CHARACTERIZING AND ASSESSING SEASONAL VARIATION OF THE BENTHIC MACROINVERTEBRATE COMMUNITY IN THE BIGHORN RIVER IN SE MONTANA

by

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DEDICATION

This one is for Shirley. Thank you for teaching me the importance of loving landscapes. I hope the red rocks are treating you well.

ACKNOWLEDGEMENTS

I would like to thank my advisor, Dr. Adam Sigler, for his guidance and insight and for challenging me to improve my writing skills and produce a paper I am proud of. I would also like to thank all of the LRES faculty whom I've had the privilege of learning from for challenging me to grow and work through rigorous, exciting material in their courses. Thank you to Erin Teichrow for her patience and support on my statistical approach and with my R code. I would also like to thank the wonderful staff at the MSU Library for connecting me with resources that are more challenging to access as a remote student.

I would not have been able to complete this project without the partnership, patience, and expertise of those with the Bighorn River Alliance. The BHRA Research Committee was supportive of this project and the Alliance's Research Associate, William McMahan, was instrumental in the completion of this project and shared his wealth of knowledge on macroinvertebrates openly. I am appreciative too of the BHRA Executive Director, Anne Marie Emery, for entertaining my proposition to partner with their organization and for her endless encouragement and support.

Thank you to my parents and family for their patience as I navigated work and school. Above all, thank you to the guy I married for his endless support, encouragement, and dedication towards keeping me fed over the past two and a half years.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Freshwater is a limited resource essential to the survival of humans and many other organisms and ecosystems on Earth, making its understanding and protection imperative. Benthic macroinvertebrates (BMI) are aquatic organisms found in these freshwater environments and they have long been used to characterize stream health as they demonstrate variable response to environmental conditions and play a large role as an intermediate trophic level within their food web, demonstrating both top-down and bottom-up effects. The Bighorn River Alliance (BHRA) is a nonprofit dedicated to the long-term health and viability of the Bighorn River in southeast Montana. They launched a research initiative in 2018 which includes biannual, seasonal collections of BMIs at several locations along the Bighorn River. Using data collected by the BHRA, I assessed BMI taxonomic diversity for differences in richness, alpha diversity, beta diversity, and rank abundance from eight collections sampled from fall 2020-fall 2024 across seven sites. I assessed this BMI community to set a baseline of expected seasonal variation in BMI assemblages, explore the impact of variable discharge and related factors on the BMI community, and as a tool to assess stream health. Seasonal differences are marked by increased species richness in fall, with abundant Naididae (sludge worms), *Parakiefferiella spp*. (midge), and *Baetis tricaudatus* (blue-winged olives) in spring and abundant *Caecidotea sp.* (aquatic sowbugs)*,* Naididae, and *Cricotopus spp*. (midge) in fall. The Bighorn River in this region is a tailwater river with hydrology primarily regulated by the Yellowtail Dam. While dams typically reduce hydrological variation, the system saw record flows in the summer of 2023. Collections after this event demonstrated reduced evenness, dominance by *Caecidotea sp.*, and more dissimilarity in community composition compared to collections prior to summer 2023. This shift appears to be concentrated to upper collection sites while lower collection sites demonstrated a shift towards more EPT (Ephemeroptera, Plecoptera, Trichoptera*)* taxa which are more sensitive to pollution than *Caecidotea sp.* This suggests that stream health may vary down the length of the river, but a more explicit site-wise analysis is needed.

Introduction

Across many ecosystems and civilizations, rivers are central figures providing fresh and accessible water. Less than 1% of all water on Earth is fresh and accessible (U.S. EPA, 2025a) and it provides crucial habitat for a diverse array of organisms and is vital to sustain life and support essential ecosystem functions and services. Humans rely on fresh water for drinking, agriculture, and industry. However, rivers may face challenges that threaten their health including degradation from dams, pollution, climate change, and poor water allocation management. Benthic macroinvertebrates (BMI) are an important biological component of river systems and can provide insight to understanding stream health as they vary in their response to pollutants, oxygen, and other factors (Goodnight, 1973). Diversity among aquatic macroinvertebrates has been shown to aid in essential functioning of streams because of their large role at the intermediate levels of a larger food-web within their environment (Wallace and Webster, 1996). They can impact ecosystem processes including nutrient cycles by influencing primary productivity, decomposition, and material transport and are also an important food source for fish (Wallace and Webster, 1996, p.115). Because BMI reside in the river, they can provide insight about river health from a longer period that isolated chemical water tests cannot.

BMI differ in their response to discharge, pollutants, and temperature with varying food and habitat needs; anthropogenic forces are known to alter these conditions in aquatic systems, which includes the construction of dams. Dams are built to meet various human needs including flood prevention, irrigation water, recreation opportunities, and hydropower. Water in the reservoir behind tall dams is often stratified by temperature, with warmer water near the surface and colder water at the base and dams with basal water releases can lower the temperature of the tailwaters below the dam during summer and maintain warmer temperatures during winter (Zaidel et al., 2021). Dam flow regulations typically decrease downstream hydrological variation by decreasing the timing and magnitude of peak flows, increasing base flows, and reducing the range of daily discharge (Foster et al., 2024; Petts, 1980; Merritt & Cooper, 2000; Graf, 2006). Sediment transportation is also significantly reduced as dams physically trap sediment in the reservoir, disrupting downstream transport (Foster et al., 2024). Sediment starvation coupled with reduced flood events associated with dams (Mei et al., 2017) can result in higher proportions of course sediments in the riverbed below the dam (Foster et al., 2024). The lack of sediment input can contribute to reduced geomorphic complexity which can be further amplified when floods do occur in sediment starved systems leading to increased channelization (Foster et al., 2024; Petts, 1980).

In an extensive literature review exploring ecological responses to altered flow regimes, Poff and Zimmerman (2009) found that macroinvertebrates typically experience a decline in abundance and diversity in response to both increased and decreased flow magnitude relative to reference sites or pre-impact conditions, but in some cases abundance and diversity did increase. In contrast, fish abundance and diversity consistently showed a negative response to changes in flow magnitude. When assessing biodiversity among macroinvertebrates, fish, and riparian species Poff and Zimmerman found that decreased peak flows, or flow stabilization, was generally associated with reduced diversity, altered assemblages, and a loss of sensitive taxa. When more extreme flow conditions occurred (both high and low), species richness typically declined in addition to assemblage alterations and impacts to sensitive species occurring. Most studies assessed in Poff and Zimmerman’s 2009 review were studies where dams and impoundments were the source of flow alterations.

Floods may cause initial decreases in macroinvertebrate abundance and richness, but most populations rebound relatively quickly (Le et al., 2020; Gholizadeh, 2021; Chattopadhyay et al., 2021). This initial decline is often associated with substrate disturbance and movement associated with high flows while the subsequent rebound is associated with substrate deposition occurring at reduced flows, recreating the habitat and conditions that support BMI populations post-flood (Chattopadhyay et al., 2021). Flood events also have the potential to mobilize and redistribute pollutants in some watersheds which could impact sensitive taxa (Ciszewski and Grygar, 2016). On the other hand, low flow or drought conditions are also known to impact BMI communities. In a comparison of short-term and long-term drought conditions, BMI communities demonstrated resilience to short-term droughts and returned to a community composition that closely resembled pre-drought assemblages (Kim et al., 2025). However, during long-term drought conditions, BMI functional evenness decreased, multivoltine species (species producing multiple generation of offspring per year) fared better, and similarity to pre-drought conditions were lowest suggesting that community composition is most impacted under long-term drought conditions (Kim et al., 2025). The decline in aquatic insect richness and abundance during low flows may be more closely associated with water temperature as opposed to the decreased discharge (Stubbington et al., 2009).

While dams inherently reduce hydrological variation in their role to mitigate both flood and drought conditions, climate change driven hydrological regime alterations can make it increasingly difficult for dams to hit their operating targets and have predictable releases

(California Department of Water Resources, n.d.). Understanding the extent and direction of BMI response to altered flow regimes may provide insight into how rivers can be managed to promote resiliency within BMI communities, which could help safeguard more flow-sensitive taxa, like the fish that feed on them, in response to hydrological changes and climate change.

Understanding the ecologic impacts of controlled dam releases is of primary concern for stakeholders relying on and studying the Lower Bighorn Watershed. The Bighorn River is the primary body of water in this watershed. The river's character is heavily influenced by the Yellowtail Dam, which is highly regulated by the Bureau of Reclamation. The Bureau of Reclamation is responsible for water releases from the dam with set minimum outflows and target lake depth levels for the reservoir across the year (Bureau of Reclamation, 2020). The release formula and process are complex and must comply with hierarchical water rights held by downstream users. This river, like many tailwater rivers, has demonstrated a reduction in both peak flows and flow variability since the construction of the dam was completed in 1967 (Klumpp, 1997). However, there is still noticeable variation in discharge and peak flow between years and the extent to which flow modifications impact macroinvertebrate communities is currently unknown. The river historically lacks long-term datasets on water quality, biodiversity, and geomorphology. The 2018 Research Initiative by the Bighorn River Alliance (BHRA) aims to close this gap through the development and implementation of sampling and analysis plans to better understand how natural and human drivers are influencing the river's health and function to help direct future projects (Kellogg et al., 2025). Recently, the BHRA has drawn attention to the considerable hydrological variation that has been occurring. My project aims to support the BHRA's research initiative by analyzing a portion of their benthic macroinvertebrate data collected from 2020-2024 to better understand factors that may influence BMI biodiversity and community composition in the Bighorn River.

