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ASSESSING MACROINVERTEBRATE COMMUNITY RECOVERY IN POST  
RESTORATION SILVER BOW CREEK, MONTANA

By

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Bachelor of Arts, University of Montana, Missoula, Montana, 2003

Thesis

Presented in partial fulfillment of the requirements  
for the degree of

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in Environmental Studies, Environmental Science

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ASSESSING MACROINVERTEBRATE COMMUNITY RECOVERY IN POST  
RESTORATION SILVER BOW CREEK, MONTANA

Chairperson: Dr. Vicki Watson

Since the turn of the twentieth century, mining activities have contaminated the floodplain and streambed of Silver Bow Creek, Montana, resulting in a streambed devoid of life and severely contaminated with heavy metals. In the mid nineteen seventies, up-stream water treatment facilities were upgraded and water quality improved, bringing benthic invertebrates back to reaches of Silver Bow Creek. The extent and concentration of toxicants in and around the streams of the Upper Clark Fork River Basin resulted in the designation of over 100 miles of river as Federal Superfund sites. Since 1999 reclamation and restoration efforts have been implemented on Silver Bow Creek. This analysis evaluates changes in benthic biotic community composition throughout the period of record (1986 to 2009). Transformations of historical data were necessary to standardize community information and calculate indices of biotic integrity. A multivariate method, Classification Strength (CS), used in conjunction with non-parametric tests of significance, demonstrated data comparability over the period of record both taxonomically and ecologically. Biotic index results indicate that remedial efforts to remove metals laden sediment from the stream bed and surrounding floodplain have resulted in a decline in the numbers of metal-tolerant organisms. Generalized indices of biotic integrity show no significant changes throughout the period, while specialized indices demonstrate increases in organic-pollutant-tolerant taxa. Multivariate analysis of community composition demonstrates taxonomic changes to the resident community throughout the period of record, and Indicator Species Analysis corroborates the results of the biotic indices. Using these methodologies as a template to measure change throughout the restored reaches of Silver Bow Creek will increase the ability of resource managers to measure the success of restoration of the 'Last Best Disturbance'.

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## INTRODUCTION

Reclamation and restoration of streams and waterways has become common place throughout the world (Palmer 2009). Billions of dollars are spent annually, in the United State alone, on river restoration (Palmer 2003, Malakoff 2004). Many efforts have been made to restore areas damaged by historic activities in a holistic ecologically sound manner; however, monitoring and assessment have been neglected until recently (Palmer et al. 2005). The Rocky Mountain West is no exception, and is often faced with the burden of reclaiming areas damaged by acid mine drainage and subsequent metal pollution in and around streams (Fore 2003). As a result of historic mining practices, biotic communities in streams may suffer deleterious effects; moreover the removal or treatment of contaminants may continue to have effects on the benthic community for years (Chadwick 1973).

Many attempts have been made to measure the response of biological communities to restoration techniques (Lepori 2005, Griffith 2001, Muotka 2002, Hassett 2005). However, with the exception of game fishes, little is known about the effects of restoration on stream biota (Muotka 2002). Using community metrics for assessing the degree of recovery is commonplace, but may not explain the causal mechanisms responsible for recovery ( Adams 2002).

Determining the success of a river restoration project typically requires the analysis of pre-restoration conditions with respect to the target population (Palmer et al. 2005). Due to the extent of damage, both spatially and temporally, the Clark Fork River basin is an ideal landscape from which to measure the effects restorative practices have on a variety of ecological communities. Silver Bow Creek has received reclamation and restoration, and certain reaches are in the process of recovery. This analysis aims to evaluate the changes in the benthic community measured by annual monitoring.

### Site History

The copper, gold and silver veins that ran through the mountains of the Continental Divide near the town of Butte, Montana, were tapped for their valuable resources over a century ago, decades prior to the implementation of the United States Clean Water Act. Heavy rain events washed toxicant laden

burden into the surrounding waterways. Silver Bow Creek, historically originated near the continental divide near what is now known as the Yankee-Doodle Tailings and Berkley Pit; mining activities channelized and manipulated the creek to serve as a conduit of waste from both private and public sources. Silver Bow Creek extends from Butte approximately 23 miles to the Warm Springs Ponds, a water treatment facility located at the headwaters of the Clark Fork River. Since the late 1800s, tailings and other mine wastes containing elevated concentrations of metals have been discharged to or otherwise entered Silver Bow Creek through flood events. These toxic discharges contaminated the stream and floodplain with heavy metals and eliminated the majority of aquatic life in the stream. Tailings deposited in the floodplain are toxic to plants and have resulted in a floodplain that is largely devoid of vegetation and is generally incapable of supporting wildlife (NRDP 2005).

In accordance with the Clean Water Act of 1972, the mining operations were instructed to implement waste water treatment in and around the city of Butte in the fall of 1972. Early benthic ecologists, James Chadwick and Steven Canton, saw this as an opportunity to conduct a study on benthic invertebrates' response to water treatment upgrades in the area. Sampling was initiated at 5 sites along the contamination gradient from the Butte District Discharge to what is now known as the Mill/Willow Creek Bypass. From 1972 to 1975, no invertebrates were detected in Silver Bow Creek (Chadwick 1986). Chadwick and Canton (1986) found that in the 10 years after the improved water treatment, the invertebrate community was just starting to recover, and metals in the substrate likely limited the recovery of the benthic community.

More recently, since 1986 benthic invertebrate sampling has been conducted as part of the Clark Fork River Bioassessment funded by the Environmental Protection Agency (EPA) and conducted by McGuire Consulting, Inc. The Clark Fork River monitoring was conducted on a large watershed scale, ranging from Silver Bow creek in the headwaters to below the confluence with the Flathead River over 200 river miles away.

In 1983, after the passing of the Comprehensive Environmental Response, Compensation and Liability Act (CERCLA), the State of Montana filed a natural resource damage lawsuit against the Atlantic Richfield Company for damages to water, soils, fish, and wildlife in the basin. As part of the 1999 (Montana v. ARCO) settlement, 230 million dollars were earmarked for the reclamation and subsequent restoration of Silver Bow Creek (NRDP 2005). The Natural Resource Damage Program (NRDP), a division of the Montana Department of Justice, has been monitoring stream biota on an annual basis with an increased effort within the Stream Side Tailings Operable Unit (SSTOU) to aid in evaluating the progress of the reclamation and restoration of the Clark Fork River Basin. In addition to chemical, soil, and vegetation sampling, benthic invertebrates have been collected in this process.

Benthic invertebrates are effective aquatic ecological assessment tools due to their ubiquitous nature and sampling ease (Rosenberg 1993; Chu and Karr 1999). Specific restoration targets for benthic communities have been established for Silver Bow Creek in an effort to monitor the response of this vital ecological community (NRDP 2004). Biotic indices, such as the Revised Montana Foothill/Valley and Prairie Ecoregion (Bollman 1998) and Montana Intermontane Valleys Index (Bukantis, 1998), have been chosen by the Natural Resource Damage Program as the primary measures of benthic invertebrate community health (NRDP 2007) due to the robustness of the indices to measure a wide variety of environmental conditions in the benthic community. The goal, as stated in the comprehensive long term monitoring plan for the Silver Bow Creek SSTOU is to achieve a Biotic Index score of 75% of reference condition (fully supporting aquatic life) for two consecutive annual monitoring events (DEQ and NRDP, 2004). In addition to numeric goals associated with the restoration and reclamation, a narrative goal regarding the benthic community states; "Restoration will reflect a balanced, integrated, and adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of the natural habitat of the region (NRDP 2004, Karr and Dudley 1981)." The narrative goal mirrors the conditions often found in reference stream systems.



As stated earlier, annual assessment of biotic communities in the SSTOU has been undertaken by the state, however the goals of restoration fail to take into account the nearly 20 continuous years of pre-restoration data (dating back to the mid 1970's) as a baseline from which to measure restoration success. In addition to tracking changes towards a reference condition, bioassessment functions as a tool to track changes away from contaminated conditions.

## SCOPE AND PURPOSE

Bioassessment type monitoring has, as mentioned above, taken place since the early 1970's in Silver Bow Creek. Throughout this time, collection methods, sampling effort, taxonomic rigor, study designs and bioassessment tools have changed resulting in a quagmire of data spanning a quarter of a century. Through an extensive review I have found no evidence of efforts to compare historical and present benthic community data from Silver Bow Creek using identical bioassessment tools. The goal of this thesis is to determine the effect, to date, of remediation and restoration on benthic invertebrate communities of Silver Bow Creek. Determining the effect of restoration in Silver Bow Creek on aquatic biota will be assisted by answering the following: 1) Have significant changes occurred in the measure of biotic indices? 2) Has community composition been significantly altered since reclamation and restoration? To answer the aforementioned questions a sequential list of objectives were identified:

- 1) Obtain annual monitoring data from historical and current monitoring efforts.
- 2) Evaluate data for comparability and period of record analysis.
- 3) Identify a single site within the network of monitoring sites to be used in this study.
- 4) Homogenize (make comparable) historical and current data and test methods of homogenization.
- 5) Using previously developed indices, determine trends and track changes in aquatic invertebrate index scores pre and post reclamation in Silver Bow Creek.
- 6) Evaluate and describe changes in community composition over the period of record: 1986 to 2009.

## METHODS

Prior to detailing each of the objectives' methods, a brief description of the Silver Bow Creek watershed is provided.

### Description of Silver Bow Creek Watershed:

Silver Bow Creek is a first order stream located in western Montana and is a major tributary of the headwaters of Upper Clark Fork River. Silver Bow creek is approximately 22 miles long and spans two distinct level IV ecoregions of Montana with the upper reaches flowing through the Dry Intermontane Sagebrush Valleys (17aa) and lower reaches flowing through the Intermontane Hills and Valleys (17ak) (Woods et al. 2002). The upper reaches of Silver Bow Creek drain 267 square kilometers with an elevation of 1649m with elevation dropping to 1460 meters and draining an additional 958 square kilometers approaching the confluence with Warm Springs Creek (USGS 2010). Review of the annual hydrographs from the gauging stations on Silver Bow creek reveals a hydrograph that is dominated by spring snow melt.

The Silver Bow Creek watershed is dominated by the Cretaceous granitic rocks of the Boulder Batholith; granitic rocks typically form fine grained sediments in streams and do not contain an abundance of gravels ( NRDP 2005)(Figure 2). Soils in the area have suffered deposition, both aerial and fluvial, of phytotoxic contaminants for over a century and little is known about the pre-contamination conditions of the soil or stream substrate. (NRDP 2005).

### Objective #1: Obtain annual monitoring data from historical and current monitoring efforts

Bioassessment produces several types of data. Raw data is in the form of taxa lists with associated abundances. Raw data is often summarized using community metrics and indices, which help describe certain attributes of the community present in each sample. Also important is metadata-- the information describing the procedures for collection and analysis of samples-- which allows the researcher to verify locations, analyze comparability, and assess repeatability.

The data mined for these analyses come from two main sources; the Montana Department of Environmental Qualities' Annual Clark Fork River Monitoring (ACFRM) (1986 to 2001), and the Montana Department of Justice, Natural Resource Damage Program (NRDP) Annual SSTOU monitoring (2003 to 2009). The two data sources differ in their taxonomic contractors, sampling effort, collection and analytical methods, and ecological interpretation tools. Specific differences in methods and procedures will be discussed in following section of this analysis. No additional sampling was conducted for this analysis, and all data are public accessible through requests made to the Montana Department of Environmental Quality Library, Helena, Montana.

#### Objective #2: Evaluate data for comparability and long term analysis

Data sampling and analysis methods between the two contracting laboratories varied highly. ACFRM used a different method for sample collection and analysis than did the Natural Resource Damage Program. Data produced by the ACFRM effort was collected ,processed , and ecological interpretations made by McGuire Consulting. Data produced by the monitoring effort funded by the NRDP of Montana was collected by Confluence Consulting and taxonomic determinations made by qualified taxonomists at Rhithron Associates, Inc. This thesis will evaluate all data throughout the period of record for ecological analyses.

#### Collection and Sample Analysis Methodologies:

The ACFRM dataset was obtained using the following methodology. Four replicate quantitative samples were taken at each site using a Hess sampler and preserved in 95% Ethanol until sample processing could take place. Samples were processed by removing all organisms from the collected substrate; those organisms were then identified to the lowest practical taxonomic resolution, in most cases genus or species. Laboratory method descriptions indicate that reference collections were made and delivered to the Montana Department of Environmental Quality, as a measure of quality assurance.

