

EFFECTS OF SEDIMENT ACCUMULATION ON  
MACROINVERTEBRATE ASSEMBLAGES NEAR ROAD CROSSINGS  
IN THE UPPER PENINSULA OF MICHIGAN

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## LITERATURE REVIEW

### I. Introduction

The road/stream boundary is one of the main pathways for sediment to reach waterways (Croke *et al.*, 2005), and thus is an important aspect of stream sediment processes. The purpose of this review is to describe the components of sedimentation, from its generation and transport, to how it is measured, and finally, to the effects of sediment on biota.

### II. Definition

Sediment is broken down material from weathering and erosion, also known as dirt, soil, or eroded material, composed of both organic and inorganic contents of various sizes. Particles less than 2 mm in diameter, such as sand, silt, and clay, are classified as fine sediment or fines (Rosenberg and Wiens, 1978; Jones *et al.*, 2012 and others). Suspended and bedded sediments (SABS) are organic and inorganic particulates that are suspended in the water column or deposited on the stream bed (USEPA, 2003; Berry *et al.*, 2003). Sedimentation, or accumulated sediment, is a rate by which sediment is deposited or accumulated in an area over time, given in a mass per area per time unit.

### III. Creation of sediment

Sediment deposition and erosion is a natural process in fluvial ecosystems (Berry *et al.*, 2003). Sources of sediment are defined as either channel sources, which derive sediment from within the stream channel, or non-channel sources, which originate outside of the stream channel. Channel derived sediments are sourced from banks and channel margins, point bars, fines stored in interstitial pore spaces or sequestered in vegetation, and pools or backwater areas. Outside of the stream channel, sources of sediment encompass leaf and litter fall, biological pseudofeces, exposed or unvegetated soils, landslides, particles from atmospheric deposition, and in general, anthropogenic activities (Wood and Armitage, 1997). Transport of non-channel sediment to the water body depends on the source of sediment as well as the path of transport, which is highly variable. Stream derived sediment transport is less varied, and depends on hydrological and hydraulic characteristics, such as stream discharge and stream bed stability (Waters, 1995; Wood and Armitage, 1997).

### IV. Sediment as a pollutant

Sediment becomes a concern for water quality and biota when the natural cycle of sedimentation is altered, either by increasing or decreasing natural levels (Jones *et al.*, 2012). In the U.S., sediment in water bodies in greater amounts than what would occur naturally, primarily originate from non-channel sources created by a variety of anthropogenic activities (USEPA, 2013; Waters, 1995). While agricultural practices, such as raising livestock and row-crop cultivation, are the largest contributor of

sediments, forestry and mining are also sources. Other anthropogenic activities, including dredging, dam construction, hydrologic and hydraulic alterations, and urban development, also alter natural sedimentation and deposition cycles (Wilber and Clarke, 2001; Waters, 1995).

Sediment can cause physical and chemical problems for the biological integrity of water bodies. Physically, suspended sediment increases turbidity and limits UV penetration. In great excess, bed load sediment can smother the stream bed and alter channel morphology by reducing stream depth or eliminating heterogeneous habitats (Walser and Bart, 2006). Sediment with large quantities of organic matter can deplete oxygen levels through decay (Jones *et al.*, 2012). Particles of certain sizes, such as silt and clays (< 63 $\mu$ m), which make up the majority of suspended sediments, absorb metals and chemicals more readily, creating a transport pathway for toxics (Waters, 1995; Wood and Armitage, 1997).

#### V. Roadways as a source of sediment

Unpaved roads, or unsealed/dirt roads, generate sediment from erosion of the road surface primarily from precipitation, known as rain splash, and related water flows (Ramos-Scharrón and MacDonald, 2005; MacDonald and Coe, 2008). Compared to vegetated forest, unpaved roads are high in sediment production (Macdonald and Coe, 2008). Road derived sediment reaches water bodies directly from road crossings (direct hydrological connectivity) or by runoff transport over land (diffuse hydrological connectivity) (Croke *et al.*, 2005). In forested areas, diffuse connectivity is typically low (Macdonald and Coe, 2008). Although landslides and debris flows are rare events that can contribute a great deal of sediment in forested regions, the greatest source of normal, low level sediment in forested regions is the erosion of unpaved roads (Megahan and Kidd, 1972). At road crossings, the majority of sediment that is created reaches the stream (Lane and Sheridan, 2002; Croke *et al.*, 2005). Eaglin and Hubert (1993) assessed 28 reaches with a range of stream and culvert densities, and found that fine sediment deposition and embeddedness increased as the number of road crossings increased.

Headwater catchments with unpaved roads have been shown to have greater bed load sediment than those without unpaved roads (Ramos-Scharrón and Macdonald, 2007; Kreutzweiser and Capell, 2001), but this is not always the case or always detectable (Studinski *et al.*, 2012). During and immediately following a disturbance, such as road building or reshaping/grading the road to remove irregularities, erosion rates can be high (Luce and Black, 1999; Fu *et al.*, 2010), but are likely to subside in the following years (Cline, 1982; Ketcheson and Megahan, 1996; Hinderer, 2012; Ramos-Scharrón and Macdonald, 2005). In a survey of three headwater watersheds in Idaho, Ketcheson and Megahan (1996) found that 70% of the sediment created from road building occurred in the first year after construction, with the remaining 30% in the following 3 years of the study.

The amount of sediment that erosion creates from unpaved roads as well as the likelihood that it will reach a water body depends on many factors. The amount and type of traffic, precipitation amounts and intensity, climate, road material types, slope of the landscape, characteristics of the drainage network, distance from erosion source to the water body, and surrounding ecosystem characteristics all effect the creation and transport of sediment to water bodies (Macdonald and Coe, 2008; Ketcheson and Megahan, 1996; Fu *et al.*, 2010).

#### VI. Measuring erosion

Radionuclides have been used to follow the movement of eroded particles to water bodies; however, factors such as water chemistry can affect results and therefore limit the reliability of these methods (Fu *et al.*, 2006, Fu *et al.*, 2010). Erosion pins/plots, sediment traps and weirs, empirical sediment budgets and sediment fences have also been used to estimate rate of erosion and degree of sedimentation (Megahan and Kidd, 1972; Luce and Black, 1999). Numerous physics-based and empirical models, using a modification of Universal Soil Loss Equation (USDOI, 2006) calculate and trace catchment wide erosion of past, present and possible future events, and further research will lead to improvements in this type of erosion modeling (Fu *et al.*, 2010). Sediment yield models calculate a sediment load for a watershed based on similar variables as the amount sediment created by unpaved roads (USDOI, 2006).

#### VII. Preventing erosion from reaching water bodies

Erosion control practices are varied and some are more effective at limiting the amount of eroded sediment that reaches water bodies (Ketcheson and Megahan; 1996). Barriers, such as straw bale or silt fences, collect sediment before it reaches water bodies. Sediment basins allow sediment to settle out into the basin before merging with the stream, but current practiced designs suggest they are not as effective as possible (USEPA, 2012; Kalainesan *et al.*, 2009). The US EPA recommends a variety of temporary and permanent best management practices to control erosion and sediment run-off from roads and highways (USEPA, 2012). Most practices include adding vegetation (e.g. grasses, wildflowers, wetlands) to prevent bankside erosion or to collect/slow the movement of over land water/sediment flows (USEPA, 2012).

#### VIII. Measuring sediment in fluvial systems

Suspended sediments are primarily measured directly as total suspended solids (TSS concentration) or indirectly as turbidity (NTU or FTU), but there are a variety of methods to measure and assess depth of deposited sediments and rate of deposition, from which most impacts are derived (Jones *et al.*, 2012). A common field method is a visual assessment of the percent of an area covered in fines (e.g. Davies and Nelson, 1994; Zweig and Rabeni, 2001; Larsen *et al.*, 2009), but is subjective to the assessor and becomes problematic when comparing results from

different assessors. Centrifuge tubes placed in the stream have also been used to measure accumulated sediment for a more precise measurement (Kreutzweiser and Capell, 2001). Another quantitative method is to calculate an embeddedness score by measuring the height of each stone perpendicular to the streambed area and the height of the silt line in a particular area (Zweig and Rabeni, 2001).

