GEI Consultants

Consulting Engineers and Scientists

2021 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine

Lawrence County, South Dakota

April 2022

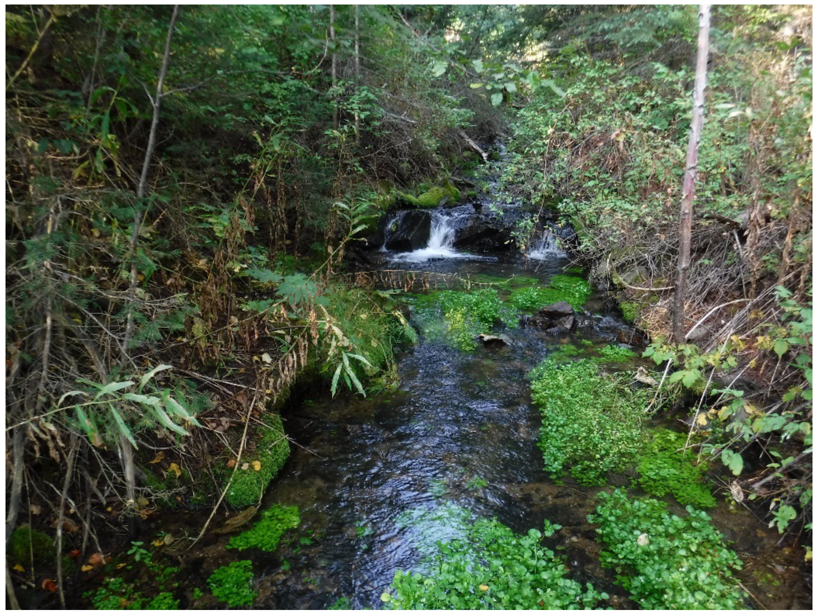
This page intentionally left blank

2021 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine

Lawrence County, South Dakota

GEI Consultants

Consulting Engineers and Scientists



Submitted to:
Coeur Wharf
10928 Wharf Road
Lead, SD 57754

Submitted by:

GEI Consultants, Inc.
4601 DTC Boulevard, Suite 900
Denver, CO 80237

April 2022

Craig Wolf

Craig Wolf, Project Manager

Jeniffer J. Lynch

Jeniffer Lynch, Reviewer

This page intentionally left blank

Table of Contents

Figures

Tables

Photos

Appendices

This page intentionally left blank

1. Introduction

The purpose of this study is to monitor aquatic habitat, fish, benthic macroinvertebrates, and periphyton in streams near the Wharf Mine in the northern Black Hills of South Dakota (Figure 1-1). The Wharf Mine study area includes sites in Annie Creek, Ross Valley, Lost Camp Gulch, Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek. The Golden Reward mine area includes sites in Fantail Creek, Nevada Gulch, and Stewart Gulch. This sampling is part of an annual monitoring of biological populations in these streams as they relate to current mining activities at Coeur Wharf, which includes the Golden Reward Mine, the Expansion Areas, and the Processing Facilities, as required in their National Pollutant Discharge Elimination System (NPDES) discharge permit. Current data are compared to corresponding reference sites, which are not affected by mining activities but may be influenced by historical and current human activities. Current data are also compared to data from previous years to evaluate possible relationships between the aquatic populations and mining activities.

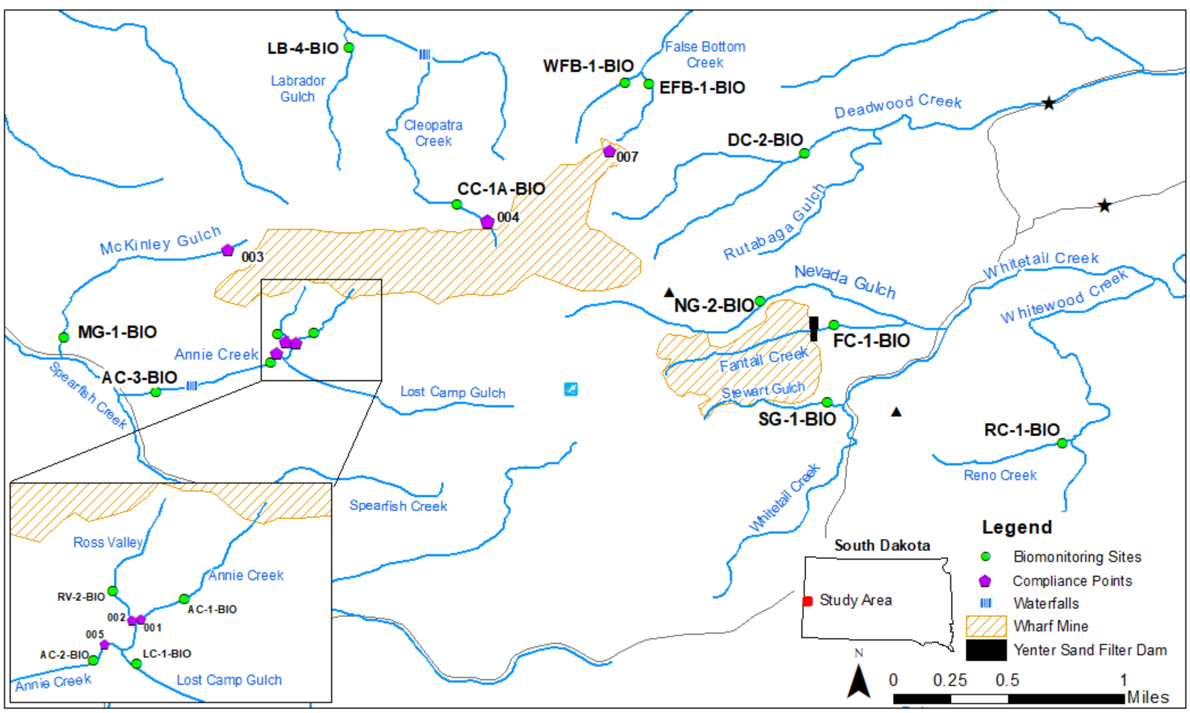


Figure 1-1: Aquatic biological monitoring sites on streams near the Wharf mine, Lead, South Dakota in August 2021.

This report presents results from the annual aquatic biological monitoring conducted in August 2021 by GEI Consultants, Inc. (GEI, formerly Chadwick Ecological Consultants, Inc. [CEC]), in Annie Creek, Ross Valley, Lost Camp Gulch, Deadwood Creek, False Bottom Creek, McKinley Gulch, Cleopatra Creek, Fantail Creek, Nevada Gulch, Stewart Gulch, Reno Creek, and Labrador Gulch. Sites on Labrador Gulch and Reno Creek serve as reference sites for comparison to sites downgradient of mining activities. Biological parameters evaluated in these streams in August 2021 included fish, benthic macroinvertebrates, and periphyton populations. Aquatic habitat parameters were also measured and summarized.

1.1 Monitoring History

1.1.1 Labrador Gulch

Labrador Gulch was added to the Wharf Sampling and Analysis Plan (SAP) in 2018 and has been part of the Richmond Hill Mine's monitoring program since 1993. Labrador Gulch does not receive any mine discharge and has served as a reference site (with fish) for the Richmond Hill Mine. This stream has a cascading waterfall immediately upstream of the confluence with Cleopatra Creek that isolates the brook trout population from downstream populations. The fish habitat is relatively complex with a variety of small scour pools, low and high gradient riffles, runs, and cascading habitats that support the self-sustaining brook trout population. From 1995 to 2003, habitat and macroinvertebrate sampling occurred on an annual basis, including fish sampling in most years (Table 1-1). Since 2005, habitat, benthic macroinvertebrate, and periphyton monitoring occurred in all years, while fish populations were only monitored on a 3-year cycle until 2017 when sampling became annual.

1.1.2 Reno Creek

In 2017, a site on Reno Creek was added as a reference site for the project. Personnel from GEI, South Dakota Game, Fish, and Parks (SDGFP), and South Dakota Department of Agriculture and Natural Resources (SDDANR) collaborated on site selection and location. This site has no known influence from past historical mining activities or past human impacts within its drainage, although the Mickelson Trail and a water supply pipe for the town of Lead cross Reno Creek at two locations. The upper part of the watershed contains the Powder House Pass subdivision which is a large lot residential district with a permitted (SD0028615) onsite wastewater treatment system. A review of available discharge monitoring reports indicates limited numeric violations (2018 and 2019) of the total ammonia nitrogen permit limit, although each exceedance was resolved by post event compliance. There is evidence of past forest management activities (i.e., slash piles) by the United States Forest Service (USFS) in the area, and in July 2020, extensive treefall damage occurred during a storm event. As a result, the site was moved 21 meters (m) upstream from the original downstream boundary to avoid a deadfall tree. Wharf will continue to sample Site RC-1-BIO on a yearly basis (Table 1-1).

Table 1-1: Aquatic biological monitoring summary for sites on Labrador Gulch, Reno Creek, Annie Creek, Ross Valley, Lost Camp Gulch, Deadwood Creek, False Bottom Creek, Cleopatra Creek, Fantail Creek, Nevada Gulch, Stewart Gulch, and Whitetail Creek from 1986 through 2021. Parameters: H = habitat, F = fish populations, B = benthic macroinvertebrate populations, and P = periphyton populations.

Date	Labrador Gulch Reference	Reno Creek Reference	Annie Creek			Ross Valley	Lost Camp Gulch	Deadwood Creek		False Bottom Creek		Cleopatra Creek	Fantail Creek	Upper Nevada Gulch	Nevada Gulch	Stewart Gulch	Whitetail Creek
	LB-4-BIO-BIO	RC-1-BIO	AC-1-BIO	AC-2-BIO	AC-3-BIO	RV-2-BIO	LC-1-BIO	DC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO	CC-1A-BIO	FC-1-BIO	NG-1-BIO	NG-2-BIO	SG-1-BIO	--
1986	--	--	--	--	--	--	--	--	--	--	--	--	H, F, B, P	--	--	H, F, B, P	--
1987	--	--	--	--	--	--	--	--	--	--	--	--	B, P	H, F, B, P	H, F, B, P	B, P	H, F, B, P
1988	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--	--
1989	--	--	--	--	--	--	--	--	--	--	--	--	H, B, P	H, B, P	H, B, P	H, B, P	H, B, P
1990	--	--	H, B	H, F, B	--	--	--	--	--	--	--	--	--	--	--	--	--
1991	--	--	--	B, P	--	--	--	--	--	--	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
1992	--	--	F, B, P	F, B, P	H, F, B	--	--	--	--	--	--	--	B, P	B, P	B, P	B, P	B, P
1993	F	--	H, B, P	H, B, P	--	--	--	--	--	--	--	--	B, P	B, P	B, P	B, P	B, P
1994	F	--	B, P	B, P	--	--	--	--	--	--	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
1995	H, F, B	--	H, F, B, P	H, F, B, P	F, B	--	--	H, F, B, P	--	H, F, B, P	--	--	B, P	B, P	B, P	B, P	B, P
1996	H, F, B	--	B, P	B, P	--	--	--	H, F, B, P	--	H, F, B, P	--	--	B, P	B, P	B, P	B, P	B, P
1997	H, F, B	--	B, P	B, P	--	--	--	B, P	--	B, P	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
1998	H, F, B	--	H, F, B, P	H, F, B, P	--	--	--	H, F, B, P	--	H, F, B, P	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
1999	H, B	--	B, P	B, P	--	--	--	B, P	--	B, P	--	--	B, P	B, P	B, P	B, P	B, P
2000	H, F, B	--	H, B, P	H, B, P	H, B, P	--	--	Dry	H, B, P	H, B, P	--	--	B, P	B, P	B, P	B, P	B, P
2001	H, B	--	H, F, B, P	H, F, B, P	H, F, B, P	--	--	Dry	H, F, B, P	H, F, B, P	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
2002	H, F, B	--	H, F, B, P	H, F, B, P	H, F, B, P	--	--	Dry	H, F, B, P	H, F, B, P	--	--	B, P	B, P	B, P	B, P	B, P
2003	H, B	--	H, F, B, P	H, F, B, P	H, F, B, P	--	--	Dry	H, F, B, P	H, F, B, P	--	--	B, P	B, P	B, P	B, P	B, P

Date	Labrador Gulch Reference	Reno Creek Reference	Annie Creek			Ross Valley	Lost Camp Gulch	Deadwood Creek		False Bottom Creek		Cleopatra Creek	Fantail Creek	Upper Nevada Gulch	Nevada Gulch	Stewart Gulch	Whitetail Creek
	LB-4-BIO-BIO	RC-1-BIO	AC-1-BIO	AC-2-BIO	AC-3-BIO	RV-2-BIO	LC-1-BIO	DC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO	CC-1A-BIO	FC-1-BIO	NG-1-BIO	NG-2-BIO	SG-1-BIO	--
2004	--	--	H, F, B, P	H, F, B, P	H, F, B, P	--	--	Dry	H, F, B, P	H, F, B, P	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
2005	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	--	Dry	H, F, B, P	H, F, B, P	--	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
2006	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	Dry	--	H, F, B, P	--	H, F, B, P	B, P	--	B, P	B, P	B, P
2007	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	Dry	--	--	H, F, B, P	H, F, B, P	B, P	--	B, P	B, P	B, P
2008	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	Dry	--	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2009	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	--	H, F, B, P	B, P	--	B, P	B, P	B, P
2010	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P
2011	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2012	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2013	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2014	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2015	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2016	H, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	DRY	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2017	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	--	H, F, B, P	DRY	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P
2018	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	--
2019	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	DRY	H, F, B, P	--	H, F, B, P	H, F, B, P	--
2020	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	F*	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	DRY	H, F, B, P	--	H, F, B, P	H, F, B, P	--
2021	H, F, B, P	H, F, B, P	H, F, B, P	H, F, B, P	F*	H, F, B, P	H, F, B, P	--	H, F, B, P	H, F, B, P	H, F, B, P	DRY	H, F, B, P	--	H, F, B, P	H, F, B, P	--

*Only qualitative sampling in portions of creek due to tornado deadfall limiting access.

1.1.3 Annie Creek, Ross Valley, McKinley Gulch, and Lost Camp Gulch

Limited sampling began on Annie Creek in 1990 (Table 1-1). In 1992, habitat was measured, and fish and benthic macroinvertebrates were extensively sampled for the Annie Creek/Reliance Tailings Project (Chadwick & Associates, Inc. [C&A] 1993). This sampling provided data on the existing fish populations at six sites in Annie Creek. Brook Trout (*Salvelinus fontinalis*) and Brown Trout (*Salmo trutta*) were limited to the lower portion of Annie Creek, just upstream of the confluence with Spearfish Creek and downstream of the falls on Annie Creek (Figure 1-1). The continued absence of trout in the upper portion is due to the falls, which act as a barrier to upstream fish movement.

Historically, a population of Mountain Suckers (*Catostomus platyrhynchus*) was found in Annie Creek at monitoring Site AC-2-BIO. In 1990 and 1992, Mountain Suckers were abundant at Site AC-2-BIO, with 380 and 127 collected, respectively, and multiple age classes present (C&A 1993). Density estimates during these years were over 15,000 fish per hectare (C&A 1993). A few individuals were also found in Annie Creek above the confluence with Lost Camp Gulch and in the vicinity of the confluence with Ross Valley, but none were found further upstream at Site AC-1-BIO or within Lost Camp Gulch (C&A 1993). Electrofishing at two locations on Annie Creek further downstream of Site AC-2-BIO also found abundant Mountain Suckers, with high numbers collected in the pool immediately below the culvert that passes under Annie Creek Road as well as the pool below Annie Creek Falls (C&A 1993; CEC 2001). In 1995, habitat measurements indicated that a 100-year flood event had altered Annie Creek by increasing channel widths and causing erosion of the banks (CEC 1996b).

In 1995, an ammonia and cyanide release occurred via Ross Valley into Annie Creek, and Mountain Sucker and macroinvertebrate numbers were reduced (CEC 1996a, 1996b). Only 3 live Mountain Suckers (density estimate: 102 Mountain Suckers per ha) were collected in Site AC-2-BIO, and 68 dead Mountain Suckers were observed (CEC 1996a, 1996b). Complete details of the effects of this event and sampling for recovery patterns were presented previously (CEC 1996c). Macroinvertebrate monitoring from 1996 through 2001 indicated that the benthic community had recovered from the 1995 release (CEC 1997a, 1998a, 1999a, 2000a, 2001a, and 2002a). However, no Mountain Suckers were found during the 1998 and 1999 surveys, indicating that populations had still not recovered from the ammonia and cyanide release in 1995 (CEC 2001). Elevated ammonia levels were also recorded in February 2002 and April 2004. From 2001 to 2006, density estimates at Site AC-2-BIO ranged from 818 Mountain Suckers per hectare to 500 Mountain Suckers per hectare, with a small population persisting within the site (CEC 2002, 2003, 2004, 2005, 2006, and 2007). During these years, annual electrofishing surveys found from 8 to 18 Mountain Suckers within Site AC-2-BIO.

High biological oxygen demand (BOD) water that exceeded standards was inadvertently released into upper Annie Creek in 2007 by Wharf Resources, Inc. In addition, ammonia and cyanide standards were exceeded downstream of mining activities in Annie Creek in 2007 (GEI 2008b). Biomass accumulations were observed during annual monitoring on Annie Creek in August 2007 by GEI personnel (GEI 2008a). Biomass accumulations on the stream bottom were also observed by SDDANR personnel during a site inspection on November 27, 2007. Benthic invertebrate communities in middle and upper Annie Creek appeared stressed in August 2007, typical of communities tolerant of low dissolved oxygen (GEI 2008a). Furthermore, no Mountain Suckers were collected from Site AC-2-BIO, where a population had existed in past years (GEI 2008a). On April 8, 2008, Wharf Resources, Inc received a violation of their mining permit from SDDANR.

As a result of the absence of Mountain Suckers in August 2007 and the failure to meet water quality permit limits, Wharf Resources, Inc. was ordered to clean up the biomass accumulations by August 1, 2008, in an amended order for the violations of the surface water discharge and mining permit (GEI 2008b). The clean-up effort was conducted on July 15 and 16, 2008 and supervised by GEI personnel. The clean-up process included the use of a vacuum truck to collect the biomass and affected sediments from the surface of the riparian areas and the streambed in Annie Creek.