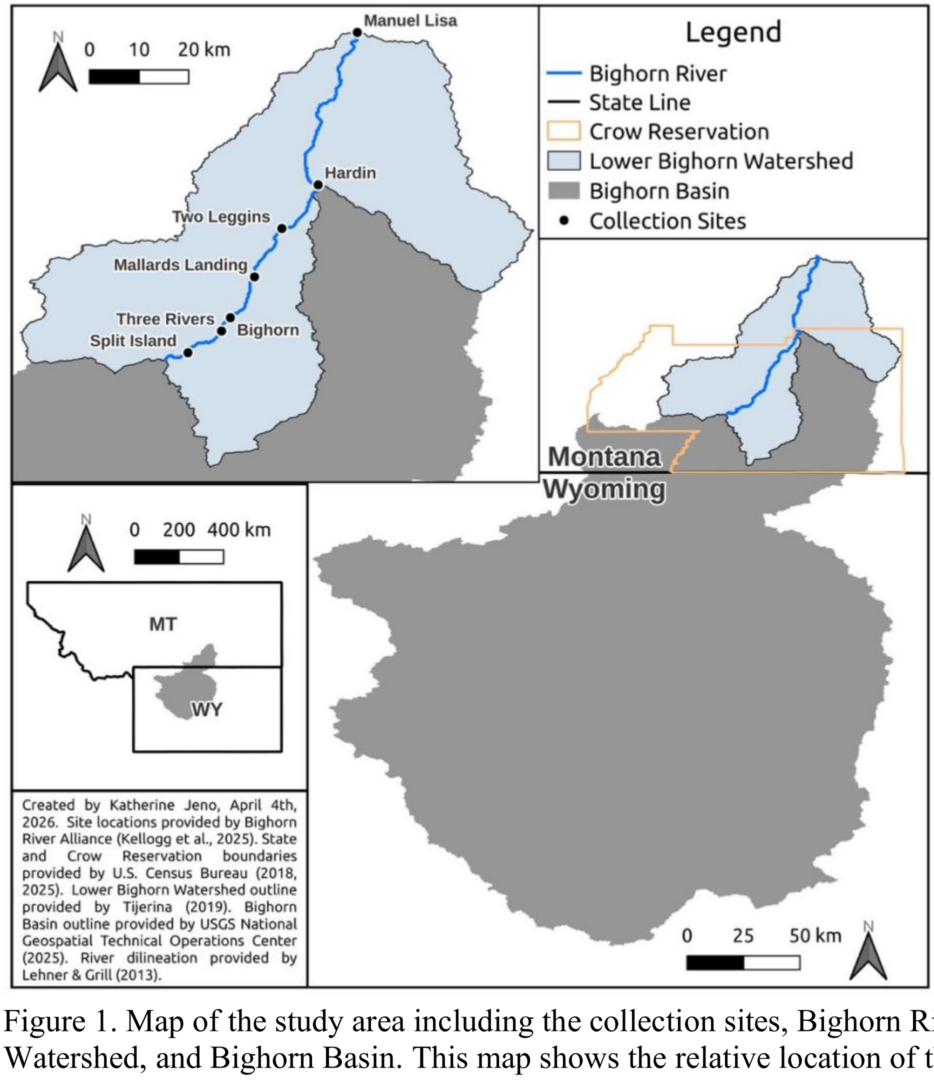
The goal of my study is to determine if there are significant differences in aquatic macroinvertebrate diversity in the Bighorn River from the fall of 2020 through the fall of 2024. The objectives for this work are to: 1) determine if benthic macroinvertebrate diversity in the Bighorn River differs between each collection period (spring and fall samples across the years) and each season (spring and fall collectively) as assessed by richness, alpha-diversity metrics, beta-diversity metrics, and rank abundance and 2) determine to what extent environmental conditions, such as discharge, are associated with benthic macroinvertebrate community composition and structure and to assess the relationship between biotic characteristics and treatment groups. While it's expected that fall and spring seasons will exhibit differences due to the unique life histories of BMI species in the Bighorn River, objective 1 serves to determine if those differences are consistent across all years regardless of the discharge variation or if consistent patterns are observed between years with higher or lower peak and median discharge. Objective 2 serves to further establish if any associations between discharge and macroinvertebrate diversity exist and provide insight for future areas of study.

Methods

Study Site

The Bighorn River in southeast Montana is an important resource for the region providing recreation, irrigation, and wildlife habitat. This river is the primary body of water in the Lower Bighorn Watershed (HUC 10080015), which spans 5,082 km^2 (1,255,731 acres) in southeastern Montana, lying primarily within Big Horn County, MT (U.S. Geological Survey

[USGS], 2025). The Lower Bighorn Watershed begins at the outlet of the Yellowtail Dam where the Bighorn River flows north for approximately 145 km (90 mi) before its confluence with the Yellowstone River near Bighorn, MT (National Park Service [NPS], 2019). The river originates at the Boysen Reservoir, 235 km (146 mi) upstream of the dam. The Lower Bighorn Watershed is the final catchment within the greater Bighorn Basin (HUC 100800) which spans from northwest Wyoming into southern Montana (Figure 1).



Created by Katherine Jeno, April 4th, 2026. Site locations provided by Bighorn River Alliance (Kellogg et al., 2025). State and Crow Reservation boundaries provided by U.S. Census Bureau (2018, 2025). Lower Bighorn Watershed outline provided by Tijerina (2019). Bighorn Basin outline provided by USGS National Geospatial Technical Operations Center (2025). River dilineation provided by Lehner & Grill (2013).

Figure 1. Map of the study area including the collection sites, Bighorn River, Lower Bighorn Watershed, and Bighorn Basin. This map shows the relative location of the study area within the Bighorn Basin and the states of Montana and Wyoming as well as the boundary of the Crow Reservation.

Yellowtail Dam, built in the 60s, is a multi-purpose dam that provides power generation, storage for irrigation, flood control, and recreation opportunities (U.S. Bureau of Reclamation, n.d.). The dam sits at the base of the Bighorn Canyon. For the first 45 miles from Yellowtail Dam to just outside of Hardin, MT the Bighorn River flows through the lands of the Crow Reservation. The Bighorn Canyon is a sacred site for the Crow people and as such, the construction of the dam was controversial in the area (NPS, 2022). Contentions rose after the completion of the dam over the responsibilities of managing the river and the newly created cold-water fishery in the tailwaters. In 1981, after lengthy legal battles, the Bighorn River was turned over to the control of the state of Montana (Montana Department of Fish, Wildlife, and Parks [Montana FWP], n.d). Today, the river is managed by the Montana Department of Fish, Wildlife, and Parks.

The Bureau of Reclamation is responsible for managing water releases from the dam and these releases heavily moderate the more natural flow regime of the upper Bighorn River from the base of the dam to Hardin, with increasing input from tributaries and run-off in the lower river from Hardin to the Bighorn's confluence with the Yellowstone River (Klumpp, 1997). While rivers below dams typically exhibit decreased peak flows compared to their undammed counterparts (Foster et al., 2024), the Bighorn River has experienced notable, unpredictable fluctuations in discharge over the past few years and the impacts this has on aquatic macroinvertebrates is currently being explored.

Bighorn River Alliance Research Initiative

The Bighorn River Alliance formed in the 1990s when a group of concerned anglers came together to protect, preserve, and enhance the health of the Bighorn River (BHRA, n.d.).

Today, the BHRA has become a respectable organization collaborating with reservation, federal, and state agencies as well as local stakeholders, like farmers and fishermen, to create solutions to watershed problems and improve the viability of the river (BHRA, n.d.). One hurdle posing a challenge for the Alliance is to develop solutions that preserve and enhance a system that is no longer operating under entirely natural controls. For example, the dam has altered the river's hydrology by reducing sediment input, impacting discharge (Foster et al., 2024), and converting the upper river below the dam from a warm to a cold, tailwater fishery which now supports an abundant trout population. In response to these challenges, the BHRA developed a Research Initiative in 2018 to better understand the various natural and human factors that influence this ecosystem (Kellogg et al., 2025). Beginning in 2020, the BHRA began monitoring and sampling the benthic macroinvertebrate community of the Bighorn River.

Study Period & Sampling Locations

Data used in this study was collected by the BHRA from 2020 to 2024. The intention of the BHRA was to sample for macroinvertebrates bi-annually in the spring and fall of each year. However, data was only collected in both spring and fall of 2021-2023 with 2020 and 2024 lacking a spring data set. In total, there are 8 datasets available from the macroinvertebrate sampling during this time: fall 2020, spring 2021, fall 2021, spring 2022, fall 2022, spring 2023, fall 2023, and fall 2024.

Throughout the study period, peak discharge varied from a low peak of 3,390 cfs in 2021 to a high peak of 19,500 cfs in 2023 and median peak discharge over the past 12 years was 5,700 cfs (Table 1; Figure 2; USGS, 2001). Median discharge during the study period was around 2,790 cfs with far less range than the peak discharge (Table 1; Figure 2). This data is from USGS gauge site 06287800 Bighorn River at bridge, at St. Xavier, MT (USGS, 2001) which reflects the combined discharge of dam releases, tributary contributions, and run-off.

Table 1. Peak and Median Discharge from USGS gauge site 06287800 Bighorn River at bridge, at St. Xavier, MT 2020-2024 (USGS, 2001).

Year	Peak Discharge (cfs)	Median Discharge (cfs)	Minimum Discharge (cfs)
2020	5530	3230	1630
2021	3390	2220	1480
2022	7500	2350	1560
2023	19500	3000	2260
2024	6180	3170	2080

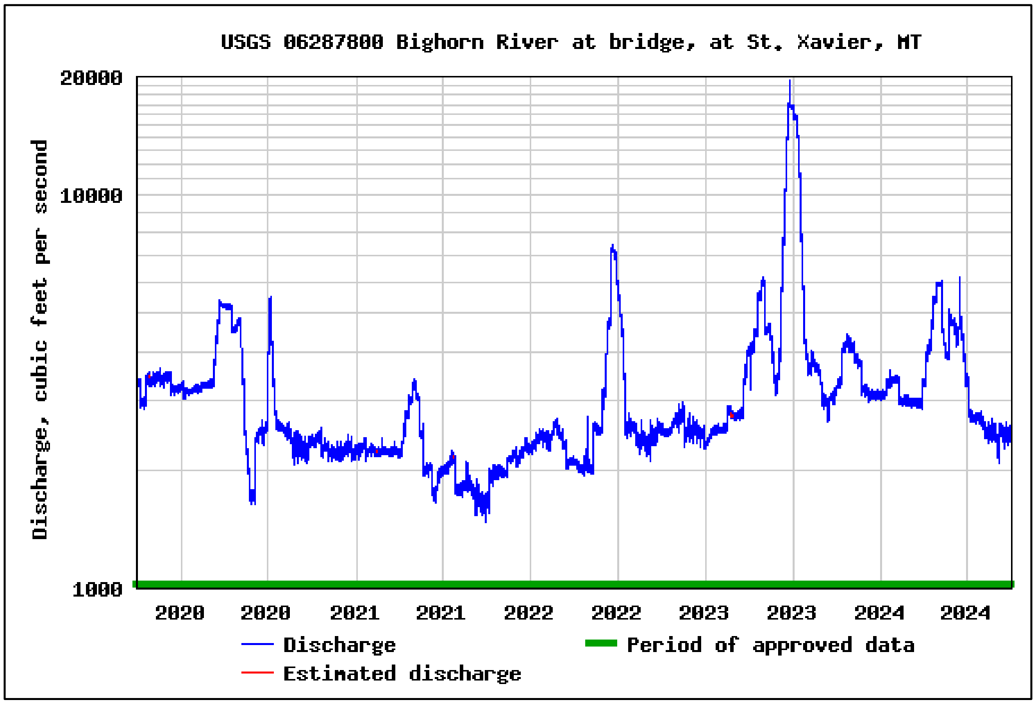


Figure 2. Hydrograph from October 2019-September 2024 at USGS 06287800 Bighorn River at bridge, at St. Xavier, MT. (USGS, 2001).