The NRDP dataset was generated using a single index sample collected using Montana Department of Environmental Quality standard operating procedures for collection of macroinvertebrates

( MTDEQ 2006). This methodology employs a single transecting, multi-habitat, Kick-net sample preserved in 95% Ethanol. Samples were delivered to Rhithron Associates for processing. Samples were processed using the MTDEQ standard procedures for macroinvertebrate sample analysis. A target number (300) of individuals is randomly selected from the substrate. The “blocked” random selection of individuals is conducted by a trained biological technician by placing the collected substrate into a Caton tray, an evenly divided mesh tray, and whole portions are randomly selected and organisms removed until the target number of individuals is met or the whole sample is processed. The organisms are then identified and enumerated to the lowest practical resolution (genus/species). Quality assurance measures are implemented at both the subsampling process and the identification process. As a measure of quality assurance, ten percent of the sorted residue sampled by a technician is re-sampled by another technician. Any organisms found in the re-sorted substrate are then included in the sample for identification and a Sorting Efficiency is calculated (Equation 1). Taxonomic determinations also undergo a similar quality assurance step. Ten percent of the samples are randomly selected for identification and enumeration by another taxonomist. Sample taxa lists are then compared using a Bray–Curtis Similarity index (Equation 2). Any samples failing to have greater than or equal to 95 percent similarity are rectified by the two taxonomists and data corrections are made accordingly.

Objective #3: Identify a single site within the network of monitoring sites to be used for this analysis.

In order to answer the primary research question of this analysis, a site (or sites) must be selected that has long term records of invertebrate community composition both pre and post reclamation efforts. In 2002 Sub-Area one of the SSTOU received the final treatment of restoration following the massive re-channelization and remediation of metals contaminated soils. Since this area has the longest post restoration data record, sites within this area are the primary targets of this analysis. Three sites within SSTOU Sub-Area one have been monitored for invertebrates throughout the majority of the study period and are located on Figure 1 as SS-06(Silver Bow Creek at the WWTP),SS-07( Silver Bow Creek below the WWTP),and SS-08 (Silver Bow Creek at Rocker).

Silver Bow Creek above the Waste Water Treatment Plant (WWTP) , was monitored almost continuously prior to remediation efforts, but data acquisition demonstrates significant gaps in data following 2002, making this a poor choice for analysis. Silver Bow Creek below the WWTP, SS-07, is located almost directly downstream of the effluent of the Butte-Metro Storm Drain and the fallout of the Butte WWTP. Influences of the WWTP effluent and the Butte Metro Storm Drain could be significant at this site and pose a potential confounding factor in determining change as a result of remedial activities. However, Silver Bow Creek at Rocker is located almost 1.0 miles from SS-07, and no significant surface flow contributes to the stream discharge within the reach, potentially isolating the effects of remediation and restoration on the benthic community.

Remedial activities and restoration efforts were completed in 2002 at Rocker. The data set shows that ACFRM data at the site ended in 2001, and NRDP monitoring started in the summer of 2003; therefore, this site has the oldest and most continuous dataset throughout the period of record, making it an ideal candidate site for these analyses.

Throughout the remainder of this analysis, the site Silver Bow Creek at Rocker will be the subject of all analyses, unless otherwise stated. It is recognized that periods of transition, i.e. 1999-2002, may present some variability in the data due to the activities on site throughout this time frame, however, the year 2003 will be the date used to mark the difference between pre and post restoration conditions.

#### Objective #4: Homogenize data and test methods of homogenization

Due to the varied methods used for collection and analysis of invertebrate communities, it is necessary to perform systematic alterations to the data collected between the years of 1986 and 2001 in order to make the data comparable and evaluate changes and trends in community composition and indices.

#### Data Homogenization Method:

Sample data were first evaluated for taxonomic consistency, and the taxonomic resolution was adjusted where needed. For example, in the ACFRM data, species level determinations were made for the genus *Hydropsyche*. In many cases, taxa reported with a species determination were followed by a “?” indicating a low level of taxonomic certainty. Conversely, data produced for the NRDP left *Hydropsyche* at genus, thus all species level identifications for *Hydropsyche* were consolidated to genus. Similar examples exist throughout the dataset, and all resolution discrepancies were rectified by consolidating the taxon to the higher resolution so as to avoid bias between taxonomic efforts. Additionally, any “suspect” identifications were flagged and taxonomic resolution was reduced (i.e. *Baetis punctiventrus* = *Baetis*).

Differences in sampling area, sample size, and sampling effort require the standardization of data to a fixed count method for comparison (Cao 2005). A random sample of 300 individuals was systematically selected from the first of the four replicate samples taken for the ACFRM effort. A random selection of the individuals allows for no bias in selecting a specific individual, thus some taxa were “lost” to the re-sampling process. The inherent nature of rare taxa limits the probability of a rare or unique taxon to be selected during re-sampling, and has been shown to be consistent with fixed count sub-sampling efforts (Cao 2003, Hawkins 1996, Rai 2010).

### Testing the Data Homogenization Method:

As with any data manipulation, it is important to test the data's sensitivity to alteration. Samples were not taken from the site selected during the period 2003-2009 using the ACFRM methodologies; therefore a direct intra-site comparison could not be executed. However, sampling using both methods, ACFRM and MTDEQ, was conducted intermittently post reclamation at the site known as Silver Bow Creek at Opportunity, thus presenting an ideal candidate to test the data alteration method used in this analysis. Sampling coincided using both methodologies in the years 2004, 2006, 2007 and 2008, with both collections happening in mid-late August of each year. Data reduction methods, as described above, were used to reduce the number of individuals in each sample. Taxa lists were generated as a result of this sub-sampling and community metrics calculated.

Two methods were used to validate the alteration of historical data to a fixed count methodology, Classification Strength (Van Sickle 1997, Cao et al. 2005) of taxonomic similarity and Wilcoxon Sign Rank tests (Zar 2010) of index scores between the two data sets .

Classification Strength: Classification Strength (CS) was developed as a tool for the characterization of ecoregions by using environmental data and multivariate statistics (Van Sickle 1997). Cao et al. 2005, evaluated this measure for use in comparing methodologies of aquatic biota sampling, and found that samples using differing methods of collection ranged between 77-99% mean similarities when based on the Bray-Cutis Index. Studies indicate that the use of community similarity measures, based solely on taxonomic structure is a preferred alternative to assessing similarity , rather than comparing metric or index scores, due to the lack of direct taxonomic comparison of the latter (Cao 2005, Van Sickle 1997). Characterization strength is calculated by first computing all known distance measures between all pairs of samples. In this case I have chosen to use the Bray- Curtis distance as the measure of similarity due to the widespread use of this similarity index in community ecology. Using PC-ORD v.5.1 (McCune 2010) to calculate the matrix of distance measures , three descriptive statistics are then calculated 1) mean similarity between groups, 2) mean similarity within group one ( reduced ACFRM data) and 3) mean similarity within group 2 (NRDP data ). The calculated statistics are then placed into



(Equation 3) to determine the Classification Strength. The resulting statistic may be interpreted as a percent comparability (Cao 2005). No standard has been set for the acceptable comparison of methods using this classification tool, therefore if the CS value is greater than 77%, as found by Cao et al. (2005), then the methods will be considered comparable.

Non-Parametric Paired Sample Test: During this analysis, the primary question targets trends in biotic indices before and after restoration; therefore it is appropriate to compare the index scores generated by the two methodologies. Metrics and indices were calculated for each of the samples taken using each of the sampling methods, ACFRM and NRDP, and compared using a paired sample t-test. It is common for ecologists evaluating community data to use non-parametric tests due to the inability to ensure all assumptions are met for parametric analysis (Zar 2010, McCune and Grace 2002). The test, Wilcoxon Signed Rank Test, will evaluate the two-tailed null hypothesis: “Index scores derived from the two methodologies scores are not significantly different.” All statistical tests were performed with an alpha level set at .05.

Objective #5: Analyze trends and change in aquatic invertebrate communities pre and post reclamation in Silver Bow Creek, using selected indices.

Restoration success for Silver Bow Creek is to be evaluated using two common biotic indices produced for Montana ecoregions, the Montana Revised Valley/Foothills Index (Bollman 1998) and the Montana Intermontane Valleys Index (Bukantis 1998) (NRDP 2004). Multimetric indices use a host of candidate metrics to compute a unit-less score based on the metrics selected and their known response to anthropogenic impacts (Barbour 1998).

A multimetric index combines tested and calibrated metrics or indicators and transforms them to a unit-less score; often this score is then used to assess the benthic integrity of a stream's community (Karr and Chu 1999). The multimetric index produced by Bollman in 1998 uses 6 metrics to evaluate community integrity: Ephemeroptera Richness, Plecoptera Richness, Trichoptera Richness, Sensitive Taxa Richness, Percent Filterer and Percent Tolerant. The Montana Intermontane Valleys Index

produced by Bukantis et al. (1998) uses 8 metrics to evaluate community integrity: Taxa Richness, EPT Richness, Hilsenhoff Biotic Index, percent Dominant, Percent Collectors, Percent Scrapers and Shredders, percent Hydropsychinae of Trichoptera, and percent EPT. A side by side comparison of the metrics contained in each candidate index is shown in (Table 1).

This analysis will focus on the use of the Revised Montana Valley/ Foothills Index( RMVFI) (Bollman 1998) for three primary reasons, 1) No metrics used in the Bollman index are potentially highly correlated with each other (e.g EPT richness to % EPT, as in the Montana Intermontane Valleys Index), 2) Historical use of these indices demonstrates that the RMVFI is more conservative than MIVI for Silver Bow Creek, providing a stronger test of significant differences between pre and post restoration conditions ( NRDP 2008),3) The RMVFI was developed and tested in the same ecoregion as the Silver Bow Creek watershed and has shown strength in discriminating impaired and non-impaired sites for environmental stresses commonly found within the ecoregion (e.g. grazing, riparian integrity, deforestation, and mining). RMVFI scores derived from this analysis are reported as a percent of total possible, matching the stated goals with the restoration goals of the Natural Resource Damage Program. When comparing indices of varied authors and constituents, ‘percent of possible’ normalizes the scores on a scale of 0-100, and streams scoring near 100 are typically identified as “reference” streams.

In addition to the RMVFI ,this analysis will include two additional indices that are used to analyze specific environmental perturbations, metals and low oxygen conditions. Both the Metals Tolerance Index (McGuire 2001) and the Hilsenhoff Biotic Index (Hilsenhoff 1987) use numeric tolerance values assigned to each taxon to help describe the tolerance of the community present in relation to their respective environmental stress. For example the HBI is calculated by summing the relative abundance of each taxon multiplied by that taxon’s tolerance value (Equation 4). The Metals Tolerance Index and the Hilsenhoff Biotic Index are evaluated using a range of scores, 0 being the least tolerant and 10 being the most tolerant. For each index, as seen in Tables 2 and 3, qualitative associations of water quality and severity of impairment have been defined. This analysis will include both of these additional

indices due to the known relevance of each of these stressors in the Upper Clark Fork River Watershed (Ingman and Kerr 1989).

#### Change and Trend Assessment Methods:

In determining the trends of each of these indices over time, two methods were used for each of the indices of concern. In order to describe the scores of each index over time, the index score was plotted against time. This analysis provides a subjective and visual assessment of benthic data, after homogenization of methods, of index scores over time. Secondly, a time-series analysis test was conducted on each of the indices measured in this thesis using a common water quality trend test, The Sen's Slope (Samli 2002). This analysis is preferred for use of annual stream monitoring data to determine the significance of trends in equally space intervals (Ingman, G.L. *Personal Communication*, May 4<sup>th</sup>, 2010).

In addition to the above trend analyses, a non-parametric ranking test (Mann-Whitney U- test) was used to test the one-tailed null hypothesis: pre-restoration and post restoration index scores are the same. All statistical tests were implemented using a set alpha level of .05.

#### Objective #6: Evaluate and describe changes in community composition pre and post restoration.

An additional goal of the restoration occurring on Silver Bow creek is to achieve a community that “reflects a balanced, integrated, and adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of the natural habitat of the region (NRDP 2004, Karr and Dudley 1981).” In order to evaluate and describe the changes in the community composition of Silver Bow Creek, several methods were used.