Longing *et al.* (2010) utilized a systematic random sampling method to measure sedimentation by averaging three measured variables (percent fines, percent embeddedness, and the  $D_{50}$  particle size), ranking from 1-3, then averaging for a high, medium or low sediment score. While Longing *et al.*'s method is useful in limiting the assessor's subjectivity, the simplification of results to a categorical ranking reduces the strength of the initial sediment measurement. In other studies, sediment in specific quantities or duration, were added to a stream or to trays placed in natural or artificial channels (Connolly and Pearson, 2004; Larsen and Ormerod, 2010; Bond and Downes, 2003; Rosenberg and Wiens, 1978; Angradi, 1999; Larsen *et al.*, 2011). Experiments that add specific quantities of sediment to streams result in a range of sedimentation values, useful for investigating impacts along a stressor gradient. Angradi (1999) added sediment to trays with natural substrate in 5% increments up to 30% coverage, creating a form of a dose-response curve to sediment deposition.

Macroinvertebrate community samples, collected with typical techniques such as kick nets, Surber samples or D-nets, are not always collected simultaneously or from the identical patch of streambed as sedimentation measurements (Kaller and Hartman 2004; Longing *et al.*, 2010), which may reduce the practicality of the combined measurements. Some studies have combined invertebrate samples from several different patches in a reach into a composite sample and compared it with overall sedimentation levels of the reach (Longing *et al.*, 2010). Composite samples or non-concurrent sampling can make it difficult to find strong stressor-response relationships by adding in taxa from patches with sediment levels that are higher or lower than the sedimentation level of the reach (Longing *et al.*, 2010). In laboratories, dose response and toxicity tests have been used to assess sediment impacts on individual macroinvertebrate taxa (Wood *et al.*, 2005; Suren, 2005), although the usefulness to real world results may be limited because direct mortality by sediment is less likely than mortality by indirect effects (e.g. reduced feeding rates) (Jones *et al.*, 2012). Others have correlated data from sedimentation and insect populations to investigate impacts (Jones *et al.*, 2012), which may be useful when looking at large scale impacts. Because many macroinvertebrates are micro-habitat specialists, adjacent patches of a stream may contain very different assemblages; therefore, patch scale studies that take both macroinvertebrate samples and sediment measurements concurrently may increase the strength of the results. For example, experiments whereby insects colonize in trays placed in streams can provide data on the relationship between sedimentation levels and the macroinvertebrate community at the patch level (Larsen *et al.*, 2011; Angradi, 1999).

## IX. Sedimentation impacts on biota

When the supply of sediment in a stream is reduced or eliminated, the stream will scour sediment from banks or stream bed to meet its capacity to carry sediment. For example, scouring occurs downstream of a dam, where sediment has been trapped upstream (Grant *et al.*, 2003). Sediment provides spawning habitat, refuge from predators, and locations to search for food, among other benefits, therefore insufficient sediment may impact biota. However, in the U.S., most problems arise from excess sediment, as approximately 45% of surveyed streams are rated 'poor' or 'fair' for excess sediment (USEPA, 2013).

Biotic effects of excess suspended and depositional fine sediment on primary producers, macro-invertebrate and fish has received substantial coverage in the scientific literature (Jones *et al.*, 2012; Suren, 2005; Broekhuizen *et al.*, 2001; Wood and Armitage, 1997). There are both direct and indirect effects from increased sediment deposition. The effects on biota from suspended sediment are a factor of both the amount of sediment deposition and the duration of exposure (Newcombe and Macdonald, 1991). Changes to habitat, availability, type, and nutritional component of food sources (Cline *et al.*, 1982) and overall food web changes are also possible indirect consequences to fine sediment deposition (Jones *et al.*, 2012). In general, these impacts can affect certain species more than others depending on the individual mobility, feeding and breathing method, habitat preference, and overall life history, as well as the depth of sediment, size of particles, and duration of impact.

An increase in turbidity due to increased suspended sediment primarily limits photosynthesis and primary productivity for macrophytes. In addition, reduced organic content, physical damage, and elimination of basal species has been observed with increased fine sediment deposition (Wood and Armitage, 1997). Following an increase in suspended sediment in high-elevation streams due to road construction in Colorado, Cline *et al.* (1982) observed a decrease in diversity of algal species and reduced organic content of periphyton cells compared to unimpacted sites. Primary producers have also been shown to recover less quickly after the impact, compared to sedimentation levels which are more apt to return to normal once the disturbance has ceased (Cline *et al.*, 1982). When organic content of periphyton, food for some macroinvertebrates, decreases, bottom-up effects to the food web are likely.

Fine sediment can affect macroinvertebrates both directly and indirectly. Jones *et al.* (2012) identified 4 main physical impacts; physical damage, clogging of organs, smothering or burial, and habitat alteration. Because of the life history diversity of macroinvertebrates, sediment may impact certain organisms more than others. Bodily harm is caused by the physical impact, or abrasion of particles, from both suspended sediment and saltation. The particle impact may also cause increased drift by dislodging insects, but it is challenging to accurately measure (Jones *et al.*, 2010). Suspended sediments may also cause a change in behavior, such as switching to alternative feeding methods to protect body parts from impact or seeking refuge

instead of other normal behaviors (Wood and Armitage, 1997). Clogging, where fine sediment infiltrates organs, can impact an organism's feeding and breathing ability. Sediment blockage is especially harmful to filter feeders and groups that are exposed to sediment as they attach to exposed, relatively clean substrate. The Diptera, Simuliidae, which attaches to hard surfaces in streams, has been shown to increase in drift immediately after fine sediment additions (Rosenberg and Wiens, 1978). Burial by sediment and subsequent effects can also impact macroinvertebrates survival. Jones *et al.*, (2012) suggests that burial can contribute to mortality by preventing access to food sources, but more profound effects of sediment burial are caused by the depletion of oxygen. Some species, such as burrowing mayflies, will not colonize in areas with depleted oxygen levels in the hyporheic zone (Krieger *et al.*, 2006). Finally, excess sediment physically alters the substrate. For example, fines can fill in interstitial pore space and alter the flow of water through the hyporheic zone. In addition, organisms use this zone with pore space for refuge from predators or during high flow events (Jones *et al.*, 2012). Finer sediment and sand creates unstable substrate, which is unsuitable habitat for species that must attach to hard surfaces. This in turn can induce behavioral responses, such as drifting to a new location with a more suitable habitat (Connolly and Pearson, 2004).

Perhaps due to the diversity of macroinvertebrate life histories, studies have reported a range of invertebrate metric responses to increased dispositional sediment. Common findings are reduced diversity, abundance, biomass, and density, decrease in EPT richness, change in composition of assemblages, and an increase in drift density (Waters, 1995; Jones *et al.*, 2012; Larsen *et al.*, 2011; Angradi, 1999; Wood and Armitage, 1997; Rosenberg and Wiens, 1978). However, metric responses are not always consistent, nor detectable. While one metric may be a strong indicator in one stream, it may be weak for another. Following an road improvement causing an increase in fine sediment in a forested headwater stream, Kruetzwiser *et al.*, (2001) found no significant changes in biomass, although other metrics were significant. Contrarily, after adding specific amounts of sediment to trays, Angradi (1999) examined colonized insects and found that most relationships were fairly weak and subtle, except for biomass (and density).

In a review of sedimentation, Wood and Armitage (1997) identified five mechanisms where SABS can impact fish. 1) Clog gill rakers and gill filaments causing increased susceptibility to disease, reduced growth rates or death. 2) Altering the suitability of spawning habitat and impacting the early stages of development and life (e.g. egg, larvae and juvenile). 3) Disruption of natural migration patterns. 4) Reduced food sources, either primary producers or altering habitat space for benthos food sources. 5) Impacting their ability to successfully capture prey because of poor visibility. The main impacts to fish can cause changes in fish assemblages and community structure, as Sutherland *et al.* (2002) found that an increase in 10% of non-forested land cover increased bed load and suspended sediment and resulted in different fish assemblages. Higher sedimentation increased abundance of fish

utilizing soft sediments and decreased abundance in fish utilizing cobbles for spawning and nest building (Sutherland *et al.*, 2002).

## X. Conclusions

Sediment in streams, while unavoidable, can be a pollutant in excess. Anthropogenic activities, such as agricultural and forestry practices, are the primary cause of excess sediment in water bodies. In forested regions, road crossings are a common source of excess sediments that are transported into the stream as suspended sediment and bed load. As development of forested areas and the number of road/stream crossings may increase in the future, it is important to study road/stream crossings and establish methods to prevent unfavorable impacts from excess sediment on fluvial systems.