Sampling of aquatic biological populations in 2008 was conducted in June, prior to clean-up activities, and in August, after clean-up activities. Overall, annual monitoring data collected in August 2008 indicated that Site AC-1-BIO was not fully recovered from the release of high BOD water into Annie Creek, while Site AC-2-BIO appeared largely recovered when compared to data from August 2007. Four Mountain Suckers were collected during June 2008, and two were collected in August 2008, with a density estimates of 182 and 71 Mountain Suckers per hectare at Site AC-2-BIO during these two surveys, respectively. During the August 2009 survey, no Mountain Suckers were collected (GEI 2010). One Mountain Sucker was collected during the 2010 survey (GEI 2011), and since 2011, no Mountain Suckers have been observed in the upper portion of Annie Creek or its tributaries (GEI 2012, 2013, 2014, 2015, 2016, 2017, 2018, 2019, 2020, and 2021). During the water quality upset in upper Annie Creek, Site AC-3-BIO appeared healthy in 2008, similar to 2007 conditions. Continued monitoring has shown improvement in the aquatic habitat and benthic macroinvertebrate conditions at sites AC-1-BIO and AC-2-BIO since 2007.

Data were collected on Annie Creek by C&A in 1993 and 1994, by CEC in 1995 through 2005, and by GEI in 2006 through 2021. Sampling has usually been conducted in late August or early September. Benthic macroinvertebrate and periphyton data have been collected nearly every year and habitat and fish population data were collected every two to three years from 1990 through 2000 based on SDGFP wildlife monitoring guidelines. Habitat measurements and fish, benthic macroinvertebrate, and periphyton populations were sampled at Annie Creek sites from 2001 to 2021. At Site AC-1-BIO, data have been collected in every year since 1990 except for 1991 (Mariah Associates, Inc. 1990, 1992a, and 1992b; C&A 1993, 1994a, and 1995a; CEC 1996b, 1997a, 1998a, 1999a, 2000a, 2001a, 2002a, 2003a, 2004a, 2005a, and 2006a; GEI 2007a, 2008a, 2009a, 2010a, 2011a, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). In terms of data continuity, Site AC-2-BIO and Site AC-2, established in 1990 and sampled through 1999, are considered the same site due to their proximity. When data are combined for these two sites, this reach of Annie Creek has been sampled every year since 1990. Site AC-3-BIO was not sampled as part of the monitoring study prior to 2000. However, at this site (formerly Site AC-6), habitat measurements, and fish and benthic invertebrates were sampled as part of another study in June and October 1992 (C&A 1993) and data are used for comparison with Site AC-3-BIO. Additionally, fish and benthic macroinvertebrates were sampled in 1995 at this site as part of the investigation to determine the effects of an accidental ammonia and cyanide release into Annie Creek in that year and are used as long-term comparison data. In 2020 and 2021, access to Site AC-3-BIO for routine sampling was extremely limited due to the extensive tree deadfall covering the creek caused by the July 8, 2020 tornado activity. Only qualitative fish sampling occurred in these years and other biomonitoring tasks could not be performed.

In 2004, two temporary sampling locations were established on Ross Valley in response to the release of ammonia (CEC 2005a). Following the initial investigation, both sites were abandoned, and in 2006, Site RV-2-BIO was established as the permanent biomonitoring location on Ross Valley which has maintained perennial flows. McKinley Gulch was also included in the Wharf monitoring program in 2006 to address the monitoring needs downstream of compliance point 003. However, a physical location for this site has not been established due to the ephemeral nature of this stream. Site RV-2-BIO has been sampled for fish, benthic invertebrates, periphyton, and habitat every year since 2006 (Table 1-1).

Lost Camp Gulch was included in the Wharf monitoring program in 2010 (Table 1-1), as part of the supplementary sampling necessary for the potential mine expansion. Specifically, this site was used to establish baseline aquatic resource conditions in a basin before mine expansion occurred, per the requirements of the SDDANR. The headwaters are located near Terry Peak, and there is a residential sub-division located in a sub-basin that drains into Lost Camp Gulch. In the past, the Lost Camp Gulch sub-basin receives heavy recreational ATV traffic, and the trail adjacent to the stream influences sediment conditions in both Lost Camp Gulch and Annie Creek, although recent forest management efforts by the USFS have limited access to the Annie Creek Basin. Habitat measurements and fish, benthic macroinvertebrate, and periphyton populations have been sampled at the Lost Camp Gulch site from 2010 to 2021.

The McKinley Gulch site has never been formally established but the channel is visited each year at the culvert crossing on Highway 14 to determine the presence of flowing water. Similarly, the compliance point in McKinley Gulch is located immediately downgradient of a lined storage pond which has no direct discharge to the channel. However, in extremely wet years both storage ponds receive surface water runoff that may exceed the holding capacity of the pond, allowing surface water and contaminants to flow downgradient in the channel.

Five Brook Trout were collected at Site AC-3-BIO for selenium whole-body tissue analysis in 2011 to 2019. The fish population was not sampled in 2020 due to extensive deadfall covering the stream caused by the July 2020 tornado activity. As a result, no fish could be collected for whole-body tissue selenium analysis. Fish populations in the creek could not be assessed again in 2021. However, five Brown Trout were collected in accessible pools and analyzed.

1.1.4 Deadwood Creek and False Bottom Creek

In 1995, Wharf Resources initiated a baseline aquatic biological monitoring study for upper Deadwood Creek and False Bottom Creek in anticipation of a possible mine expansion (CEC 1996a). This study evaluated the existing status of aquatic habitat, fish, benthic macroinvertebrate, and periphyton populations in these streams. The Wharf Resources expansion was approved in June 1998. The Expansion Area is located on the east side of Foley Ridge and is west and southwest of the Deadwood and False Bottom drainages (Figure 1-1). Development of the Portland pit began in March 1999, and development of the Trojan pit began in 2000.

Data collected in 1995 (Site DC-1-BIO) indicated that aquatic habitat in upper Deadwood Creek was very limited due to low, interrupted flows (Table 1-1; CEC 1996a). No fish were found in this upper section of the stream, but benthic macroinvertebrate communities appeared healthy and included species considered sensitive to pollutants. Periphyton communities were similar to those found in other streams in the area. Results from additional baseline sampling from 1996 through 2001 also indicated limited aquatic habitat and no fish, but healthy benthic macroinvertebrate and periphyton populations. The original site on upper Deadwood Creek was dry most years since 2000, but a small amount of water was present in August 2009 and 2010. An additional site (Site DC-2-BIO) further downstream was added in 2000 to provide a site with perennial flow and fish populations (CEC 1997b, 1998c, 1999c, 2000a, 2001a, 2002a, 2003a, 2004a, 2005a, and 2006a). This additional site was sampled from 2000 through 2005 but was discontinued from the monitoring plan from 2006 to 2009 because sampling of the benthic macroinvertebrate and periphyton populations still occurred further upstream on Deadwood Creek when practicable. Monitoring habitat and fish, benthic macroinvertebrates, and periphyton populations began again at Site DC-2-BIO in 2010 as part of the Wharf expansion permit. Monitoring at this site has continued on a yearly basis given the perennial flow which supports a resident Brook Trout population.

In 1995, two sites were sampled on False Bottom Creek. One was located upstream of the confluence on East Fork False Bottom Creek (formerly FB-1; currently EFB-1-BIO), and one was located immediately downstream of the confluence with West Fork False Bottom Creek (historical Site FB-2). Aquatic habitat in 1995 at two sites on False Bottom Creek supported fish, and healthy populations of Brook Trout were found at both study sites. Benthic macroinvertebrate and periphyton populations were also healthy in False Bottom Creek in 1995 (CEC 1996a). Additional sampling from 1996 through 2000 provided similar conclusions

(CEC 1997b, 1998c, 1999c, 2000a, and 2001a). Sampling was reduced to one site on the East Fork False Bottom Creek (FB-1-BIO) in 2000 because of the similarity between the two sites (CEC 2002a, 2003a).

In August 2017, during a site visit to False Bottom Creek, SDGFP raised the issue that the current biological monitoring location was not in the original location as established in 1995. In 2017 a new rebar sign was posted on the West Fork identifying the water quality monitoring location as Site FB-2, which caused further confusion. In recent years, the biological monitoring occurred on the West Fork which is not consistent with past Site FB-1-BIO conditions.

These observations precipitated a Root Cause Analysis (RCA) to identify the root causes of the event or decision-making process that resulted in this error and to develop corrective actions. Wharf and GEI personnel concluded that Site FB-1-BIO was relocated to the West Fork rather than the East Fork and developed corrective actions. The factors contributing to the changes in site location in each year are discussed in detail in a memo written by GEI (2018a). In summary, East Fork False Bottom Creek (EFB) was sampled in 2000 through 2006, 2009, and 2010, while West Fork False Bottom Creek (WFB) was sampled in 2007, 2008, and 2011 through 2017. Both sites have been sampled since 2018.

Five Brook Trout were collected at Site EFB-1-BIO for selenium whole-body tissue analysis in 2021.

1.1.5 Cleopatra Creek

Sampling on Cleopatra Creek began in 1985. In 1991, as per an agreement between LAC Minerals, Wharf Resources, and SDGFP, LAC Minerals and Wharf Resources shared an aquatic sampling location, designated Site CC-1. Previous designations for this site have been SQ-1 (Knudson 2003) and SQ-1-BIO (CEC 2000c). Sampling on this stream had previously been conducted by OEA Research, Inc. and KNK Aquatic Ecology (Knudson 2003). In 2006, the Wharf monitoring site on Cleopatra Creek was moved upstream of its former location to the headwaters of Cleopatra Creek (Figure 1-1) between Monitoring Well 41 and Compliance Point 004 and designated as Site CC-1A-BIO (GEI 2007a). When sufficient surface water present, the 2006 through 2015 and 2018 monitoring results are used to evaluate the status of the aquatic biological populations in Cleopatra Creek in relation to ongoing mine operations (Table 1-1; GEI 2007a, 2008a, 2009a, 2010a, 2011a, 2012, 2013, 2014, 2015, 2016 and 2019). However, Cleopatra Creek was dry in five out of the last six years which has limited the suitability of Site CC-1A-BIO as a biomonitoring site (GEI 2017a, 2018a, 2020, and 2021).

1.1.6 Fantail Creek, Nevada Gulch, and Stewart Gulch

Sampling of these streams is a continuation of a long-term aquatic biological monitoring program that was initiated in 1986 to collect baseline data on the aquatic resources of Fantail Creek and Stewart Gulch prior to establishment of the Golden Reward gold mine, in compliance with South Dakota Mined Land Reclamation Regulations (Golden Reward Mining Company [GRMC] 1987). Nevada Gulch was added to the scope of the monitoring program in 1987. Mining commenced in 1989 and continued until 1996. GRMC received approval for temporary cessation of mining in 1996 and remained in temporary cessation until the end of 2001. GRMC entered final reclamation and reclaimed approximately 189 acres from April through November 2002. All but 5.23 acres of the total area of disturbed land (approximately 403 acres) has been reclaimed since 2002. In January 2009, the South Dakota Board of Minerals and Environment approved the reclamation and placed the site into Post Mine Closure and Monitoring status.

The 1986 and 1987 surveys provided baseline data on habitat, benthic macroinvertebrates, fish, and periphyton for these streams prior to operation of the gold mine. Streams were then surveyed semiregularly from 1989 to 2010 and annually through 2021 (Table 1-1; GRMC 1990, 1992, and 1993; Chadwick & Associates, Inc. 1994b and 1995b; Chadwick Ecological Consultants, Inc. 1996d, 1997c, 1998b, 1999b, 2000b, 2001b, 2002b, 2003b, 2004b, 2005b, and 2006b; GEI 2007b, 2008c, 2009b, 2010b, 2011b, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). This schedule was revised in 1998 to synchronize sampling with the schedule for the Wharf Mine. Sampling followed the activities and methods outlined in the most recent Sampling and Analysis Plan (GEI 2018b).

The historical study site on Fantail Creek was near the confluence with Nevada Gulch, approximately 1.0 km downstream of the Yenter Sand Filter Dam. Construction of the filter dam began in 2002 due to sedimentation concerns from the reclaimed mine site, GRMC installed a sand filter dam in Fantail Creek. Rock was added to the face of the dam in August 2003 to increase the area of sediment filtration (Kim Schultz, personal communication). In 2005, at the request of SDGFP, the Fantail study site was moved upstream so that the top of the study reach was at the filter dam outfall. In 2008, the Gilded Mountain Road was constructed over Fantail Creek in the study reach, approximately 70 m downstream from the top of the site. The site was moved downstream of the road crossing with the top of the site just downstream of the road culvert.

The study site on Nevada Gulch (Lower Nevada Gulch) replaced a site further upstream (Upper Nevada Gulch) and served as the background control site from 2006 through 2016 (CEC 2006d). The site runs directly alongside the paved Nevada Gulch Road. From 2017 to 2021, the Lower Nevada Gulch site (Site NG-2-BIO) was sampled but is no longer considered a background control site due to mining activities occurring upstream in the watershed.

The Stewart Gulch site is located near the confluence with Whitetail Creek downstream of Reno Creek, Nevada Gulch, and Fantail Creek. The site is near Highway 14 and contains historic flow control structures and a modern stream gage. A large portion of the flow in Stewart Gulch comes from an adit located in abandoned mine workings in Whitetail Creek.

This page intentionally left blank

2. Study Area

Currently, there are fifteen monitoring sites in the Wharf study area (Table 2-1). Other monitoring sites have existed but were either moved or are no longer included in the monitoring program. All sites in the Wharf Mine area are located in Black Hills Core Highlands of the Middle Rockies Ecoregion (Omernik 1987; Omernik and Gallant 1987).

Table 2-1: GPS coordinates for Wharf sites.

Stream/Site	Latitude and longitude	Elevation (m)
Reference Sites		
Labrador Gulch, LB-4-BIO	44°22.523’ -103°51.893’	1,683
Reno Creek, RC-1-BIO	44°19.267’ -103°46.264’	1,647
Mining Activity Sites		
Annie Creek, Site AC-1-BIO	44°20.245' -103°52.058'	1,762
Annie Creek, Site AC-2-BIO	44°19.951' -103°52.420'	1,691
Annie Creek, Site AC-3-BIO	44°19.642' -103°53.628'	1,576
Ross Valley, Site RV-2-BIO	44°20.088' -103°52.380'	1,730
Lost Camp Gulch, Site LC-1-BIO	44°19.921' -103°52.381'	1,698
Deadwood Creek, Site DC-2-BIO	44º21.587' -103º48.258'	1,623
East Fork False Bottom Creek, Site EFB-1-BIO	44°22.207' -103°49.558'	1,673
West Fork False Bottom Creek, Site WFB-1-BIO	44°22.205' -103°49.574'	1,677
McKinley Gulch, MG-1-BIO	Not established, approx. 44°20.073' -103°54.162	1,593
Cleopatra Creek, Site CC-1A-BIO	44°21.161' -103°51.106'	1,808
Fantail Creek, FC-1-BIO	44°20.205' -103°48.028'	1,684
Nevada Gulch, NG-2-BIO	44°20.432' -103°48.564'	1,726
Stewart Gulch, SG-1-BIO	44°19.576' -103°47.984'	1,695

2.1 Reference Sites

The Wharf Mine is located in the headwaters of many drainages, therefore, all monitoring sites for this project are located downgradient of mining activities. As a result, it is not possible to establish upstream reference or control sites to evaluate possible impacts from mining activities. In 2017, the sites on Whitetail Creek and Nevada Gulch were discontinued as background control sites for aquatic life use assessment and two new reference site locations were established on Labrador Gulch and Reno Creek with the assistance from SDGFP and SDDANR. These sites are located in adjacent drainage basins and are intended to be used as a tool to evaluate whether patterns in the data downstream of the mining areas reveal similar patterns to the reference sites. These reference sites should also help tease out the effects of regional climatic conditions from other patterns in the data downgradient of the Wharf's influence.

2.1.1 Labrador Gulch

The headwaters of Labrador Gulch are located approximately 8.7 kilometers (km) west of Lead, South Dakota, and flow northeast into Cleopatra Creek downstream of Site CC-1A-BIO. Labrador Gulch contains a resident trout population for comparison to larger streams that contain fish populations (lower Annie Creek, Deadwood Creek, False Bottom Creek, and Stewart Gulch). Labrador Gulch is not currently classified in the administrative rules, although the reach of Cleopatra Creek that it enters is classified with the standard beneficial uses and coldwater permanent fish life propagation water, immersion recreation water, and limited contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site LB-4-BIO: This site corresponds to Richmond Hill Mine designation LB-4, although the acronym BIO has been added for consistency with naming conventions used for the Wharf Mine. The bottom of the site is located approximately 25 m upstream from the confluence with Cleopatra Creek, above a small cascade that is a barrier to upstream movement by fish, at an elevation of 1,683 m above sea level.

2.1.2 Reno Creek

Reno Creek is located 3.3 km south of Lead, South Dakota and is used for comparison with the smaller (narrower) streams that do not typically support fish populations (upper Annie Creek, Ross Valley, Lost Camp Gulch, McKinley Gulch, Cleopatra Creek, and Fantail Creek sites). This site serves as a baseline representation of relatively non-impacted conditions for a small, headwater stream in the Black Hills. Reno Creek is a tributary to Whitewood Creek and is classified with the standard beneficial uses and coldwater permanent fish life propagation water, immersion recreation water, and limited contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site RC-1-BIO: This site is located approximately 170 m upstream of the confluence with Whitewood Creek, at an elevation of 1,647 m above sea level.

2.2 Mining Activity Sites

2.2.1 Annie Creek

Annie Creek is a tributary to Spearfish Creek in the Black Hills National Forest, approximately 5.5 km west of Lead, South Dakota. As part of the annual biological monitoring activities in 2021, three study sites on Annie Creek were sampled (Figure 1-1,

Table 1-1). The study sites were renamed in 2000 with site names corresponding to the Aquatic Biological Monitoring Plan (CEC 2000c), as was required by the renewal of the Wharf Resources Surface Water Discharge Permit in 1999. The study sites and corresponding site names from previous monitoring efforts are described below.

Site AC-1-BIO: This site is located near the headwaters of Annie Creek, at an elevation of 1,762 m above sea level, approximately 0.5 km upstream of Ross Valley and 0.7 km upstream from Lost Camp Gulch. Site AC-1-BIO is downstream of Outfall 006A and upstream of Compliance Points 001 and 005.

Site AC-2-BIO: This site corresponds to Site AC-4, historically, and is located approximately 0.4 km downstream of Ross Valley, at an elevation of 1,691 m above sea level, just downstream of the confluence with Lost Camp Gulch and upstream of the falls. Site AC-2-BIO is approximately 0.5 km downstream of the original monitoring Site AC-2. This site is downstream of Outfall 006A, and Compliance Point 001. This site is upstream of Compliance Point 005.