BMI collections included in this study were taken from seven locations along the Bighorn River. Starting at the Bighorn's confluence with the Yellowstone River and working south towards the dam, the sampling sites are as follows: Manuel Lisa Fishing Access Site (FAS) at river mile (RM) 1.8, near Hardin at RM 40, Two Leggins FAS at RM 52, Mallards Landing FAS at RM 63, Bighorn FAS at RM 72, Three-Rivers Junction at RM 75, and Split Island at RM 82.

North of Hardin, the river contains both cold and warm water fish species and south of Hardin the river primarily contains cold water fish due to the hypolimnetic release of cold water from far below the dam surface (Montana FWP, 2023).

Data Collection

BMI samples were collected from designated riffles in near-bank habitats from downstream to upstream at each site. Three replicates were taken at each site using a Hess sampler at appropriate water depths for the equipment. The river substrate area ($0.1m^2$) in the Hess sampler was carefully raked for one minute until all the organic matter was flushed into the collection net of the sampler. After elutriation and careful examination of inorganic matter, this material was then transferred into collection containers filled with 95% ethanol. Samples were then sent to the lab for processing where BMI were counted and identified to the lowest taxonomic level possible following MDEQ (2012) protocols. This provided the number of individuals per taxa per $0.1m^2$ per sample which was then be extrapolated to calculate the average number of individuals/taxa/m^2 at each site. More detailed methodology can be found in BHRA's Water Quality and Macroinvertebrate Monitoring Sampling and Analysis Plan (Kellogg et al., 2025).

Data Processing

Data were provided to me by BHRA for all eight collections included in this study. Each collection's data was provided in a separate Excel spreadsheet. For collections from 2023 on, raw counts were provided and required extrapolation from $0.1m^2$ to $1m^2$ so that BMI counts were assessed with the same units for all collections. I combined this abundance data into one master data set in Excel with all data in a standardized format that would be appropriate for statistical analysis. I then standardized taxa between all years while keeping all data at the lowest taxonomic level possible. In some cases, multiple taxa were combined, such as the instance where the family of Tubificidae was reclassified and absorbed into the larger family of Naididae.

Once all counts were finalized for each site and each collection, I then researched each species to identify its tolerance value, functional feeding group, functional habitat group, taxonomic classification, and voltinism characteristics primarily utilizing Merritt et al. (2019) in addition to The Watershed Institute (n.d.), Mandaville (2002), and Squam Lakes Association (n.d.). Using this information, I then calculated total abundance, the Hilsenhoff Biotic Index (HBI) using Equation 1, and the percent of individuals belonging to the following groups: Ephemeroptera, Plecoptera, and Trichoptera (EPT), swimmers, collector-gatherers, scrapers, predators, collector-filterers, piercers, shredders, divers, clingers, climbers, burrowers, sprawlers, univoltine, bivoltine, trivoltine, multivoltine, semivoltine, merovoltine, non-insects, and chironomids.

$$HBI = \frac{\Sigma n_i a_i}{N}$$

Equation 1. Hilsenhoff Biotic Index. Where *n* is the number of specimens in taxa *i*, *a* is the tolerance value of taxa *i*, *N* is the total number of specimens in the sample. A greater HBI value corresponds with an increased abundance of pollution tolerant taxa present in the sample.

These were calculated for each site at each collection. This data represents all biotic characteristics I used in addressing the final objective. For abiotic data, I downloaded the continuous (15 minute) discharge data from the USGS monitoring station USGS-06287800 Bighorn River at bridge, at St. Xavier, Montana (USGS, 2001) and calculated the median and maximum discharge each year. Then, I exported each data set as its own .csv file for a total of three data sets: abundance data, biotic characteristics, and abiotic data. These data sets were then in the proper format for import into RStudio (R Core Team, 2023) where I could perform the statistical analysis.

Statistical Methods

Objective 1

The first objective aims to compare richness, alpha diversity, beta diversity, and rank abundance between each collection and each season. To analyze richness and alpha diversity, I used the year and season as explanatory variables and my treatment groups were each collection period (fall 2020, spring 2021, fall 2021, spring 2022, fall 2022, spring 2023, fall 2023, and fall 2024) and each season (spring and fall). Using the abundance data, I then calculated the species richness in R. Richness provided the number of distinct taxa present at each of the seven sites for each collection period, for a total of 56 values. I evaluated species richness differences between the treatment groups using an additive generalized linear model with Poisson distribution, and an analysis of variance (ANOVA) test followed by a pairwise comparison with Tukey adjustment.

For alpha diversity, I calculated Shannon's and Simpson's diversity and evenness indices using the R package 'vegan' (Oksanen et al., 2022). However, I displayed inverse-Simpson's (1-D) diversity index in figures as it creates standardization between the interpretation of both indices with higher values indicating greater diversity. Shannon's diversity index places more emphasis on rare species whereas Simpson's diversity index puts more emphasis on dominant species (Magurran & McGill, 2011). To assess differences in alpha diversity among the treatments I used an additive linear regression model and ANOVA, again followed by a pairwise comparison with Tukey adjustment for each of the four indices.

To assess beta diversity, I calculated the percent similarity using the R package ‘vegan’ (Oksanen et al., 2022). This was done both between and within collections and seasons using Jaccard, Bray-Curtis, and Morisita-Horn’s dissimilarity indices. Jaccard’s dissimilarity index uses only presence and absence data whereas the Bray-Curtis and Morisita-Horn indices use abundance data (Magurran & McGill, 2011). The Morisita-Horn index can handle different sample sizes and is resistant to under sampling (Oksanen et al., 2022; Magurran & McGill, 2011). An Adonis test was used for each beta metric to assess if there was a difference between treatments, regarding their dissimilarity. To visualize this data, I used the R package ‘labdsv’ (Roberts, 2023) to generate an NMDS ordination plot using the Bray-Curtis dissimilarity index for all eight collections and for both seasons.

Finally, I subset my abundance data by each collection and each season and ranked each species from highest to lowest abundance. I then relativized this abundance data and created relative rank abundance plots for collections and seasons. I accomplished this using the R packages ‘Biodiverity R’, ‘ggplot2’ and ‘ggrepel’ (Kindt and Coe, 2005; Slowikowski, 2024; Wickham, 2016). Calculations for all biodiversity indices can be located in Magurran & McGill (2011).

Objective 2

The next analysis aims to better understand the impact of environmental variables on BMI diversity and community structure for each collection and season as well as the relationship between biotic characteristics and community composition. I generated an NMDS ordination plot using the R package ‘vegan’ and the Bray-Curtis dissimilarity index for each collection (Oksanen et al., 2022). I also used this package to create vectors of the species abundance, biotic characteristics, and abiotic factors. I then subset the significant vectors using a threshold of $p<0.005$ for species vectors, $p<0.001$ for biotic characteristics, and $p<0.05$ for abiotic vectors for ease of interpretation and legibility on the NMDS plots. These vectors were then fitted to the NMDS plot and illustrate the correlation between factors and ordination points with the length of the vector arrows corresponding to the strength of the correlation.

I used R version 4.2.3 for all analyses (R Core Team, 2023).

Results

Objective 1

Richness

Spring collections combined had a normal distribution with mean richness near 34 and fall collections combined were skewed slightly left with a median richness near 40 with one outlier above and below the median (Figure 3). Both fall and spring exhibit similar ranges for richness. Fall 2020 is heavily skewed right with median richness near 35 and on outlier near 24 (Figure 3). Spring 2021 is skewed right with median richness near 34. Fall 2021 is skewed left with median richness near 36 and one outlier near 48. Spring 2021 has a slight rightward skew with median richness near 34 and an outlier above and below the mean. Fall 2022 is skewed right with median richness near 40 and one outlier near 33. Spring 2023 has a fairly normal distribution with median richness near 33 with no outliers. Fall 2023 is skewed left with median richness near 41. Fall 2024 has a near normal distribution with median richness near 39 and the greatest spread of all collections. In general, spread was greatest in the three most recent collections (Figure 3). The regression model suggested that richness was greater across fall collections of all years compared to spring collections ($p=0.014$) and the ANOVA test indicated there was a difference in species richness between seasons ($p=0.014$) but not between years ($p>0.1$). However, the Tukey pairwise comparison indicated there was no evidence of differences between species richness in each possible combinate of collections ($p>0.1$ all).