## Chapter 2: Manuscript

### ABSTRACT

Recently widened unpaved roads and increased traffic due to a new mining operation have the potential to increase sedimentation and impact fluvial biology in the Yellow Dog Plains region of the Upper Peninsula of Michigan. To assess sediment accumulation around road crossings, partially open chambers filled with natural rocky substrate were deployed in similar river habitats upstream and downstream of road crossings in three reaches to allow sediment accumulation and insect colonization. We observed spatial variability in sedimentation with significantly greater deposition upstream of road crossings and the lowest sediment accumulation at the site with recently widened roads. Proportion Ephemeroptera, Chironomidae, and EPT, EPT: C, HBI and family richness metrics varied spatially and were affected by sediment accumulation. The two diversity indices (Simpson's and Shannon-Wiener) were not affected by sediment accumulation and only one functional family group (% predators) was affected by sediment accumulation. Insect abundance was significantly greater at downstream chamber location and significantly increased with greater sediment accumulation at all sites. This study provides evidence that sediment deposition may increase around road crossings, but invertebrates are not necessarily negatively affected by increase sedimentation. It also suggests that even if intensive road construction activities occur near streams, excess sedimentation and impacted stream invertebrate communities may not always be the outcome.

### INTRODUCTION

Land altering activities, such as logging, mining, agriculture, and urban development, as well as channel alteration activities, such as dykes, dams, and culverts, cause changes to the natural sedimentation and erosion processes in water bodies (Waters, 1995; Wilber and Clarke, 2001). When natural resource extraction occurs in undeveloped areas, obtaining access to the site may require road construction near or over water bodies, potentially exposing rivers and streams to fresh sediment sources. Previous studies have found that the majority of sediment derived from unpaved roadways enters water bodies directly at road crossings (Lane and Sheridan, 2002; Croke *et al.*, 2005). Stream density also influences the amount of sedimentation, as increased stream density is tied to the number of road crossings, where sediment is delivered (Eaglin and Hubert, 1993; MacDonald and Cue, 2008).

Sediment can negatively affect both invertebrate producers and consumers through many distinct mechanisms including physical abrasion, clogging of breathing or feeding apparatuses, burial by fine sediments, and alteration of the habitat (Wood and Armitage, 1997; Jones *et al.*, 2012). Experimental manipulations and case studies of fine sediment demonstrate a variety of responses by the macroinvertebrate community, such as reduced diversity, density, and biomass, as well as increased drift (Angradi, 1999; Broekhuizen *et al.*, 2001; Kaller and Hartman, 2004; Larsen and Ormerod, 2010; Zweig and Rabeni, 2001). Thus, it has generally been concluded

that excess fine sediment is unfavorable to macroinvertebrates as a whole (Jones *et al.*, 2012; Waters, 1995). However, not all taxa respond negatively to sedimentation, but rather it depends on their primary functional group and life history. For example, certain Chironomidae, which utilize finer sediments in protective case building and avoid areas with little sediment (Mclachlan and Cantrell, 2006), may benefit from an increase in sediment deposition.

A newly constructed underground nickel and copper mine (Eagle Mine) is located in a sparsely populated, forested area of the Upper Peninsula of Michigan known as the Yellow Dog Plains, which serves as the headwaters for a number of high quality streams, such as the Salmon Trout River. Road construction and increased traffic in conjunction with the mining operation has the potential to increase sedimentation in streams in the Yellow Dogs Plains region. While some attention has been given to the impacts from sand on macroinvertebrates in the Salmon Trout River (Eggert *et al.*, 2011), we wished to assess the sedimentation derived impacts from road crossings on stream quality in this region.

To determine if streams near the Eagle Mine are being affected by sedimentation near road crossings, we conducted a study to measure sediment accumulation and macroinvertebrate colonization in chambers near three stream crossings. We hypothesized that, within each site, sedimentation would be greater immediately adjacent to road crossings, as road crossings have been identified as the primary route for road derived sediments to reach water bodies in prior studies (Croke *et al.*, 2005). When comparing among sites, we expected to observe greatest sediment accumulation at the site located on a recently widened road because it has been shown that sedimentation usually increases greatly during and immediately following a disturbance (Cline *et al.*, 1982; Hinderer, 2012; Ramos-Scharrón and MacDonald, 2005; Kreutzweiser and Capell, 2001). Further, we expected the colonizing macroinvertebrate community composition to differ across sites as well as within each site, with decreased abundance, lower richness (family and EPT), lower diversity metrics (Simpson's Index of Diversity, Shannon-Wiener Index), reduced proportion of Ephemeroptera, Plecoptera and Trichoptera taxa, and decreased Hilsenhoff Biotic Indices with increased sediment deposition in the chamber. Because some Chironomidae species respond favorably and others poorly to increased sedimentation, we expected no change in Chironomidae proportion (Zweig and Rabeni, 2001).

## METHODS

### *Study Area*

This experiment was completed within the Dead-Kelsey Watershed, in a region of glacial sands deposit known as the Yellow Dog Plains in Marquette County, Michigan (USEPA, 2013). The Yellow Dog Plains contains the headwaters for a number of high-quality groundwater-fed streams including the Salmon Trout River. The watershed of the Salmon Trout River (12,823 hectares) contains mostly sandy

soils, dominated by forest (88.2%), wetlands (6.2%), and igneous outcroppings (4.4%) (Superior Watershed Partnership, 2007). We selected the central branch of the Salmon Trout River and farthest east tributary of the East Branch of the Salmon Trout River for this experiment as both encompassed a road crossing, were near the Eagle Mine, and had relatively similar geomorphology. A reference site, Pine River, was selected primarily for its isolation from the Eagle Mine. It is located in the Pine sub-watershed of the Dead-Kelsey Watershed.

The two first-order reaches (East-culvert and Central-culvert) in the Salmon Trout sub-watershed are characteristic of low order streams; cool, swift waters (4-21 ° C, 1-3 m wide, ~1 cfs), dense canopy cover, and a food web primarily dependent upon allochthonous organic carbon input from coarse particulate organic matter. The East-culvert site contains a 6.2 m long culvert crossing located on a recently-widened unpaved public road, directly on the designated Eagle Mine transportation route (Table 1). The substrate at East-culvert was relatively similar throughout the surveyed reach, consisting of gravel (40%), cobbles (25%), and sand (25%). Silt, clay, and boulders (10%) comprised the remaining substrate in the sampling reach. Coniferous trees were the dominant vegetation present in the riparian buffer throughout riparian area in East-culvert.

Central-culvert was located on the same unpaved public road containing a 5.5 m long culvert, approximately 2.1 km west of the Eagle mine. Upstream of the culvert, the riparian vegetation contained primarily shrubs with less canopy cover. The substrate on the reach upstream of Central-culvert was a mix of sand and silt (70%), less cobble and gravel (25%), and minimal boulders and clay substrate (5%). The downstream reach of the Central-culvert was comprised mostly of boulders and cobbles (65%), sand and silt (25%), and gravel (10%) and had a denser canopy cover than the upstream reach. The section of road near Central-culvert was not recently widened, nor is it on the current transportation route.

The reference site, North-bridge, a lake-outflow and groundwater fed stream (Pine River), was on private property in the Pine sub-watershed, approximately 16 km north of the Eagle Mine with no access for commercial vehicles related to the Eagle Mine. North-bridge site had a 6.3 m wide bridge on an unpaved private road. The substrate consisted of cobble and gravel (70%), sand and silt (25%), and few boulders (5%). A deep pool (>2 m) was located downstream of the bridge. The North-bridge site had relatively similar temperature, average depth, and velocity as the reaches on the Salmon Trout, but stream width and discharge was greater (approximately 8 m, ~3 cfs). North-bridge appeared to be a primarily autochthonous system, with an open canopy and a mix of conifers and shrubs in the riparian area.

### *Experimental Design*

We constructed sedimentation and colonization chambers (13 cm long x 6.5 diameter, with two 4.5 cm x 10.5 cm windows) from polyurethane tubing with partially-open end caps, to provide a habitat for colonization and collection of settling

particles (Fig. 1) (Kochersberger *et al.*, 2012). The chambers were filled halfway with coarse natural substrate that was collected on site, sieved to remove particles < 2 mm, placed into the chambers and secured with coarse mesh. Three chambers in each of three locations were deployed parallel to flow in relatively similar habitats along a 100 m reach extending across the culvert or bridge. Each reach comprised three locations; directly upstream and downstream of the road crossing and a reference location farther upstream (~ 40 – 50 m) of the road crossing. Water depth at each chamber location was less than 0.5 m (Fig. 2).