#### Principal Component Analysis:

A Principal Component Analysis (PCA) is a data reduction tool. The goal of a PCA is to express covariation in many variables in a smaller number of composite variables (McCune and Grace 2002). Silver Bow Creek data are limited by the number of samples (23 years) that have been obtained in

comparison to the number of variables (taxon), thus the number of variables must be reduced to prevent confounding covariance measures. Variables were reduced from over 63 variables (unique reported taxa) to 6 by assessing the number of non-zero values within each variable (taxa). All taxa with less than 13 non-zero values (based on abundances of taxa) were selected and resulted in the 6 most common taxa throughout the dataset. A general rule of sample size for PCA has been stated as five sample units for each observed variable (Tabachnick 1989). Although the variable sample ratio of 6 :23 falls outside this guidance, no additional reductions of variables could be made without potential ecological bias. Using PC-ORD (McCune 2010) PCA was completed on the dataset from Silver Bow Creek, with the modifications as mentioned above and using a variance-covariance cross products matrix. Data were then plotted in species space and were used as a descriptive tool in determining the variation in taxonomic composition throughout the period of record. Because PCA was used as a descriptive tool, no additional randomizations were conducted to discern significance of the axis.

#### Indicator Species Analysis:

Indicator Species Analysis (ISA) is a multivariate tool used to describe the relationship species have to *a priori* defined groups (McCune and Grace 2002). ISA is based on the comparison of relative abundances and relative frequencies of occurrence of taxa in different groups of sites, and identifies taxa that vary more between groups than would be expected by chance. Indicator Values (IV) vary between 0 and 100, and reflect the percent strength of the discrimination between groups. In analyzing Silver Bow Creek, ISA assists in describing the significance a particular species plays in distinguishing the two pre-defined groups. In this analysis the two groups were determined to be pre-restoration (1986-2001) and post restoration (2003-2009). The ISA assisted in verifying the exploratory analysis conducted using the PCA. Using PC-ORD (McCune 2010) to conduct the Indicator Species Analysis developed by Dufrene and Legendre (1997), indicator species were identified as a measure of Indicator Value (IV) based on the observed and expected similarities of each taxon in relation to the group in which they belong. Ideal indicator species of a particular group maintain two characteristics-- presence and exclusivity within and

between groups ( McCune and Grace 2002). A Monte Carlo test of significance, with 1000 randomizations, evaluated the null hypothesis: there is no significant difference between the groups, with an alpha set at .05. The p-value is based on the proportion of randomized trials with the indicator value equal to or exceeding the observed indicator value (McCune and Grace 2002)

#### Relative Abundance:

Relative abundance can be calculated in a variety of ways (MacArthur 1960). In this analysis relative abundance was represented by the percent composition of major taxonomic groups (Order). The percent of each taxonomic group (Order) present in each sampling event was plotted against time to assist in describing the changes in community composition on a very broad scale. In addition to examining the relative abundance of the major orders, relative abundance of each Functional Feeding Group (FFG) (Merritt et al. 2008) was evaluated over time to describe the historical and present composition of each feeding group. Analysis of the relative abundance of each taxonomic group and FFG will demonstrate changes in the structure of the resident community through time. Detecting change using this measure will require dramatic changes in community composition over the period of record, due to the taxonomic resolution of the analysis.

## RESULTS

### Data Homogenization Method Analyses:

#### Classification Strength:

The matrix of Bray–Curtis Index scores are displayed in Table 4. Group one consisted of the data that were re-sampled to a fixed count ,and Group two contained taxa information collected using the current DEQ methodologies. The mean similarity between the two groups was 64 percent. The mean similarity within group 1 was 89%, and the mean similarity within group 2 was 72%. The Classification Strength derived from equation 2 is 80%. Using the standard set in the methods section of this analysis ( $\geq 77\%$  CS), we can conclude that the two data sets are taxonomically comparable, and the methods used for adjustment to a fixed count of 300 individuals are valid for analysis.

#### Wilcoxon Signed Rank Test:

After re-sampling the ACFRM dataset, a two-tailed Wilcoxon test was conducted to test the null hypothesis: biotic index scores are the same between samples collected via the MTDEQ method and those collected via ACFRM methods and rarified. Table 5 displays the results of the three indices score comparisons for all comparisons and the null hypotheses is not rejected. There is no evidence (RMVFI  $P=.357$  ,MTI  $P=.465$ , HBI  $P=.273$ ) to suggest that there is a difference between scores obtained via the two methods. Thus we can conclude that, once randomly sampled to a fixed count, the ACFRM data and the NRDP data yield comparable index values.

### Change and Trend Assessments

#### Revised Montana Valleys/Foothills Index (Bollman 1998):

Figure 3 shows the scores of the Revised Montana Valleys Foothills index, reported as percent of possible, plotted over time demonstrating the annual variation in sample scores throughout the period of record. The trend assessment conducted using Sen’s slope found no significant trend ( $p=0.561$ ) in

RMVFI scores over the period of record, 1986 to 2009, and no significant trend ( $p=0.67$ ) over the pre-restoration period, 1986 to 2001. There was insufficient post restoration data to perform the analysis.

Throughout the period of record, the scores for RMVFI ranged from 11 % to 42% with a standard deviation of 8%. When stratified by the grouping variable of restoration, scores ranged 11% to 33% and 11% to 42% of possible index score for pre-restoration and post restoration respectively. Mean RMVFI scores are 19 % in the pre-restoration group and 21% post restoration. A complete list of descriptive statistics is found in Tables 6 and 7 of Appendix A. Figure 4, a box plot of the RMVFI scores, demonstrates the spread of the scores in each group and corroborates the lack of a significant difference between the two groups.

The results of the one-tailed Mann-Whitney U test performed on the index scores for RMVFI (testing the null hypothesis: RMFVI scores are not greater in samples taken after restoration in Silver Bow Creek at Rocker) indicate that there is absolutely no evidence (one tailed  $P=.50$ ) to suggest that scores in Silver Bow Creek are higher post restoration (Tables 8 and 9). Thus we accept the null hypothesis and conclude that RMVFI scores are not higher in samples taken after restoration.

#### Metals Tolerance Index (McGuire 2001)

Metals Tolerance Index scores ranged from 3.8 (no metals impact) to 9.1 (severe metals impact) throughout the period of record (Table 10). Once the data were stratified by restoration group, metals tolerance ranged from 5.2 to 9.1 during the pre-restoration period and 3.8 to 4.9 post restorations (Table 11). Figure 6 depicts the values for MTI scores over the period of record and suggests two periods of positive response to metals-- the early 1990's and a more sustained response after reclamation. These values indicate that once restoration/reclamation was implemented, the MTI scores declined dramatically. The removal of contaminated sediments from the floodplain and streambed is associated with the decrease in metals tolerance values.

Sen's slope estimate shows a statistically significant negative trend in metals tolerant organisms over the 23 year period ( $P<.001$ ). Additionally a significant trend ( $P=.003$ ) was detected during a trend test of pre restoration data alone, suggesting the effects of water treatment and upstream activities are

decreasing the metals tolerant community. Although insufficient data exist post restoration to determine a statistically significant trend, observationally data suggest a decline in MTI scores. The negative trend in MTI scores over the period suggest that throughout the period MTI values have been declining, resulting in a less metals tolerant community in Silver Bow creek at Rocker.

Figure 7 demonstrates the variation seen in scores from the two treatment groups suggesting that there is a significant difference between the pre restoration and post restoration Metals Tolerance Index scores. Mann-Whitney test results, Tables 12 and 13, indicate that there is overwhelming evidence ( $P < .001$ ) to reject the null hypothesis: “Metals Tolerance Index values are the same between samples taken pre and post restoration in Silver Bow Creek at Rocker”. Therefore, one can conclude that the MTI scores from samples taken after restoration are significantly less than the MTI scores sampled prior to restoration.

#### Hilsenhoff Biotic Index (Hilsenhoff 1987):

Hilsenhoff Biotic Index scores ranged from 4.8 (some organic pollution) to 8.4 (very significant organic pollution) throughout the period of record, 1986-2009 (Table 14). Stratified by treatment group (Table 15), the HBI values ranged from 4.8 to 7.1 and 7.2 to 8.4, for pre and post restoration groups respectively. Visual inspection of Figure 8 suggests an increase over time of tolerance to organic enrichment as measured by the Hilsenhoff Biotic Index (Hilsenhoff 1987). However, testing the significance of this trend resulted in a statistically insignificant positive trend in HBI scores throughout the period of record ( $P = .057$ ). When stratified by time period (pre and post restoration), data acquired from 1986-2001 also show no significant trend, and data post restoration are insufficient to calculate statistical significance.

Figure 9, captures the variation of HBI scores between the treatment groups of pre and post restoration, indicating a significant increase in HBI scores post restoration. Non-parametric analysis of HBI scores shows that there is sufficient evidence to reject the null hypothesis ( $P < .001$ ), and conclude that the HBI scores are significantly greater post restoration than pre restoration (Tables 16 and 17).



## Community Composition

### Principal Component Analysis:

The number of variables (taxa) was reduced, by eliminating those taxa with less than 13 non-zero values throughout the period of record. The resulting 6 taxa selected via this filter were: *Cardiocladius* sp., *Cricotopus* sp., *Eukiefferiella* sp., *Phaenopsectra* sp., *Simulium* sp. And *Tubificidae*. These taxa represent the 6 most common taxa throughout the period of record. An ordination of these taxa and their abundances can be seen in Figure 10. The ordination of these taxa and their abundances indicates that the presence of *Tubificidae* worms could be the dominant factor allowing for the discrimination of the pre and post restoration groups. The cluster of post restoration points indicates that there has been a taxonomic composition change after restoration, based on the 6 most common taxa in the data set. The shotgun style scatter of the pre-restoration points is primarily driven by the two midge genera *Cricotopus* and *Cardiocladius*. As shown in Figure 10 Axis one explains 67 % of the variation and Axis 2 explains 15 percent of the variation in clusters.

### Indicator Species Analysis:

Using all taxa collected throughout the study period, an Indicator Species analysis was conducted, resulting in the isolation of 16 significant indicator taxa within the dataset. Table 18 shows these taxa and their significance values as well as the values of all other taxa. Three of the 16 significant indicator taxa (*Simulium*, *Cardiocladius*, and *Pagastia*) were identified by the ISA as indicative of pre restoration data. The remaining 13 taxa with significant IV scores were indicative of post restoration conditions. Significant indicator species within the post restoration group included the hemoglobin bearing taxa (*Cryptochironomus* sp., *Chironomus* sp. and *Tubificidae*), and the warm water taxon (*Helobdella stagnalis*) dominated the species list, suggesting low oxygen and high temperature environments (Rossaro 1991 and Klemm 1972).

### Relative Abundance:

The percent composition of each major taxonomic group was calculated based on the taxa lists derived for this analysis. Using the relative importance of the Chironomidae family in the Order Diptera , the order was separated into two groups: Non-Chironomidae Diptera and Chironomidae. The area graph in figure 11 demonstrates the percent composition throughout the study period. Groups with less than 5% composition were composited into the group labeled “other”. Non- Insects dominate the composition of samples after restoration in 2003; the two non-insect taxa that create this dramatic shift are the leech *Helobdella stagnalis* and the Oligochaeta family *Tubificidae*-- both of which are indicative of oxygen poor waters. Functional Feeding Group (FFG) characteristics were calculated, and percent composition of each group was determined for each year in this analysis. Figure 12 shows the percent composition of each FFG through the study period. Similar to the relative abundance area chart shown in figure 11, a noticeable change occurs. An increase in the relative abundance of Collector-Filterers is present after restoration, consistent with an increase in organic enrichment and reduced riparian cover(Barbour 1998).

## DISCUSSION

Many studies have attempted to assess the response of invertebrate communities following an instream disturbance such as restoration (Brooks et al. 2002, Moerke and Lamberti 2004), often following specific study designs aimed at evaluating the effects of restoration (Stewart-Oaten 1986). These specific study designs allow for a comparative analysis between before and after restoration conditions and are often related to a “reference” reach to consider natural variability within the region. This thesis was able to use available data to examine pre and post restoration conditions; however, does not use reference reaches to compare natural variability within the region. Montana has a large network of reference streams within the state, and evaluation of candidate reference reaches displayed little promise for a “natural” comparison for Silver Bow Creek. Changes in community composition reflect the recovery of benthic biota from what will hopefully be considered the ‘Last Best Disturbance’ of Silver Bow Creek.