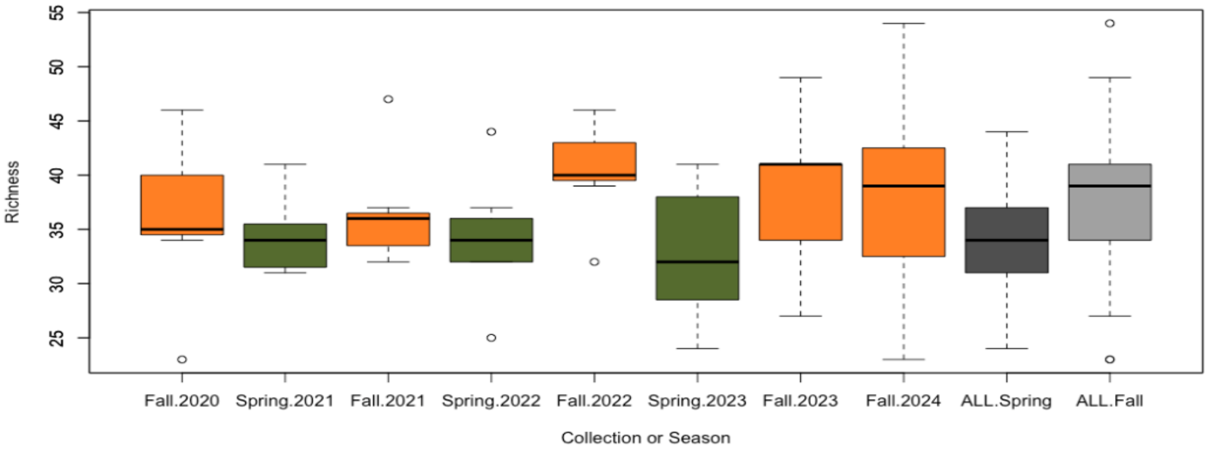
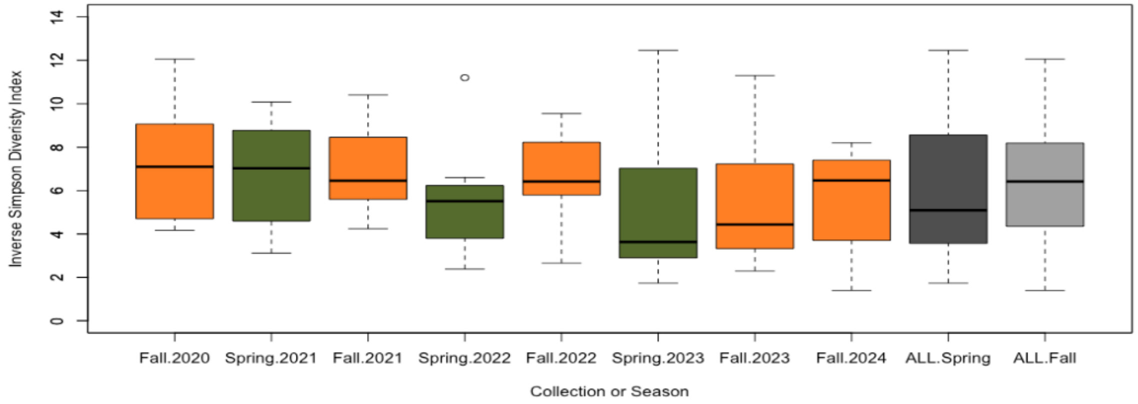


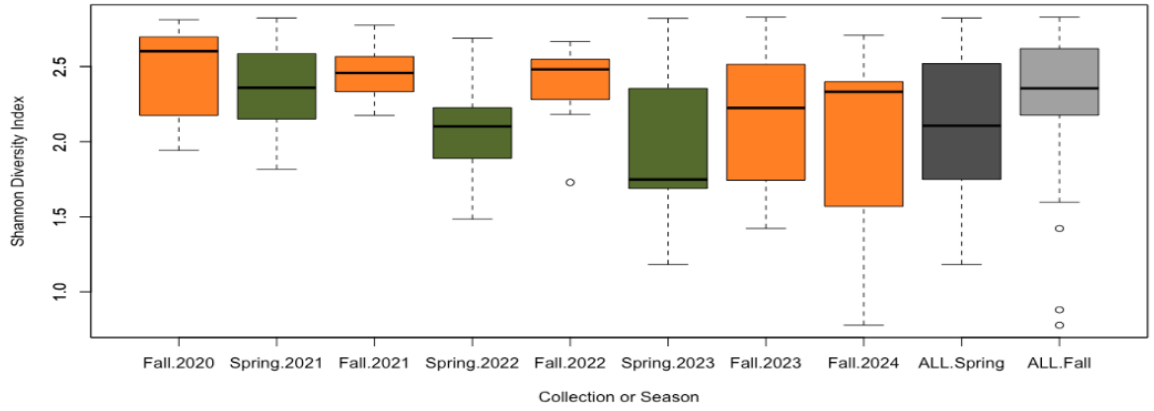
Figure 3. Species richness for each collection period and for seasons collectively. Species richness was calculated for each season each year where data was collected and species richness was calculated for both seasons collectively, without regard to year. The boxplots show the spread of the means within each timeframe (n= 7 sites for each collection, $n_{spring}=21$, and $n_{fall}=35$). The horizontal line is the median, each box represents 50% of the data, and the whiskers extend to the farthest point within 1.5 times the interquartile range from the box, circles are outliers.

Alpha Diversity

Of the eight collections, the first three (fall 2020, spring 2021, and fall 2021) differed regarding their relative patterns between Shannon and Simpson diversity but the remaining collections followed relatively similar patterns (Figure 4). Fall 2020 and spring 2021 had similar medians with Simpson's diversity with fall 2020 being right skewed and spring 2020 being left skewed and with a smaller range. With Shannon's diversity, spring 2021 was normally distributed with a lower median than fall 2020 with a greater range while fall 2020 was skewed left. Collectively, spring had a lower median than fall for both Simpson's and Shannon's diversity indices (Figure 4). Spring 2022 had one outlier above the median with Simpson's index and fall 2022 had one outlier below the median with Shannon's index. When fall was combined, it exhibited several outliers below the median with Shannon's index. For Simpson's index, the regression model (p>0.1 all), ANOVA test (p>0.1 all), and Tukey pairwise comparison (p>0.1 all) indicated there was no evidence of a difference in alpha diversity between years, seasons, or between each possible combinate of collection pairs. For Shannon's diversity index, the regression model had moderate evidence (p=0.059) that the fall 2024 collection period had lower diversity than collections in other years, but neither the ANOVA test nor Tukey's pairwise comparison suggested that diversity differed between years or in each possible collection pair comparisons (p>0.1 all).



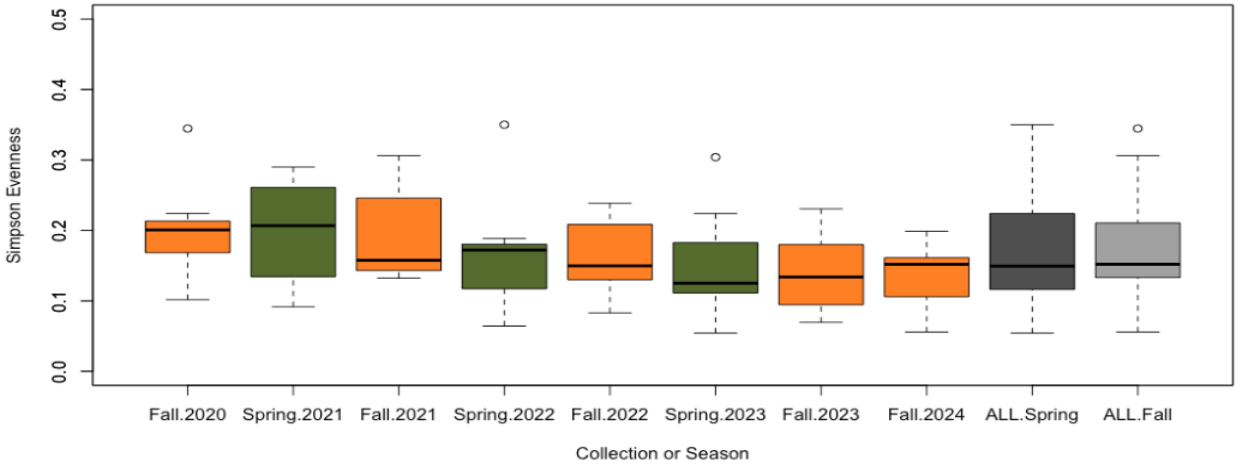
a. Inverse Simpson's Diversity



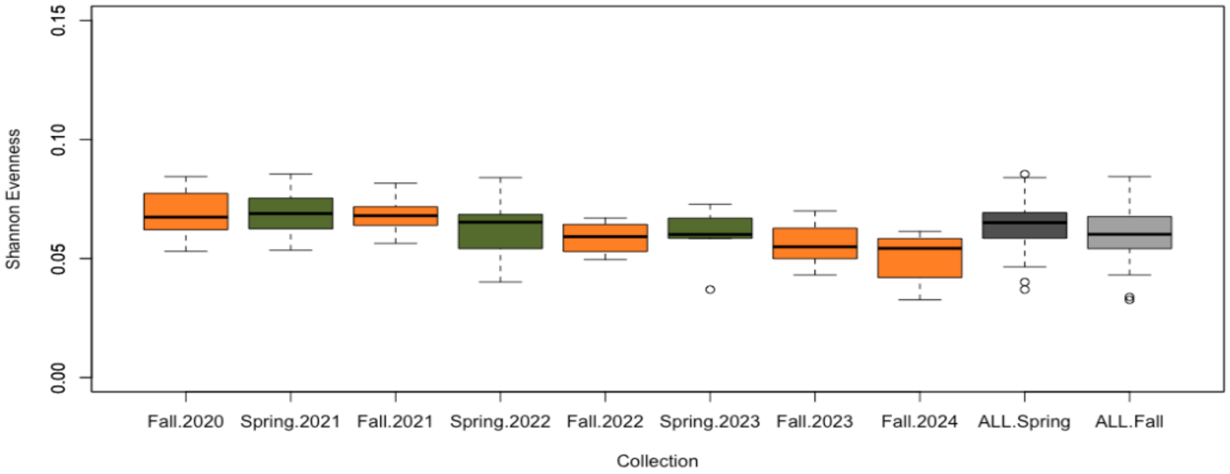
b. Shannon's Diversity

Figure 4. Shannon's and Simpson's inverse diversity indices. Simpson's inverse (a) and Shannon's diversity (b) indices are displayed for each collection period and each season. Both indices were calculated for each season each year where data was collected and each index was calculated for both seasons collectively, without regard to year. The boxplots show the spread of the means within each timeframe (n= 7 sites for each collection, n_{spring}=21, and n_{fall}=35). The horizontal line is the median, each box represents 50% of the data, and the whiskers extend to the farthest point within 1.5 times the interquartile range from the box, circles are outliers.