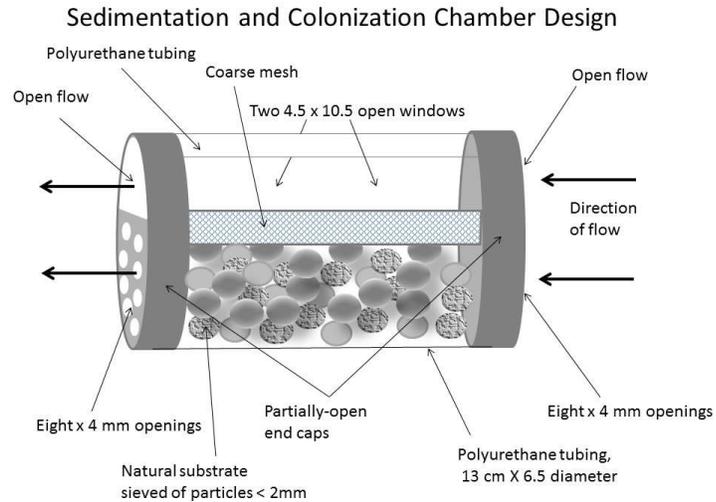


Fig. 1: Sedimentation and colonization chambers were designed to create an open flow habitat to allow both particles and invertebrates to enter the chamber. The natural substrate was collected on site and sieved to remove particles less than 2 mm. Design follows a similar technique as Korchersberger *et al.* (2012).

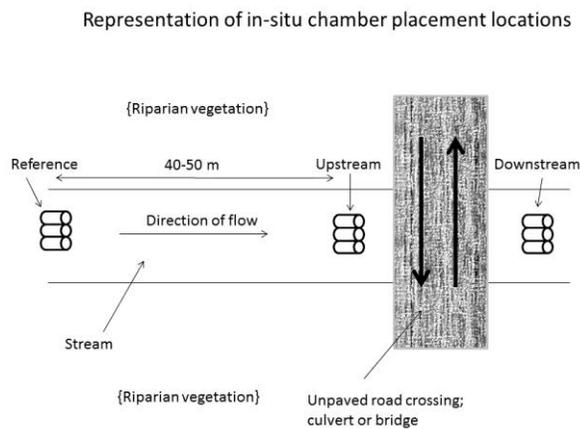


Fig. 2: A representation shows the three stream locations where chambers were placed in three reaches near Yellow Dog Plains region in order to accumulate sediment and allow invertebrate colonization.

Trays were left in the stream for 40 days (July 14 – August 23, 2011) to allow sediment accumulation and invertebrate colonization. At deployment and retrieval, we measured water velocity, temperature, total suspended solids, and depth. On day 40, each chamber was collected underwater into a plastic bag to avoid losing any material.

In the laboratory, the contents of each bag were passed through a 1-mm sieve to separate macroinvertebrates and coarse substrate from fine sediments. The water and sediment slurry that passed through the sieve was collected and diluted to a final volume of 4400 mL. From that slurry, three 50 mL subsamples were taken, collected on a filter (0.7  $\mu\text{m}$  glass fiber), dried, ashed and weighed to obtain a measure of sedimentation. Invertebrates collected on the sieve were separated from the remaining debris and preserved in 70% ethanol, and later counted and identified to family level (Merritt and Cummins, 1996; Hilsenhoff, 1995; Bouchard, 2004).

### *Data Analysis*

To quantify sediment accumulation, we converted mean subsample weight to an areal deposition rate per day based on cross-sectional surface area of the chamber (84.5  $\text{cm}^2$ ). Site and chamber locations effect on sediment deposition were identified with a two-way ANOVA, with post-hoc Tukey's HSD test.

Invertebrate abundance data was converted to 17 metrics describing community composition in each chamber: proportion of each functional group (i.e. percent filterers), Ephemeroptera proportion, Trichoptera proportional abundance, total EPT proportional abundance, EPT richness, Chironomidae proportional abundance, the ratio of EPT abundance to Chironomidae abundance, family richness, total abundance, diversity indices (Simpson's Index of Diversity (1-D) and Shannon-Wiener Diversity Index), and Hilsenhoff's Family Biotic Index. The Hilsenhoff's Biotic Index, manipulated for family data, is an index used to evaluate the tolerance level of organisms to organic pollution. Family tolerance values were obtained from the U.S.EPA Rapid Bioassessment protocols (Barbour *et al.*, 1999), and supplemented with a secondary source when required (Hauer and Lamberti, 2006). Metrics were selected for statistical analysis due to composition of the community (e.g. no shredders present) and the ability to detect differences between reference and impaired ecosystems (Carlisle and Clements, 1999; Barbour *et al.*, 1999). We removed the four observations where the ratio of EPT to Chironomidae was undefined due to no Chironomidae present. For calculations of the Simpson's Index of Diversity and Shannon-Wiener Index, two outliers were removed to normalize the data. Metrics were transformed when appropriate.

We performed ANCOVA to assess if sediment accumulation, chamber location, or stream (site) was influencing each invertebrate metric. ANCOVA models were simplified by removing main effects (site and chamber location), the covariate (sediment accumulation), and interactions that did not account for significant

variation ( $p > 0.05$ ) in each invertebrate metric. All statistics were completed using R (3.0.0) (R Core Team, 2013).

## RESULTS

### *Physical conditions*

A USGS gauge at a location on a second order stream of the main branch of the Salmon Trout River showed that flow was relatively consistent over the five week period as there were no significant storm events (USGS Flow gauge). At the North-bridge site, filamentous green algae (presumably *Cladophora sp.*) was covering the downstream chamber location and the surrounding area upstream of the chambers, serving as a potential sediment trap. Table 1 displays basic stream hydrological values for the three sites. Although TSS varied in the three sites, the reference accumulation was similar across all locations.

Table 1: Summary of location, hydrology and sediment habitat from three sampled reaches near the Yellow Dog Plains, in the Dead-Kelsey Watershed.

Site	North-bridge	Central-culvert	East-culvert
Latitude (N)	46°52'58.16"	46°45'3.25"	46°45'18.94"
Longitude (W)	87°52'8.13"	87°54'26.96"	87°48'16.69"
Flow ( $\text{m}\cdot\text{s}^{-1}$ ) <sup>a</sup>	0.11 (0.09)	0.18 (0.02)	0.07 (0.02)
TSS ( $\text{mg}\cdot\text{L}^{-1}$ ) <sup>b</sup>	0.56	0.26	0.30
Reference sediment accumulation ( $\text{kg}\cdot\text{m}^2/\text{day}$ )	0.331	0.306	0.375
Upstream embeddedness <sup>d</sup>	50%	50%	30%
Downstream embeddedness <sup>e</sup>	35%	50%	70%

<sup>a</sup> Mean flow (SE) is averaged from three locations in each reach taken at deployment and collection. <sup>b</sup> Total suspended solids is estimated from one measurement taken at the reference chamber location during collection.

<sup>c</sup> Reference accumulation is averaged from three chambers at the reference chamber location. <sup>d,e</sup> Embeddedness was visually estimated as the percent of a one square foot area covered by fines along a transect upstream and downstream of each road crossing (Platts *et al.*, 1983).

### *Sediment Accumulation*

Both site ( $p = 0.007$ ) and chamber location ( $p = 0.009$ ) had a significant effect on sediment accumulation, without a significant interaction between site and chamber location ( $p = 0.244$ ). Upstream chambers accumulated significantly more sediment than reference chamber locations ( $p = 0.007$ ) (Fig. 3). The higher accumulation in upstream locations was driven primarily by large amounts of sediment at North-bridge and Central-culvert (Table 2). North-bridge had significantly greater sediment accumulation than East-culvert ( $p = 0.005$ ), driven primarily by upstream and downstream accumulation differences between the two sites (Fig. 3, Table 2).

Table 2: Mean sediment accumulation for reference, upstream, and downstream chamber locations in three stream reaches in the Yellow Dog Plains region. Proportional accumulation to reference location is shown for upstream and downstream chamber locations.