An exhaustive literature search yielded no studies that focused on macroinvertebrate community’s response to both remediation and restoration. Studies have often focused on specific attributes of a benthic community like diversity and taxa richness (Palmer 1997) as measures of recovery in systems affected by restoration or remediation. Silver Bow Creek restoration plans include the narrative descriptions of diversity and richness as goals of restoration while specifying numeric goals for certain biotic indices, thus presenting a unique learning opportunity in the field of restoration ecology. Recovery of macroinvertebrate communities within disturbed streams depends on many factors including dispersal, colonization and perturbation persistence (Spanhoff 2007). The severity and persistence of a disturbance are major limiting factors for colonization of invertebrates in streams post restoration (Yount and Niemi 1990). Restoration, especially when coupled with soil and sediment removal for remediation, acts as a disturbance on the stream biota. Varying degrees of severity exist within each project, and restoration requiring heavy machinery and channel reconstruction can drastically affect stream invertebrate communities during sensitive stages of their life cycle (Spanhoff 2006).

Current restoration projects often set specific goals to assess the success of the project. Instream projects aimed at environmental cleanup or habitat restoration often include biodiversity increases as key measures of success (Palmer 1997), requiring recolonization of the constructed streambed. Studies indicate that restored reaches are rapidly recolonized after restoration is complete (Niemi et al. 1990, Tikkanen et al. 1994). However, studies suggest after initial recolonization, diversity failed to significantly increase relative to pre-restoration conditions after 5 years (Lepori et al. 2005).

Recolonization of disturbed reaches is facilitated by three major mechanisms: downstream drift, aerial dispersal, and up-stream migration (Bilton 2001). Invertebrate communities depend on the local species pool to recolonize the disturbed reaches, and therefore will be of similar taxonomic structure and will reflect the ecology of adjacent streams. The temporal scale of new species immigration is highly dependent on the dispersal capabilities of the taxon and the connectivity to the restored stream (Niemi et al. 1990). Additionally, the arrival and successful establishment of new species is dependent on species specific life-cycle traits, presence of competitors and environmental condition (Palmer et al. 1997).

Silver Bow Creek at Rocker is one of many locations within the immediate area (SSTOU) that has undergone, or is undergoing restoration. Throughout Silver Bow creek, restoration will be preceded by reclamation and the removal of contaminated soils and substrate. The disturbance persistence within these stream reaches, as seen at Rocker, may last for several years. Channel reconfiguration and dewatering may limit the refugia potential for colonizers thus increasing the amount of time necessary for recolonization from the local species pool. The mechanism which most affects community composition immediately after restoration is downstream drift (Tikkanen et al. 1994). Regional species pools contributing to recolonization from down-stream drift are well adapted to high temperatures and the low oxygen conditions within those reaches. Current community composition in Silver Bow Creek reflects the environmental conditions upstream. Furthermore, due to the continued restoration and remedial activities downstream of the site, other key mechanisms of recolonization may be inhibiting community composition changes.

Wallace (1990) suggests that recovery time from chronic impacts such as mining related metals contamination is indeterminate and relies heavily on the duration and extent of the impact. Previous long term monitoring efforts on a metals-contaminated river demonstrate that, 10 years after passive reclamation, communities failed to “recover” from metal pollution but exhibited long term changes in relative abundance and community composition ( Zanella 1982).

The term “recovery” implies a return to a previous state (Sheldon 1984). In the case of Silver Bow Creek, recovery will be determined as a divergence from the previous pre- restoration state and a return to a pre-contamination state as typified by regional reference streams. As mentioned in previous sections, Silver Bow Creek was devoid of aquatic insects until the mid 1970s. Since then water treatment upgrades, reclamation and restoration have influenced the composition of the invertebrate community. As time moves on and restoration efforts are completed downstream, mechanisms allowing for “recovery” may be enhanced, resulting in a functional and robust community similar to communities within the region unaffected by the mining operations of the last century.

Studies assessing the response of benthic invertebrates to instream disturbances such as restoration and reclamation often employ a reference condition approach (Ross et al. 2008, Hoiland et al. 1994, Spanhoff 2006,2007, Wallace 1990), claiming that “recovery” is achieved when communities are more similar to their undisturbed counterparts (Lepori 2005). Biological monitoring in Silver Bow Creek could benefit from the identification and use of a reference condition to measure community similarity and variance in comparison to undisturbed systems within the region. Montana Department of Environmental Quality has been monitoring candidate reference reaches throughout the state since 1992 and has identified sites that function as reference streams through a strict rubric for scoring ( Suplee et al. 2005). Rock Creek, near Clinton Montana, has been identified and frequently monitored as part of the reference stream project, and could prove to be a good candidate to compare with Silver Bow Creek. Rock Creek varies highly in drainage area, slope, precipitation and land use as compared to Silver Bow Creek; however, invertebrate communities found in Rock Creek could reflect the natural variation and undisturbed conditions of area streams for restoration comparisons.

## CONCLUSION AND RECOMMENDATIONS

Statistically significant changes have occurred in the macroinvertebrate community following restoration efforts on Silver Bow Creek at Rocker, Montana, as measured by perturbation specific indices and community composition analysis. These observed changes demonstrate that invertebrate communities have responded to the remediation of metals laden sediment and subsequent restoration in the stream channel. Compositional changes in the invertebrate community suggest a decrease in the abundance of metals tolerant taxa, with dramatic declines in Metals Tolerance Index scores, coinciding with the removal of contaminated material from the floodplain. Conversely, organic enrichment in Silver Bow Creek at Rocker appears to have significantly increased since reclamation as measured by the Hilsenhoff Biotic Index. However, this statistically significant observation may be a product of “unmasking” existing organic enrichment by removing toxic metal conditions.

Taxonomic composition analysis of Silver Bow Creek depicts change throughout the period of study. Dominant taxa shifted from metals-tolerant taxa to high temperature and low oxygen tolerant taxa, suggesting a potential shift in the limiting factors of benthic integrity of Silver Bow Creek.

Although some indices selected for this analysis show significant change, the Revised Montana Valleys/Foothills Index shows no significant change in pre and post restoration Silver Bow Creek at Rocker. No significant change in RMVFI suggests no change in broad scale biotic integrity. Three of the 6 metrics used in the RMVFI are dependent on the richness of Ephemeroptera, Plecoptera and Trichoptera. The absence of any taxa from these sensitive orders contributes to the insignificant changes in index scores over time. Additionally, the decrease in MTI and increase in HBI may assist in explaining the insignificant changes in RMVFI scores.

Community composition analyses demonstrate change in the community after restoration that seems in general agreement with the index data; both suggest a more organic enrichment tolerant community. Significant changes in selected index scores and measured change in community composition validate that a change post restoration has occurred in the benthic community. Principal

Component Analysis suggests that, of the 6 most common taxa sampled, three taxa explain most of the variation between pre and post restoration community composition. Indicator Species Analysis confirmed the presence of indicator species that delineate the pre and post restoration communities. It is no surprise that the taxa identified by the ISA were indicative of the stressors identified by both the Metals Tolerance Index and the Hilsenhoff Biotic Index. Relative abundance of major taxonomic groups and Functional Feeding Groups also indicated a strong change in composition following the restoration of Silver Bow creek, both indicating an increased response to organic enrichment (typical of municipal waste water).

Although these data suggest an increase in organic enrichment, it is quite reasonable to conclude that the effect of metals laden sediment on the invertebrate community prior to restoration overpowered the effect of organic enrichment in Silver Bow Creek at Rocker. Trend analyses, throughout the period of record, indicate that no significant trends exist for the Revised Montana Valleys Foothills Index and the Hilsenhoff Biotic Index. However, significant negative trends were detected for the Metals Tolerance Index throughout the study period and within the period prior to restoration. These trends suggest factors in addition to the local removal of metals laden sediment have contributed to the decrease in metals tolerant organisms in Silver Bow Creek at Rocker. Earlier upstream cleanup efforts may also have benefited the creek.

The study site was selected partly for its distance from the WWTP Silver Bow Creek at Rocker; however, it is still fairly close to the Butte WWTP and is heavily influenced by the effluent. Other sites currently receiving the prescribed restoration may respond differently than the Rocker site given their geographic distance from waste water discharge and diluting flows contributed by adjacent tributaries. Factors such as recruitment and continued reach scale disturbances may be contributing to the insignificant changes of the RMFVI scores. The Rocker site receives all colonization resulting from drift from nutrient rich up-stream reaches, which may explain the significant increase in HBI scores post restoration. Currently, up-stream immigration from downstream communities may be precluded by the

restoration practices and breaks in connectivity resulting from the reclamation and restoration practices downstream.

It is my recommendation, based on the results presented in this analysis, that efforts similar to those illustrated in this thesis be used to evaluate long term trends and assess community composition changes within restored sites of the SSTOU. Such an effort may assist us to better understand the temporal effects of reclamation and restoration in the Upper Clark Fork River Basin on benthic invertebrate communities. A data homogenization tool, like the methods used in this thesis, should be used throughout the basin where methodologies differ throughout the period of record. Given the propensity for sampling methodologies to change throughout time, a homogenization method will ensure that a measureable degree of comparability is present when evaluating long term trends. Additionally, measures of organic enrichment, like the HBI, should be reported in future monitoring efforts to better quantify the spatial extent to which organic enrichment may be limiting benthic biotic recovery.

Nearly 7 years after restoration, specific goals for the benthic community have not yet been met, and data show no evidence of an upward trend in RMVFI values. Downstream restoration and restored connectivity may ultimately enhance the biotic integrity of Silver Bow Creek. Continued monitoring throughout the SSTOU will assist in determining that target goals are met within Silver Bow Creek. Continued efforts to evaluate the benthic community both up-stream and down should provide sufficient data to determine additional changes in community composition. The institution of a fixed monitoring station at a location within a similar and less disturbed watershed (i.e. upper reaches of Warm Springs Creek, or Blacktail Creek above Butte) would provide a realistic reference for community recovery. The sampling regime could benefit from an additional change. Increasing the replicates per site would allow agencies and restoration managers to capture the intra-annual variation of biotic communities at each site. Although cost typically prohibits the increase of replicates at each site, a strategic reduction in the number of sites accompanied by an increase in replicate samples will increase the statistical power of annual sampling data with no net cost to the monitoring budget.



The RMFVI, selected as a restoration goal measure, shows no significant trend at Rocker. The broad sensitivity to environmental perturbation and dependence on sensitive taxa of this index could prove to be a robust measure of recovery once it occurs. The extensive restoration efforts, to date, have resulted in decreased MTI values. Given time for recolonization and with continued monitoring, evaluation, and adaptive management, it is reasonable to believe that Silver Bow Creek will one day support a noticeably more diverse, intact and functional benthic community.