Site AC-3-BIO: This site corresponds to the historical Site AC-6 and is located approximately 0.2 km upstream of the confluence with Spearfish Creek at an elevation of 1,576 m above sea level. This site is downstream of the falls and provides information on the lower reach of Annie Creek. This site is downstream of Outfall 006A and Compliance Points 001 and 005.

The upper portion of Annie Creek, including the reaches where sites AC-1-BIO and AC-2-BIO are located, is classified with the standard beneficial uses that are applied to all streams by the SDDANR. These uses are irrigation, fish and wildlife propagation, recreation, and stock watering (SDDANR 2021, Administrative Rules, Chapter 74:51:03:01). Lower Annie Creek from the confluence with Spearfish Creek upstream to Township 4N, Range 2E, Section 3 (T4N, R2E, S3), which includes Site AC-3-BIO, has two additional beneficial uses: coldwater marginal fish life propagation waters and limited-contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

2.2.2 Ross Valley

The headwaters of Ross Valley are located approximately 7.7 km west of Lead, South Dakota (Figure 1-1), at an elevation of 1,805 m above sea level. The stream flows from the Ross Valley Ore Depository and water treatment pond at the head of the valley and enters Annie Creek between sites AC-1-BIO and AC-2-BIO. Ross Valley Creek is classified with the standard beneficial uses that are applied to all streams by the SDDANR. These uses are irrigation, fish and wildlife propagation, recreation, and stock watering (SDDANR 2021, Administrative Rules, Chapter 74:51:03:01).

Site RV-2-BIO: This site is approximately 200 m upstream of the confluence with Annie Creek and Compliance Point 002 at an elevation of 1,740 m. Historically, Outfall 006B was located in the headwaters of this basin, but functionally ceased operation when changes to the water treatment systems were made in both Ross Valley and Annie Creek.

2.2.3 Lost Camp Gulch

The headwaters of Lost Camp Gulch are located approximately 7 km west of Lead, South Dakota (Figure 1-1), at an elevation of approximately 1,747 m above sea level. The stream enters Annie Creek from the east between sites AC-1-BIO and AC-2-BIO just downstream of the Ross Valley confluence. Lost Camp Gulch is classified with the standard beneficial uses that are applied to all streams by the SDDANR. These uses are irrigation, fish and wildlife propagation, recreation, and stock watering (SDDANR 2021, Administrative Rules, Chapter 74:51:03:01).

Site LC-1-BIO: This site is located about 200 m upstream of the confluence with Annie Creek at an elevation of 1,694 m.

2.2.4 Deadwood Creek

The headwaters of Deadwood Creek are located approximately 4.8 km west of Lead at an elevation of 1,740 m above sea level. The stream flows to the northeast and enters Whitewood Creek near the town of Deadwood (Figure 1-1). Near its headwaters, Deadwood Creek has a dense canopy cover with extensive woody debris and abundant riparian vegetation. Downstream of this area, the stream channel and vegetative canopy widen slightly, making the stream more accessible. Deadwood Creek in this upper section has the standard beneficial uses as designated by SDDANR for irrigation, fish and wildlife propagation, recreation, and stock watering. Lower Deadwood Creek from the confluence with Whitewood Creek up to Township 5N, Range 3E, Section 30 (T5N, R3E, S30) is additionally classified as coldwater marginal fish life propagation waters, immersion recreation waters, and limited-contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site DC-2-BIO: This site is approximately 300 m downstream of the confluence of the North and South forks of Deadwood Creek and downstream of Site DC-1-BIO at an elevation of 1,624 m.

2.2.5 False Bottom Creek

The headwaters of False Bottom Creek are located 5.3 km northwest of Lead (Figure 1-1), at an elevation of 1,673 m above sea level. False Bottom Creek flows north, joining the Belle Fourche River between the towns of Spearfish and Whitewood, South Dakota. Near its headwaters, False Bottom Creek is characterized by a semi-open vegetative canopy. False Bottom Creek is classified with the standard beneficial uses and additionally as coldwater marginal fish life propagation water and limited-contact recreation waters in the study area (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site EFB-1-BIO: This site is located just upstream of the confluence with West Fork False Bottom Creek at an elevation of 1,672 m, approximately 1 km downstream from its headwaters.

Site WFB-1-BIO: This site is also located just upstream of the confluence with the East Fork False Bottom Creek at an elevation of 1,669 m, approximately 1 km downstream from its headwaters.

2.2.6 McKinley Gulch

The headwaters of McKinley Gulch are located approximately 8.5 km west of Lead, South Dakota at an elevation of approximately 1,558 m. McKinley Gulch is a small, ephemeral stream that flows into Spearfish Creek approximately 0.8 km downstream of the inflow from Annie Creek (Figure 1-1). McKinley Gulch was dry in 2021, as it has been historically, and the exact location of Site MG-1-BIO has not been established. McKinley Gulch is not currently classified in the administrative rules, although the reach of Spearfish Creek that it enters is classified with the standard beneficial uses and coldwater permanent fish life propagation water, immersion recreation water, and limited contact recreation water (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

2.2.7 Cleopatra Creek

Cleopatra Creek is a tributary to Spearfish Creek, with headwaters located approximately 8.0 km west of Lead, South Dakota. Cleopatra Creek is a tributary to Spearfish Creek with its confluence at the community of Maurice (Figure 1-1). The headwaters of Cleopatra Creek are located in the vicinity of mining operations by Wharf and flows near the LAC Minerals Richmond Hill Mine. Cleopatra Creek is classified with the standard beneficial uses and with coldwater permanent fish life propagation water, immersion recreation water, and limited contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site CC-1A-BIO: This site is located between Monitoring Well 41 and Compliance Point 004 and is in the headwaters of Cleopatra Creek, just downstream of the toe of mining activities. It is located at an elevation of 1,808 m above sea level. The previous location of Site CC-1-BIO was downstream of Compliance Point 004 between the East Fork and Labrador Gulch.

2.2.8 Fantail Creek

The headwaters of Fantail Creek are located approximately 4.9 km from Lead, South Dakota (Figure 1-1). Fantail Creek flows northeast from the base of Terry Peak toward Lead for approximately 3 km before it joins Nevada Gulch directly upstream of the confluence of Nevada Gulch and Whitetail Creek. Fantail Creek flows through a narrow valley for most of its length. The upper portion of the Fantail Creek drainage basin contains portions of the Terry Peak Ski Area and the Golden Reward Mine operations, while the lower portion of the basin contains several private residences. Flow in Fantail Creek is ephemeral in its headwaters from Terry Peak to the former location of the GRMC guard house (Kim Schultz, personal communication). Fantail Creek is classified with the standard beneficial uses and with coldwater permanent fish life propagation water, immersion recreation water, and limited contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site FC-1-BIO: The Fantail Creek site is located immediately downstream of the intersection between Fantail Creek Road and Gilded Mountain Road, at an elevation of 1,684 m above sea level.

2.2.9 Nevada Gulch

The headwaters of Nevada Gulch are approximately 5.3 km from Lead, South Dakota (Figure 1-1). Nevada Gulch flows east from its headwaters on the northeast slopes of Terry Peak to its confluence with Whitetail Creek. The drainage is in a narrow valley that contains a paved state road and several private residences. Nevada Gulch is classified with the standard beneficial uses and coldwater marginal fish life propagation water and limited contact recreation waters (SDDANR 2021, Administrative Rules, Chapter 74:51:03:10).

Site NG-2-BIO: This site is located 2.0 km west of the intersection between Highway 85 and Nevada Gulch Road, at an elevation of 1,726 m above sea level. This site served as a background control until 2017.

2.2.10 Stewart Gulch

The headwaters of Stewart Gulch are located approximately 4.7 km from Lead, South Dakota. Stewart Gulch is located south of the mine and flows due east for approximately 1.5 km before joining Whitetail Creek (Figure 1-1). The majority of the flow in Stewart Gulch comes from an adit located in abandoned mine workings approximately 0.4 km upstream of the confluence with Whitetail Creek. Stewart Gulch is classified with the standard beneficial uses and coldwater permanent fish life propagation water and limited contact recreation waters (SDDANR 2021, Administration Rules, Chapter 74:51:03:10).

Site SG-1-BIO: This is located approximately 9 m upstream of the confluence with Whitetail Creek near monitoring well PW02 and extends upstream approximately 100 m, just downstream of the groundwater seep. The site is located at an elevation of 1,695 m above sea level.

3. Methods

3.1 Habitat Assessment

Physical habitat data were collected in August 2021 at the biological monitoring sites using a standard method that has been consistently used by GEI Consultants, Inc. (GEI) for this region and includes a subset of metrics from the U.S. Forest Service, as described by Platts et al. (1983) and Overton et al. (1997).

Once the downstream site boundary was identified (transect one), transects were established every 10 m to achieve the 11 total transacts for each study site. The section of stream sampled was chosen to be representative of the habitat present in that stream reach in terms of pool to riffle ratio, shading, streamside vegetation, bank stability, etc. The upstream and downstream boundaries were often located at physical habitat features that restricted fish movement. In-stream habitat units (e.g., riffle, run, pool, etc.) were identified, working from the downstream end to the upstream end of each monitoring site. Measurements for the following metrics were made within each habitat unit over the entire site length of approximately 100 meters:

1. **Channel Width** – measurement of surface water width plus width of left and right banks collected at each transect.
2. **Water Width** – measurement of the surface water width collected at each transect within the habitat unit.
3. **Water Depth** – measurement of water depth collected at 25, 50, and 75% of the water width along each transect.
4. **Maximum Water Depth** – measurement collected at the deepest point in each habitat unit.
5. **Water Velocity** – measurements collected at 25, 50, and 75% of the water width along each transect.
6. **Percent Surface Fines** – substrate measurement based on a grid sampling device, as described in Overton et al. (1997). Measurements are collected at three or more individual locations in each habitat unit, and the mean value is reported.
7. **Eroding Streambank** – length of eroding streambank along each bank for entire length of habitat unit.
8. **Streambank Vegetation** – describes dominant streambank vegetation at the transect.
9. **Streambank Cover** – visual estimate of percentage of streambank covered by different vegetation types, along entire length of habitat unit.

10. **Streambank Stability** – rating of whether streambank is stable or exhibits erosional or depositional characteristics at the transect; where a rating of 1 represents stable bank, 2 represents cut/sloughing bank, or 3 represents a depositional bank.
11. **Streambank Angle** – rating of whether streambank is sloping, vertical, or undercut at the transect, where a rating of 1 represents an undercut bank, 2 represents a sloping bank, or 3 represents a vertical bank.
12. **Streambank Undercut** – depth of undercut bank for each bank at the transect. Values expressed in this report represent the mean undercut streambank for the entire reach.
13. **Vegetation Overhang** – measurement of vegetation overhanging water column providing fish cover at each transect. Values expressed in this report represent the mean overhanging vegetation for the entire site length.
14. **Substrate Composition** – estimate of the percentage of the stream bottom covered by bedrock and boulder (> 304 millimeter [mm]), rubble (76 - 304 mm), gravel (4.8 - 75 mm), coarse sediment (0.8 - 4.8 mm), or fine sediment (≤ 0.8 mm) within each habitat unit.

Additionally, water depths and velocities were measured at one transect at each study site using an OTT MF Pro flow meter to allow calculation of discharge. Lastly, precipitation data from GHCND:USC00394834 in nearby Lead, SD (NOAA 2020) was downloaded to evaluate wet-dry year type conditions since the Annie Creek stream gage was decommissioned in 2018.

Select habitat metrics are reported in the Results section. Total length and percent eroding bank are calculated as the sums of all units within the respective habitat types, average water width and depth are calculated as the averages of all units within the respective habitat types, and average maximum depth is calculated as the average of the maximum depths within each unit of a habitat type.

3.2 Fish Populations

Fish populations were quantitatively sampled (consistent with SDGFP guidelines) by electrofishing at all monitoring sites that contained flowing water in August 2021 to determine presence/absence, species composition, density, biomass, size structure, and condition (i.e., body "plumpness," as an indicator of overall health) of the fish assemblage. Sampling consisted of making at least three passes through the representative stream section (approximately 100 m reaches) using a Smith-Root LSR4 electrofishing backpack set to approximately 150 volts and 18 percent duty cycle. If an adequate proportion of the total number of fish sampled were collected on the first and second passes, three passes were typically adequate. If not, a fourth pass was performed to provide a more accurate estimate of the total fish population within the site. If no fish were collected during the first pass, no more passes were conducted, and fish were considered absent from the reach. When physical habitat features did not limit fish movement, the upper and lower boundaries at each site were blocked with nets to reduce fish immigration or emigration during sampling.

Fish captured during each pass were retained separately to allow for estimates of population density. All fish were identified by species, counted, measured for total length (mm), weighed (gram, g), and released. Any obvious injuries, deformities, or signs of disease were noted in the comments section of the data sheet. Population density was estimated by using a maximum likelihood estimator and the MicroFish program developed for the U.S. Forest Service (Van Deventer and Platts 1983 and 1986). These sampling procedures provided species lists and estimates of density (number per hectare, #/ha) and biomass (kilogram per hectare, kg/ha). In addition, length-frequency data were used to analyze the size structure of the fish populations to determine whether recruitment likely occurred from natural reproduction within or near the reach, or by immigration from populations outside of the reach (Everhart and Youngs 1981; Anderson and Neumann 1996). Zero-year age class are fish in the first year of life, aka Young-of-Year (YOY) and are generally less than 100 mm, first year age class includes juveniles and are generally 100 - 150 mm, and second year-plus age class are in their second year of life and are generally greater than >150 mm. However, these ranges can vary by location and exact breaks were determined using the histograms.

3.2.1 Metric Calculations

The condition of trout was evaluated using the relative weight index (W_r) (Anderson and Neumann 1996; Neuman et al. 2012) as well as Fulton's condition factor (K) (Anderson and Neumann 1996; Neumann et al. 2012). Values for these indices were compared among sites to evaluate the health of the fish and to identify potential environmental stressors that may be affecting the populations. To determine relative weight, measured fish weights were compared to length-specific standard weights constructed to represent the species. Relative weight values were only calculated for Brook Trout greater than 120 mm and Brown Trout greater than 140 mm. Expected values of the relative weight index have the same general range across species. Relative weight values generally fall between 70 and 130 (Murphy and Willis 1991). Relative weight values between 95 and 105 are the optimal management target range for most species. Relative weights below 95 represent fish below the optimum weight (i.e., underweight) and relative weights above 105 represent those above the optimum weight (i.e., overweight; Anderson 1980; Anderson and Neumann 1996). Low relative weights can indicate a lack of suitable prey or other stressors that may have a negative influence on fish health, while high relative weights may indicate an overabundance of prey in proportion to predators.

Additionally, Fulton's condition factor was calculated to further evaluate fish health among sites. This metric does not have a minimum size requirement for individuals to be included in the calculation. In fish populations dominated by smaller, first year age class fish, as is often the case at the Wharf sites, a much greater proportion of the population can be included in the calculation of Fulton's condition factor than can be in the calculation of the relative weight index. Thus, Fulton's condition factor may better evaluate the overall health of some fish populations, despite the associated limitations. Fulton's condition factor is not standardized to allow comparisons among different species of fish, or between populations with greatly different size structures. The condition factor of many trout populations in the western United States (US) averages approximately 1.00 (Carlander 1969), so a condition value at or near 1.00 is considered desirable and indicative of a healthy population.

Further analysis of fish populations across all sites was conducted using quartile analysis that examines 2021 density and biomass at each site compared to the 1990-2020 density and biomass values categorized by quartiles: Q1 = minimum value to 25th centile, Q2 = 25-50th centile, Q3 = 50-75th centile, and Q4 = 75th centile to maximum value. Fish population quartiles were split into fish less than 150 mm and fish greater than or equal to 150 mm, which roughly corresponds to a lower threshold for the second year age class or greater.

3.2.2 Fish Tissue Selenium

Selenium is an essential micronutrient required by most aquatic and terrestrial species to maintain metabolic function (EPA 2015). It occurs in virtually all environmental media at trace concentrations, including rocks, soils, water, and living organisms. Mining activities can bring selenium-enriched deposits to the surface where waste materials are crushed and stored as overburden. This process increases the exposure to weathering and can lead to the mobilization of selenium in watersheds (Presser et al. 2004). Other Anthropogenic activities, such as irrigating seleniferous soils, operation of coal-fired power plants, and oil refining, have increased selenium beyond background concentrations in many aquatic ecosystems (Lemly 1997).

Given the role of selenium as an essential micronutrient, aquatic organisms readily bioaccumulate organic forms of selenium (e.g., selenomethionine), yet are not able to excrete selenium at the same rate of consumption when water/prey concentrations are elevated. This imbalance of intake and excretion can lead to elevated tissue concentrations that can be toxic to the organism.

Five brook Trout were collected for fish tissue analysis during fish population sampling at sites AC-3-BIO and EFB-1-BIO. Attempts were made to collect fish of similar sizes, with the smallest fish being at least 75 percent of the length of the largest fish retained for tissue analyses. The fish targeted for these analyses were at least second-year age class or greater and generally between 145 mm and 165 mm in length.

Whole-body fish samples were placed in separate zip locking plastic bags stored on ice in the field and then frozen. Upon completion of field sampling, fish samples were shipped overnight to ACZ Laboratories (ACZ) in Steamboat Springs, Colorado, where they were analyzed for total recoverable selenium (wet weight) and percent solids using the EPA Method M6020 ICP-MS, and ASTM D2216-80, respectively. The minimum detection limit using this method was 0.01 µg/g wet weight. Dry weight selenium concentrations were calculated by dividing the wet weight selenium concentration by the percent solids for each sample. Both forms of whole-body concentrations are presented in this report.

In June 2016, EPA published the final updated national chronic aquatic life criterion for selenium in fresh water (EPA 2016). This criterion consists of multiple elements: whole-body fish tissue, fish muscle, fish egg-ovary, and water column criteria. Fish tissue concentrations are given precedence over the water column concentrations when data for both are available. The fish collected from Annie Creek and East False Bottom Creek were analyzed as whole-body fish tissues and are thus comparable to the whole-body fish tissue criterion of 8.5 µg/g dry weight (dw) value included in the final document. Calculation of this criterion utilized data from nine fish species and included some taxa not found in South Dakota. The whole-body selenium values for Rainbow Trout (*Oncorhynchus mykiss*), Brown Trout (*Salmo trutta*), Dolly Varden (*Salvelinus malma*), and Cutthroat Trout (*Oncorhynchus clarkii*) were included in the EPA selenium dataset. The whole-body Genus Mean Chronic Values (GMCVs) for *Oncorhynchus*, *Salmo*, and *Salvelinus* are 11.6, 13.2, and 34.9 µg/g (dw), respectively (EPA 2016). Although naturally reproducing fish from the genus *Oncorhynchus* are not currently found in Annie Creek or East False Bottom Creek, the data from studies of this species were retained for use since they represent sensitive coldwater salmonid species found in other coldwater streams in the Black Hills. Also, the value for *Oncorhynchus* was chosen for comparison as it would be more protective than the GMCV for *Salmo* and *Salvelinus*. Therefore, a selenium concentration of 11.6 µg/g (dw) was used as a whole-body fish tissue threshold for comparison to Brook and Brown Trout tissues, in addition to the more protective whole-body fish tissue threshold value of 8.5 µg/g established by the EPA (2016).