Site	Mean reference location accumulation (kg/m <sup>2</sup> /day)	Mean upstream accumulation (kg/m <sup>2</sup> /day)	Mean downstream sediment accumulation (kg/m <sup>2</sup> /day)	Mean upstream proportional accumulation to reference location	Mean downstream proportional accumulation to reference location
North-bridge	0.331	2.124	2.883	6.41	8.70
Central-culvert	0.306	1.635	0.471	5.33	1.54
East-culvert	0.375	0.364	0.271	0.97	0.72

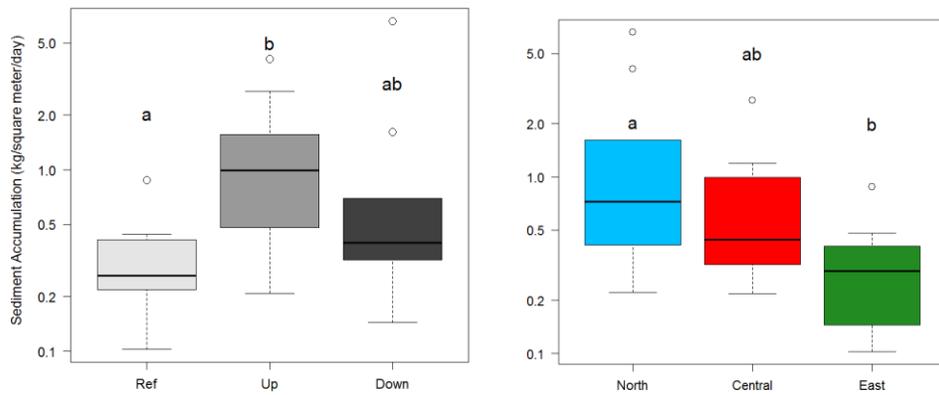


Fig. 3: Fine sediment accumulation in three streams in the Dead-Kelsey Watershed, near the Yellow Dog Plains, displayed by chamber location (left) and site location (right). Boxplots depict maximum and minimum (whiskers), median (bold horizontal line), interquartile range (box), and outliers (circles). Bars with the same letter above are not significantly different from each other (Tukey's HSD). Ref = Reference, Up = Upstream, Down = Downstream. Note log y-axis. All statistics were performed on appropriately transformed metrics.

### *Invertebrate colonization*

From 27 chambers, we identified 1184 individuals from 42 families. Abundance ranged from 1,300 to 13,100 individuals/m<sup>2</sup> (11-111 individuals per chamber), representing a range of 2 to 19 families per chamber. Although our most commonly identified family was Chironomidae, we also identified many families from the order Ephemeroptera (Table 3).

We observed families in the order Plecoptera only at East-culvert and even at that site they were in low abundances (<4 individuals). Chironomidae (Diptera) were present in 85% of chambers. A clutch (116 individuals) of recently hatched Hirudinae (Annelida) found in one chamber were omitted from our enumeration due to the absence of this taxa from any other chambers.

Table 3: The top five family groups in the Insecta class of Arthropoda ranked by abundance, with the number of chambers each family was observed in out of 27 total chambers. Chambers were deployed in three streams in the Yellow Dog Plains region of Michigan.

Rank by abundance	Number of chambers observed (out of 27)	Family	Order
1	24	Chironomidae	Diptera
2	20	Leptophlebiidae	Ephemeroptera
3	9	Leptohyphidae	Ephemeroptera
4	18	Baetidae	Ephemeroptera
5	9	Hydropsychidae	Trichoptera

Our invertebrate metrics were strongly influenced by sediment accumulation and stream location (site) (Table 4). Sediment accumulation was an important covariate, as it affected most metrics (8 of 14). Greater sediment accumulation coincided with increased proportion of Chironomidae (Fig. 4) and predators, and increased family richness (Fig.4), abundance (Fig. 7), and Hilsenhoff Biotic Index values (HBI). Proportion Ephemeroptera, EPT, EPT: C had a negative relationship with sediment accumulation; as sedimentation increased, the proportion of Ephemeroptera and EPT, and EPT: C decreased (Fig. 5).

Site location was a significant main factor or interacted with chamber location (i.e., scrapers and filterers) for all invertebrate metrics except abundance (Table 4). East-culvert had the greatest proportion of both Ephemeroptera and EPT, and highest EPT: C ( $p < 0.05$ ) and the lowest proportions of Chironomidae and family richness ( $p < 0.05$ ) (Fig. 5).

We found that only site affected the percent of Trichoptera, percent of collectors, Simpson's Index of Diversity, and Shannon-Weiner Index (Table 4). East-culvert had the lowest diversity indices of the three sites ( $p < 0.05$ ) (Fig. 6). EPT richness was not associated with chamber location or site, and was unaffected by sediment accumulation ( $p > 0.05$ ).

Abundance was significantly affected by chamber location (Table 4). The slopes of regression lines of chamber abundance based on sediment accumulation were homogenous and statistically significant ( $\beta = 0.77$ ,  $p\text{-value} = 0.014$ ,  $R^2 = 0.56$ , see Fig. 7). The downstream chamber locations were significantly higher in abundance than the upstream and reference location ( $p < 0.05$ ). There was no statistical difference in abundance between upstream and reference locations (Fig. 7).

Table 4: ANCOVA of the effects on sediment accumulation, site location, and chamber location (where significant) on 14 invertebrate metrics from data obtained from colonization chambers in the Yellow Dog Plain region. Metrics without significant factors are not shown.

<b>Dependent Variables</b>	<b>Source of Variation</b>	<b>d.f.<sup>a</sup></b>	<b>MS<sup>b</sup></b>	<b>F-value</b>	<b>P- value</b>
a) % Chironomidae	Covariate (Accumulation)	1	0.445	25.02	<b>&lt;0.0001</b>
	Site	2	0.414	23.29	<b>&lt;0.0001</b>
	Error	23	0.018		
b) % Ephemeroptera	Covariate (Accumulation)	1	0.224	7.9	<b>&lt;0.01</b>
	Site	2	0.931	32.87	<b>&lt;0.0001</b>
	Error	23	0.028		
c) % Trichoptera	Site	2	0.129	6.75	<b>&lt;0.01</b>
	Error	24	0.019		
d) % EPT	Covariate (Accumulation)	1	0.381	16.98	<b>&lt;0.001</b>
	Site	2	0.67	39.83	<b>&lt;0.0001</b>
	Error	23	0.022		
e) EPT:C	Covariate (Accumulation)	1	4.887	7.54	<b>&lt;0.05</b>
	Site	2	26.107	20.13	<b>&lt;0.0001</b>
	Error	19	12.32		
f) Family Richness	Covariate (Accumulation)	1	1.696	7.65	<b>&lt;0.05</b>
	Site	2	1.523	6.87	<b>&lt;0.01</b>
	Error	23	0.222		
g) Abundance	Covariate (Accumulation)	1	19.194	11.33	<b>&lt;0.01</b>
	Chamber	2	15.48	9.14	<b>&lt;0.01</b>
	Error	23	38.95	1.70	
h) HBI	Covariate (Accumulation)	1	0.384	21.29	<b>&lt;0.001</b>
	Site	2	0.93	51.53	<b>&lt;0.0001</b>
	Error	23	0.018		
i) Simpson's Index of Diversity	Site	2	0.730	4.81	<b>&lt;0.05</b>
	Error	24	0.152		
j) Shannon-Weiner Index	Site	2	0.961	5.43	<b>&lt;0.05</b>
	Error	24	0.177		
k) % Collectors	Site	2	0.326	8.86	<b>&lt;0.01</b>
	Error	24	0.037		

Table 4 cont.