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**APPENDIX A: TABLES and FIGURES**

Figure 1: Stream Side Tailings Operable Unit Sampling sites  
 Source: 2008 Annual Monitoring Report , Confluence Consulting

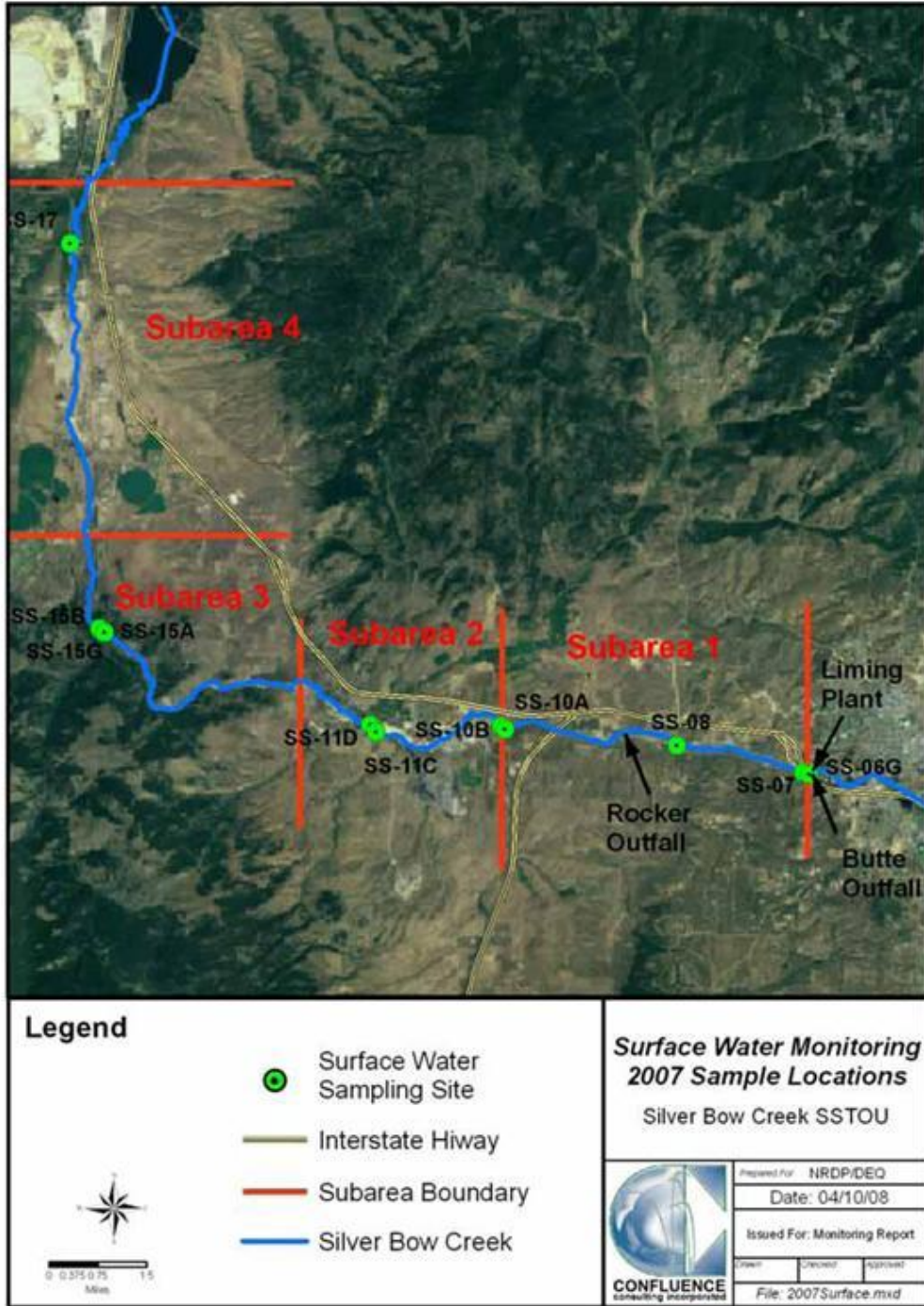
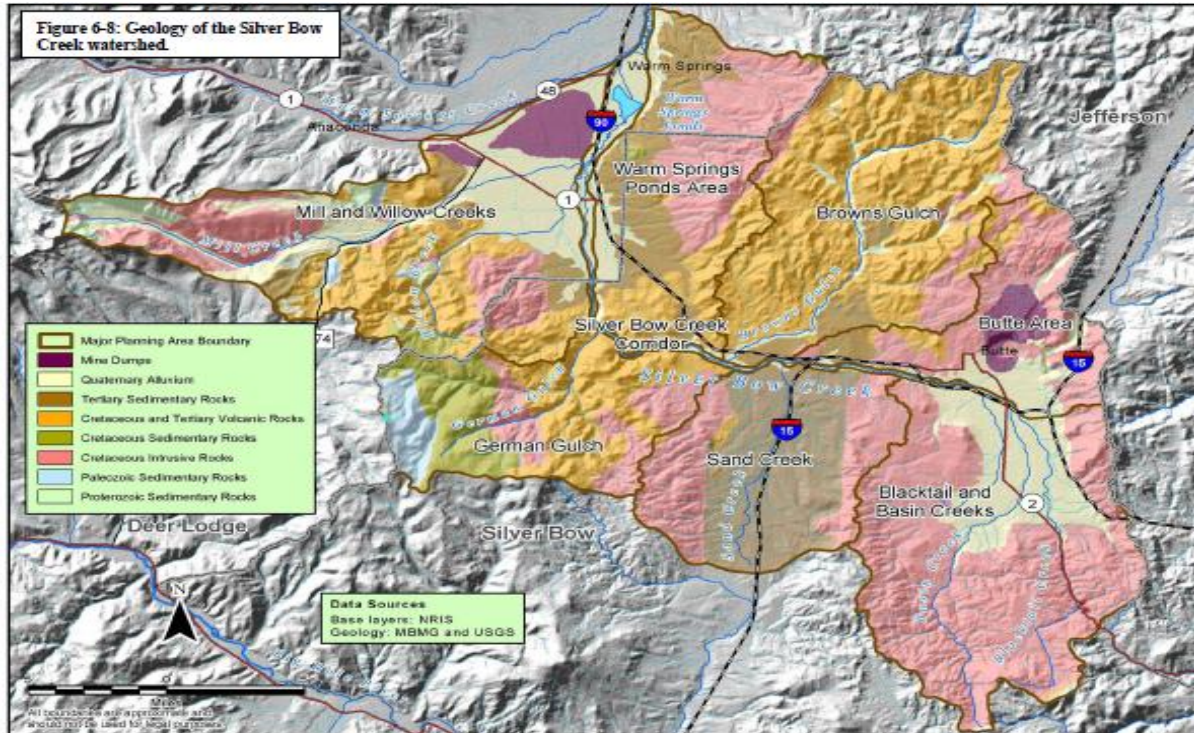


Figure 2: Geologic map of Silver Bow Creek watershed (source NRDP Final Restoration Plan, Figure 6-8, 2005)



Equation 1: Sorting Efficiency

$$\text{Sorting Efficiency} = \frac{(n_o * \%QA Vol)}{\text{Total } n}$$

Where:

$n_o$  is equal to the number of organisms found in the QA

%QAVol is equal to the percentage of substrate checked by the additional technician

Total n is equal to the total number of organisms found by the original technician

Equation 2: Bray Curtis Similarity

$$BC_{ij} = \sum \frac{|n_{ik} - n_{jk}|}{(n_{ik} + n_{jk})}$$

Equation 3: Classification Strength as defined by Cao et al. 2005

$$\text{Classification Strength} = \frac{2(\bar{S}_B)}{(\bar{S}_{W_1} + \bar{S}_{W_2})}$$

Where:

$\bar{S}_B$  is equal to the Mean Similarity Between Groups

$\bar{S}_{W_1}$  is equal to the Mean Similarity within Group 1

$\bar{S}_{W_2}$  is equal to the Mean Similarity within Group 2

Equation 4: Generalized equation for calculation of an Index using Tolerance Values

$$Index\ Score = \sum (\%RA_i * T_i)$$

Where:

%RA<sub>i</sub> is equal to the percent relative abundance of species <sub>i</sub>

T<sub>i</sub> is equal to the tolerance value of species <sub>i</sub>

Table 1: Metric composition of candidate multimetric indices.

Montana Revised Foothills/Valleys ( Bollman 1998)	Montana Intermontane valleys and Foothills (Bukantis 1998)
Ephemeroptera Richness	Taxa Richness
Plecoptera Richness	EPT Richness
Trichoptera Richness	Hilsenhoff Biotic Index
Sensitive Taxa Richness	% Dominant
Percent Filterer	% collectors
Percent Tolerant	% Scrapers + Shredders
	% Hydropsychinae of Trichoptera
	% EPT

Table 2: Metals Tolerance Index Score Interpretation

Metals Tolerance Index Score Evaluation		
Score	Water Quality	Degree of Organic Enrichment
<4.0	Excellent	No Impairment
4.1-8.9	Fair	Impairment
>8.9	Poor	Severe Impairment

Table 3: Hilsenhoff Biotic Index Score Interpretation

Hilsenhoff Biotic Index Score Evaluation		
Score	Water Quality	Degree of Organic Enrichment
0.00-3.50	Excellent	No Apparent Organic Pollution
3.51-4.50	Very Good	Slight Organic Pollution
4.51-5.50	Good	Some Organic Pollution
5.51-6.50	Fair	Fairly Significant Organic Pollution
6.51-7.50	Fairly Poor	Significant Organic Pollution
7.51-8.50	Poor	Very Significant Organic Pollution
8.51-10.00	Very Poor	Severe Organic Pollution



Table 4: Matrix of Similarity Scores and Classification Strength Results

Bray - Curtis Similarity Index Matrix Silver Bow Creek at Opportunity (2004,2006,2007,2008)								
Sample	2008 Mod	2006 Mod	2004 Mod	2007 Mod	2008 NRDP	2006 NRDP	2004 NRDP	2007 NRDP
SBC at Opportunity 2008 Modified	1.00							
SBC at Opportunity 2006 Modified	0.91	1.00						
SBC at Opportunity 2004 Modified	0.72	0.75	1.00					
SBC at Opportunity 2007 Modified	0.84	0.89	0.83	1.00				
SBC at Opportunity 2008 NRDP	0.40	0.38	0.47	0.43	1.00			
SBC at Opportunity 2006 NRDP	0.60	0.63	0.76	0.69	0.62	1.00		
SBC at Opportunity 2004 NRDP	0.59	0.63	0.84	0.72	0.55	0.80	1.00	
SBC at Opportunity 2007 NRDP	0.84	0.85	0.67	0.79	0.46	0.58	0.56	1.00
Shaded Cells represent the Similarity Measures Between Groups								
Classification Strength	Mean Similarity Between ( $S_B$ )			Mean Similarity Within ( $S_{W1}$ )		Mean Similarity Within ( $S_{W2}$ )		
79.98%	64.42%			89.44%		71.65%		

Table 5: SPSS output for Wilcoxon Signed Rank Test (Comparability analysis)

**Test Statistics<sup>b</sup>**

	Montana Revised Valleys/foothills_s_M-Montana Revised Valleys/foothills_s	Metals Tolerance index (McGuire)_M-Metals Tolerance index (McGuire)	Hilsenhoff Biotic Index-M - Hilsenhoff Biotic Index
Z	-.921 <sup>a</sup>	-.730 <sup>a</sup>	-1.095 <sup>a</sup>
Asymp. Sig. (2-tailed)	.357	.465	.273

a. Based on positive ranks.

b. Wilcoxon Signed Ranks Test

Table 6: RMVFI Descriptive statistics of Silver Bow Creek at Rocker between the years 1986 and 2009

**Descriptive Statistics**

	N	Minimum	Maximum	Mean	Std. Deviation
Montana Revised Valleys/foothills	23	11.11	41.67	19.1552	8.38861
Valid N (listwise)	23				

Table 7: RMVFI Descriptive Statistics Stratified by group (Pre-restoration/ Post Restoration)

		Descriptives		Statistic	Std. Error
Montana Revised Valleys/foothills	Restoration				
	Pre-Restoration	Mean		18.6673	1.83108
		95% Confidence Interval for Mean	Lower Bound	14.7401	
			Upper Bound	22.5946	
		5% Trimmed Mean		18.2726	
		Median		16.6700	
		Variance		50.293	
		Std. Deviation		7.09173	
		Minimum		11.11	
		Maximum		33.33	
		Range		22.22	
		Interquartile Range		11.11	
		Skewness		.850	.580
		Kurtosis		-.160	1.121
		Post Restoration	Mean		20.5557
	95% Confidence Interval for Mean		Lower Bound	9.7403	
			Upper Bound	31.3711	
	5% Trimmed Mean			19.9075	
	Median			16.6700	
	Variance			136.757	
Std. Deviation			11.69431		
Minimum			11.11		
Maximum			41.67		
Range			30.56		
Interquartile Range		18.89			
Skewness		1.119	.794		
Kurtosis		.360	1.587		

Figure 3: Silver Bow Creek at Rocker RMVFI scores (Percent of possible) over time