3.3 Benthic Macroinvertebrate Populations

Benthic macroinvertebrate population sampling was conducted at all monitoring sites with flowing water present in August 2021. Consistent with the SDDANR protocol (SDDENR 2017), a 0.1 m^2 area was sampled using a kick net (20 × 50 centimeter [cm] opening and 500 micrometer [µm] mesh size) beginning with Transect 1 and proceeding upstream to each of the 11 transects delineated during the habitat assessment. At Transect 1, a randomly selected location (25, 50, or 75% of the water width) was identified for macroinvertebrate collection with collection location systematically rotating at subsequent transects. In erosional habitat, loose rocks and large substrates were kicked vigorously for 30 seconds to dislodge organisms into the net. In depositional habitats, similar techniques were used, except that the net was dragged through the standing water within the 0.1 m^2 area to capture suspended benthic organisms. In habitats with dense vegetation (i.e., aquatic plants or filamentous algae), the net was swept through the vegetation or strands of filaments were removed and placed in the sample. The collected organisms were combined into a single, "reach-wide" composition sample for each site. All samples were transferred to appropriately labeled sample containers, preserved with 95% ethyl alcohol, and returned with a Chain of Custody form to the GEI laboratory for processing.

In the laboratory, organisms were sorted from the debris. If the number of organisms was excessive (i.e., >300 organisms/sample), the sample was subsampled such that a minimum of 300 organisms in a minimum of 1/10 of the sample was sorted (Vinson and Hawkins 1996; Carter and Resh 2001). For quality assurance, an experienced technician or taxonomist checked all sorted samples, and the results were documented for 10% of the samples. These procedures indicated over 97% thoroughness for sorting from sample debris.

The sorted specimens were then identified to the lowest practical taxonomic level using available keys (dependent upon the age and condition of each specimen) and counted by taxon (Carter and Resh 2001). Quality assurance for identifications and counts (Whittaker 1975; Stribling et al. 2003) were randomly conducted on 10% of the samples and indicated 99% or higher agreement for taxonomic and count accuracy of identified taxa.

Oligochaetes were mounted on glass slides prior to identification, and chironomids were identified under a dissecting microscope. If the number of chironomids or oligochaetes was excessive (i.e., >30 organisms/sample), they were subsampled prior to mounting such that 10% of the total number (minimum of 30 individuals each) were mounted.

These procedures provided species lists and estimates of abundance. Further analyses were conducted to calculate additional population metrics including measures of species richness, community composition, tolerance, trophic habit, and life history.

3.3.1 Metric Calculations

Many metrics are available for evaluating benthic macroinvertebrate populations with most belonging to one of five categories: richness, composition, tolerance, trophic habit, and life history. The large number of available metrics necessitates a focus on those that are most useful in the region or state of interest (Barbour et al. 1999). The most useful metrics are those that best distinguish impacted and unimpacted sites and include “reference” conditions established using 20 to 50 unimpacted sites (Bowman and Somers 2005; Grafe 2002). These references have not been determined for the Wharf study, due to the challenges of identifying “reference” conditions in the Black Hills mining region. As agreed to by Wharf, SDGFP, and SDDANR, several metrics used on other Black Hills monitoring projects and/or previously used in biomonitoring projects were used in this study to analyze benthic macroinvertebrate data and to compare study sites with reference sites (Table 3-1).

The metrics listed below were calculated for Wharf study sites to allow comparisons between the current data and previous years and between study sites and their respective reference sites. Some metrics have established ranges or values that can indicate the occurrence of a disturbance that affects benthic macroinvertebrate communities while other metrics are evaluated within the context of historical ranges in the Black Hills. These metrics are described as “poor” or “low”, “fair” or “moderate”, and “good” or “high” in this text.

Table 3-1: Summary of benthic macroinvertebrate metrics calculated for the Wharf biomonitoring sites.

Metric	Type of Metric	Definition	Change Expected Following Environmental Disturbance
Density	Richness	Total abundance of invertebrates (#/sample).	Decrease
Number of Taxa	Richness	Number of distinct taxa	Usually Decrease
Number of EPT Taxa	Richness	Number of taxa in the orders Ephemeroptera, Plecoptera, and Trichoptera (EPT).	Decrease
Shannon-Weaver Diversity Index	Composition	The extent that density is spread among a wide number of species.	Decrease
Percent Sensitive EPT Taxa	Composition	Percent of total taxa comprised of EPT taxa with tolerance values between 0 and 4.	Decrease
EPT Index	Composition	Percent of total taxa comprised of EPTs.	Decrease
Percent *Baetis sp.*	Composition	Percent of Ephemeroptera abundance comprised of individuals representing *Baetis sp.*	Increase
Number of non-*Baetis* Ephemeroptera	Composition	Number of Ephemeroptera individuals not in the genus *Baetis*.	Decrease
Percent Chironomidae	Composition	Percent of midge larvae	Increase
Number of Plecoptera Taxa	Composition	Number of taxa within the order Plecoptera.	Decrease
Percent Abundance of Oligochaetes & Hirudinea	Composition	Percentage of total abundance comprised of oligochaetes (segmented worms) and Hirudinea (leeches).	Variable
Dominant Taxon	Composition	Species name of most abundant taxon.	--
Percent Dominant Taxon	Composition	Measures the dominance of the single most abundant taxon.	Increase
Number of Common Taxa	Composition	Number of taxa common to both reference and non- reference sites.	Decrease
Community Loss Index	Composition	Percent of species at the reference site not present at the non-reference site.	Increase
Hilsenhoff Biotic Index (HBI)	Tolerance	Abundance-weighted mean of the tolerance values.	Increase
Percent Tolerant Taxa	Tolerance	Percent of total taxa comprised of taxa with tolerance values ranging from 7 to 10.	Increase
Percent Intolerant Taxa	Tolerance	Percent of total taxa comprised of taxa with tolerance values ranging from 0 to 4.	Decrease
Number of Intolerant Taxa	Tolerance	Count of the total taxa with tolerance values ranging from 0 to 4.	Decrease
Number of Predator Taxa	Trophic Habit	Number of taxa belonging to this functional feeding group.	Decrease
Percent Collector-Gatherers	Trophic Habit	Relative abundance belonging to this functional feeding group.	Variable
Number of Shredder Taxa	Trophic Habit	Number of taxa belonging to this functional feeding group.	Decrease
Number of Univoltine Taxa	Life History	Number of taxa classified as having a life history of 1 year.	Increase
Number of Semivoltine Taxa	Life History	Number of taxa classified as having a life history of greater than 1 year.	Decrease
Percent Semivoltine taxa	Life History	Percentage of total taxa comprised of taxa classified as having a life history of greater than 1 year.	Decrease
Number of Merovoltine Taxa	Life History	Number of taxa classified as having a merovoltine (three or more years) life history.	Decrease

3.3.1.1 Richness Metrics

Three metrics were calculated for richness: Density, Number of Taxa, and the Number of EPT Taxa. The Number of Taxa is commonly used to represent invertebrate species richness at a site, and higher richness usually indicates better water quality. In mountain streams, such as those in the northern Black Hills, the presence of mayfly (Ephemeroptera), stonefly (Plecoptera), and caddisfly (Trichoptera) taxa (collectively referred to as the EPT taxa) are generally an indicator of good water quality because most of these taxa are sensitive to a wide range of pollutants (Hynes 1970; Wiederholm 1984; Klemm et al. 1990; Merritt et al. 2008; Barbour et al. 1999). The Number of Taxa and the Number of EPT Taxa would be expected to be higher in unimpacted sites than in impacted sites. However, in some cases, the Number of Taxa can increase due to increases in the number of non-insect taxa or to tolerant insect taxa which indicate poor water quality. Therefore, changes in the Number of Taxa were also evaluated with respect to species composition. The Number of Taxa and the Number of EPT Taxa were determined for each study site and compared to the reference sites to determine whether any sites were negatively impacted.

3.3.1.2 Composition Metrics

Composition analyses included the calculation of twelve metrics: the Shannon-Weaver Diversity Index (Diversity), Percent Sensitive EPT Taxa, EPT Index, Percent *Baetis sp.*, number of non-*Baetis* Ephemeroptera, Percent Chironomidae, Number of Plecoptera Taxa, Percent Abundance of Oligochaetes & Hirudinea, the Dominant Taxon and Percent Dominant Taxon, Number of Common Taxa between the reference and non-reference sites, and the Community Loss Index.

The US Environmental Protection Agency (EPA) recommends Diversity for measuring the stress level of invertebrate communities (Klemm et al. 1990). This index ranges from 0 to greater than 4. Values greater than 2.5 are indicative of a healthy invertebrate community (Wilhm 1970; Klemm et al. 1990).

In addition to the Number of EPT Taxa (a richness Metric discussed above), two other EPT metrics were calculated. The Percent Sensitive EPT Taxa metric characterizes the EPT taxa that have low tolerance to perturbation (organic, inorganic, nutrient, and metal pollution and physical disturbance) out of all taxa present. Tolerance values for each taxon are obtained from the Northwest (Idaho) Regional Tolerance Value database (see additional explanation below), with sensitive taxa ranging from 0 to 4 on a scale of 0 to 10. The percent of sensitive EPT taxa would be expected to be higher in unimpacted sites (Wiederholm 1984; Klemm et al. 1990; Barbour et al. 1999). The EPT Index was also calculated as the percent of the total taxa that is comprised of EPT taxa. The EPT Index is expected to decrease with increasing environmental perturbation. The other mayfly composition metrics include Percent *Baetis sp.* and Number of non-*Baetis* mayflies. Percent *Baetis sp.* is calculated as the abundance of *Baetis* mayflies divided by the abundance of all mayflies. *Baetis* mayflies are relatively tolerant of environmental stressors, and high abundances of these groups can indicate conditions less suitable for more sensitive taxa. Number of non-*Baetis* Mayfly individuals indicates the abundance of other mayfly species.

The composition-based metric of Percent Chironomidae describes relatively high abundances of one family of dipterans (true flies) that are typically tolerant of less suitable environmental conditions. In addition, the Percent Dominant Taxon metric is expected to increase with environmental perturbation, as a high relative abundance of a single taxon is correlated with low diversity within the macroinvertebrate community and can indicate stressors are present. The Dominant Taxon was also identified, and its tolerance value noted. The Number of Plecoptera (stonefly) taxa was also included, as stoneflies can be sensitive to certain environmental stressors. Percent Abundance of Oligochaetes & Hirudinea was also calculated.

The Community Loss Index uses the Number of Common Taxa metric to measure the changes in benthic invertebrate communities between reference sites and potentially impaired sites. This metric is calculated by taking the Number of Taxa at the reference site, subtracting the Number of Common Taxa between the two sites, and then dividing the remaining number by the total taxa present at the potentially impaired site. The calculated values are dimensionless, and values increase with increasing dissimilarity with the reference site (Plafkin et al. 1989). Because this metric is only evaluating dissimilarity, the reason for dissimilarity should be determined in cases where large differences were identified between reference sites and potentially impaired sites.

3.3.1.3 Tolerance Metrics

Four metrics were also calculated for perturbation tolerance: The Hilsenhoff Biotic Index (HBI, Hilsenhoff 1987), Percent Tolerant Taxa, Percent Intolerant (or sensitive) Taxa, and Number of Intolerant Taxa. The HBI was originally designed to gauge the effects of organic pollution. The Idaho Department of Environmental Quality compiled a set of updated values in the Northwest Regional Tolerance Value database (Appendix B of Barbour et al. 1999) which measure sensitivity to general environmental stress (Grafe 2002). Although multiple tolerance databases are available (Barbour et al. 1999), benthic invertebrate communities in the Black Hills Core Highlands have the most taxa in common with the communities used to develop the Northwest Regional Tolerance Value database. The updated tolerance values range from 0 (sensitive, intolerant organisms) to 10 (highly tolerant organisms) and were assigned to each identified taxon. If an identified taxon was not listed in Appendix B of Barbour et al. (1999) or a tolerance value was not given for that taxon, best available literature was used to determine a tolerance value. The final HBI value is an abundance-weighted mean of the tolerance values. The HBI is expected to be higher at impacted sites because the community is expected to be comprised of more tolerant (higher scoring) organisms.

Hilsenhoff Biotic Index scores are also used to categorize benthic macroinvertebrate communities with scores of 0.00 - 3.50 considered as “Excellent,” 3.51 - 4.50 are considered “Very Good,” 4.51 - 5.50 are considered “Good,” 5.51 - 6.50 are considered “Fair,” 6.51 - 7.50 are considered “Fairly Poor,” 7.51 - 8.50 are considered “Poor,” and 8.51 - 10.00 are considered “Very Poor” (Hilsenhoff 1987). HBI values and ratings were compared between reference sites and the mining activity sites to determine if there are indications of environmental stress present downstream of the mining area.

The proportion of the community composed of tolerant or intolerant taxa was also used to evaluate community sensitivity to environmental stress. Tolerant taxa are defined as those that have been assigned tolerance values of 7, 8, 9, or 10. Intolerant taxa are those that have been assigned values of 0, 1, 2, 3, or 4. Stressed sites tend to support communities dominated by tolerant taxa (Barbour et al. 1999; Grafe 2002), so the percentage of tolerant taxa tends to increase, and the percentage of intolerant taxa tends to decrease with increasing environmental stress. The proportions of tolerant and intolerant taxa were compared between sites to evaluate whether individual sites showed signs of environmental stress.

3.3.1.4 Trophic Habit Metrics

Trophic functional feeding metrics – Number of Predator Taxa, Percent Collector-Gatherers, and Number of Shredder Taxa were also determined for each community based on Merritt et al. (2008). Predators rely upon a persistent prey base for survival, so patterns in populations often correspond to water quality and environmental disturbances that affect the prey base (Merritt et al. 2008). Specialized feeders, such as predators, are often considered sensitive organisms and are usually well represented in healthy streams (Barbour et al. 1999). Predator metrics are expected to decrease with increased disturbance.

Fine particulate organic matter is the primary food source of collector-gatherers, and their relative abundance can indicate disturbances associated with sedimentation and/or nutrient enrichment (Hargett 2011). These species are generalist feeders and can adjust to a broader range of food materials than specialist feeders and tolerate a broader range of conditions. Disturbances that increase organic matter in the stream, such as nutrient enrichment, may result in a community shift favoring the dominance of collector-gatherers (Hargett 2011). However, physical disturbances, such as increased sedimentation, can reduce these species. High percentages or large changes in the Percent Collector-Gatherers metric over time are often indicative of increased nutrient enrichment or sedimentation. Conversely, shredders often feed upon leaf litter and other unstable substrates; therefore, their low abundance or absence can be indicative of recent disturbance or lack of leaf litter or other suitable forage within a given site.

3.3.1.5 Life History Metric

Life history metrics listed include the Number of Univoltine Taxa, the Number of Semivoltine Taxa, the Percent Semivoltine taxa, and the Number of Merovoltine Taxa. Univoltine, semivoltine, and merovoltine are terms used to describe benthic macroinvertebrates that take one year, more than one year, and three or more years to complete a life cycle (generation), respectively. Short-term disruptions in suitable aquatic conditions, either chemical or physical, can reduce the Number of Taxa with longer life history traits (semivoltine and merovoltine organisms), and increase the relative abundance of more short-lived (univoltine) taxa. Therefore, as anthropogenic stressors increase, a community often shifts towards taxa with a shorter life history strategy, because longer-lived organisms are unable to persist long enough to reproduce.

3.4 Periphyton Populations

Periphyton population sampling was conducted at monitoring sites with flowing water present in August 2021 following SDDANR methods (SDDENR 2017). Using this protocol, one piece of substrate was sampled at each of the 11 transects delineated during the habitat assessment. Within Transect 1, a randomly selected location (25, 50, or 75% of the water width) was identified for periphyton collection which systematically rotated at subsequent transects. In erosional habitats, a piece of substrate was collected, and an area of 12 cm^2 was scrubbed with a stiff-bristled toothbrush for 30 seconds and washed into a 500 milliliter (mL) sample bottle. In depositional habitats, the top 1 cm of sediment from a 12 cm^2 area was collected using a 60 mL syringe and added to the 500 mL sample bottle.

The 11 periphyton samples were combined to create a single “reach-wide” composition sample for the site, and this composition sample was brought up to a total of 500 ml. After thorough mixing, a 50 mL aliquot was removed for taxonomic identification and enumeration and preserved with Lugol’s solution. A second aliquot of 25 mL was filtered onto a Whatman GF/F filter for chlorophyll-a determination and stored wrapped in foil in the dark on ice in the field and then placed in a freezer. A third aliquot of 25 mL for biomass determination was filtered onto a pre-combusted Whatman GF/F filter for ash-free dry mass (AFDM) determination.

All samples were labeled with the appropriate site name, sample type, and date, and returned to the GEI laboratory with a Chain of Custody form. Samples for identification and enumeration (SM 10200 C.2, D.2, E.4, and F.2.c) were sent to Aquatic Analysts, Friday Harbor, WA for processing. Samples for chlorophyll-a (SM 10200 H) and AFDM (SM 10300 C.5 (modified)) determination were processed by GEI (APHA 2005).

3.4.1 Metric Calculations

As with the benthic macroinvertebrate data, additional metrics were calculated from the periphyton data including richness, composition, tolerance, and trophic habit categories (Table 3-2). Candidate metrics were selected, based upon input from SDGFP, from Barbour et al. (1999), and elimination of redundant and unresponsive metrics also followed Barbour et al. (1999). Some metrics have established ranges or values that can indicate the occurrence of a disturbance that affects periphyton communities while other metrics are evaluated within the context of historical ranges in the Black Hills. These metrics are described as “poor” or “low”, “fair” or “moderate”, and “good” or “high” in this text.

Table 3-2: Summary of periphyton metrics calculated for the Wharf biomonitoring sites.