For evenness, there was more range in Simpson's evenness index for all collections and seasons than there was in Shannon's evenness index (Figure 5). Evenness was low across all collections and years for both indices. The collections followed similar relative patterns between both indices except for fall 2021, which was skewed heavily right with Simpson's evenness index but was normally distributed with Shannon's evenness index (Figure 5). For the seasons, spring and fall both had a median near 0.15 for Simpson's evenness index with a slight rightward skew but for Shannon's evenness index the median for spring was slightly higher than the median for fall and both were normally distributed (Figure 5). For Simpson's evenness index, the linear regression suggested moderate evidence that collections in 2023 (p=0.099) and 2024 (p=0.082) had less evenness than collections from the other years but no difference existed between seasons (p>0.1). However, both the ANOVA test and Tukey pairwise comparison for Simpson's evenness index suggested there was not a difference between years, seasons, or collections (p>0.1 all). For Shannon's evenness index, the linear regression suggested moderate evidence that evenness was lower in collection from 2022 (p=0.060) and stronger evidence from this model suggested evenness was lower in collections from 2023 (p=0.023) and 2024 (p=0.002) compared to collections from 2020 and 2021. It did not suggest a difference between seasons (p>0.1). The ANOVA had strong evidence that there was a difference in Shannon's evenness index between years (p=0.003) but not between seasons (p>0.1). Tukey's pairwise comparison had evidence that there was a difference between fall 2020 and fall 2024 (p=0.049), fall 2021 and fall 2024 (p=0.053), spring 2021 and fall 2024 (p=0.013) with fall 2020, fall 2021, and spring 2021 having greater evenness than fall 2024.



a. Simpson's Evenness



b. Shannon's Evenness

Figure 5. Shannon's and Simpson's evenness indices. Simpson's inverse (a) and Shannon's evenness (b) indices are displayed for both collections and for seasons. Both indices were calculated for each season, each year where data was collected and each index was calculated for both seasons collectively, without regard to year. The boxplots show the spread of the means within each timeframe (n= 7 sites for each collection, n_{spring}=21, and n_{fall}=35). The horizontal line is the median, each box represents 50% of the data, and the whiskers extend to the farthest point within 1.5 times the interquartile range from the box, circles are outliers.

Beta Diversity.

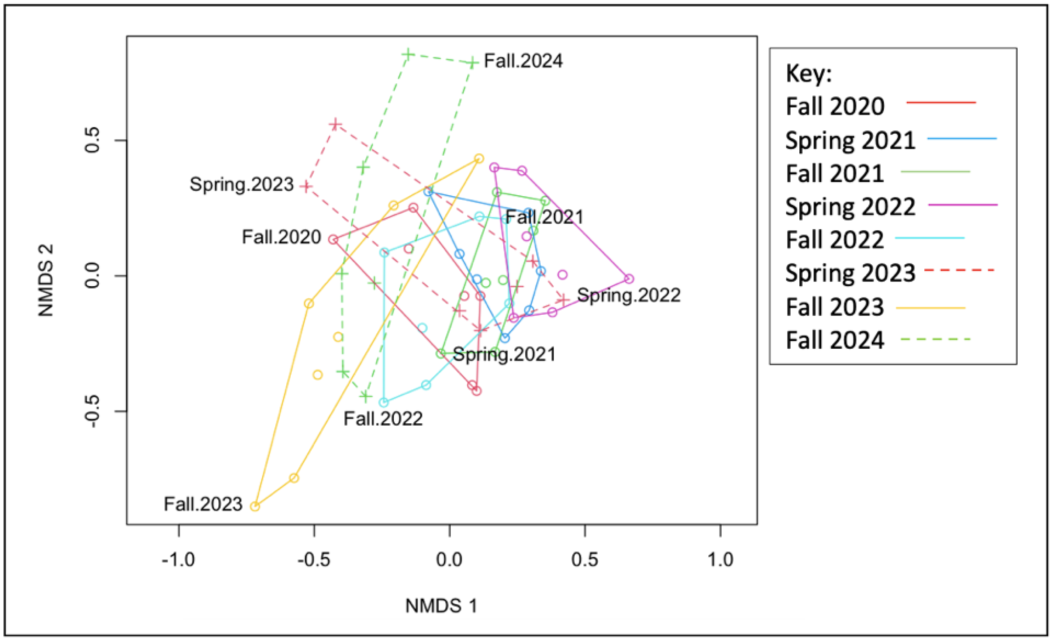
There was strong evidence (p<0.03 all) of a difference between community composition within each Jaccard, Bray-Curtis, and Morisita-Horn dissimilarity indices between each year, each season, and each collection (Table 2). Percent similarity was typically higher between each spring collection than between each fall collection (Table 3) and all spring collections were more similar to each other (36.4%) than all fall collections (24.0%). Percent similarity was lowest between seasons (22.6%). The NMDS plot for Bray-Curtis' dissimilarity does show an extent of overlap between seasons and collections, but generally spring collections were more clustered while fall collections exhibited more spread (Figure 6). Fall 2023 and fall 2024 had the least overlap with other collections, contributed most to the spread of fall data points on the NMDS plot, and tended to have lower percent similarity compared to other fall collections.

Table 2. Adonis test results for Jaccard, Bray-Curtis, and Morisita-Horn dissimilarity for community composition assessed at the level of year, season, and collection.

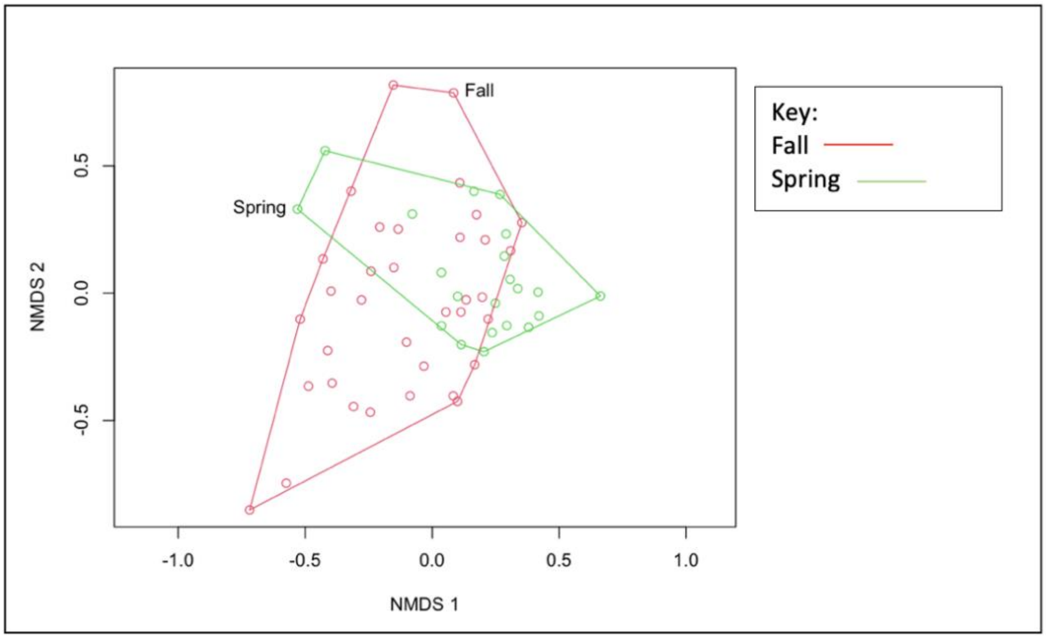
Dissimilarity Index	**p-value**		
	Year	Season	Collection
Jaccard	0.001	0.001	0.006
Bray-Curtis	0.001	0.001	0.004
Morisita-Horn	0.001	0.001	0.027

Table 3. Percent similarity based on the inverse of Bray-Curtis' dissimilarity index calculated for aquatic macroinvertebrate communities of the Bighorn River within and between collections. Percent similarity explains how similar sites are to each other within the same collection and how similar sites are collectively between collections. Green values have greater similarity and as values become more yellow and red they indicate decreasing similarity.

Collection	Fall.2020	Fall.2021	Spring.2021	Fall.2022	Spring.2022	Fall.2023	Spring.2023	Fall.2024
Fall.2020	34.3%							
Fall.2021	33.7%	39.0%						
Spring.2021	31.4%	35.6%	41.7%					
Fall.2022	32.0%	30.6%	29.8%	32.2%				
Spring.2022	23.9%	34.6%	37.9%	26.1%	37.1%			
Fall.2023	21.2%	16.1%	14.7%	19.7%	10.8%	19.9%		
Spring.2023	25.1%	30.4%	37.3%	23.3%	31.2%	12.6%	37.1%	
Fall.2024	20.7%	18.2%	15.3%	17.7%	12.3%	18.3%	12.5%	23.0%



a. Collection



b. Season

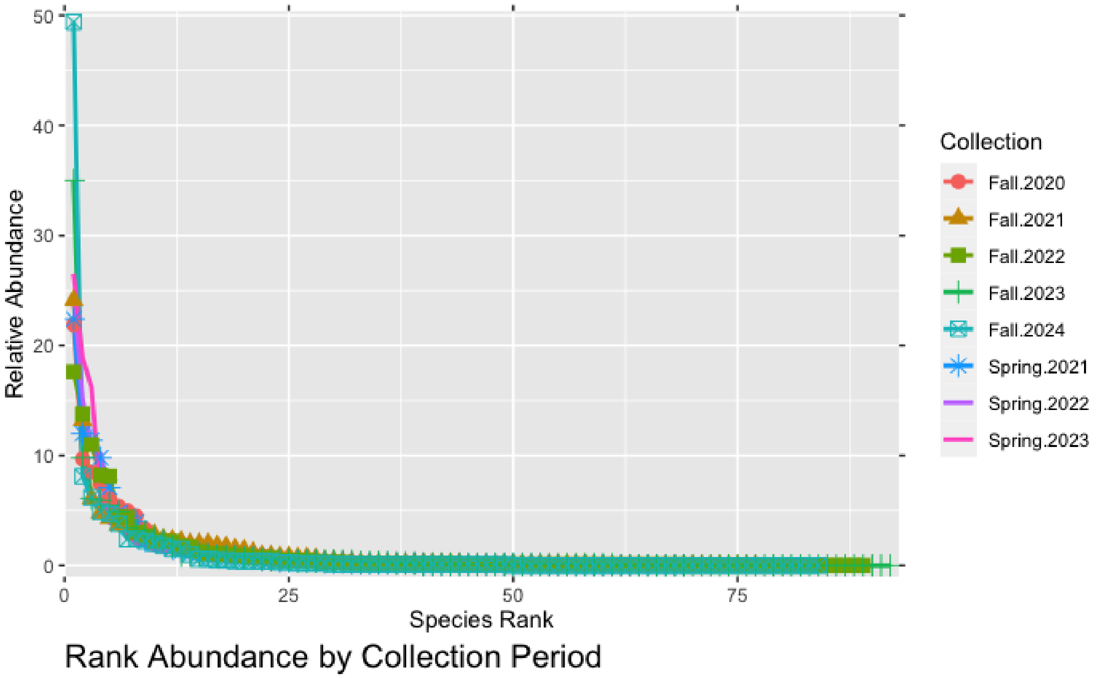
Figure 6. NMDS Ordination plot of all collections (a) and seasons (b) based on Bray-Curtis's dissimilarity index. Community composition differed between collections and seasons.

