index (HBI)*—October (autumn). In the up section, the HBI was  $6.09 \pm 0.40$ , which represented a water quality rating of 'fair' and a degree of environmental stress categorized as 'fairly significant'. Macroinvertebrate taxa sensitive to environmental stress, e.g., mayflies and water pennies (tolerance values of 0.0–4.5) were  $13.6\% \pm 9.1$  of the abundance, whereas stress-tolerant taxa, e.g., midges and aquatic worms (values of 6.6-10) were  $30.4\% \pm 8.0$  of the abundance. Macroinvertebrate taxa with intermediate tolerance (4.6–6.5), e.g., pea clams, dragonflies and damselflies, were  $56.0\% \pm 9.3$  of the abundance.

In the down section, the HBI was  $7.04 \pm 0.15$ , which represented a water quality rating of 'fairly poor' and a degree of environmental stress categorized as 'significant'. Macroinvertebrate taxa sensitive to environmental stress were  $1.8\% \pm 1.4$  of the abundance, whereas stress-tolerant taxa were  $67.3\% \pm 6.0$ , and taxa with intermediate tolerance were  $30.9\% \pm 5.7$ .

In the unmined stream, the HBI was  $4.47 \pm 0.13$ , which represented a water quality rating of 'very good' and a degree of environmental stress categorized as

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'possible slight'. Macroinvertebrate taxa sensitive to environmental stress were  $57.0\% \pm 3.2$  of the abundance, whereas stress-tolerant taxa were  $26.0\% \pm 3.8$  and taxa with intermediate tolerance were  $16.8\% \pm 1.1$ .

*Hilsenhoff biotic index (HB1)*—January (winter). In the up section, the HBI was  $6.42 \pm 0.09$ , which represented a water quality rating of 'fair' and a degree of environmental stress categorized as 'fairly significant'. Macroinvertebrate taxa sensitive to environmental stress, e.g., mayflies and water pennies (tolerance values of 0.0-4.5) were  $8.7\% \pm 1.1$  of the abundance, whereas stress-tolerant taxa, e.g., midges and aquatic worms (values of 6.6-10) were  $44.8\% \pm 6.6$  of the abundance. Macroinvertebrate taxa with intermediate tolerance scores (4.6-6.5), e.g., pea clams, dragonflies and damselflies, were  $46.4\% \pm 6.3$  of the abundance.

In the down section, the HBI was  $6.78 \pm 0.15$ , which represented a water quality rating of 'fairly poor' and a degree of environmental stress categorized as 'significant'. Macroinvertebrate taxa sensitive to environmental stress were  $4.5\% \pm 1.9$  of the abundance, whereas stress-tolerant taxa were  $64.1\% \pm 8.7$  and taxa with intermediate tolerance were  $31.4\% \pm 7.5$ .

In the unmined stream, the HBI was  $3.68 \pm 0.25$ , which represented a water quality rating of 'very good' and a degree of environmental stress categorized as 'possible slight'. Macroinvertebrate taxa sensitive to environmental stress were  $66.2\% \pm$ 4.9 of the abundance, whereas stress-tolerant taxa were  $26.7\% \pm 3.3$  and taxa with intermediate tolerance were  $7.1\% \pm 2.2$ .

## CURRICULUM VITAE

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Education	
Ph.D., Biology	Aug. 2021
University of Louisville, Louisville, KY	
Advisor: James E. Alexander, Ph.D.	
Dissertation: Benthic Macroinvertebrate Dynamics in a Retrofitted Watersl	hed
	<b>D</b>
M.S. Non-Thesis, Biology	Dec. 2015
University of Louisville, Louisville, KY	

Dec. 2011

Advisor: Margaret M. Carreiro, Ph.D.

Bachelor of Science, Biology University of Louisville, Louisville, KY Ecology and Evolutionary Biology

## **Research Experience**

University of Louisville, Louisville, KY Ph.D. Student; Advisors: Margaret M. Carreiro, Ph.D., and James E. Alexander, Ph.D. Community structure and dynamics of benthic macroinvertebrates in a recreated headwater stream system on a valley fill in a retrofitted watershed located in the Appalachian coalfields of Southeastern Kentucky, U.S.A. Skills gained: Data analysis using R programming language for statistical computing and graphics, microscopy, benthic invertebrate identifications

## **Presentations**

Bailey SW, 2018. Temporal dynamics of benthic macroinvertebrate communities in a headwater stream reconstructed on top of a valley fill created from mountain mining in the Appalachian coalfields of Southeastern Kentucky. Poster Presentation at the Graduate Student Regional Research Conference (GSRRC), University of Louisville, Louisville, KY.

Bailey SW, 2016. Legacy effects of mountain mining on headwater streams. Oral presentation at the Biology Awards Day, University of Louisville, Louisville, KY.

Bailey SW, 2016. Legacy effects of mountain mining on headwater streams. Guest speaker invitation, Biology Department, Centre College, Danville, KY.