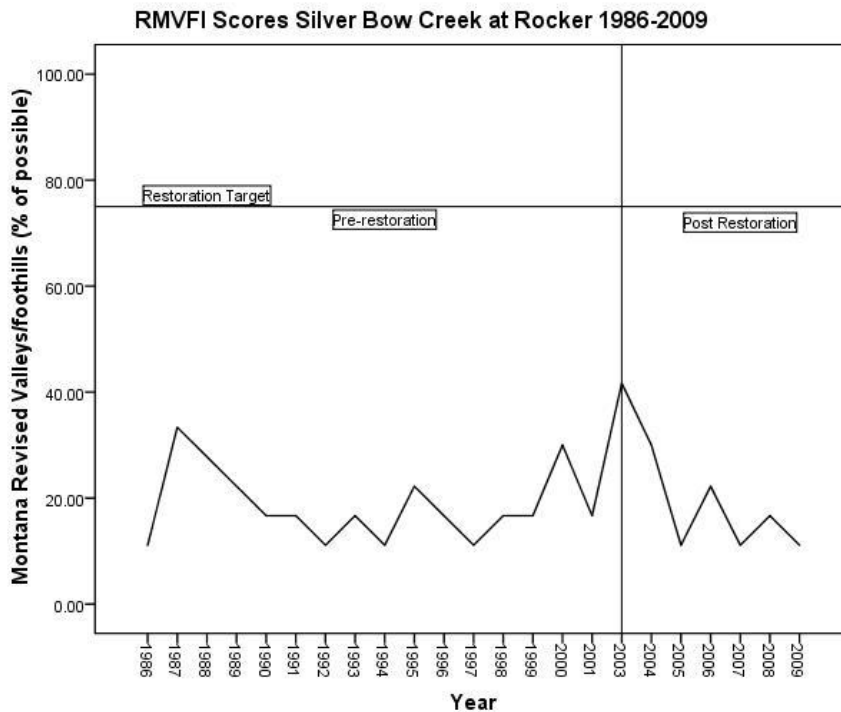


Figure 4: Box Plot comparing the spread of RMVFI scores in each treatment Group

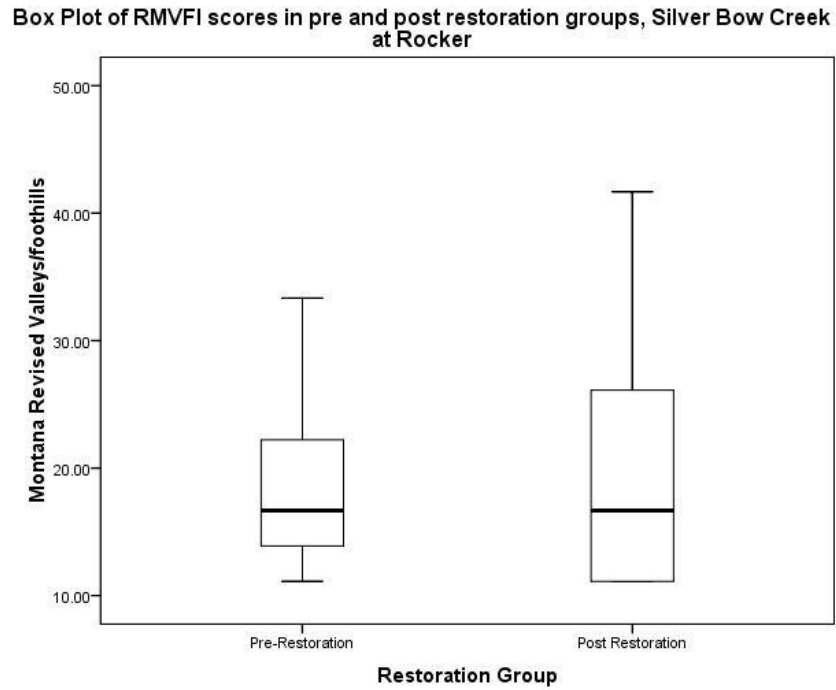


Table 8: SPSS output Mann-Whitney Test on RMVFI scores

Ranks				
	Restoration	N	Mean Rank	Sum of Ranks
Montana Revised Valleys/foothills	Pre-Restoration	15	11.50	172.50
	Post Restoration	7	11.50	80.50
	Total	22		

Table 9: SPSS output Mann-Whitney Test on RMVFI Scores (test statistics)

Test Statistics <sup>b</sup>	
	Montana Revised Valleys/foothills
Mann-Whitney U	52.500
Wilcoxon W	80.500
Z	.000
Asymp. Sig. (2-tailed)	1.000
Exact Sig. [2*(1-tailed Sig.)]	1.000 <sup>a</sup>

a. Not corrected for ties.

b. Grouping Variable: Restoration

\*\*\*one tailed significance .500

Table 10: Descriptive statistics for Metals Tolerance Index (McGuire 2001) 1986-2009.

**Descriptive Statistics**

	N	Minimum	Maximum	Mean	Std. Deviation
Metals Tolerance Index (McGuire 2001)	23	3.76	9.10	6.4652	1.91579
Valid N (listwise)	23				

Table 11: Descriptive statistics stratified by group Metals Tolerance Index (McGuire 2001)

**Descriptives**

Restoration			Statistic	Std. Error		
Metals Tolerance Index (McGuire 2001)	Pre-Restoration	Mean	7.5400	.33637		
		95% Confidence Interval for Mean	Lower Bound	6.8185		
			Upper Bound	8.2615		
		5% Trimmed Mean	7.5806			
		Median	7.6400			
		Variance	1.697			
		Std. Deviation	1.30277			
		Minimum	5.25			
		Maximum	9.10			
		Range	3.85			
		Interquartile Range	2.33			
		Skewness	-.358	.580		
		Kurtosis	-1.378	1.121		
		Post Restoration	Post Restoration	Mean	4.1143	.14283
				95% Confidence Interval for Mean	Lower Bound	3.7648
Upper Bound	4.4638					
5% Trimmed Mean	4.0903					
Median	4.0700					
Variance	.143					
Std. Deviation	.37788					
Minimum	3.76					
Maximum	4.90					
Range	1.14					
Interquartile Range	.33					
Skewness	1.773			.794		
Kurtosis	3.887			1.587		

Figure 6: Metals Tolerance Index Scores throughout the period of record 1986-2009

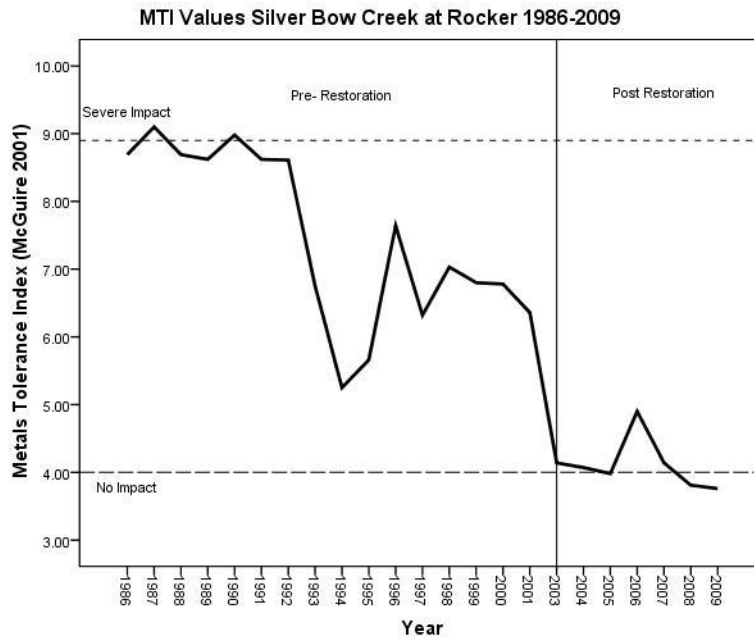


Figure 7: Box Plot of MTI scores by treatment group Silver Bow Creek at Rocker

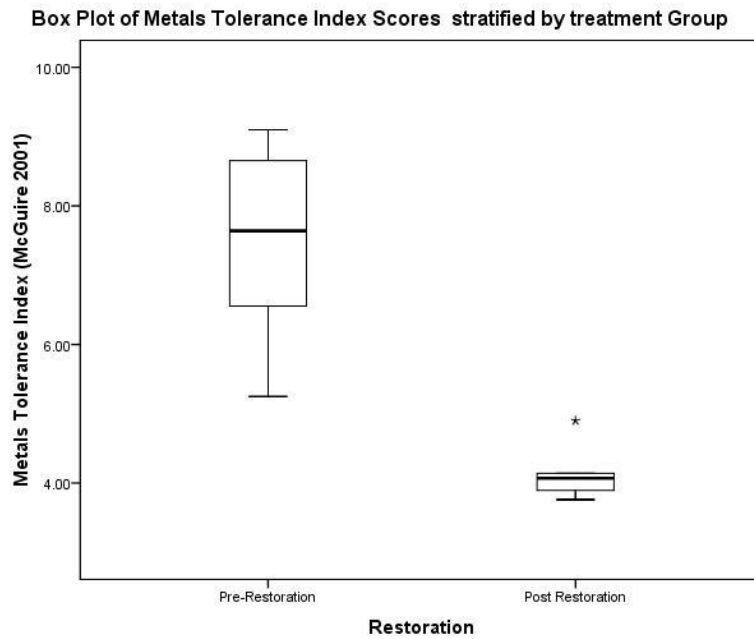


Table 12: SPSS output for Mann-Whitney U Metals tolerance Index

Ranks				
	Restoration	N	Mean Rank	Sum of Ranks
Metals Tolerance Index (McGuire 2001)	Pre-Restoration	15	15.00	225.00
	Post Restoration	7	4.00	28.00
	Total	22		

Table 13: SPSS output Mann-Whitney U test statistics Metals Tolerance Index

Test Statistics <sup>b</sup>	
	Metals Tolerance Index (McGuire 2001)
Mann-Whitney U	.000
Wilcoxon W	28.000
Z	-3.704
Asymp. Sig. (2-tailed)	.000
Exact Sig. [2*(1-tailed Sig.)]	.000 <sup>a</sup>

a. Not corrected for ties.

b. Grouping Variable: Restoration

\*\*\*\* one tailed significance  $p < .001$

Table 14: Hilsenhoff Biotic Index Descriptive statistics

Descriptive Statistics					
	N	Minimum	Maximum	Mean	Std. Deviation
Hilsenhoff Biotic Index	23	4.84	8.42	6.7565	.81886
Valid N (listwise)	23				

Table 15: Hilsenhoff Biotic Index Descriptive statistics stratified by treatment group

				Descriptives	
Restoration				Statistic	Std. Error
Hilsenhoff Biotic Index	Pre-Restoration	Mean		6.3647	.14490
		95% Confidence Interval for Mean	Lower Bound	6.0539	
			Upper Bound	6.6754	
		5% Trimmed Mean		6.4085	
		Median		6.5000	
		Variance		.315	
		Std. Deviation		.56120	
		Minimum		4.84	
		Maximum		7.10	
		Range		2.26	
		Interquartile Range		.54	
		Skewness		-1.365	.580
		Kurtosis		3.126	1.121
	Post Restoration	Mean		7.6900	.18951
		95% Confidence Interval for Mean	Lower Bound	7.2263	
			Upper Bound	8.1537	
		5% Trimmed Mean		7.6761	
		Median		7.4500	
		Variance		.251	
Std. Deviation			.50140		
Minimum			7.21		
Maximum			8.42		
Range			1.21		
Interquartile Range		1.03			
Skewness		.894	.794		
Kurtosis		-1.187	1.587		