Metric	Type of Metric	Definition	Change Expected Following Environmental Disturbance
Density	Richness	Number of periphyton cells/cm	Variable
Relative Diatom Density	Richness	Ratio of diatom taxa to the total number of taxa	Variable
Number of Taxa (species)	Richness	Number of distinct periphyton taxa	Decrease
Number of Diatom Taxa	Richness	Number of distinct diatom taxa	Decrease
Number of Periphyton Divisions	Richness	Number of periphyton divisions represented in the sample	Variable
Number of Periphyton Genera	Richness	Number of Periphyton genera represented in the sample	Variable
Shannon-Weaver Diversity Index for Diatoms	Composition	The extent that density is spread among a wide number of species.	Decrease
Bahls Similarity Index	Composition	Proportion of periphyton taxa and density that is shared between study and reference sites.	Variable
Autotrophic Index	Composition	Ratio of AFDM to chl-a	Increase
Percent Tolerant Diatoms	Tolerance	Percent relative abundance of diatom taxa with tolerance value of 1.	Increase
Lange-Bertalot Pollution Index	Tolerance	Cumulative index of pollution tolerance values of all taxa sampled; separated into class 1 (tolerant), class 2, and class 3 (sensitive).	Decrease
Percent Eutrophic Diatoms	Trophic Habit	Percentage of taxa comprised of eutrophic diatoms	Variable
Percent Acidiphilic Diatoms	Trophic Habit	Percentage of taxa comprised of acidiphilic diatoms	Variable
Percent Alkaliphilic Diatoms	Trophic Habit	Percentage of taxa comprised of alkaliphilic diatoms	Variable
Percent Nitrogen Heterotrophs	Trophic Habit	Percentage of taxa comprised of Nitrogen heterotrophs	Variable
Percent High Oxygen Diatoms	Trophic Habit	Percentage of taxa comprised of high oxygen diatoms	Variable
Percent Motile Diatoms	Trophic Habit	Percentage of taxa comprised of motile diatoms	Increase
Percent Saprobic Diatoms	Trophic Habit	Percentage of taxa comprised of saprobic diatoms	Variable

3.4.1.1 Richness Metrics

Six metrics were calculated to describe richness – Density, Relative Diatom Density, Number of Taxa, Number of Diatom Taxa, Number of Periphyton Divisions, and Number of Periphyton Genera. Density is a measure of the number of algae cells per unit area of substrate sampled, and relatively high values of density often indicate nutrient enrichment. However, other stressors such as extended periods of low flow can increase the density of periphyton, whereas high flow events that scour the substrate can reduce the density of periphyton. Therefore, this metric is often evaluated in the context of other supporting data. The Number of Taxa represents the biological diversity at a given site. This measure includes taxa from all algal Divisions present, although it should be recognized that several taxa within some Divisions are often too small to be identified during routine examinations (e.g., several Cyanophyta). Diatoms (Bacillariophyta) are generally larger, have more resilient physical architecture, and have a more stable taxonomy (Patrick and Reimer 1966; 1975; Wehr and Sheath 2003). Both the Number of Taxa and the Number of Diatom Taxa would be expected to decrease with increased perturbation.

3.4.1.2 Composition Metric

The Diversity, Bahls Similarity Index, and Autotrophic Index metrics were calculated to describe composition. Diversity is a function of both the Number of Taxa and the abundance of each taxon and often ranges from 0 to greater than 4. Because diatom species richness and composition often vary independently depending on environmental conditions, the changes in this metric over time is a useful tool to identify the presence of stressors. The diatoms are considered to be the most sensitive taxa to changes in water quality (Barbour et al. 1999). Stressed sites often are dominated by a few taxa with lower diversity.

The similarities between periphyton community at the reference site and those at other sites were evaluated using the Bahls Similarity Index (1993). This index compares the appropriate reference site to its respective site downstream of mining activities by calculating the relative abundances of each taxon common to both sites. The smaller relative abundance value for each common taxon is summed for an index that evaluates percent similarity of the periphyton community between sites. This index varies from 0 (different communities) to 100 (identical communities). Ratings for this index are Very Similar (>60), Somewhat Similar (60 - 40), Somewhat Dissimilar (40 - 20), and Very Dissimilar (<20) (Bahls 1993). Dissimilarity between sites can be expected due to habitat differences even if neither is affected by water quality issues or excessive environmental disturbance. If diatom communities are dissimilar, other metrics are carefully considered to determine whether the dissimilarity is due to perturbation or other differences between sites.

The Autotrophic Index was also calculated using the laboratory derived biomass estimates (SM 10300 C.6). This metric is calculated by dividing the AFDM value by the chlorophyll-a value and is used to indicate proportions of the assemblage composed of either heterotrophic (outside sources of organic matter, such as leaf litter) or autotrophic (in-stream sources such as periphyton) material. Communities less disturbed by organic pollution and dominated by algae usually contain Autotrophic Index values ranging from 50-100. Values greater than 400 often indicate communities affected by organic pollution. Values of approximately 250 are more typical for streams enriched with nitrogen or phosphorus and show a potential for increased algal growth (Watson and Gestring 1996; Biggs 1996). However, the Autotrophic Index should be cautiously interpreted because dead organic matter may artificially inflate the ratio. This phenomenon is commonly observed in streams with low flow conditions that allow for the accumulation of dead organic matter over time due to the infrequent high flow scouring events.

3.4.1.3 Tolerance Metrics

Two tolerance metrics were calculated – Percent Tolerant Diatoms and the Lange-Bertalot Pollution Index (Pollution Index). The Percent Tolerant Diatoms metric is the sum of the relative abundances of all pollution-tolerant species. Tolerance values are based on values in Bahls (1993), which incorporated previously published tolerance values that range from 1 to 3 (Lowe 1974; Lange-Bertalot 1979). Tolerant diatoms are defined as those diatoms with a tolerance value of 1, whereas sensitive diatoms receive a tolerance value of 3 (Bahls 1993). This metric is often insightful when evaluating water quality of low-order streams where primary productivity may be naturally low (Barbour et al. 1999), such as for the streams near Wharf.

The Pollution Index was also calculated. The Pollution Index is calculated by multiplying the relative abundance of each taxon by its pollution tolerance value. The sum for all taxa is the Pollution Index, which ranges from 1.0 (all tolerant taxa) to 3.0 (all sensitive taxa). The scores are rated according to Bahls (1993) as No Organic Enrichment (>2.50), Minor Organic Enrichment (2.01 to 2.50), Moderate Organic Enrichment (1.50 to 2.00), and Severe Organic Enrichment (<1.50).

3.4.1.4 Trophic Habit Metric

Percent motile, eutrophic, acidiphilic, alkaliphilic, nitrogen heterotrophs, high oxygen diatoms, and saprobic diatoms were also identified. Eutrophic diatoms are adapted to waters with nutrient enrichment, while acidiphilic and alkaliphilic diatoms are adapted to acidic and alkaline waters, respectively. nitrogen heterotrophs are able to utilize other sources of nitrogen in low-light environments as a source of nutrients, and high-oxygen diatoms require habitats with high levels of dissolved oxygen. Saprobic diatoms are able to utilize decaying organic matter and may increase in abundance following a disturbance that kills other, more sensitive genera. The diatom genera *Navicula*, *Nitzschia*, and *Surirella* are relatively mobile organisms that work their way to the benthic surface when covered by silt (Wehr and Sheath 2003). Because of their mobility, the combined relative abundance of these three genera and others is thought to reflect the amount and frequency of siltation at a site (Barbour et al. 1999; Bahls

1993). Therefore, the Percent Motile Diatoms metric is a surrogate siltation index and was calculated as the sum of the relative abundances of all motile genera. The Percent Motile Diatoms metric is expected to be greater at sites with more silt.

In 2021, GEI completed an extensive update of the periphyton taxa database used to calculate metrics. The original database contained tolerance, composition, and trophic habit (autecological metrics) gleaned from Bahls (1993), Van Dam et al. (1994), Barbour et al. (1999), Hill et al. (2000), and Fore and Grafe (2002) for over 1900 species. The update incorporated an extensive U.S. Geological Survey database (Porter 2008) and database from the Southern California Coastal Water Research Project (TR#0730), including online sources (e.g., www.diatoms.org) that supplemented many pieces of missing information. The Wharf periphyton taxonomic list includes 155 taxa and the updated database greatly increased (e.g., 17 to 325%) the ecological information for the Wharf taxa. As a result, all periphyton metrics were recalculated for the Wharf biomonitoring sites (active or abandoned sites) from 2006 to the present and were used to evaluate the long-term trends and site comparisons provided herein.

3.5 Water Quality

Water quality samples were collected by Wharf mine personnel from all active biomonitoring locations, including the reference sites, within 30 days of the biological sampling event. Water quality analyses included a suite of physicochemical and metals parameters analyzed by Midcontinent Testing Laboratories, Inc. in Rapid City, SD. The suite of analyses includes:

Physicochemical Analyses
- Discharge, field current meter
- pH, field SM 4500-H^+ B
- Temperature, field SM 2550 B
- Hardness, SM 2300 B
- Total dissolved solids, SM 2540 C
- Total suspended solids, SM 2540 D

Inorganic Analyses
- Calcium, SM 3111 B
- Cyanide (weak acid dissociable), Kelada 01
- Magnesium, SM 3111 B
- Nitrate as nitrogen, SM 4500-NO3 F
- Phosphorus (dissolved), SM 4500-P E

Metals Analyses
- Arsenic (Trec), EPA 200.8
- Cadmium (Trec), EPA 200.8
- Chromium (Trec), EPA 200.8
- Copper (Trec), EPA 200.8
- Iron (Trec), EPA 200.8
- Lead (Trec), EPA 200.8
- Mercury (Tot), EPA 200.8
- Nickel (Trec), EPA 200.8
- Selenium (Trec), EPA 200.8
- Selenate (Se^{6+}), IF Trec Se > 12 µg/L
- Selenite (Se^{4+}), IF Trec Se > 12 µg/L
- Silver (Trec), EPA 200.8
- Zinc (Trec), EPA 200.8

3.6 Data Analysis

Aquatic biological monitoring data were summarized and analyzed in relation to mining activities and natural occurrences, such as unusual flows and weather events. When appropriate, fish, benthic macroinvertebrate, and periphyton data were qualitatively correlated with stream habitat and flow data to explain temporal and spatial variation in the aquatic community. The data collected in August 2021 from sites downgradient of mining activities were also compared to data from the reference sites (GEI 2018b).

Long term analyses were limited to data beginning in 2006 as the SDDANR methods and laboratories used have been consistent since this time (GEI 2018b). The fish population density and biomass estimates were compared qualitatively between years and sites. Species composition and size structure were examined within sites to determine if fish are naturally reproducing at the site or are being recruited from other sources.

Least-squares regression analysis was performed on fish K and Wr and all benthic macroinvertebrate and periphyton metric data to evaluate any increasing or decreasing trends at each site. This parametric test is robust to deviations in the assumptions for parametric tests when used to evaluate whether the metric of interest is trending. The Mann Whitney U test, a nonparametric test, was used to evaluate the differences in the long-term median values for a subset of macroinvertebrate metrics (Abundance, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, Hilsenhoff Biotic Index) and periphyton metrics (Density, Number of Taxa, Number of Diatom Taxa, Diversity, Autotrophic Index, Percent Tolerant Diatoms, Pollution Index, Trophic Habit, and Percent Motile Diatoms) for a mining activity site and its comparative reference site. The Mann Whitney U test was performed using the fish metrics (K and Wr) for mining activity sites and compared to the Labrador Gulch reference site. Long-term statistical comparisons (Mann Whitney U test) were also performed for sites that use Reno Creek as their reference site. However, the results were cautiously interpreted due to the small sample size for the reference site (i.e., $n = 4$ for RC-1-BIO). The Kruskal-Wallis multiple comparison test was used to evaluate the differences in the long-term metrics among the three sites on Annie Creek which allows for comparisons among data sets that do not follow a normal distribution. This test is a modification of the Mann Whitney U test which allows for multiple comparisons (Hintze 2004). A 95% confidence level ($\alpha = 0.05$) was used to determine significant differences among sites for all statistical comparisons, including trend analyses.

4. Results and Discussion

4.1 Habitat Assessment

4.1.1 Labrador Gulch and Reno Creek

In 2020, Site RC-1-BIO was moved 21 m upstream to avoid a blown down tree that covered the bottom of the site. This change had little effect on the overall assessment of the stream in 2020 and 2021. Sites LB-4-BIO and RC-1-BIO on Labrador Gulch and Reno Creek, contained 11 and 13 habitat units, respectively, during 2021 surveys (Table 4-1). A large portion of both sites were comprised of fast water habitat, although each site also contained ample slow-water pool habitat. Average stream depth and widths were similar at both site (Table 4-1) with Site LB-4-BIO containing two pools roughly twice the size of pools found at Site RC-1-BIO. Eroding banks were absent from both sites. Overall, Site LB-4-BIO was wider and slightly deeper on average than Site RC-1-BIO.

Table 4-1: Habitat characteristics for Labrador Gulch and Reno Creek, August 2021. HGR = high gradient riffle; LGR = low gradient riffle; DMB, SLB, SLM, SMB, SMW, SPB, and STR = types of pools; STP = step pool complex.

Site/ Habitat Type	No. of Units	Total Length (m)	Average Water Width (m)	Average Water Depth (cm)	Average Maximum Depth (cm)	Eroding Bank (%)
LB-4-BIO						
HGR	3	45.1	1.6	10	21	0
LGR	3	24.7	1.5	9	18	0
DMB	1	3.2	2.7	14	20	0
SLB	1	2.8	2.2	15	22	0
SMB	2	9.5	1.9	19	33	0
STP	1	14.7	1.4	17	24	0
RC-1-BIO						
HGR	1	11.9	1.2	7	20	0
LGR	4	53.6	0.9	5	10	0
SLM	1	7.1	1.0	7	11	0
SMB	2	5.4	1.4	15	30	0
SMW	2	9.0	1.3	15	25	0
SPB	2	6.0	1.2	19	28	0
STR	1	10.0	1.8	5	15	0

Substrate compositions varied at both sites, with Site LB-4-BIO dominated by boulder/ bedrock and rubble, while Site RC-1-BIO was mainly comprised of gravel substrate

(Table 4-2). Differences in substrate composition are due to the differing geologies and geomorphologies at these two sites. The Labrador Gulch site is a high-gradient stream that flows through a steep, bedrock canyon (Photo 4-1). Sources of fine sediment are generally absent within the reach and would likely flow into Site LB-4-BIO from upstream, accumulating in slower, deeper sections of pools. Fine sediments and surface fines at Site LB-4-BIO were low but greater than other years since the site was added to Wharf monitoring in 2018 and correspond with the lowest observed flow. In contrast, Site RC-1-BIO on Reno Creek is a lower gradient stream, in a vegetated and forested valley, with ample sources of fine sediments (Photo 4-2). Fine sediments and surface fines were moderate. However, well vegetated banks at the site, which was more extensive in 2021 than in previous years, prevented bank erosion and did not add to sedimentation.

Table 4-2: Average substrate characteristics for all habitat types at sites LB-4-BIO and RC-1-BIO, August 2021.

Site/ Habitat Type	Flow (cfs)	Average % Surface Fines	Substrate Composition (%)				
			Fine Sediment (≤ 0.8 mm)	Coarse Sediment (0.8 - 4.8 mm)	Gravel (4.8 - 75 mm)	Rubble (76 - 304 mm)	Boulder and Bedrock (> 304 mm)
LB-4-BIO	0.37	10	1	3	18	38	40
RC-1-BIO	0.08	38	20	16	29	23	12

Photo 4-1: High gradient and canyon walls in Labrador Gulch.

Photo 4-2: Low gradient, vegetated stream banks along Reno Creek.

4.1.2 Annie Creek, Ross Valley, Lost Camp Gulch

The Annie Creek Basin contains five of the fifteen monitoring sites, and each site was influenced to some degree by the tornado activity in July 2020. Lower Annie Creek (Site AC-3-BIO) received the most storm disturbance and performing the majority of biomonitoring tasks were not practicable in 2020 and 2021 due to the extensive deadfalls covering the stream (Photo 4-3). Only large sections in the upper half of the reach were accessible and consisted of runs and pools, formed primarily from the wooden debris (Photo 4-5). No habitat measurements were collected, and only a qualitative fish survey was conducted to collect fish tissue. Similarly, on Lost Camp Gulch, a massive pile of deadfalls covered 13.3 m of stream in the middle of Site LC-1-BIO (Photo 4-5). This tree covered section was excluded from the habitat assessment at Lost Camp Gulch.

Photo 4-3: Fallen trees from tornado activity at Site AC-3-BIO. Annie Creek is below trees

Photo 4-4: Short section of Site AC-3-BIO that was accessible.

In August 2021, the numbers of habitat units observed at sites AC-1-BIO and AC-2-BIO were 16 and 13, respectively, while the numbers of habitat units at sites RV-2-BIO and LC-1-BIO were much lower at 3 and 9, respectively (Table 4-3). Fast water habitat types (riffles and/ runs) comprised most of all sites and no pools were found at Site RV-2-BIO. Multiple pools were found at both of the Annie Creek sites with pool forming features including both large woody debris and boulders in the stream. Water widths and depths varied among all sites and in different habitat units (Table 4-3).

Table 4-3: Habitat characteristics for sites on Annie Creek, Ross Valley, and Lost Camp Gulch, August 2021. Habitat types: CAS = Cascade; HGR = high gradient riffle; LGR = low gradient riffle; RUN = run; DMB, DMW, SLB, SMB, and SMW = types of pools; STP = step pool complex.

Site/ Habitat Type	No. of Units	Total Length (m)	Average Water Width (m)	Average Water Depth (cm)	Average Maximum Depth (cm)	Eroding Bank (%)
AC-1-BIO						
CAS	1	1.6	1.5	2	3	0
HGR	4	20.5	0.9	5	13	0
LGR	5	51.0	1.3	5	10	0
RUN	1	4.6	1.2	7	20	0
SMB	3	10.1	1.8	13	24	0
SLB	1	5.7	1.7	21	25	0
SMW	1	3.8	1.3	10	17	0
AC-2-BIO						
HGR	1	3.7	2.4	6	18	0
LGR	5	48.6	1.5	10	21	0
RUN	2	11.5	2.1	19	25	0
DMB	1	5.8	2.6	24	35	0
DMW	1	5.2	2.3	11	21	0
SMB	2	11.5	1.8	18	39	0
STP	1	15.7	1.7	21	34	0
RV-2-BIO						
HGR	1	8.4	0.4	5	10	0
LGR	2	91.6	0.6	5	15	1
LC-1-BIO						
LGR	6	78.7	1.0	6	12	0
RUN	1	5.4	1.3	16	23	0
DMB	1	2.6	1.6	17	30	0
STP	1	11.7	0.9	10	34	0

Sites RV-2-BIO and LC-1-BIO sites are both located in the headwaters of comparatively small creeks and are characterized by relatively narrow water widths and shallow depths (Table 4-3). Site RV-2-BIO was narrower and shallower than other sites and sites AC-1-BIO and LC-1-BIO were narrower than at AC-2-BIO. Riffle and pool depths were similar at sites AC-1-BIO, AC-2-BIO, and LC-1-BIO while runs were much shallower at AC-1-BIO than the other sites. Maximum depths ranged from 3 cm in a cascade flowing over steep bedrock at Site AC-1-BIO, to 39 cm in a pool formed by boulders at Site AC-2-BIO.