Rank Abundance

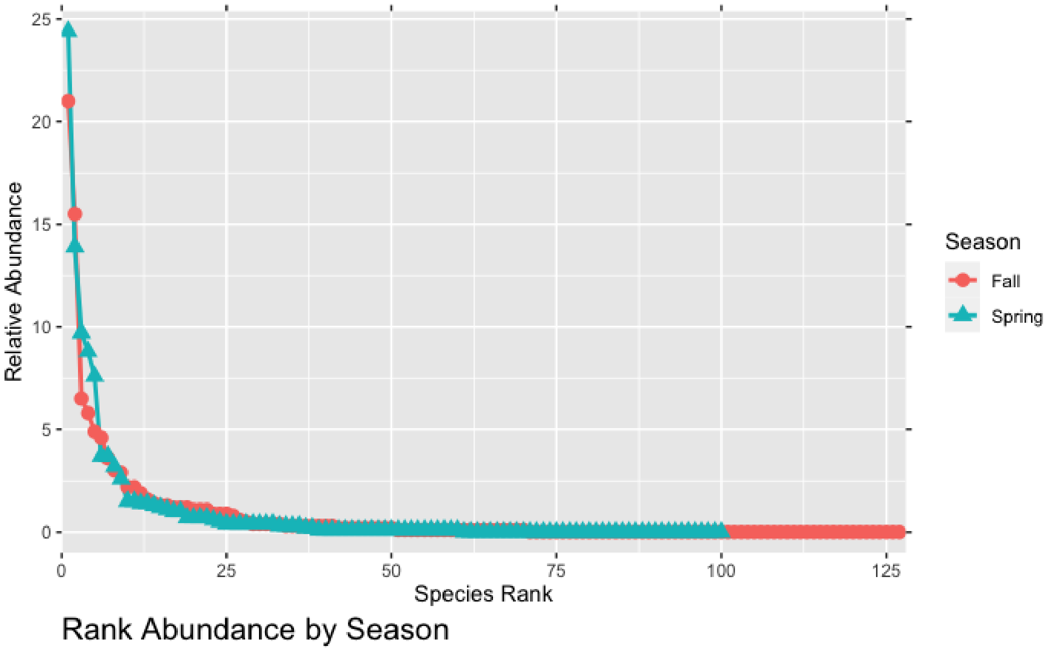
The rank abundance curves were similar between spring and fall (Figure 7) except the species occupying the greatest proportion in spring was 3.4% higher than in fall but fall had more species overall. For each collection, the rank abundance curve was also similar for fall 2020-spring 2023 (Figure 7.a) but fall 2023 and 2024 had a species dominating a higher percent compared to the rest of the collections. Fall 2023 had the most species followed by fall 2022 and then fall 2024 (Figure 7.a). For all collections from fall 2020 through spring 2023 Naididae was the top taxon of the BMI community ranging from 17.6%-26.5% (Table 4). In fall 2023 and 2024 the top-ranking taxon was *Caecidotea sp.* (35.0% and 49.4%, respectively). The contribution of *Caecidotea sp.* from fall of 2023 and 2024 was so great that it was also the top taxon of all falls collectively (21.0%) and the second highest taxon across all falls was Naididae (15.5%). *Caecidotea sp.* was present in all collections except spring 2023 (Table 4). Spring and fall had four taxa in common: Naididae*, Caecidotea sp., Baetis tricaudatus,* and *Cricotopus spp*. The other two top taxa in spring were *Parakiefferiella spp.* (13.9%) and *Diamesa spp.* (8.8%) which did not occupy a top six taxa in any fall collection. The other two top taxa in fall were *Tricorythodes explicatus* (5.8%) and Turbellaria (4.9%) which did not occupy a top six taxa in any spring collection.

Table 4. Top six ranking species of each benthic macroinvertebrate collection and each season in the Bighorn River. For each period aquatic macroinvertebrate orders were ranked in terms of their abundance. The proportion of that collection year occupied by any given order was calculated and relativized between all collections and seasons, presented below as a percent.

Collection	**Species**	**Percent**	**Collection**	**Species**	**Percent**
Fall 2020	*Naididae*	21.9	**Fall 2022**	*Naididae*	17.6
	Caecidotea sp.	9.7		*Baetis tricaudatus*	13.8
	Turbellaria	8.5		*Hydroptila spp.*	11.0
	Tricorythodes explicatus	7.4		*Tricorythodes explicatus*	8.2
	Physella sp.	6.1		*Caecidotea sp.*	8.1
	Hydroptila spp.	5.4		*Cricotopus spp.*	4.4
Spring 2021	*Naididae*	22.4	**Spring 2023**	*Naididae*	26.5
	Baetis tricaudatus	12.0		*Diamesa spp.*	18.9
	Diamesa spp.	11.4		*Parakiefferiella spp.*	16.2
	Parakiefferiella spp.	9.8		*Cricotopus spp.*	5.9
	Orthocladius spp.	7.1		*Baetis tricaudatus*	5.0
	Caecidotea sp.	4.8		*Phaenopsectra sp.*	4.6
Fall 2021	*Naididae*	24.1	**Fall 2023**	*Caecidotea sp.*	35.0
	Cricotopus spp.	13.2		*Naididae*	9.8
	Caecidotea sp.	6.1		*Acentrella insignificans*	6.1
	Psectrocladius sp.	4.8		*Polypedilum spp.*	5.9
	Dicrotendipes sp.	4.3		*Tricorythodes explicatus*	5.5
	Turbellaria	3.7		*Turbellaria*	4.9
Spring 2022	*Naididae*	24.7	**Fall 2024**	*Caecidotea sp.*	49.4
	Parakiefferiella spp.	15.2		*Hydropsyche occidentalis*	8.1
	Baetis tricaudatus	10.6		*Tricorythodes explicatus*	6.2
	Cricotopus spp.	10.2		*Turbellaria*	4.9
	Caecidotea sp.	3.9		*Petrophila sp.*	4.7
	Lumbricidae	3.8		*Physella sp.*	3.8
Season	Species	Percent	Season	Species	Percent
Spring	*Naididae*	24.4	**Fall**	*Caecidotea sp.*	21.0
	Parakiefferiella spp.	13.9		*Naididae*	15.5
	Baetis tricaudatus	9.7		*Cricotopus spp.*	6.5
	Diamesa spp.	8.8		*Tricorythodes explicatus*	5.8
	Cricotopus spp.	7.6		*Baetis tricaudatus*	4.9
	Caecidotea sp.	3.7		*Turbellaria*	4.6