<b>Dependent Variables</b>	<b>Source of Variation</b>	<b>d.f.<sup>a</sup></b>	<b>MS<sup>b</sup></b>	<b>F-value</b>	<b>P- value</b>
l) % Predators	Covariate (Accumulation)	1	0.081	8.36	<b>&lt;0.01</b>
	Site	2	0.228	23.62	<b>&lt;0.0001</b>
	Error	23	0.01		
m) % Scrapers	Site	2	0.044	2.64	0.099
	Chamber	2	0.015	0.92	0.416
	Site x Chamber	4	0.070	4.18	<b>&lt;0.05</b>
	Error	18	0.017		
n) % Filterers	Site	2	0.049	2.32	0.127
	Chamber	2	0.003	0.14	0.872
	Site x Chamber	4	0.087	4.14	<b>&lt;0.05</b>
	Error	18	0.021		

<sup>a</sup> d.f.= degrees of freedom <sup>b</sup> MS= mean square

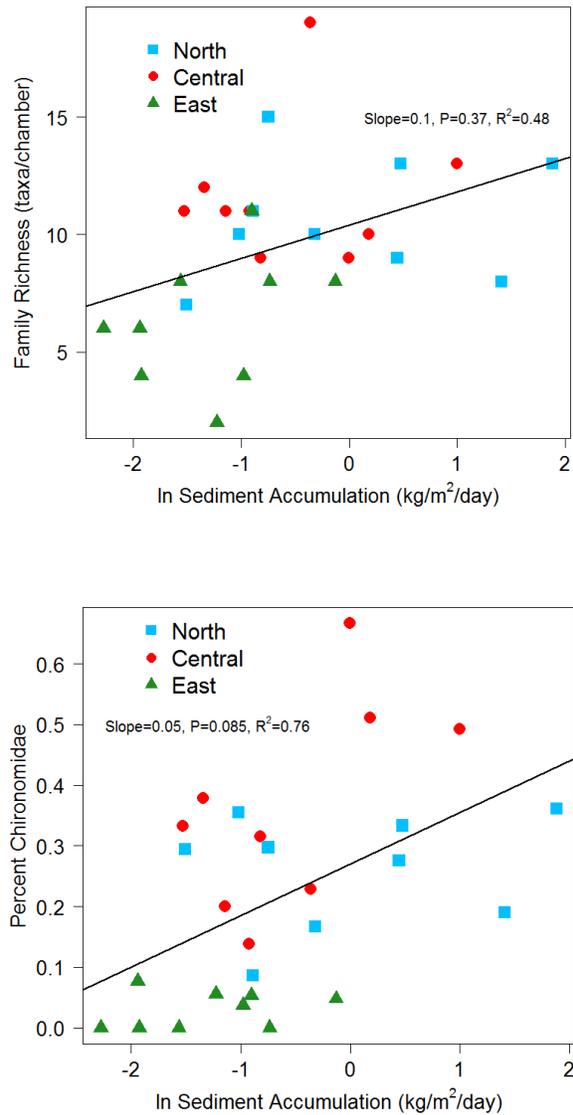


Fig. 4: Scatterplots of colonized invertebrate response to fine sediment accumulation in three streams for the Yellow Dog Plains region. Increased sedimentation was associated with a greater family richness and proportion of Chironomidae and (positive slope values). Statistics on the graph represent the slope of the homogenous regression line and its associated p-value, with the R<sup>2</sup> value of the ANCOVA model. Metrics were obtained from data collected in the Yellow Dog Plains region of the Upper Peninsula of Michigan. All statistics were performed on appropriately transformed data.

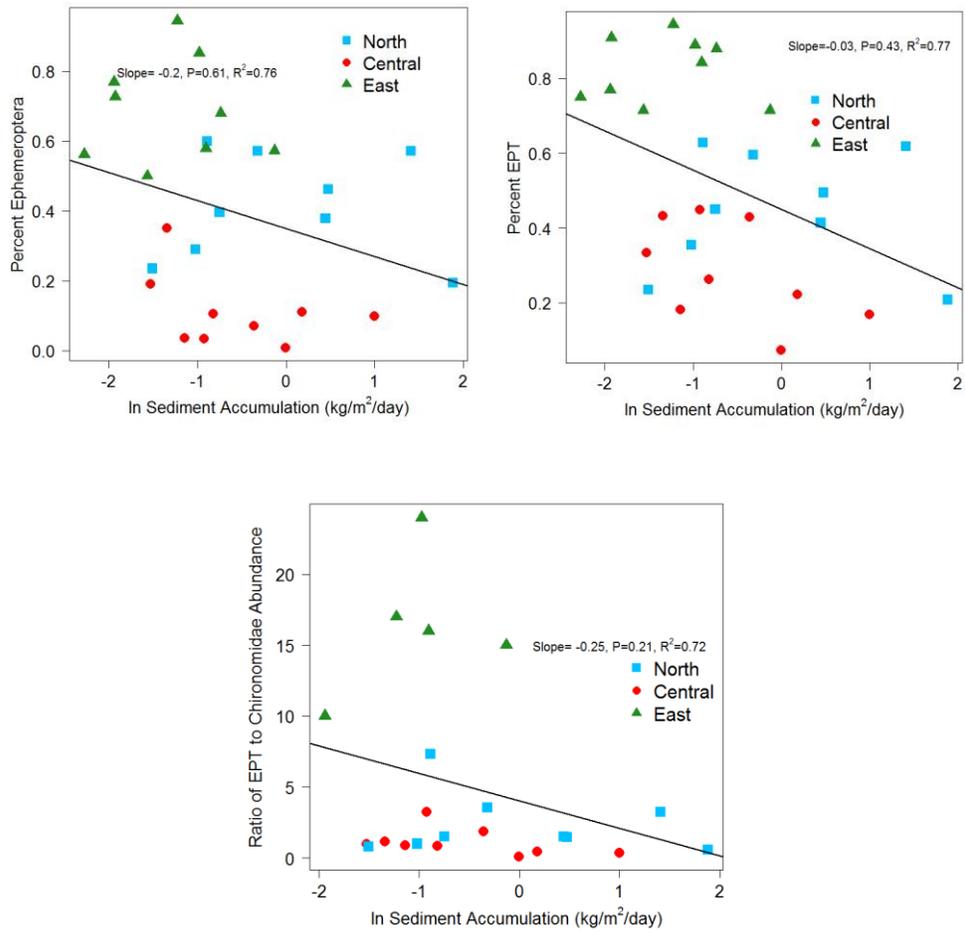


Fig. 5: Scatterplots show the relationship between sediment accumulation and three invertebrate metrics; percent Ephemeroptera, percent EPT, and ratio of EPT to Chironomidae abundance. Statistics on the graph represent the slope of the homogenous regression line and its associated p-value, with the R<sup>2</sup> value of the ANCOVA model. Metrics were obtained from data collected in the Yellow Dog Plains region of the Upper Peninsula of Michigan. All statistics were performed on appropriately transformed data.

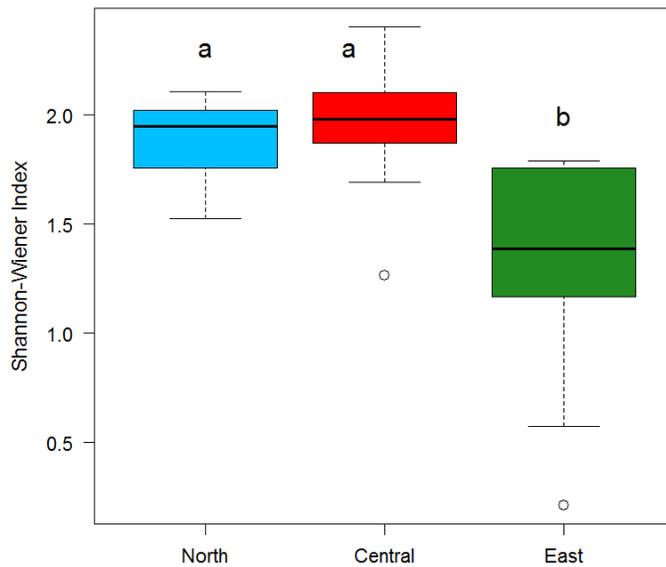
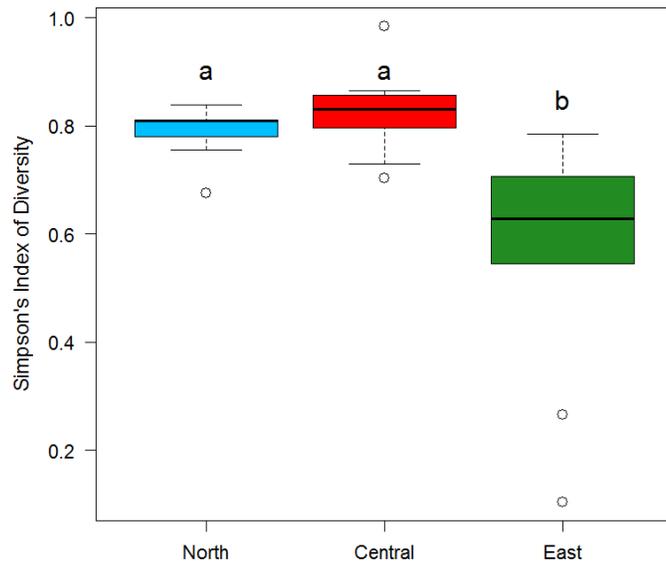


Fig. 6: Boxplots show Simpson's Index of Diversity and Shannon Wiener Index values from chambers deployed in three streams near the Yellow Dog Plains were affected by site location. Statistics on the scatterplot represent the slope of the homogenous regression line and its associated p-value, with the  $R^2$  value of the ANCOVA model. Boxplots depict maximum and minimum (whiskers), median (bold horizontal line), interquartile range (box), and outliers (open circles). Bars with the same letter above are not significantly different from each other (Tukey's HSD). All statistics were performed on appropriately transformed data.