Substrate composition at all sites included a range of rock sizes. Boulders and bedrock were the dominant substrate size classes at Site AC-1-BIO, fine sediment at Site RV-2-BIO, and rubble at Site LC-1-BIO (Table 4-4). Substrate at Site AC-2-BIO was distributed among size categories. Percent surface fines were moderate at all sites except at Site RV-2-BIO where fines were much higher. However, all sites included abundant gravel, rubble, and boulders/bedrock, which are considered desirable substrate size classes, as all three substrate sizes provide habitat for benthic macroinvertebrates, and gravel serves as favorable spawning habitat for salmonids (Waters 1995).

Photo 4-5: Deadfall at Site LC-1-BIO.

Habitat conditions at sites AC-1-BIO, AC-2-BIO, and LC-1-BIO were generally comparable to the reference site, Site RC-1-BIO (Table 4-1), with a variety of habitat units at both sites, including riffles, pools, and runs, and similar average widths, depths, and abundance of fines. Water widths and depths at Site RV-2-BIO were similar or lower than those measured at the reference site. Site RV-2-BIO also contained a much smaller variety of habitat unit types and more fines than Site RC-1-BIO.

Table 4-4: Average substrate characteristics for all habitat types at each site on Annie Creek, Ross Valley, and Lost Camp Gulch, August 2021.

Site/ Habitat Type	Flow (cfs)	Average % Surface Fines	Substrate Composition (%)				
			Fine Sediment (≤ 0.8 mm)	Coarse Sediment (0.8 - 4.8 mm)	Gravel (4.8 - 75 mm)	Rubble (76 - 304 mm)	Boulder/ Bedrock (> 304 mm)
AC-1-BIO	0.21	25	16	16	18	17	33
AC-2-BIO	0.23	23	24	20	17	22	18
RV-2-BIO	0.01	69	36	27	16	14	6
LC-1-BIO	0.04	15	11	14	20	31	25

4.1.2.1 Summary of Habitat Conditions

Overall, fish habitat was favorable at the two accessible Annie Creek sites and Site LC-1-BIO in 2021. These sites contained a variety of habitat types, and surface fines and fine sediments did not account for large proportions of the substrate. No trends in fines or surface fines were observed at these sites in recent years. Storm runoff and recreational usage of the dirt road adjacent to sites AC-1-BIO, AC-2-BIO, and LC-1-BIO likely contribute to the surface fines at these sites and not mining activities. Four-wheel drive vehicles have been observed during the 2021 and past sampling events. Road maintenance (i.e., filling in potholes) may also have contributed to fine sediment inputs into Annie Creek. No eroding banks were observed at any Annie Creek site during 2021. Eroding banks are strongly influenced by high flow events and areas of unvegetated or vulnerable streambanks are unrelated to small discharges of water into the Annie Creek drainage via the Wharf outfalls. While some differences have occurred from year to year at these sites, generally, the available habitat at these sites has been diverse, with a range of habitat types observed each year throughout the study period.

Conversely, fish habitat is not favorable at Site RV-2-BIO due to its small size, lack of habitat diversity, and large amount of surface fines and fine sediments, both of which increased from 2020. The higher percentages of fine substrates at the Ross Valley site are likely due to the low discharge typically found at this site and the accumulation of organic matter from the surrounding forest. Prolonged low flows due to drought conditions in 2020 and 2021 (Figure 4-1) reduced the flushing flow capacity to remove fine sediments.

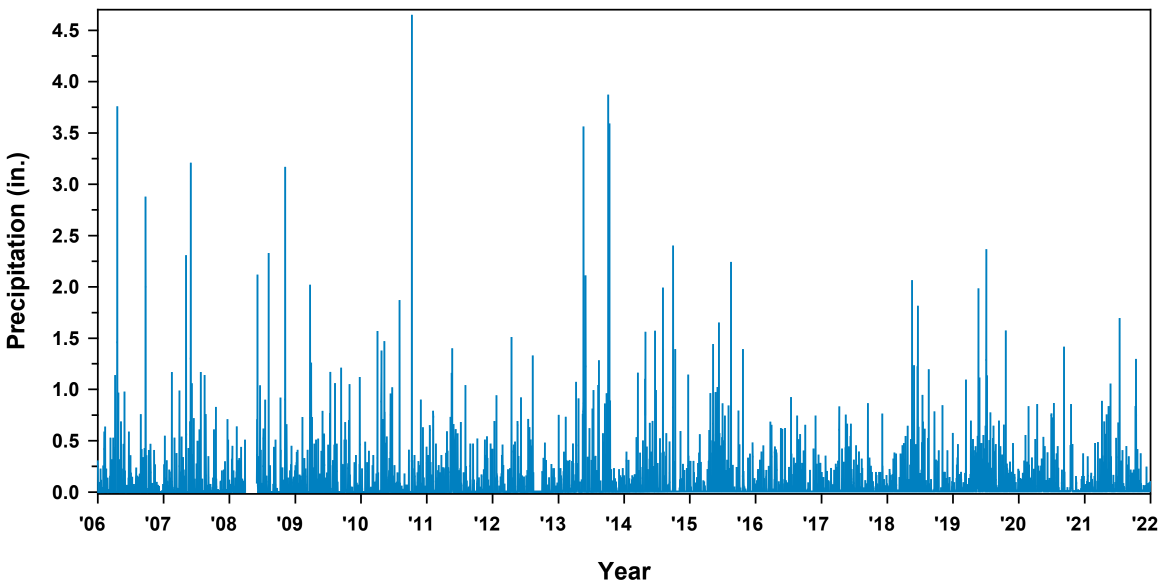


Figure 4-1: Cumulative daily precipitation in Lead, SD (GHCND:USC00394834) from 2006 to 2021 (NOAA 2020).

Precipitation has varied in the Black Hills since 1995 and changes in habitat related to flow have been observed in other years and at other sites. During the years 2007, 2013, 2018 and 2019, peak precipitation events were large and/or frequent (Figure 4-1) resulting in high discharge that caused minor changes to the habitat structure of Annie Creek, Ross Valley, and Lost Creek. In recent years, the peak precipitation in 2018 and 2019 are likely partially responsible for the general decrease in surface and fine sediments observed in Annie Creek and Lost Creek sites in those years. Peak rainfall events in the spring of 2016, 2017, 2020, and 2021 were not large and did not significantly alter habitat at Annie Creek, Ross Valley, and Lost Creek. In fact, precipitation in 2021 was similar to that in 2016, 2017, and 2020 and was accompanied by slight sedimentation at sites AC-1-BIO and RC-1-BIO. However, this relationship between flow and habitat change was not always apparent over the years and large and/or frequent precipitation in other years (i.e., 2010) did not result in habitat changes.

Habitat features at Site AC-1-BIO, Site AC-2-BIO, Site LC-1-BIO, and the Reno Creek reference site, Site RC-1-BIO, were similar. All sites were comprised mainly of low gradient riffles but contained a variety of small pools as well. Average and maximum depths were comparable at these three sites and eroding banks were absent. Overall, comparisons reveal no differences between the upper two Annie Creek sites and the reference site indicating that stream habitat in Annie Creek is not being affected by mining activities when compared to habitat at the reference site. Both sites on Annie Creek contain a mixture of habitat types, including deep water pool habitat suitable for fish. Substrate conditions are also favorable and indicate that interstitial spaces (habitat) are sufficient for macroinvertebrates. Site RV-2-BIO is not comparable to the reference site; however, these differences are due to the low discharge typically found at this site and the accumulation of organic matter from the surrounding forest.

4.1.3 Deadwood Creek, False Bottom Creek, and Cleopatra Creek

Habitat measurements were collected at Site DC-2-BIO on Deadwood Creek, and sites EFB-1-BIO and WFB-1-BIO on the East Fork and West Fork of False Bottom Creek, respectively (Table 4-5). Small residual pockets of water were present at Site CC-1A-BIO, but no flow was present, and the riffles were dry (Photo 4-6). Therefore, the decision was made to not sample Cleopatra Creek in 2021, as was the case in 2016, 2017, 2019, and 2020.

These biomonitoring sites are all located in the headwaters of comparatively small creeks and are characterized by relatively narrow water widths and shallow depths (Table 4-5). The number of habitat units at each of the sampling sites ranged from 12 to 17 units. All sites contained a mixture of fast water and slow water habitat types. Site DC-2-BIO was the only site dominated in terms of length by pool habitat, and Site WFB-1-BIO was the only site that contained eroding banks. Substrate compositions included a combination of sizes from fine sediment to boulders at all study sites in 2021 (Table 4-6). Fine sediment and boulders were the most abundant substrate at Site DC-2-BIO, gravel was most abundant at Site EFB-1-BIO, and course sediment and gravel at Site WFB-1-BIO. Surface fines and fine sediment were less abundant at Site EFB-1-BIO than at other sites. Iron oxide deposits were observed at Site WFB-1-BIO, similar to previous years of sampling on this fork, and sediment from eroding banks limiting interstitial spaces impacted the benthic invertebrates, fish, and periphyton at this site (Photo 4-7).

Table 4-5: Habitat characteristics for sites on Deadwood Creek, East Fork False Bottom Creek, West Fork False Bottom Creek, August 2021. HGR = high gradient riffle; LGR = low gradient riffle; SRN = step run complex; RUN = run; DMW, SLW, SMB, and SMW = types of pools.

Site/ Habitat Type	No. of Units	Total Length (m)	Average Water Width (m)	Average Water Depth (cm)	Average Maximum Depth (cm)	Eroding Bank (%)
DC-2-BIO						
LGR	5	29.3	1.3	6	11	0
DMW	1	8.5	0.8	7	13	0
RUN	5	30.7	1.5	9	19	0
SLW	1	3.3	1.3	21	25	0
SMW	1	4.2	2.5	22	30	0
SRN	2	33.7	1.1	7	17	0
EFB-1-BIO						
HGR	1	2.5	1.7	8	10	0
LGR	7	60.6	0.8	5	14	0
RUN	5	30.8	1.0	8	13	0
SMB	3	5.4	0.8	17	22	0
SMW	1	1.7	1.1	8	18	0
WFB-1-BIO						
LGR	4	57.6	0.6	5	10	0
DMW	1	13.1	0.7	2	12	0
SMB	2	4.5	1.1	13	20	0
SMW	3	7.4	1.0	9	18	3
RUN	2	19.4	1.4	7	12	0

Table 4-6: Substrate characteristics for sites at Deadwood Creek, West Fork False Bottom Creek, August 2021.

Site/ Habitat Type	Flow (cfs)	Average % Surface Fines	Substrate Composition (%)				
			Fine Sediment (≤ 0.8 mm)	Coarse Sediment (0.8 - 4.8 mm)	Gravel (4.8 - 75 mm)	Rubble (76 - 304 mm)	Boulder/ Bedrock (> 304 mm)
DC-2-BIO	0.05	40	23	14	18	21	24
EFB-1-BIO	0.11	19	8	23	31	28	11
WFB-1-BIO	<0.01	40	22	28	27	18	6

Photo 4-6: **Cleopatra Creek with no flowing water.**

Photo 4-7: **Iron oxide deposits at Site WFB-1-BIO.**

4.1.3.1 Summary of Habitat Conditions

Habitat characteristics have been relatively stable at sites DC-2-BIO, EFB-1-BIO and WFB-1-BIO in recent years. The location of these sites in the headwaters of small streams reduces the likelihood that high flow events leading to significant habitat changes or streambank erosion will occur. Variability in streamflow from year to year can lead to changes in average water depths and fine sediment deposition within these small streams. In fact, length of pool habitat was greater than riffle habitat in Site DC-2-BIO in 2019 to 2021 which has not been the case in many previous years. Surface fines increased at all sites from 2020 and fine sediment increased at DC-2-BIO and EFB-1-BIO which were the result of 2021 having low precipitation (Figure 4-1) and flows lower than in recent years. Conversely, surface fines and fine sediment at Site DC-2-BIO were lowest in 2019 as a result of high precipitation and flows in 2018 and 2019. The iron oxides historically observed on the substrate at WFB-1-BIO continue to be present and are due to impacts of groundwater contributions upstream of Site WFB-1-BIO.

Aquatic habitat at sites DC-2-BIO, EFB-1-BIO, and WFB-1-BIO were comparable to the Labrador Gulch reference site, LB-4-BIO, in 2021. Average widths and depths were similar between these sites and Site LB-4-BIO, although average and maximum depths were slightly greater in the pools at the Deadwood Creek and Labrador Gulch sites. Site LB-4-BIO has far less surface fines and fine sediment and more bedrock than the other sites as a result of its steep gradient and higher flows.

4.1.4 Fantail Creek, Nevada Gulch, and Stewart Gulch

The study sites at Fantail Creek, Nevada Gulch, and Stewart Gulch contained from 8 to 13 total habitat units, with low gradient riffles being the most prevalent habitat type within each site (Table 4-7). Average stream widths were narrower at Site FC-1-BIO than at sites NG-2-BIO and SG-1-BIO. Average water depths were similar between the sites except for deeper riffles at Site SG-1-BIO. This site also contained run habitats which were absent from the other sites. A range of habitat units, including riffles and pools formed by various elements, such as logs or boulders, were also present in each site. Eroding banks were not present at any site.

Substrate composition at these three sites included assorted sizes of substrate. Gravel was the most abundant substrate size class and surface fines were moderate at sites FC-1-BIO and NG-2-BIO which corresponds to low flows at these sites (Table 4-8). Flow at Site SG-1-BIO was much larger and is accompanied by low surface fines and course sediment being the most abundant size class. A large percentage of surface fines at Site FC-1-BIO in 2021 (Photo 4-7), particularly in pool habitat, indicates that sedimentation is occurring from Gilded Mountain Road and influenced to some degree by the new home construction adjacent to the creek (Photo 4-8). The location of this site in the headwaters of Fantail Creek limits the occurrence of high flow events which would flush large amounts of fine sediments downstream. A sand filter basin just north of the road captures runoff and sediment from the Golden Rewards mine area as well. Eroding banks were not observed at any site indicating good bank stability and a lack of fine sediment inputs into the site from unstable banks (Table 4-7). Differences in aquatic habitat among sites in 2021 were mainly due to the surrounding geology (substrate size) and proximity to roads, and not associated with Wharf mining activities (Table 4-8).

Table 4-7: Habitat characteristics for sites on Fantail Creek, Nevada Gulch, and Stewart Gulch, August 2021. HGR = high gradient riffle; LGR = low gradient riffle; RUN = run; SRN = step run complex; SLB, SMB, SPB, SPO, PMO, SMO and SPW = types of pools; and STP = step pool complex.

Site/ Habitat Type	No. of Units	Total Length (m)	Average Water Width (m)	Average Water Depth (cm)	Average Maximum Depth (cm)	Eroding Bank (%)
FC-1-BIO						
LGR	4	78.0	0.5	4	9	0
SMB	2	3.0	0.8	9	14	0
STP	2	19.0	0.8	21	23	0

Site/ Habitat Type	No. of Units	Total Length (m)	Average Water Width (m)	Average Water Depth (cm)	Average Maximum Depth (cm)	Eroding Bank (%)
NG-2-BIO						
HGR	1	4.6	1.0	3	12	0
LGR	6	86.8	0.9	4	13	0
SLB	1	4.3	0.6	8	18	0
SPB	1	2.1	1.0	12	20	0
SMW	1	2.2	1.5	18	24	0
SG-1-BIO						
HGR	1	3.2	1.9	11	19	0
LGR	6	54.6	1.7	11	21	0
RUN	2	19.5	1.9	17	23	0
PMO	1	5.2	2.7	27	32	0
SPB	1	3.3	2.5	19	37	0
SMO	1	1.7	2.7	18	25	0
SRN	1	10.7	1.8	6	21	0

Table 4-8: Substrate characteristics for sites on Fantail Creek, Nevada Gulch and Stewart Gulch, August 2021.

Site/ Habitat Type	Flow (cfs)	Average % Surface Fines	Substrate Composition (%)				
			Fine Sediment (≤ 0.8 mm)	Coarse Sediment (0.8 - 4.8 mm)	Gravel (4.8 - 75 mm)	Rubble (76 - 304 mm)	Boulder/ Bedrock (> 304 mm)
FC-1-BIO	0.04	41	21	21	47	9	3
NG-2-BIO	0.02	30	12	17	40	19	14
SG-1-BIO	0.96	10	18	36	20	17	8

Photo 4-8: Fine sediment accumulation in Fantail Creek channel, August 2021.

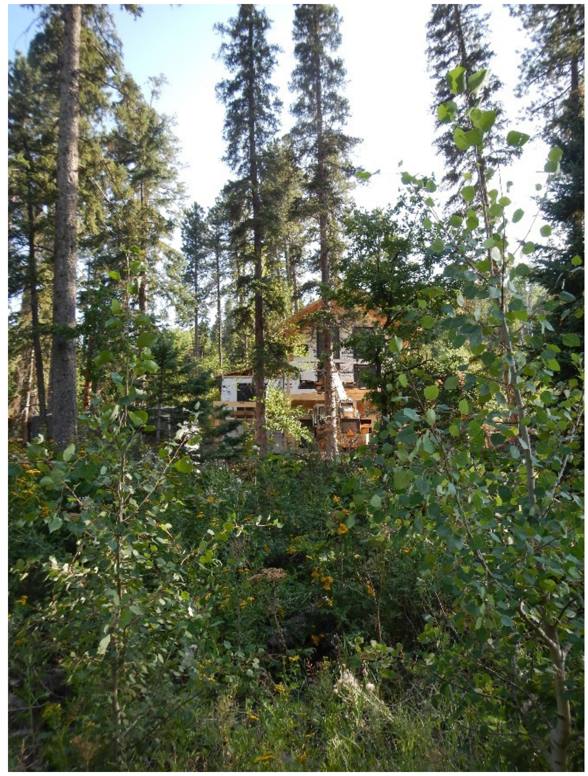


Photo 4-9: New home construction adjacent to Site FC-1-BIO likely acting as a source of sediment to the stream, August 2021.