Figure 8: Hilsenhoff Biotic Index scores Silver Bow Creek at Rocker 1986-2009

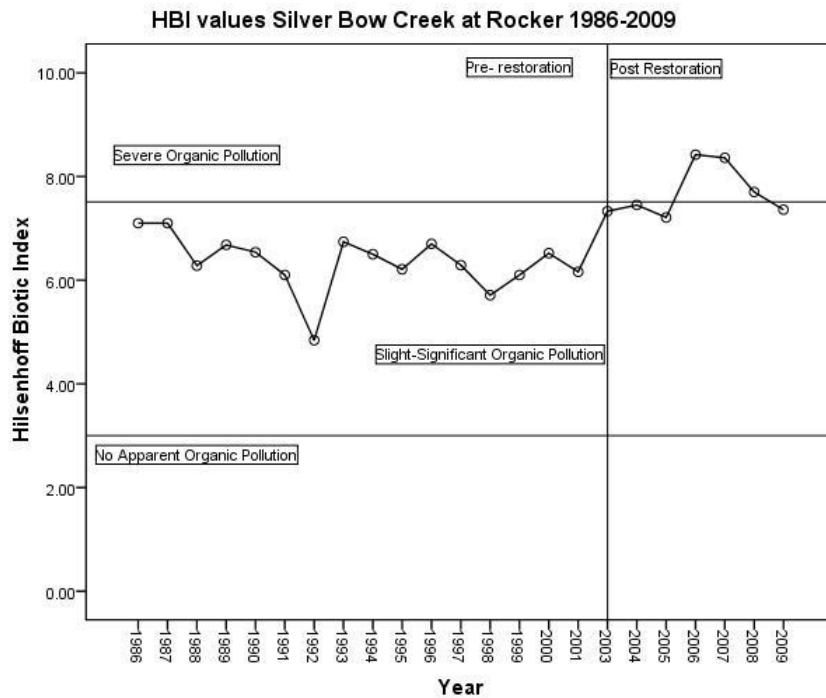


Figure 9: Box Plots of HBI values pre and post restoration

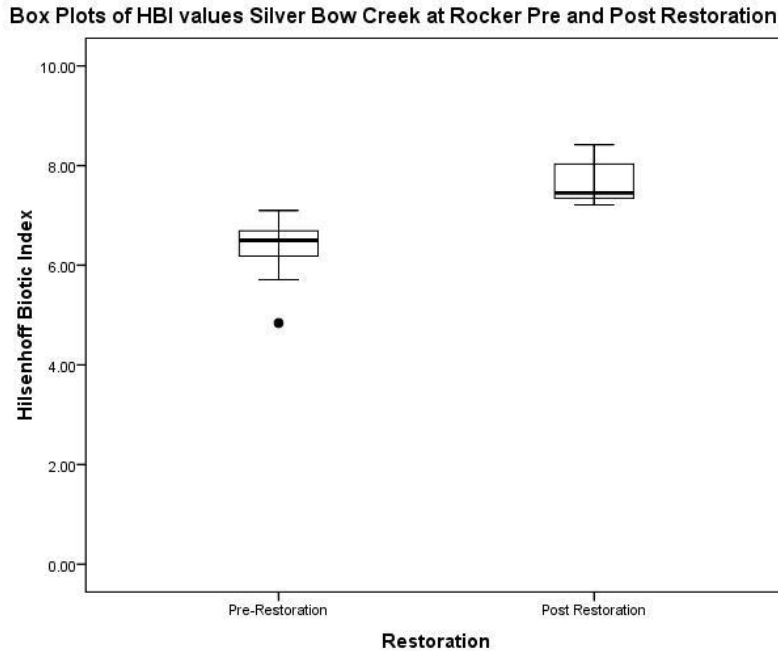


Table 16: SPSS output Mann-Whitney Ranks HBI scores

Ranks				
	Restoration	N	Mean Rank	Sum of Ranks
Hilsenhoff Biotic Index	Pre-Restoration	15	8.00	120.00
	Post Restoration	7	19.00	133.00
	Total	22		

Table 17: SPSS output Mann-Whitney U test , test statistics and p-values

Test Statistics <sup>b</sup>	
	Hilsenhoff Biotic Index
Mann-Whitney U	.000
Wilcoxon W	120.000
Z	-3.702
Asymp. Sig. (2-tailed)	.000
Exact Sig. [2*(1-tailed Sig.)]	.000 <sup>a</sup>

a. Not corrected for ties.

b. Grouping Variable: Restoration

\*\*\* one-tailed significance P<.001



Figure 10: Ordination of the 6 most common taxa in species space

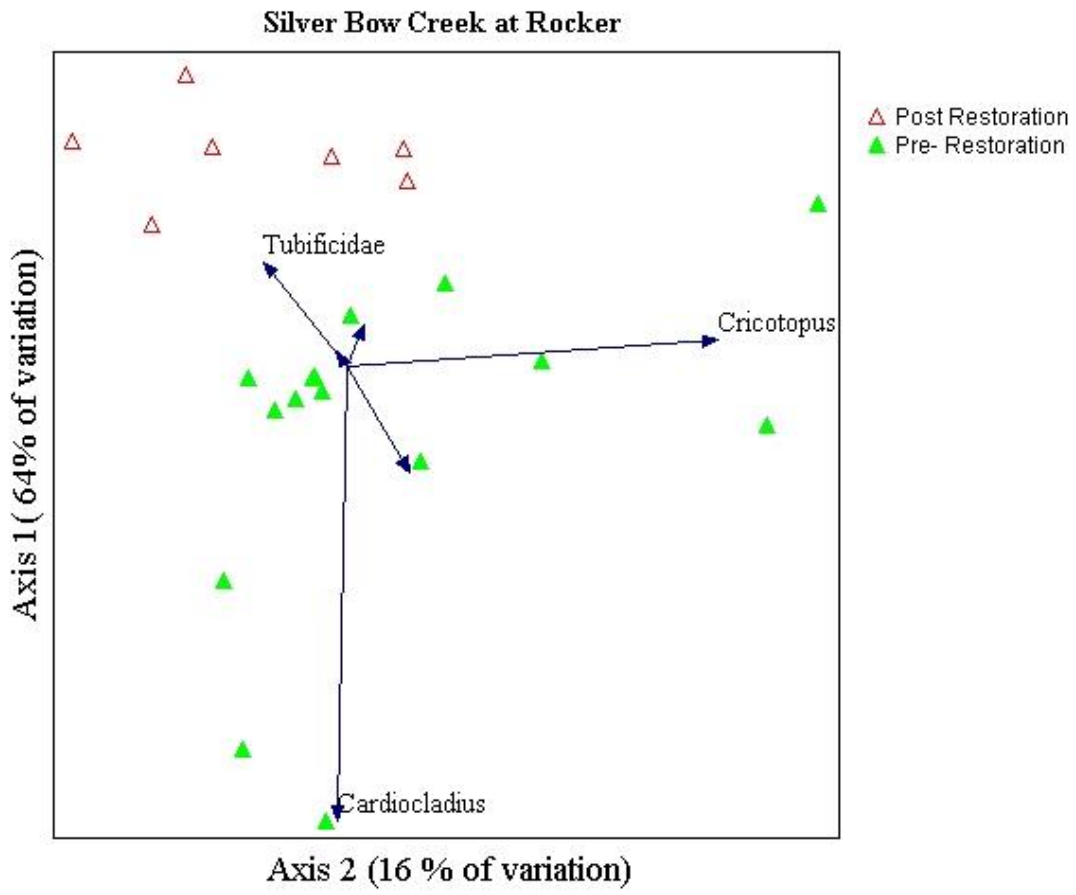


Table 18: Significance of Indicator Values based on 1000 Monte Carlo randomizations Gray shaded taxa are significant to an alpha of .05

Taxon	Group	IV value(%)	Mean	Std.Dev	P-value
Cardiocladius	Pre-Restoration	93.5	47.9	11.09	0.001
Microtendipes	Post Restoration	85.7	24	9.02	0.001
Parametriocnemus	Post Restoration	67.2	26.6	9.58	0.002
Stagicola	Post Restoration	71.4	22	8.89	0.002
Cryptochironomus	Post Restoration	57.1	18.1	7.87	0.003
Chironomus	Post Restoration	73.9	39	10.68	0.011
Acari	Post Restoration	42.9	15.7	6.75	0.017
Physa	Post Restoration	42.9	15.5	6.95	0.018
Hydra	Post Restoration	42.9	14.8	7.48	0.020
Chironomidae	Post Restoration	42.9	16.4	6.94	0.021
Silmulim	Pre-Restoration	74.1	58.1	6.3	0.023
Helobdella stagnalis	Post Restoration	42.9	15.4	7.68	0.023
Rheocricotopus	Post Restoration	42.9	15.1	7.79	0.025
Dicrotendipes	Post Restoration	42	19	8.66	0.028
Tubificidae	Post Restoration	80.1	56.9	10.57	0.032
Pagastia	Pre-Restoration	56.2	32.9	10.68	0.049
Copepoda	Post Restoration	28.6	12.2	5.21	0.071
Chaetocladius	Post Restoration	28.6	12.6	5.46	0.077
Clodocera	Post Restoration	28.6	12.2	5.61	0.077
Erpobdellidae	Post Restoration	28.6	12.4	6	0.092
Nematoda	Post Restoration	28.6	12.8	5.51	0.094
Naididae	Post Restoration	28.6	12.7	5.49	0.096
Orthocladius	Post Restoration	48	33.9	10.37	0.106
Phaenopsectra	Post Restoration	56.8	41.4	11.17	0.115
Muscidae	Pre-Restoration	43.7	27.4	10.02	0.118
Limnophora	Pre-Restoration	37.5	24.6	9.47	0.121
Tipula	Post Restoration	24.9	14.9	7.99	0.151
Haliphus	Post Restoration	22.1	15	7.33	0.196
Tvetenia	Post Restoration	14.3	8.6	3.65	0.290
Glyptendipes	Post Restoration	14.3	8.6	3.68	0.296
Ostracoda	Post Restoration	14.3	8.6	3.68	0.296

Table 18 Continued:

Taxon	Group	IV value(%)	Mean	Std.Dev	P-value
Tanytarsus	Post Restoration	14.3	8.6	3.68	0.296
Psectrocladius	Post Restoration	14.3	8.6	3.68	0.297
Gyraulus	Post Restoration	14.3	8.7	3.7	0.303
Coenagrionidae	Post Restoration	14.3	8.7	3.71	0.306
Hesperoperla pacifica	Post Restoration	14.3	8.7	3.71	0.306
Notonecta	Post Restoration	14.3	8.7	3.71	0.306
Sigara	Post Restoration	14.3	8.7	3.71	0.306
Sphaeriidae	Post Restoration	14.3	8.7	3.71	0.306
Tribelos	Post Restoration	14.3	8.7	3.71	0.306
Limnephilidae	Post Restoration	14.3	8.8	3.75	0.317
Parametricnemus	Post Restoration	14.3	8.8	3.75	0.317
Thienemannimyia grp.	Pre-Restoration	33.6	32	10.16	0.319
Apedilum	Post Restoration	14.3	8.8	3.76	0.322
Thienemanniella	Post Restoration	14.3	8.8	3.76	0.322
Cricocotopus	Pre-Restoration	58.7	57.6	5.57	0.367
Eukiefferella	Post Restoration	52.6	50.9	11.61	0.408
Chuemapsyche	Pre-Restoration	18.7	14.8	7.49	0.525
Optioservus	Pre-Restoration	21.9	22.1	8.91	0.589
Brychius	Pre-Restoration	12.5	12	5.88	0.769
Endochrionomus	Pre-Restoration	12.5	12.4	5.85	0.780
Brundiniella	Post Restoration	11.1	12.7	4.97	0.801
Aedes	Pre-Restoration	6.2	8.6	3.65	1.000
Agabus	Pre-Restoration	9.5	18.7	7.98	1.000
Baetis tricaudatus	Pre-Restoration	6.2	8.6	3.69	1.000
Ceratopogoninae	Pre-Restoration	6.2	8.7	3.71	1.000
Hydropsyche	Pre-Restoration	8.6	16	7.1	1.000
Macropelopia	Pre-Restoration	6.2	8.9	3.8	1.000
Oreodytes	Pre-Restoration	6.2	8.7	3.71	1.000
Potthastia	Pre-Restoration	6.2	8.7	3.71	1.000
Procladius	Pre-Restoration	6.2	8.7	3.72	1.000
Tricorythodes	Pre-Restoration	6.2	8.8	3.74	1.000
Radotanypus	Post Restoration	9.9	12.5	4.72	1.000
Tanypodinae	Post Restoration	9.9	12.7	4.9	1.000

Figure 11: Relative Abundance of Major groups 1986-2009 ( Values less than 5% collapsed)