a. Collection

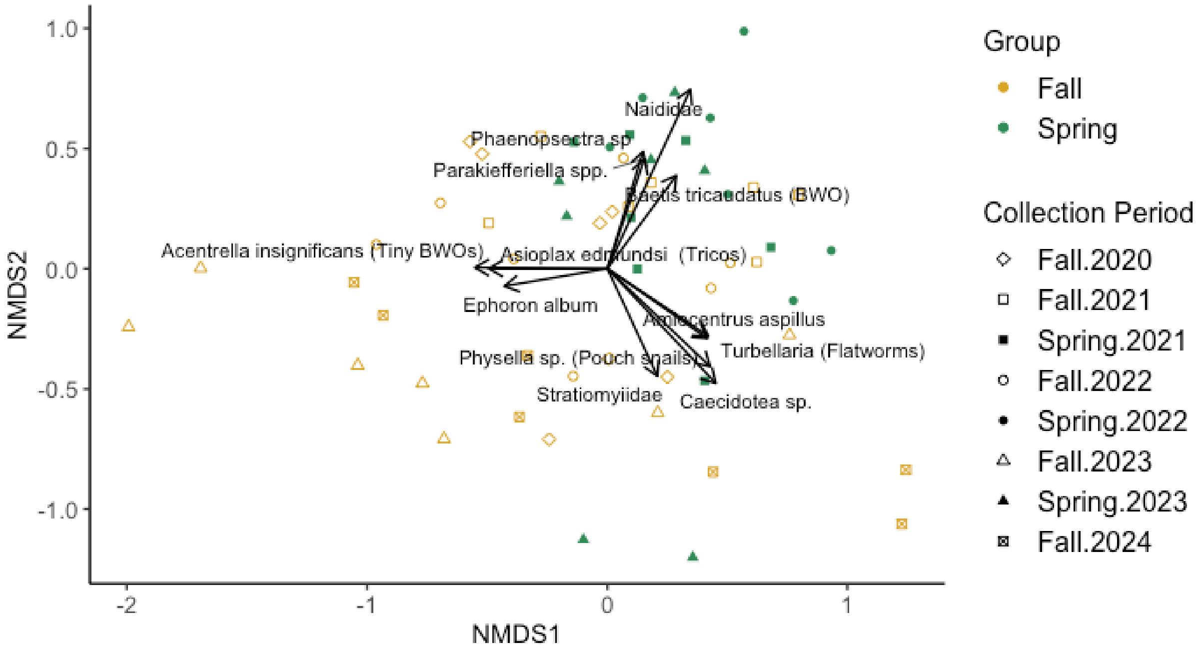


b. Season

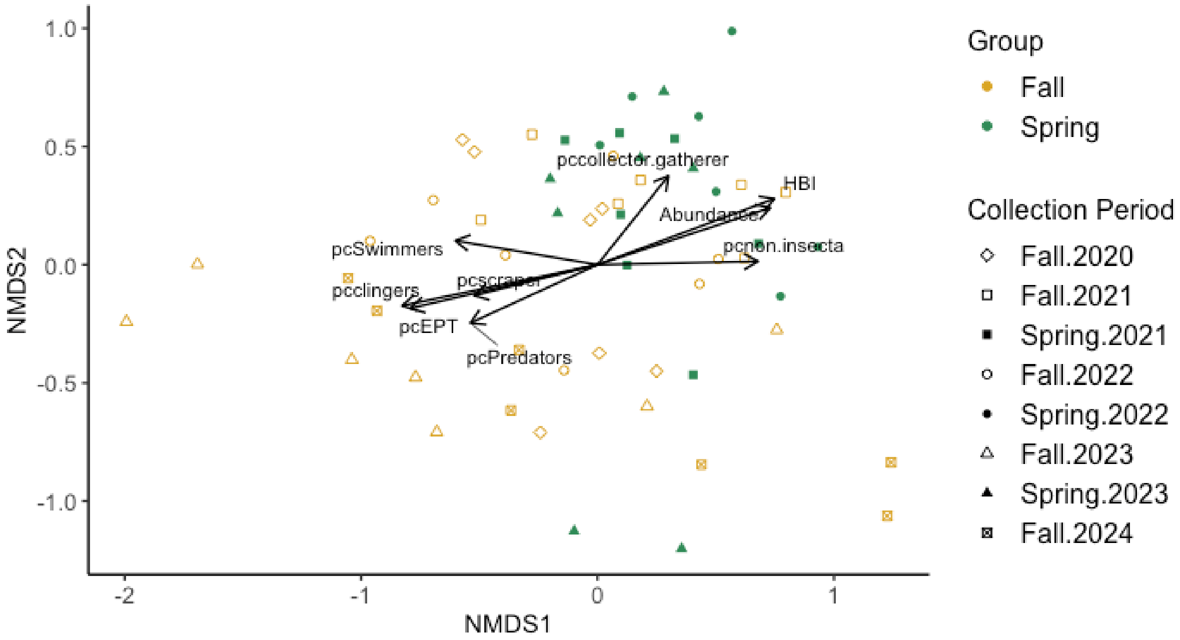
Figure 7. Relative species rank abundance for each collection (a) and for each season (b). For all periods aquatic macroinvertebrate species were ranked in terms of their abundance. The proportion of that collection year occupied by any given species was calculated and relativized between both years.

<u>Objective 2</u>

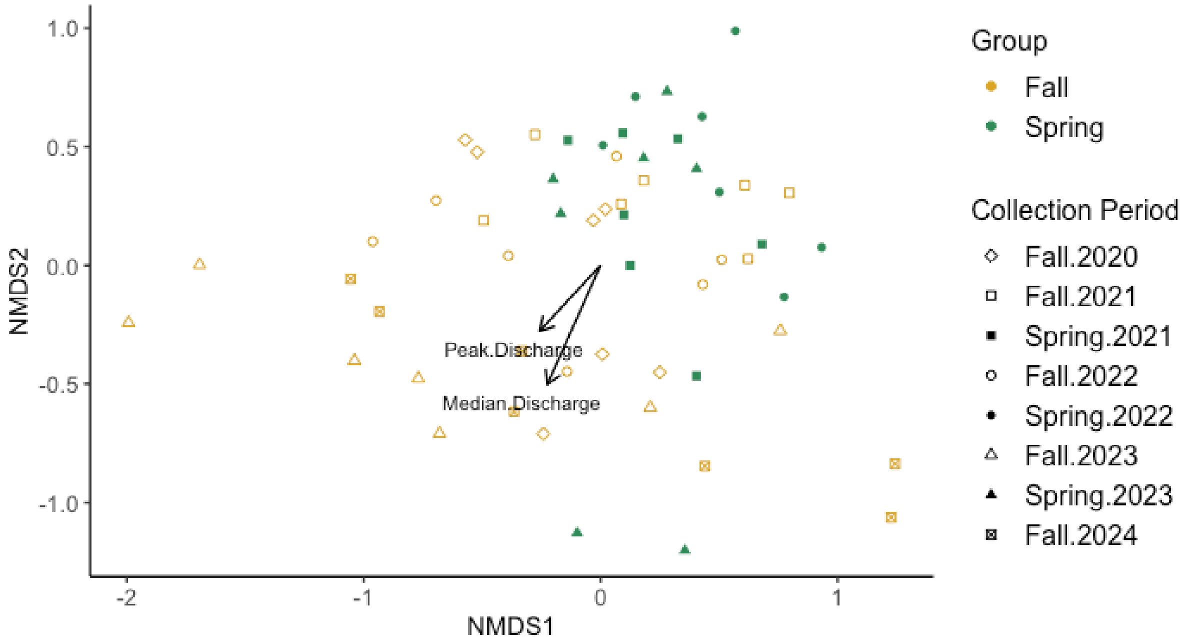
Naididae had the greatest correlation with spring collections while *Acentrella insignificans* and *Asioplax edmundsi* correlated more with fall collections (Figure 8.a). *Caecidotea sp.* contributed to both spring and fall communities. Percent clingers, percent EPT, and percent predators correlate more with fall 2022-2024 collections, with these years having more EPT taxa, clingers, and predators than other collections (Figure 8.b). There was a lesser, but significant correlation between spring collections and collector-gathers, with spring averaging more collector-gathers than in fall. Discharge tended to correlate more with fall collections than with spring collections, with median discharge having a slightly stronger correlation than peak discharge (Figure 8.c).



a. Species



b. Biotic Characteristics



c. Abiotic Factors

Figure 8. NMDS ordination plots of all collections based on Bray-Curtis's dissimilarity index with fitted vectors of species, biotic characteristics, and abiotic factors. Sites with points closer together are more similar in community composition. Vectors for species (a), biotic characteristics (b), and abiotic factors (c) have been fitted to these plots. Vector arrows represent the direction where ecological or environmental variables increase most rapidly, with longer arrows indicating stronger correlation with the ordination points. Only vectors with significant correlation with ordination points ($p<0.05$ all) were included on each figure.

Discussion

Seasonal Differences

The fall BMI community had greater richness than the spring BMI community collectively, with fall 2022-2024 having the greatest richness overall (Figure 7). The rank abundance graphs support this conclusion as well, with more species present in fall than in spring. Each fall collection had greater richness than its previous and/or subsequent spring collection with the exception of fall 2024 which was not preceded by a spring collection. Other studies demonstrate variability between whether spring or fall BMI communities in aquatic ecosystems tend to demonstrate increased richness (Baker et al., 2023; Frainer et al., 2013). In this portion of the Bighorn River, it appears as though the norm is for fall to have greater richness. Spring collections were more similar to each other while fall collections had more variation, which will be discussed in more detail below. Despite this, similarity was still lowest between seasons. Differences in community composition between seasons differed with regard to presence/absence of species and abundance of species (Table 2). Figure 8.c suggests that discharge has a stronger correlation with fall BMI communities than with spring communities.

One study exploring the seasonal drivers of BMI communities in streams found that in a stream environment experiencing wet/dry seasons the wet season was associated with increased flows, water temperature, suspended nutrients, and agriculture nutrient/organic run-off while exhibiting increased species and functional diversity (Feng et al., 2025). While the Lower Bighorn Watershed doesn’t experience seasons in the same fashion as Feng et al.’s study site, it does typically experience increased flows over the summer season (Figure 2) with active agricultural land use adjacent to the river. Experimental manipulation of in stream N and P additions have been shown to significantly impact the abundance of certain taxon and alter community composition (Demi et al., 2019). It is possible that these factors are impacting seasonal variation in Bighorn BMI, though more analysis explicitly incorporating in situ water quality metrics is needed for clarity on this topic.

The three years of data in this study showing higher BMI diversity in the fall is not sufficient to consider the pattern a long-term norm, but some observed patterns do align with anecdotal observations from fly fishers over the longer term (since the 1980s) when comparing the results of this study to fly-fishing guides from the past. For example, *Tricorythodes explicatus* was the fourth most abundant taxa across fall collections and fly fishers have known of the “maddening” “Trico mayfly activity” in late summer and early fall (Thomas, 2006, p.300). Additional alignment between study results and local common knowledge is that fly fishers on the Bighorn are known to take advantage of the spring midge hatch (Thomas, 2006) and this study found that *Parakiefferiella,* a type of midge, was the second most abundant species in spring collections. Compiling information from more of these data sets, presently collected data, and future collections may help us better differentiate between how BMI life histories and environmental factors drive the seasonal variation in the BMI community of the Bighorn River and if the observed patterns are exclusive to the study period or are an expected norm in this system. This may also allow us to assess whether ongoing climatic changes are shifting the expected BMI communities in the Bighorn.

Collection Differences

While some consistent seasonal patterns emerged across years there were notable differences observed in certain collections. Differences between species richness were consistent between season but no clear patterns in alpha diversity emerged between seasons. Fall and spring were the least similar (22.6%) in terms of beta diversity but the differences between fall collections were comparable (24.0%). This was primarily driven by the BMI communities from the fall 2023 and fall 2024 collections. Differences in alpha diversity suggest that Shannon diversity was lower in fall 2024. Collections from 2023 and 2024 had decreased evenness as measured by both Shannon and Simpson evenness. When refining the analysis to each collection, fall 2024 showed a marked decreased in evenness compared to fall 2020, spring 2021, and fall 2021. Fall 2023 and fall 2024 collections also displayed the least overlap with other collections (Figure 6.a) and contributed most to the spread of all fall collections seen in Figure 6.b. These collections also tended to have the lowest percent similarity to other collections (Table 3). When looking at the rank abundance plots (Figure 7), fall 2023 and fall 2024 had greater dominance by one species compared to all other collections. This species was *Caecidotea sp.* with 35.0% and 49.4% of the abundance in fall 2023 and fall 2024, respectively. The top-ranking species in all other collections only account for an average of 22.9% of the abundance. The dominance by *Caecidotea sp.* is likely contributing to the decreased evenness and changes in community composition observed in these collections.

The greatest peak in discharge occurred in June of 2023, prior to the shift towards a higher proportion of *Caecidotea sp*. and this peak in discharge is associated with the third greatest historic crest of the Bighorn River on record (National Oceanic and Atmospheric Administration, n.d.). *Caecidotea sp.* are a type of aquatic isopod, commonly referred to as a sowbug. *Caecidotea sp.* in the Bighorn may be either *Caecidotea communis* or *Caecidotea racovitzai* which are both known to be present in this area (Montana Natural Heritage Program, n.d.a.; Montana Natural Heritage Program, n.d.b.) and both species are likely to have been introduced from the eastern United States (Lewis, 2009). Aquatic sowbugs, like *Caecidotea sp.,* are known to fly fishers be more important for trout in this river in times of drought and longer periods without flushing flows (Thomas, 2006). Their ecology supports this, as *Caecidotea sp.* are a type of sprawler (National Science Foundation, n.d.), preferring depositional habitats with finer sediments and debris which typically get flushed away with substantial flows. Additionally, Figure 8.b suggest that the percent of clingers was greater and more correlated with fall 2023 and fall 2024, which may be due to their ability to cling more strongly to surfaces in flowing water (Merrit et al., 2019). As such, the increased contribution of *Caecidotea sp.*, a sprawler, to the BMI abundance after the increased discharge in 2023 was unexpected. It may have been expected then that *Caecidotea sp.* were flushed downstream and successfully colonized those sites, however over 95% of the *Caecidotea sp.* abundance contribution stems from the upper two sampling sites. There are several possible explanations for the increase of *Caecidotea sp.* in the fall of 2023 and 2024 though more research is needed to establish an exact cause.