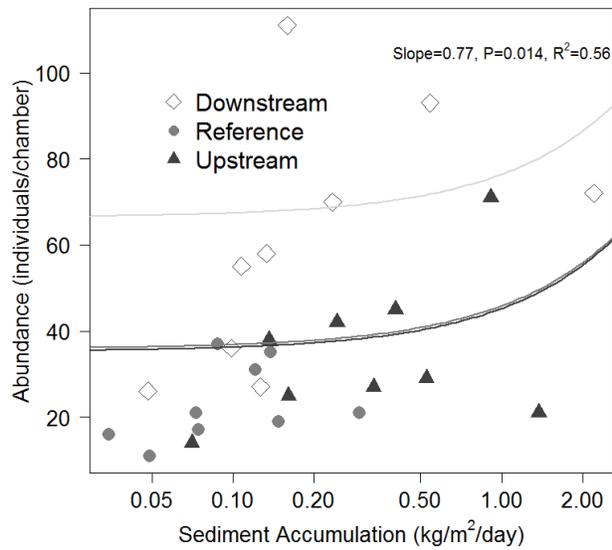
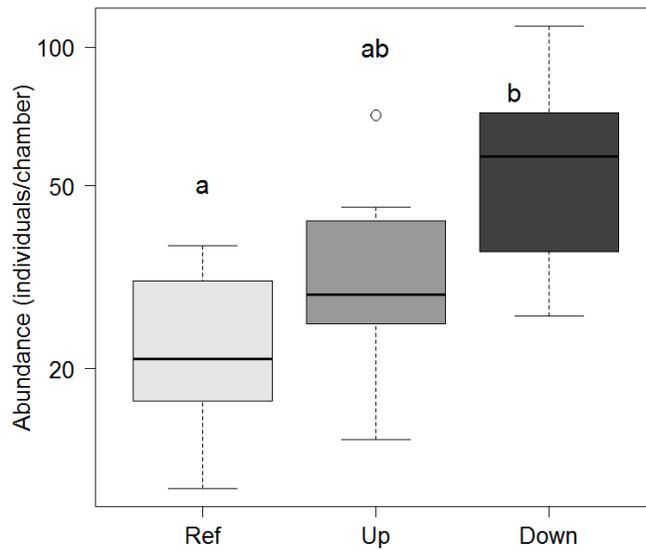


Fig. 7: Boxplots and scatterplot show insect abundance per chamber in three sampled streams near the Yellow Dog Plains region varied significantly based on chamber location and sediment accumulation. Downstream chambers had significantly higher abundances than the upstream and reference chamber ( $p < 0.05$ ). Boxplots depict maximum and minimum (whiskers), median (bold horizontal line), interquartile range (box), and outliers (circles). Bars with the same letter above are not significantly different from each other (Tukey's HSD). Note log y-axis on boxplot and log x-axis on scatterplot. All statistics were performed on appropriately transformed data.

## DISCUSSION

### *Sediment*

We detected a significant increase in sediment accumulation in chambers upstream of road crossings compared to our reference chambers. Sediment deposition can occur under all flow conditions (i.e., low, base, or post-spate flow). In low flow conditions, there is an increased deposition of fines and decaying organic matter due to decreased carrying capacity of the stream. Deposition occurs in slower moving waters close to banks, in pools, and backwater areas, including behind cobbles or boulders. Deposition can also occur at high flows, where sediment is transported into interstitial spaces and when floodwater recede. In addition, in flood conditions, bridges can slow the velocity upstream, creating a backwater area where substantial sedimentation can occur (Wood and Armitage, 1997). Because of the experimental design avoiding high depositional areas and the absence of extreme flow events (high or low), the increase in deposition is unlikely to be caused by these less-frequent flow conditions. Sediment generated from a road is most likely to reach the stream at a crossings (Cline *et al.*, 1982; Lane and Sheridan, 2002; Croke *et al.*, 2005), suggesting that the increased accumulation we observed upstream may be related to the road generated sediment and the crossing. While there was no detectable increase in accumulation found downstream of the crossings, other studies have also failed to detect a significant change in sedimentation associated with roads (Studinski *et al.*, 2011). Nonetheless, our experiment provides evidence of increased sediment accumulation adjacent to road crossings.

As with previous studies concerning road improvements, we expected to find the greatest accumulation at East-culvert, where road improvements had recently taken place (Luce and Black, 1999; Fu *et al.*, 2010; Kreutzweiser and Capell, 2001; Ketcheson and Megahan, 1996). However, East-culvert had significantly less accumulation than our reference site, North-bridge, as well as the lowest accumulation of all streams. East-culvert was the only site with active sediment collection barriers, straw bales placed next to the road above the stream to capture disturbed ground. However, the barriers appeared to be saturated, with loose sediment beyond the fences. Although we did not test the effectiveness of the sediment barriers, it appeared visually unlikely that the low sediment accumulation at East-culvert was a result of barriers.

The amount of sediment that is generated from roads depends on many different factors, such as amount and intensity of precipitation, the slope of the landscape, distance from the road to the water body, and many others. It is possible that East-culvert had the potential to receive road derived sediments, though other factors precluded the transport to the stream or sediment deposition in the chambers. Sediment that is transported aeriaily from road to stream, as opposed to overland flow during storm events, may not be substantial enough to cause an increase in sediment deposition. Because of the limited temporal span of our experiment, we

only could observe deposition under base flow conditions. If a storm event had occurred during our sampling, it is possible that deposition would have been greater than what we observed during base flow conditions.

### *Benthic Invertebrates*

Insect abundance was the only metric that varied significantly with chamber location irrespective of sediment accumulation; downstream chambers had a significantly greater number of individuals than the reference location. In addition, as sediment accumulation increased, abundance increased. However, downstream sediment accumulation was not significantly different from the reference location. Some studies submit that sediment addition can have a delay or seasonal response (Rosenberg and Wiens, 1978; Larsen and Ormerod, 2010), which is perhaps why there was no statistical difference in insect abundance between upstream and downstream chambers. Perhaps if the experiment continued for a longer period of time, we may have seen a stronger difference in abundance between the upstream and downstream chambers. Another explanation is that perhaps drifting insects from much farther upstream that need low depositional areas will not colonize the high depositional areas (upstream), and continue to drift until a preferential site has been located (downstream) (Hogg and Norris, 1991).

The increase in abundance was likely attributed to the dominant taxa, Chironomidae, which also increased with greater sediment accumulation. The response of chironomids to sedimentation can vary between sub-family groups (i.e. Orthocladiinae and Chironomae); however our family-level analysis of this group did not determine which sub-families were present in these sites (Angradi, 1999; Zweig and Rabeni, 2001). Nevertheless, Chironomidae increased in proportion with greater sediment accumulation, suggesting that the majority of chironomids were of a sediment-tolerant sub-family, such as Orthocladiinae, (Angradi, 1999).

The observed decrease in proportion of Ephemeroptera and EPT taxa with increased sediment accumulation is consistent with other studies, as these taxa tend to be more sensitive to sediment (Angradi, 1999; Hogg and Norris, 1991). Certain mayfly species have shown reduced survival and growth due to Chironomidae bioturbation, which may have affected the distribution of Ephemeroptera in the chambers with high sediment accumulation (De Haas *et al.* 2005). Irrespective of sediment accumulation, East-culvert had the greatest proportion of both EPT and Ephemeroptera. As the second most dominant taxa, Ephemeroptera strongly determined overall EPT proportion (families from Trichoptera did not respond to sediment accumulation and did not contribute strongly to the EPT metrics). The ratio of EPT to Chironomidae declined as sediment accumulation increased, which is consistent with the increase of Chironomidae and decrease of EPT proportions.