In recent years, Stewart Gulch has contained an abundant growth of watercress (*Nasturtium officinale*, Photo 4-9), which provides good habitat for first year age class trout and those less than one year old, referred to as young-of-the-year. However, in 2021, the areal extent of the watercress was less than compared to previous years. The spring upstream of the site continues to provide nitrogen-rich groundwater to the stream which can facilitate the growth of watercress. The reduced watercress coverage may be the natural cycle of the plant growth as 2021 was a dry-year with no apparent scouring precipitation events that could reduce the watercress coverage.

Photo 4-10: Abundance of watercress along margins of Stewart Gulch, August 2017 (left) and less coverage in August 2021 (right).

4.1.4.1 Summary of Habitat Conditions

Overall, habitat characteristics at sites on Fantail Creek, Nevada Gulch, and Stewart Gulch are favorable for benthic macroinvertebrates. Stewart Gulch contained a relatively low percentage of fines, while Nevada Gulch and Fantail Creek contained a larger, but moderate, amount which increased from 2020. However, all sites have more surface fines or fine sediment in 2021 as compared to 2018 and 2019 when precipitation and flows were high (Figure 4-1). Each site also contained ample amounts of riffle habitat which is preferred by many benthic macroinvertebrates. Similarly, substrate characteristics indicated sufficient amounts of gravel for mayflies, stoneflies, and caddisflies to inhabit interstitial spaces. Stream width and depth and abundance of pool habitat at Site SG-1-BIO is also sufficient to support fish. Fluctuations in habitat characteristics at these sites are, for the most part, minimal or related to natural variations in local weather events.

Habitat conditions at Fantail Creek and Nevada Gulch were similar to those on Reno Creek at the refence site, Site RC-1-BIO. All streams are small streams with low discharge, narrow widths, shallow water depths, and minimal erosion which are conditions typical of their headwater locations. These sites are all dominated by low gradient riffles but contain multiple pools as well. The percentage of surface fines was greater at Fantail Creek. All three sites contained abundant gravel and rubble substrates which provide favorable interstitial spaces for benthic macroinvertebrates to inhabit. However, flow is too low to support trout populations.

Stewart Gulch and the reference site on Labrador Gulch, Site LG-4-BIO, have comparable habitats. Both sites contain a relatively wide variety of habitat types with ample riffle and pool habitat, lack eroding banks, and have a good mixture of habitat types to support healthy populations of Brook Trout (*See* Fish Populations).

4.2 Fish Populations

4.2.1 Labrador Gulch

Thirty-three Brook Trout, ranging from 85 to 200 mm, were collected from Site LB-4-BIO (Table 4-9; Figure 4-2). Age classes sampled included YOY (<100 mm), first year age class (100 - 150 mm), and second year-plus age class (>150 mm, Figure 4-2). Abundance of these classes in 2021 were similar to recent years with fish less than 150 mm collected in 2021 being within Q2 and fish greater than 150 mm being within Q1 of fish abundance since 1993 when sampling began at this site (Figure 4-3; Table 4-10). YOY collected in 2021 and in previous years indicate this segment of the stream supports all life stages of Brook Trout. Density and biomass values have fluctuated since 1995 (Figure 4-4; Figure 4-5) but both were within the ranges previously sampled (both within Q1) in 2021 (Figure 4-6; Figure 4-7).

Table 4-9: Fish population metrics for Labrador Gulch and Reno Creek, August 2021.

Site/Species	Number Collected	Mean Length (mm)	Mean Weight (g)	Density #/ha ± 95% C.I.	Biomass (kg/ha)	Relative Weight (W_r)	Condition (K)
LB-4-BIO							
Brook Trout	33	133.5	26.2	2,063 ± 125	54.05	86.0	0.95
RC-1-BIO							
Brook Trout	8	139.6	25.7	667 ± 0	17.14	83.3	0.91
Brown Trout	3	172.3	55.3	250 ± 0	13.83	97.6	1.08

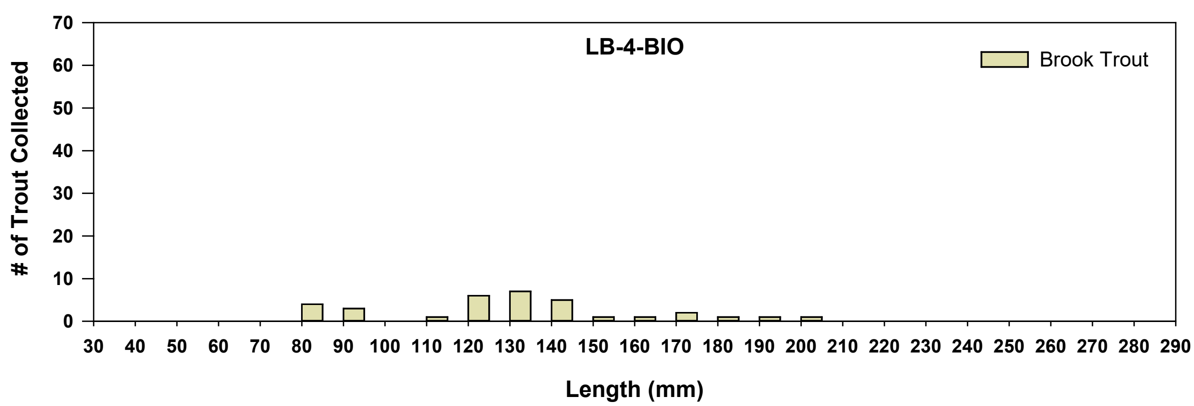


Figure 4-2: Length-frequency distribution for Brook Trout collected in Labrador Gulch at Site LB-4-BIO, August 2021.

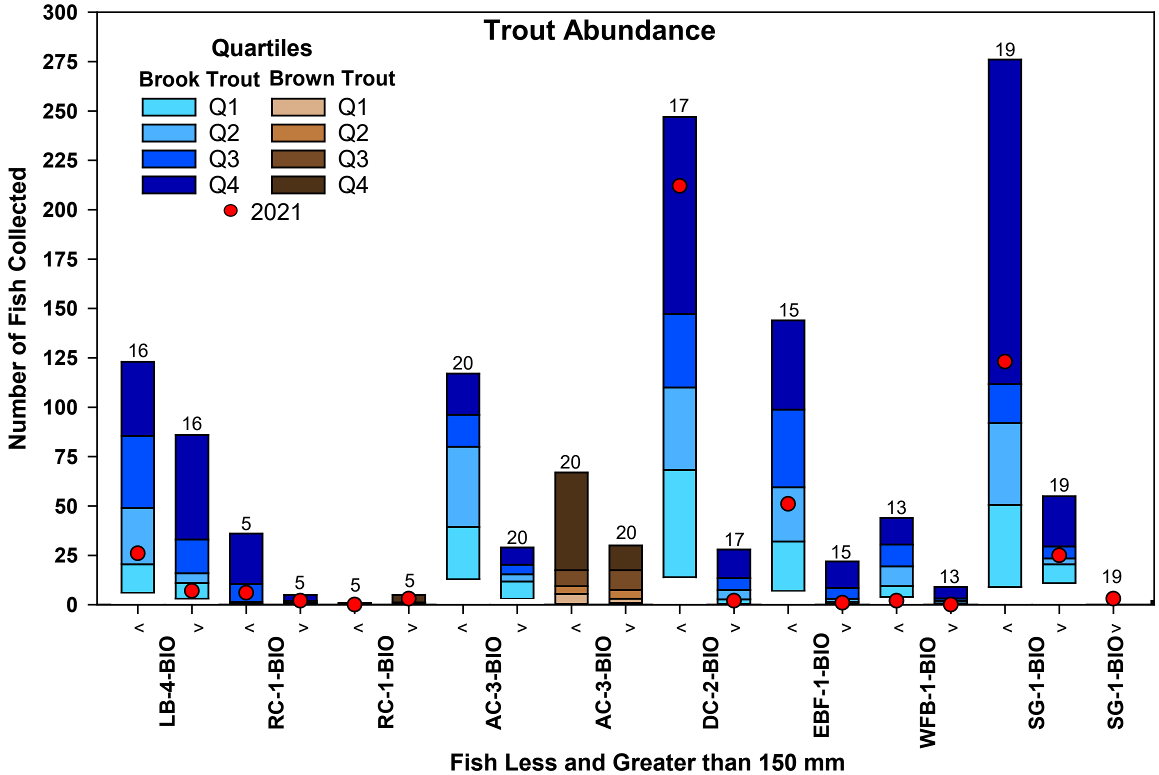


Figure 4-3: Quartile plots by length from 1990 through 2020 with overlayed 2021 data of Brook Trout and Brown Trout collected at Wharf sites. Brown Trout collected in 2019 at EFB-1-BIO (n = 1) and Brown Trout <150 mm at SG-1-BIO in 2021 (n = 1) are not displayed. Number of years sampled is displayed above each quartile and includes 2021.

Mean condition factor and relative weight values at Site LB-4-BIO were less than the optimal ranges and similar to other sites sampled in 2021, indicating that Brook Trout at all sites may be ecologically stressed. This site is not located downstream of mining activities, and lower condition factor and relative weight values indicate variations in fish condition due to natural factors such as low precipitation (i.e., lower flows) and warmer water temperatures in recent years (Figure 4-1). No increasing or decreasing trends since 2006 were observed for abundance, density, biomass, or condition factor, although, relative weight (Wr) has significantly decreased since 2006 ($p = 0.013$, slope = -1.588 Wr/year). The frequency of fish sampling at Site LB-4-BIO has been variable since 2006, as a result, some interannual variability may not be evident in the data.

Table 4-10: Quartile data (minimums, percentiles, and maximums) for Brook and Brown Trout less than or greater than 150 mm at Wharf sites from 1990 – 2020 and 2021 values. NS = Not sampled.

Site/Species	Abundance (<150 mm)						Abundance (≥150 mm)					
	Min	25th	50th	75th	Max	2021	Min	25th	50th	75th	Max	2021
LB-4-BIO												
Brook Trout	6	20.5	49.0	85.5	123	26	3	11.0	16.0	33.0	86	7
RC-1-BIO												
Brook Trout	0	0.8	1.5	10.5	36	6	1	1.0	1.0	2.0	5	2
Brown Trout	0	0.0	0.0	0.3	1	0	0	0.0	0.0	1.3	5	3
AC--BIO3												
Brook Trout	13	39.5	80.0	96.3	117	NS	3	11.8	15.5	20.3	29	NS
Brown Trout	0	5.5	9.5	17.5	67	NS	1	3.0	7.5	17.5	30	NS
DC-2-BIO												
Brook Trout	14	68.3	110.0	147.3	247	212	0	2.8	7.5	13.5	28	2
EFB-1-BIO												
Brook Trout	7	32.0	59.5	98.8	144	51	0	1.5	3.0	8.5	22	1
Brown Trout	0	0.0	0.0	0.0	1	0	0	0.0	0.0	0.0	0	0
WFB-1-BIO												
Brook Trout	4	9.5	19.5	30.5	44	2	0	0.8	2.0	3.3	9	0
SG-1-BIO												
Brook Trout	9	50.5	92.0	111.8	276	123	11	20.5	23.5	29.5	55	25
Brown Trout	0	0.0	0.0	0.0	0	1	0	0.0	0.0	0.0	2	3

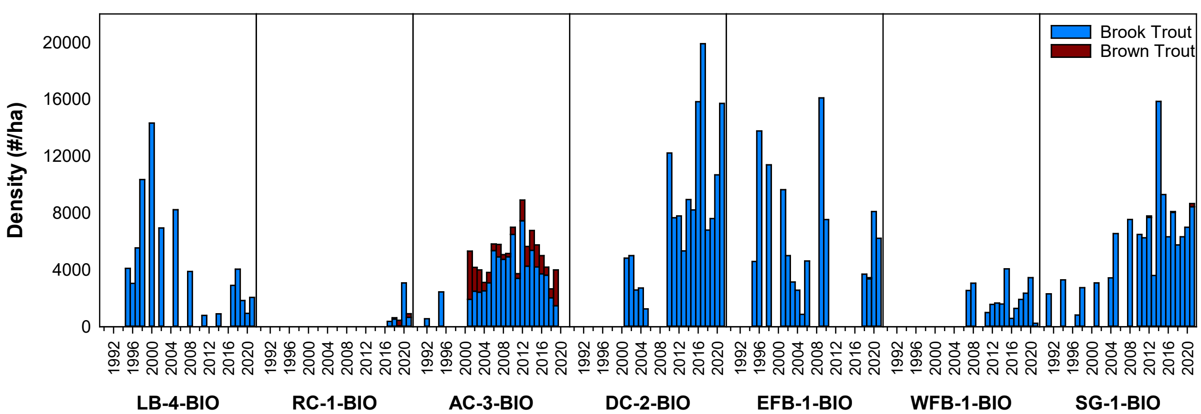


Figure 4-4: Trout population density values at the Wharf sites from 1990-2021. Data for AC-3-BIO is stacked.

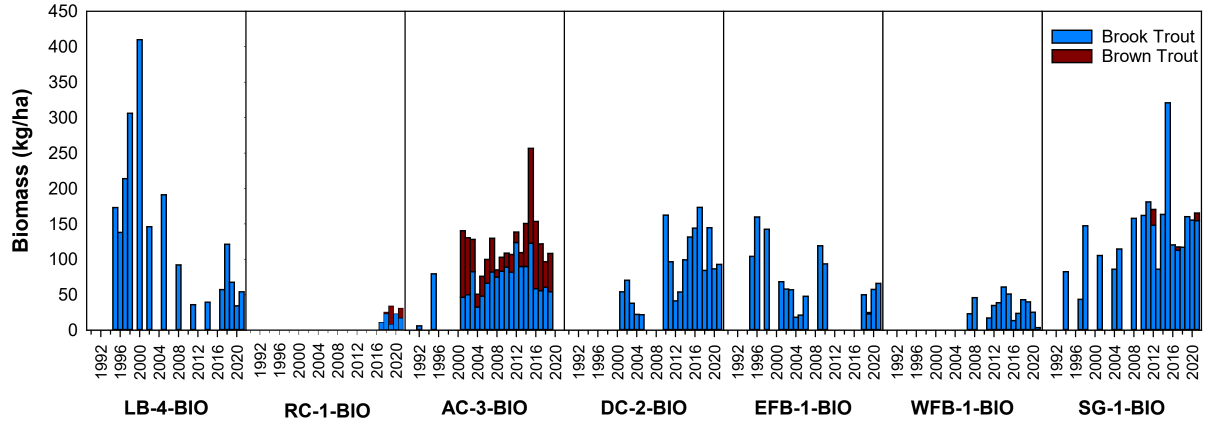


Figure 4-5: Trout population biomass values at the Wharf sites from 1990 to 2021. Data for AC-3-BIO is stacked. Brown trout collected in 2019 at EFB-1-BIO (n = 1) and in 2012, 2017, and 2021 at SG-1-BIO (n = 2, n = 1, n = 4, respectively) are not displayed.

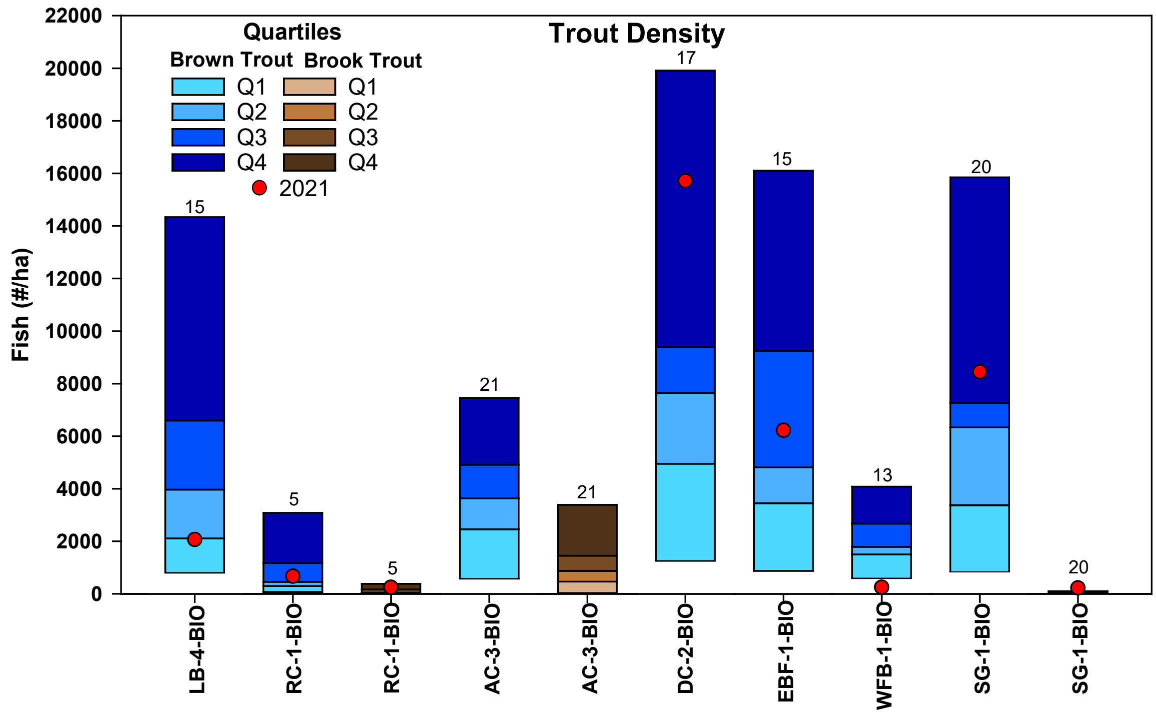


Figure 4-6: Quartile plots of population density values from 1990 through 2020 with overlayed 2021 data of Brook and Brown Trout at Wharf sites. The Brown Trout collected in 2019 at EFB-1-BIO (n = 1) is not displayed. Number of years sampled is displayed above each quartile and includes 2021.