Studies exploring the relationship between floods and BMI assemblages conclude that increased food availability in the form of organic debris post-flood may create conditions favorable to *Caecidotea sp.* or other collector guild feeding types (Chattopadhyay et al., 2021; Johnson & Colón-Gaud, 2013; Kerr, 1978). Chattopadhyay et al. (2021) also suggest that sediment deposition post-flood may support BMI recovery. However, much of the *Caecidotea sp.* abundance is located at the sites most near the dam, which traps sediment (Foster et al., 2024), and it’s uncertain whether much sedimentation occurred after the summer flows at these sites which could create suitable habitat for *Caecidotea sp*. It is also possible that because

Caecidotea sp. are known to be more tolerant of low dissolved oxygen conditions, which can be associated with flushing flows (Baran & Nasielski, 2011), that *Caecidotea sp.* in safe refugia were able to survive and act as a source to recover the population after the discharge decreased. However, Figure 8.b suggests that the percent of EPT taxa (which are highly sensitive to low DO (U.S. EPA, 2025b)) were greatest and most correlated with fall 2023 and 2024. When looking at the percent of EPT taxa at each site, the levels are higher at downstream collection sites and much lower (<10%) at upper collection sites where *Caecidotea sp.* were most abundant. Regardless of the cause, there is a clear shift in BMI diversity and community composition after the flushing flows of 2023 driven by an increase in *Caecidotea sp*. Future collections may indicate if this is a temporary disturbance or if the BMI assemblage returns to pre-fall 2023 conditions.

Figure 8.c suggests that peak discharge correlates more with the community structure of fall 2023 and 2024. Seasons and sites display clearer patterns in the NMDS figures than years, suggesting that observed differences in community composition have more to do with those factors than time with the exception of the increased variation between sites in the falls of 2023 and 2024. It is unclear if discharge itself or other associated factors are the cause of this shift in the BMI community of the Bighorn River. Additionally, only fall collections are available post summer 2023 so it's unknown how or if these flushing flows significantly impact the spring BMI community. More research is needed to understand the ecological factors that control fluxes in the diversity and community composition of BMI in this system. Studies further exploring BMI spatially and temporally may yield more insight into the observed phenomenon as sites vary greatly in their communities—as demonstrated by the difference in the percent of *Caecidotea sp.*

and EPT taxa observed in the upper and lower sample sites. Integrating these studies with water quality data (like temperature, pH, and DO), sediment surveys, and fish surveys may also provide insight on the primary drivers controlling community composition.

Implications for Stream Health

Benthic macroinvertebrates can provide insight to understanding stream health as they vary in their response to pollutants, oxygen, and other factors (Goodnight, 1973). The Bighorn River has abundant populations of *Caecidotea,* Naididae*,* and *Parakiefferiella.* Isopods, including *Caecidotea,* have long been associated with BMI communities in polluted streams as they are a tolerant group with a tolerance value (TV) around 8 (Kerr, 1976; National Science Foundation, n.d.). Other prevalent BMI fauna in the Bighorn include Naididae and *Parakiefferiella,* both of which also exhibit relatively high tolerance with TVs of 7 and 6, respectively. The remaining top species all have TVs ranging from 4.0 (fairly tolerant) to 7 (tolerant), suggesting that the Bighorn may be experiencing some level of organic pollution. Smalling et al. (2026) recently assessed two sites on the Bighorn River and found that they both had measurable levels of various organic pollutants near sites impacted by agricultural activity and wastewater, though none of these levels exceeded safety thresholds. Both of these sampling sites were in the upper portion of the Bighorn, south of Hardin, which are near those with higher proportions of *Caecidotea*. However, this doesn't necessarily represent the lower Bighorn that saw a recent increase in EPT taxa and was not assessed in the Smalling et al. (2026) study.

Stream health may be better assessed in a site-wise study, as suggested by the difference in the proportion of EPT and *Caecidotea sp*. between sites. The HBI scores don't show a strong difference between season, but it does correlate more with collections from before the 2023 flushing flows (Figure 8.b) at sites closer to the dam (Figure 9), suggesting higher HBI (more tolerant taxa present) scores prior to the flushing flows. However, when ranking each sample from greatest to lowest HBI score, the fall 2023 and 2024 samples extend to both ends of the spectrum, suggesting that a site-wise analysis may be more appropriate. HBI scores by site vary from 4.35-7.55, suggesting a wide range in organic pollution levels. EPT taxa show an inverse relationship with HBI and are more prevalent and correlated with sites further from the dam after the 2023 flushing flows. While a full site-wise assessment is not within the scope of this study, it may provide better insight into how BMI communities and stream health differ along the length of the Bighorn River in this watershed. This current analysis appears to suggest that overall stream health improved after the high discharge event of summer 2023 since the EPT taxa, which are more sensitive to pollution, increase with fall 2023 and fall 2024 collections while greater HBI scores were correlated with collections prior to that event but site-specific conditions may be skewing these results.

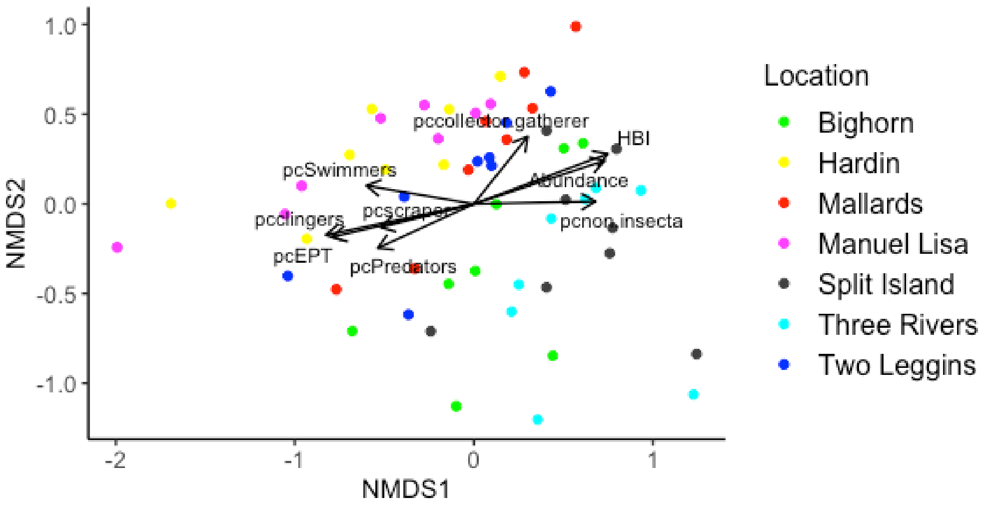


Figure 9. NMDS ordination plots of all collections based on Bray-Curtis's dissimilarity index displayed by site fitted with biotic characteristic vectors. Sites with points closer together are more similar in community composition. Vector arrows represent the direction where

characteristics increase most rapidly, with longer arrows indicating stronger correlation with the ordination points. Only vectors with significant correlation with ordination points ($p<0.05$ all) are included in the figure.

Limitations

This study was designed to broadly explore the taxonomic diversity of BMI in the Bighorn River from the fall of 2020 through the fall 2024 and there are several limitations worth discussing. The two primary objectives of this study were to determine if BMI diversity differed across the study period and to what extent environmental conditions are associated with any changes in BMI diversity. However, this study was not experimental therefor causation cannot be determined as to the cause of changes observed in BMI communities over time and it was difficult to tease out whether discharge, water quality conditions, or other confounding factors were the primary drivers behind observed shifts in diversity. The BHRA has been collecting water quality data from these sites, but this data was not incorporated into the study which further limited my ability to assess the extent to which environmental factors impact BMI communities. Incorporating chemistry data for future analysis could allow for more robust insight into objective 2. Furthermore, this study assesses BMI diversity by the collection period, rather than by site. However, clear trends were revealed in this study across sites and future studies assessing BMI diversity at the site level may be more informative and useful for targeted management efforts.

This study also focused on the Lower Bighorn Watershed, excluding the reservoir above Yellowtail Dam. In future studies, it may be worthwhile to compare the water quality data from BHRA and other groups from above the dam to water quality data below the dam to assess whether pollution or other factors that could alter BMI diversity are driven by processes within this watershed or from larger phenomena stemming from the Bighorn Basin. This study was also limited by the lack of data in spring 2020 and spring 2024, which limits the analysis for both objectives and hinders the ability to establish a baseline of what's expected in terms of seasonal difference in years of both high and low flows. The availability of long-term BMI data is lacking which limits our understanding on the ways in which the dam and how current changes in climate have impacted have altered BMI diversity, but this study creates a foundational assessment of existing data to inform future data collection and analysis. Finally, this study does not explicitly assess BMI functional diversity, which has been shown to be informative, and in some cases more insightful, than studies exclusively focusing on BMI taxanomic diversity.

Conclusion

The Bighorn River in the Lower Bighorn Watershed does demonstrate some degree of variability between seasons, with the fall season exhibiting greater richness than spring. Spring is marked by abundant Naididae*, Parakiefferiella spp.,* and *Baetis tricaudatus* while fall is more abundant with *Caeicidotea sp.,* Naididae*,* and *Cricotopus spp.* After the flushing flows in summer of 2023, the subsequent fall collections demonstrated reduced evenness, a decrease in similarity between community composition with previous collections, and greater dominance by a singular species: *Caecidotea sp.* The increase in *Caecidotea sp.* post flushing flows was unanticipated given the characteristics of this species and it's unclear whether discharge itself or other associated factors were driving this shift. However, the shift towards *Caecidotea sp.* was most prevalent in the upper collection sites while the lower collection sites had a marked increase in EPT taxa after the high-water event. Future site-wise analysis may provide more insight into the anticipated seasonal variation along the river's length while also allowing more refined insight as to how high discharge events may be impacting BMI across this system and how stream health varies along the length of the Bighorn River.

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