Diversity, as depicted by family richness, increased with greater sediment accumulation, contrary to other studies where taxa richness decreased (Larsen *et al.*, 2011; Angradi, 1999; Zweig and Rabeni, 2001). One possible explanation is that

sediment may add habitat complexity to the large-substrate filled chambers, which allows a more diverse community to colonize. Larsen and Ormerod (2010) observed a decrease in benthic diversity merely from adding fine sediment to the stream; however, the two diversity metrics we analyzed were not negatively affected by sediment accumulation. This suggests that these broad indices of biodiversity may be inadequate to evaluate sediment effects on sensitive taxa (i.e., Ephemeroptera).

The Hilsenhoff Biotic Index was significantly correlated with sediment accumulation and site. Perhaps because HBI relates to organic pollution, Zweig and Rabeni (2001) did not observe a significant relationship between HBI and sediment accumulation and consequently developed a tool for insect tolerance to sediment. The tool, Deposited Sediment Biotic Index (DSBI), is based on sub-family identification so we could not utilize it because of our identification level (family); however, it is potentially useful for future benthic invertebrate and sediment studies.

As predicted, some functional feeding groups were affected by site location or an interaction between site and chamber location. We were unable to detect sediment accumulation effects for collectors, scrapers, or filterers. Only predator proportion responded to sediment accumulation. Other studies also find weak or undetectable changes in functional groups due to sedimentation (Longing *et al.*, 2010; Angradi, 1999), do not measure functional feeding groups (Zweig and Rabeni, 2001), or measure another type of functional response (Larsen and Ormerod, 2010; Larsen *et al.*, 2011).

The streams in this region contain assorted substrate, from fine-grained sediment and sands, to cobbles and boulders. During high flows and spring runoff, the fine sediment is flushed from the stream bed, which will begin to accumulate again when flows return to normal. The communities that live in these streams may have adapted to this regime and the variety of substrate habitats, so an increase in road derived sediments may not have the impact on macroinvertebrate communities that we may see in other streams with less habitat diversity.

### *Conclusions*

This study suggests that road crossings are related to the sediment deposition around crossings in streams. In addition, it suggests that sedimentation may increase total invertebrate colonization in areas with larger substrate by adding habitat complexity. However, we did observe the loss of some sensitive taxa (i.e., families in Ephemeroptera) in response to increased sediment accumulation. The broad diversity metrics utilized in our study had no relationship with chamber location or sediment accumulation, and differed only by site. We expected East-culvert with a recent road modification to have the greatest sediment accumulation, yet it had the lowest of all sites. This suggests that even though a sediment-generating disturbance takes place adjacent to streams, excess sedimentation in the streams is not an absolute outcome. However, the short temporal scale of our sampling (40 d, no storm events) does not

account for all potential sediment deposition events such as storms, and these rarer events can be great contributors of significant sediment (Megahan and Kidd, 1972).

The limited spatial area covered by the chambers, as well as the low number of replications, appear to limit our ability to measure sedimentation impacts from a road crossing, which may be better observed from a reach perspective. While our results indicated an increase in chamber sediment accumulation upstream of the road crossings, we should use caution when scaling up results from chambers to a broader area (upstream). Perhaps a more systematic and multi-method sampling method, such as used by Longing *et al.* (2010), to develop a degree of sedimentation for a reach would be more appropriate. The chambers were useful for specifically exploring the effects of sediment accumulation on the invertebrate community; however, more chambers across a greater spatial extent in each stream would improve our understanding of the effect of road construction on these streams.

As with many field experiments in ecology over several weeks, this study was subject to uncontrolled variables and limitations. Chambers can be tampered with or removed (as was the case in this experiment which originally encompassed five sites), as well as subjected to the changing morphology of a small stream. For future studies on sedimentation and macroinvertebrates, researchers should classify invertebrates to sub-family levels in order to utilize the DSBI (Zweig and Rabeni, 2001) and assess its potential as a tool for evaluating sediment pollution impacts on macroinvertebrates.

Our experiment examined the patch scale composition of invertebrates and sediment deposition, and allowed a direct analysis of the two chamber components. Studies that examine reach scale sediment and invertebrate metrics do not always obtain strong correlations; however, increasing the sampling resolution to patch scale can possibly show stronger sediment effects (Larsen *et al.*, 2009). This experiment contributes evidence to an extensive collection of studies on the diversity of macroinvertebrate responses to sedimentation (Larsen *et al.*, 2009; Zweig and Rabini, 2001; Angradi, 1999; see also Jones *et al.*, 2012 and Wood and Armitage, 1997). In addition, while our observations did not find that East-culvert is currently impacted by excess sediment accumulation, the mining and mineral transportation has not yet commenced, so it should be monitored for potential future effects. For future research, sampling under diverse flow conditions, across a seasonal or temporal gradient may prevent sedimentation effects from going undetected in naturally sandy, yet high quality streams, such as the Salmon Trout River.

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## Appendix A:

Functional family group and tolerance scores of macroinvertebrates encountered in sampling.

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<b>Class</b>	<b>Order/subclass</b>	<b>Family</b>	<b>Functional Family Group</b>	<b>Tolerance Score</b>
Malacostraca	Amphipoda	Taltridae	GC	8
Clitellata	Hirudinea		PR	10
Clitellata	Oligochaeta	Oligochaeta	GC	8
Bivalvia	Veneroidea	Sphareiididae	FC	8
Insecta	Coleoptera	Elimidae	GC	4
Insecta	Coleoptera	Hydrophilidae	PR	5
Malacostraca	Decapoda	Cambaridae	GC	6
Insecta	Diptera	Athericidae	PR	2
Insecta	Diptera	Ceratopogonidae	GC	6
Insecta	Diptera	Chironomidae	GC	8
Insecta	Diptera	Simuliidae	FC	6
Insecta	Diptera	Tabanidae	PR	6
Insecta	Diptera	Tipulidae	SH	3
Insecta	Ephemeroptera	Ameletidae	GC	0
Insecta	Ephemeroptera	Baetidae	GC	4
Insecta	Ephemeroptera	Caenidae	GC	7
Insecta	Ephemeroptera	Ephemerellidae	GC	1
Insecta	Ephemeroptera	Heptageniidae	SC	4
Insecta	Ephemeroptera	Leptohyphidae	GC	4
Insecta	Ephemeroptera	Leptophlebiidae	GC	2
Gastropoda	Physidae		SC	8
Gastropoda	Planorbidae		SC	7
Insecta	Megaloptera	Corydalidae	PR	0
Insecta	Megaloptera	Sialidae	PR	4
Insecta	Odonata	Aeshnidae	PR	3
Insecta	Odonata	Calopterygidae	PR	5
Insecta	Odonata	Corduliidae	PR	5

<b>Class</b>	<b>Order/subclass</b>	<b>Family</b>	<b>Functional Family Group</b>	<b>Tolerance Score</b>
Insecta	Plecoptera	Capniidae	SH	1
Insecta	Plecoptera	Chloroperlidae	PR	1
Insecta	Trichoptera	Brachycentridae	GC	1
Insecta	Trichoptera	Glossosomatidae	SC	0
Insecta	Trichoptera	Hydropsychiidae	FC	4
Insecta	Trichoptera	Hydroptilidae	GC	4
Insecta	Trichoptera	Leptoceridae	GC	4
Insecta	Trichoptera	Limnephilidae	SH	4
Insecta	Trichoptera	Molannidae	SH	6
Insecta	Trichoptera	Philopotamidae	FC	3
Insecta	Trichoptera	Phyrganeidae	SH	4
Insecta	Trichoptera	Polycentropodidae	FC	6
Insecta	Trichoptera	Psychomyiidae	GC	2
Insecta	Trichoptera	Rhyacophilidae	PR	0

## Appendix B: Site locations

Location of three sampled reaches near the Yellow Dog Plains, in the Dead-Kelsey Watershed. Sites were located in Marquette County, Michigan.

Site	North-bridge	Central-culvert	East-culvert
Latitude (N)	46°52'58.16"	46°45'3.25"	46°45'18.94"
Longitude (W)	87°52'8.13"	87°54'26.96"	87°48'16.69"