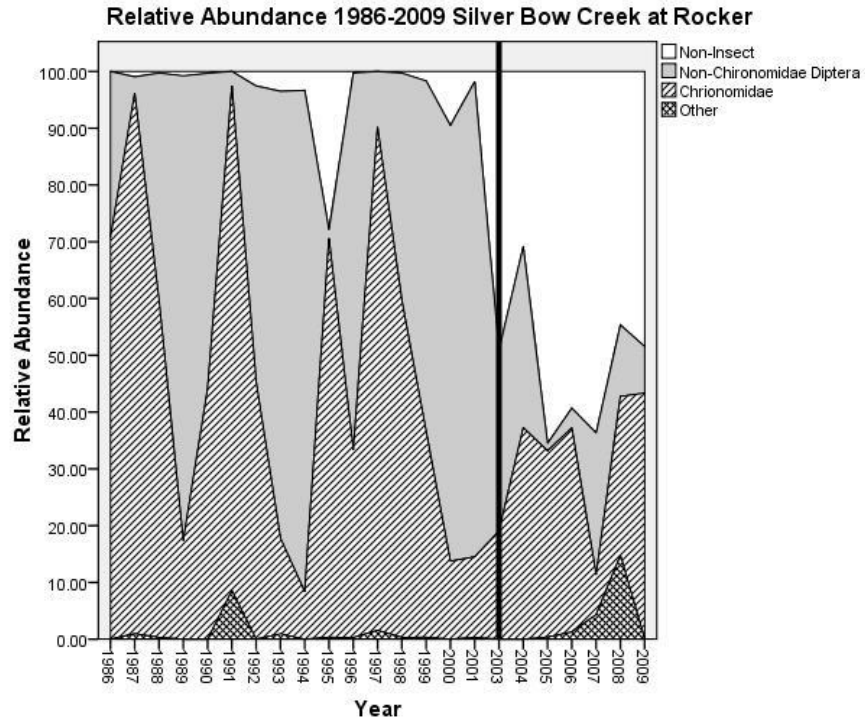


Figure 12: Relative Abundance of Functional Feeding Groups Silver Bow Creek 1986-2009

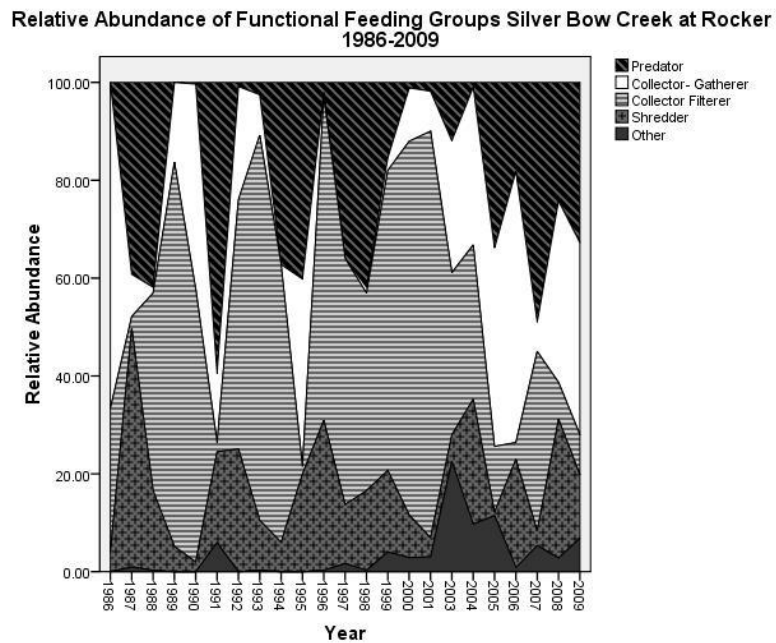
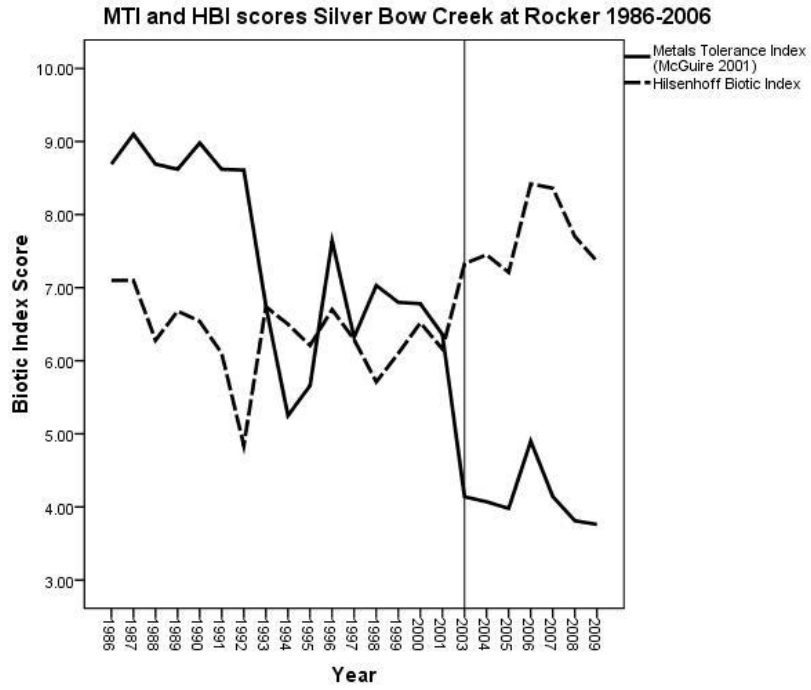


Figure 13: Metals Tolerance and Hilsenhoff Biotic Index Scores Silver Bow Creek at Rocker 1986-2009



APPENDIX B:  
RAW TAXONOMIC DATA SILVER BOW CREEK AT ROCKER 1986-2009

Taxon	1986	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999
Acari	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Aedes	0	0	0	0	0	0	2	0	0	0	0	0	0	0
Agabus	0	0	0	0	0	5	0	0	0	0	1	0	0	1
Apedillum	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Baetis tricaudatus	0	0	0	0	0	0	0	0	0	1	0	0	0	0
Brundinella	0	0	0	0	0	0	0	0	2	0	0	0	0	0
Brychius	0	0	0	0	0	5	0	0	0	0	0	0	0	0
Cardiocladius	2	81	137	55	8	149	1	5	1	127	5	71	137	1
Ceratopogoninae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Chaetocladius	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Chironomidae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Chiromus	0	0	0	0	0	0	0	1	1	1	0	0	0	1
Chuematopsyche	0	0	1	0	0	0	0	1	0	0	0	0	1	0
Clodocera	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Coenagrionidae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Copepoda	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Cricocotopus	208	102	54	48	34	50	78	32	21	63	102	204	54	50
Cryptochironomus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Dicrotendipes	0	0	0	0	0	0	1	0	0	0	0	0	0	0
Endochironomus	0	0	0	0	0	0	10	1	0	0	0	0	0	0
Eukiefferella	2	16	0	3	0	0	64	7	0	3	1	1	0	7
Glypotendipes	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Gyraulus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Halipus	0	2	0	0	0	0	0	0	0	0	0	0	0	0
Helobdella stagnalis	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Hesperoperla pacifica	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Hydra	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Hydropsyche	0	0	0	0	0	4	1	0	0	0	0	0	0	0
Limnephilidae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Limnophila	0	1	0	0	0	6	2	1	0	0	1	0	0	0
Microcylleopus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Macropelopia	0	0	0	0	0	0	0	0	0	0	0	0	0	11
Erpobdella	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Muscidae	0	1	0	0	0	6	2	1	0	0	1	0	0	1
Naididae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Nematoda	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Notonecta	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Optioservus	0	0	0	1	0	11	0	1	0	0	0	3	0	0
Oreodytes	0	0	0	0	0	0	0	1	0	0	0	0	0	0
Orthocladius	0	0	0	0	0	0	0	3	0	13	0	0	0	4
Ostracoda	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Pagastia	0	0	3	0	0	37	5	4	0	15	1	4	3	0
Parametriocnemus	0	0	0	0	0	0	0	0	0	0	0	0	0	1
Paratendipes	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Phaenopsectra	0	0	1	0	0	0	0	0	2	0	1	0	1	19
Physa	0	0	0	0	0	0	0	0	0	0	0	0	0	0

Taxon	1986	1987	1988	1989	1990	1991	1992	1993	1994	1995	1996	1997	1998	1999
Potthastia	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Procladius	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Psectrocladius	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Radotanypus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Rheocricotopus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Sigara	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Silmulim	88	5	133	7	208	1	179	247	265	5	220	31	133	184
Sphaeriidae	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Stagicola	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tanypodinae	0	0	0	0	0	0	0	0	1	0	0	0	0	0
Tanytarsus	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Thienemanniella	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Thienemannimyia grp.	1	0	2	0	0	3	0	1	0	0	0	14	2	0
Tipula	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tribelos	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Tricorythodes	0	0	0	0	0	1	0	0	0	0	0	0	0	0
Tubificidae	0	2	1	0	2	0	9	11	10	88	1	2	1	5
Tvetenia	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Taxon	2000	2001	2003	2004	2005	2006	2007	2008	2009					
Potthastia	1	0	0	0	0	0	0	0	0					
Procladius	0	1	0	0	0	0	0	0	0					
Psectrocladius	0	0	0	0	0	0	0	1	0					
Radotanypus	1	0	1	0	0	0	0	0	0					
Rheocricotopus	0	0	0	0	2	1	1	0	0					
Sigara	0	0	0	0	0	0	4	0	0					
Silmulim	273	275	93	94	29	7	73	24	24					
Sphaeriidae	0	0	0	0	0	0	37	0	0					
Stagicola	0	0	16	11	3	1	1	0	0					
Tanypodinae	0	0	0	0	1	0	0	0	0					
Tanytarsus	0	0	0	0	7	0	0	0	0					
Thienemanniella	0	0	0	0	0	0	0	0	1					
Thienemannimyia grp.	2	3	0	0	6	0	0	0	0					
Tipula	0	1	0	0	0	1	2	0	0					
Tribelos	0	0	0	0	0	0	1	0	0					
Tricorythodes	0	0	0	0	0	0	0	0	0					
Tubificidae	34	6	48	80	51	90	5	2	27					
Tvetenia	0	0	0	1	0	0	0	0	0					

Taxon	2000	2001	2003	2004	2005	2006	2007	2008	2009
Acari	0	0	0	0	1	1	0	5	0
Aedes	0	0	0	0	0	0	0	0	0
Agabus	0	0	0	0	0	0	3	0	0
Apedillum	0	0	0	0	0	0	0	0	1
Baetis tricaudatus	0	0	0	0	0	0	0	0	0
Brundinella	0	0	0	0	3	0	0	0	0
Brychius	0	1	0	0	0	0	0	0	0
Cardiocladius	0	1	0	0	0	0	0	1	0
Ceratopogoninae	1	0	0	0	0	0	0	0	0
Chaetocladius	0	0	1	0	81	0	0	0	0
Chironomidae	0	0	0	0	1	0	1	28	0
Chiromus	3	5	13	3	5	2	9	1	0
Chuematopsyche	0	0	0	0	0	0	0	0	0
Clodocera	0	0	4	0	1	0	0	0	0
Coenagrionidae	0	0	0	0	0	0	1	0	0
Copepoda	0	0	0	0	3	0	0	1	0
Cricocotopus	31	12	92	76	1	48	7	90	38
Cryptochironomus	0	0	1	1	1	0	0	0	2
Dicrotendipes	0	0	0	0	0	3	0	7	12
Endochironomus	0	0	0	0	0	0	0	0	0
Eukiefferella	0	4	5	5	1	2	0	47	15
Glyptotendipes	0	0	0	0	1	0	0	0	0
Gyraulus	0	0	1	0	0	0	0	0	0
Haliphus	0	0	2	0	0	0	0	1	0
Helobdella stagnalis	0	0	0	0	0	0	136	71	93
Hesperoperla pacifica	0	0	0	0	0	0	2	0	0
Hydra	0	0	1	1	2	0	0	0	0
Hydropsyche	0	0	0	0	0	1	0	0	0
Limnephilidae	0	0	0	0	0	2	0	0	0
Limnophila	0	1	0	0	0	0	0	0	0
Microcylleopus	0	0	18	5	45	15	0	22	46
Macropelopia	0	0	0	0	0	0	0	0	0
Erpobdella	0	0	0	0	0	0	4	0	1
Muscidae	0	1	0	0	0	0	0	0	0
Naididae	0	0	0	0	0	4	0	0	2
Nematoda	0	0	0	0	0	0	0	8	19
Notonecta	0	0	0	0	0	0	3	0	0
Optioservus	0	0	0	0	1	0	0	0	0
Oreodytes	0	0	0	0	0	0	0	0	0
Orthocladus	1	8	0	0	5	8	1	4	8
Ostracoda	0	0	0	0	1	0	0	0	0
Pagastia	0	3	0	0	0	0	0	0	0
Parametriocnemus	0	1	2	2	0	2	0	5	3
Paratendipes	0	0	0	0	0	1	0	0	0
Phaenopsectra	10	9	6	12	16	1	1	0	1
Physa	0	0	41	0	13	0	8	0	0