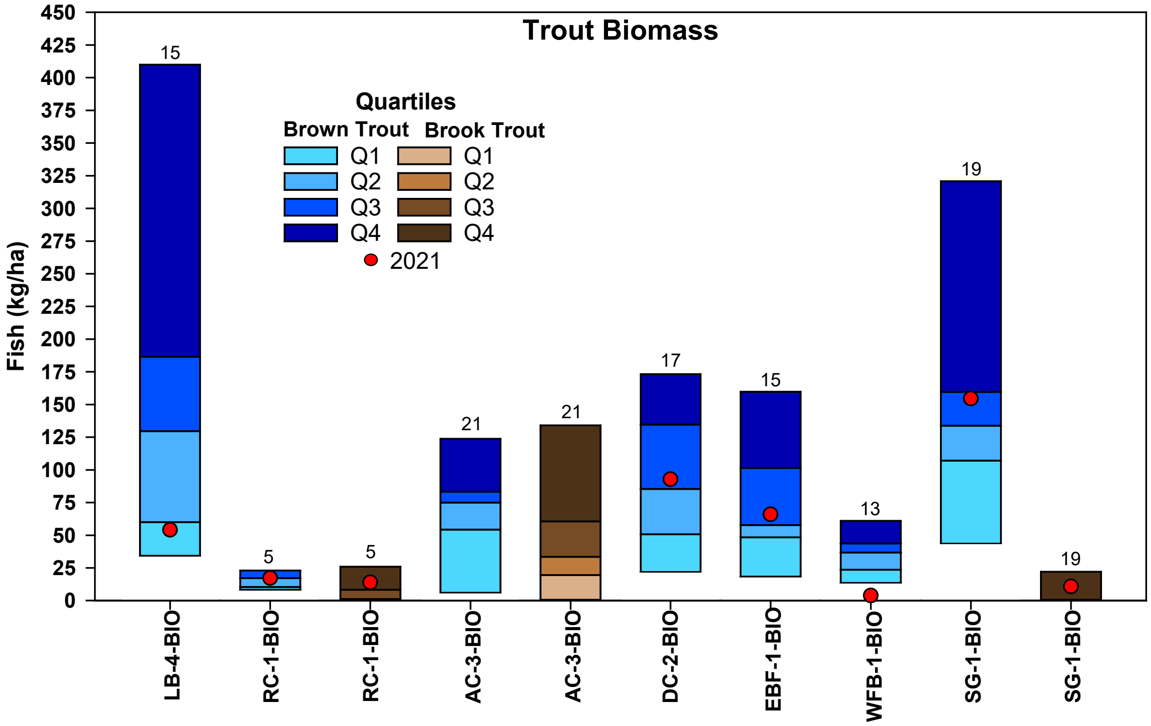


Figure 4-7: Quartile plots of population biomass values from 1990 through 2020 with overlayed 2021 data of Brook and Brown Trout at Wharf sites. The Brown Trout collected in 2019 at EFB-1-BIO (n = 1) is not displayed. Number of years sampled is displayed above each quartile and includes 2021.

4.2.2 Reno Creek

Eight Brook Trout and three Brown Trout were collected at Site RC-1-BIO in 2021 (Table 4-9; Appendix A) which is similar to the number collected in the first three years of sampling (2017-2019). In 2020, 37 Brook Trout and no Brown Trout were collected at Site RC-1-BIO. Most Brook Trout in 2021 ranged in length from 128 to 173 mm, with one fish at 95 mm, indicating that Brook Trout were all first and second year-plus age classes except for one YOY (Figure 4-8). In contrast, most fish in 2020 were YOY. The abundance of fish less than 150 mm and greater than 150 mm in 2021 was similar to that sampled since 2017 (within Q3 and on the 75th centile, respectively; Figure 4-3, Table 4-10). Brown Trout ranged from 160 to 181 mm indicating all second year-plus age class fish. This species was recently collected in 2018 and 2019 and abundance in 2021 was similar (within Q4) with all fish greater than 150 mm.

Brook Trout density and biomass values have fluctuated since 2017 (Figure 4-4; Figure 4-5). but both were similar in 2021 to previous years (both within Q3, respectively; Figure 4-6; Figure 4-7). Density in 2020 was much larger than other years. During greater flows in fall of 2019, second year-plus age class fish likely moved into small streams, such as Reno Creek, to spawn and then moved downstream. An abundant amount of YOY were then observed in 2020 but likely moved downstream by 2021. Brown Trout density and biomass was also similar to previous years (both within Q4).

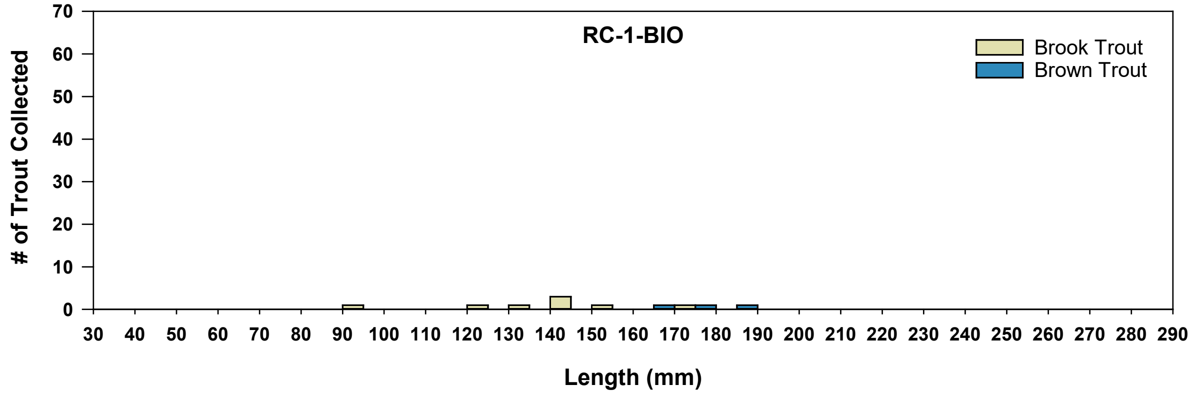


Figure 4-8: Length-frequency distribution for fish collected at Site RC-1-BIO in Reno Creek, August 2021.

As with Labrador Gulch, mean condition factor and relative weight values at Site RC-1-BIO were less than the optimal but similar to other sites sampled in 2021. Considering the reference type conditions found in Reno Creek basin, the lower condition factor and relative weight values at this site indicate variations in fish condition due to natural factors despite some development in the upper watershed.

No qualitative trends for abundance, density, or biomass or significant trends for relative weight or condition factor were observed for this site. However, trends are difficult to identify with only four years of data and should be interpreted with caution.

4.2.3 Annie Creek

4.2.3.1 Site AC-1-BIO

No fish were collected during sampling at Site AC-1-BIO in August 2021 (Appendix A). Fish have not been collected at this site since sampling began in 1992 (Mariah Associates, Inc. 1992b; CEC 1996b, 1999a, 2002a, 2003a, 2004a, 2005a, and 2006a; GEI 2007a, 2008a, 2008c, 2009a, 2010a, 2011a, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). The absence of fish at this site reflects its headwater location upstream of perennial fish habitat (C&A 1993). Also, small waterfalls that either impede or prevent upstream fish migration are common in this section of Annie Creek.

4.2.3.2 Site AC-2-BIO

No fish were collected during sampling at Site AC-2-BIO in August 2021 (Appendix A). Mountain Sucker were collected in large numbers in the early 1990s, but populations declined after the 1995 ammonia and cyanide release, high BOD in 2007, and 2008 clean up (Mariah Associates, Inc. 1990 and 1992b; CEC 1996b, 1999a, 2002a, 2003a, 2004a, 2005a, and 2006a; GEI 2007a, 2008a, 2008c, 2009a, and 2010a). No fish have been collected at this site since 2010 (GEI 2011a, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021).

4.2.3.3 Site AC-3-BIO

The majority of lower Annie Creek, including most of Site AC-3-BIO was not accessible for fish collection in 2021 due to extensive deadfall from the tornado activity on July 8, 2020. Only large sections in the upper half of the reach were accessible. This area was qualitatively sampled with electrofishing equipment, but the population size was not estimated based on the seven Brown Trout that were collected, of which five fish were retained for tissue analysis.

Sampling began at this site in 1992, with annual sampling beginning in 2001, and Brook and Brown Trout have been collected in most years. Brook Trout of all age classes were collected in all years, but YOY were more numerous than second year-plus age class fish in all years but 2018 and 2019 (Figure 4-3; Table 4-10). This change is part of an overall decreasing trend in YOY and first year class Brook Trout abundance since 2009 which was likely due to competition with Brown Trout which has increased in density since 2009. Even with this trend, the quartile data indicate that resident populations of Brook Trout utilize the lower portion of Annie Creek. Because high numbers of YOY trout are sampled during some years, indicating natural reproduction occurs in or near this reach.

YOY and first and second year-plus age class Brown Trout have been collected from Site AC-3-BIO in most years since sampling began, although second year-plus age class Brown Trout are generally present in lower numbers (Figure 4-3; Table 4-10). However, a large proportion of the Brown Trout sampled in 2014 to 2019 were second year-plus age class fish, and YOY were absent in 2014, 2015, 2017, and 2018. This indicates that in some years Site AC-3-BIO may act as a spawning and rearing stream for Brown Trout from Spearfish Creek, which is approximately 200 m downstream of the site.

The highest Brook Trout density and biomass values were observed in 2012 and values have trended downwards since (Figure 4-4; Figure 4-5). Brown Trout density at Site AC-3-BIO was greatest in 2019 while biomass was greatest in 2015. The dominant species as measured by biomass has been Brook Trout in most years, except for in 2015 through 2017, when Brown Trout comprised more of the total biomass at the site. Prior to 2016, Brown Trout were collected in a more limited size range than Brook Trout in most years, indicating that this stream served as spawning and rearing habitat for Brown Trout. However, the population included a wider size range of Brown Trout in 2016 through 2019, indicating that this species may now inhabit Site AC-3 year-round.

Condition factors and relative weights for both species have been within, or approaching, optimal ranges in most years since 2006. Brook Trout condition factor significantly improved from 2006 to 2019 ($p = 0.020$, slope = 0.012 K/year) while relative weight did not trend, and no trend was observed for Brown Trout for either metric. Increased precipitation in 2018 following the 2017 drought in the Black Hills (NOAA 2020) appears to have positively affected relative weights and condition factors, indicating that low flows and generally associated higher water temperatures during the previous summer may have been causing stress to fish inhabiting Site AC-3-BIO. The long-term median relative weight and condition factor for Brook Trout were not significantly different than the reference site, Site LB-4-BIO.

4.2.4 Ross Valley

No fish were collected in this stream during sampling at Site RV-2-BIO from 2006, when sampling began at this site, through 2021 (Table 4-11; Appendix A; GEI 2007b, 2008c, 2009b, 2010b, 2011b, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). The small stream size and habitat in Ross Valley are unsuitable to support fish and steep gradient downstream of the site prevents upstream migration.

Table 4-11: Fish population metrics for sites on Ross Valley, Lost Camp Gulch, Deadwood Creek, West Fork False Bottom Creek, and Cleopatra Creek, August 2021.

Site/Species	Number Collected	Mean Length (mm)	Mean Weight (g)	Density (#/ha) ± 95% C.I.	Biomass (kg/ha)	Relative Weight (W_r)	Condition (K)
RV-2-BIO	No Fish	--	--	--	--	--	--
LC-1-BIO	No Fish	--	--	--	--	--	--
DC-2-BIO							
Brook Trout	214	79.8	5.9	15,714 ± 500	92.71	86.1	0.85
EFB-1-BIO							
Brook Trout	52	98.9	10.6	6,222 ± 889	65.95	91.6	0.93
WFB-1-BIO							
Brook Trout	2	115.5	15.0	250 ± 0	3.75	--	0.97
CC-1A-BIO	No Fish	--	--	--	--	--	--

4.2.5 Lost Camp Gulch

No fish have been collected from Site LC-1-BIO from 2010, when sampling began at this site, through 2021 (Table 4-11; Appendix A; GEI 2011b, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). Very low flows were observed in this site during 2016 and 2017 sampling, indicating that aquatic habitat at this site may be limited during drier years. Similar to sites AC-1-BIO, AC-2-BIO, and RV-2-BIO, the physical locations of Lost Camp Gulch, upstream of fish barriers, likely precludes any establishment of fish population.

4.2.6 Deadwood Creek

In August 2021, 214 Brook Trout were collected from Site DC-2-BIO (Table 4-11). Fish ranged in length from 45 mm to 175 mm (Appendix A), indicating that multiple age classes of fish were present (Figure 4-9). Young of the year were especially abundant in 2021, as was the case in 2016 and 2017, and more numerous than second year-plus age class Brook Trout (Figure 4-3; Table 4-10) as was the case for all years. In addition, the abundance of fish less than 150 mm and greater than 150 mm were similar to the annual abundances since 2001 when fish sampling began at this site (within Q4 and Q1, respectively). Multiple size classes of Brook Trout have been present in all years in which Site DC-2-BIO was sampled, indicating consistent natural reproduction of Brook Trout in this reach of Deadwood Creek. Over the years, far fewer second year-plus age class sized fish have been observed compared to YOY and first year age class fish at this site, indicating this section of stream may serve primarily as a spawning and rearing area. Deep water habitat is also minimal at this site, limiting suitable habitat for larger, second year-plus age class trout.

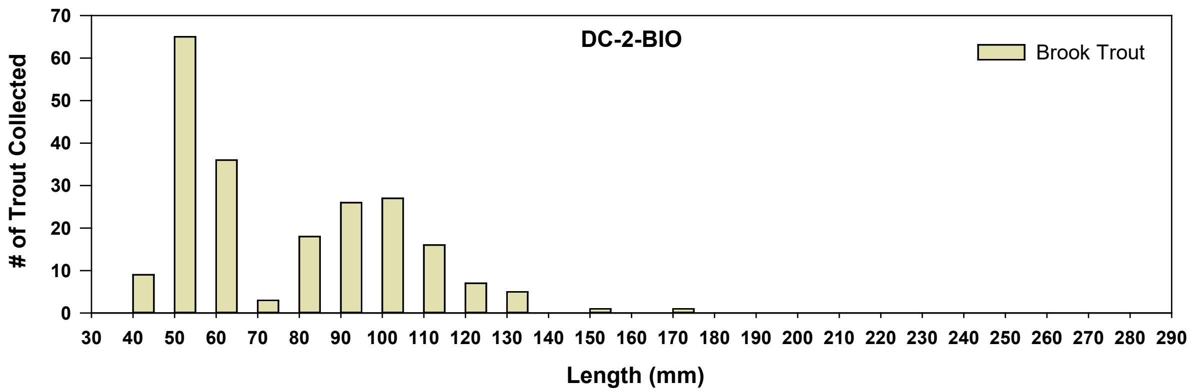


Figure 4-9: Length-frequency distribution for fish collected at Site DC-2-BIO in Deadwood Creek, August 2021.

Trout population density and biomass values have fluctuated since 2001 (Figure 4-4; Figure 4-5), and the 2021 values were within the range previously sampled (within Q4 and Q3, respectively; Figure 4-6; Figure 4-7). The highest Brook Trout density and biomass values were observed in 2017.

The relative weight and condition factor values of Brook Trout at this site were both less than the optimal ranges and have been decreasing since 2010, although not significantly. These trends may be due to low flows and the high density of fish, primarily YOY and first year age class fish, sampled at this site in previous years. The historic high densities in low flow may have led to competition for limited food and habitat resources and could increase stress, reducing the overall condition of the Brook Trout population at this site. No other trends were observed for Brook Trout abundance, density, or biomass metrics.

When compared to the reference site, LB-4-BIO, the number of total fish, density, and biomass were greater at Site DC-2-BIO, but fish were larger (mean length and weight) and the condition factor was slightly higher at Site LB-4-BIO (Table 4-9; Table 4-11), although, only condition factor was significantly different between the sites ($p = 0.039$). This indicates that the high numbers of fish may cause competition for food and habitat resources at Site DC-2-BIO.

4.2.7 False Bottom Creek

The site location on False Bottom Creek was inadvertently changed between the East Fork to the West Fork in some years; these locations are now designated as Site EFB-1-BIO and Site

WFB-1-BIO, respectively. In 2006 and 2009-2010, Site EFB-1-BIO was sampled, and in 2007-2008 and 2011-2017, Site WFB-1-BIO was sampled. Both sites have been sampled since 2018. Fish populations from these two locations are discussed separately below.

4.2.7.1 Site EFB-1-BIO

Fifty-two Brook Trout were sampled at Site EFB-1-BIO in August 2021 (Table 4-11), and sizes ranged from 63 to 156 mm. All but one fish were YOY and first year age class fish, indicating natural reproduction in or near this site (Figure 4-10; Appendix A). In addition, the abundance of fish less than and greater than 150 mm in 2021 was similar to that sampled since 1995 when sampling began at this site (within Q2 and Q1, respectively; Figure 4-3; Table 4-10). Over the years, fewer second year-plus age class fish have been observed compared to YOY and first year age class fish at this site, indicating this section of stream may serve primarily as a spawning and rearing area.

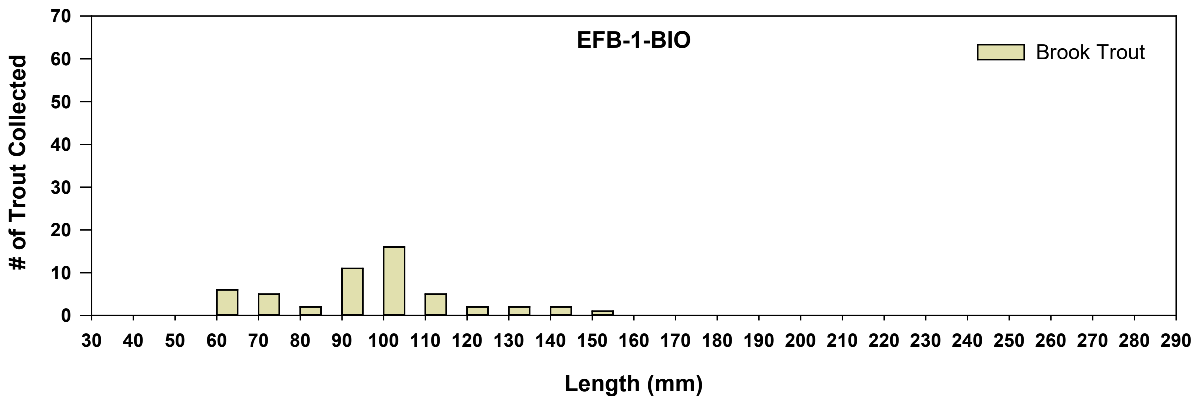


Figure 4-10: Length-frequency distribution for fish collected at Site EFB-1-BIO in False Bottom Creek, 2021.

Brook Trout population density and biomass at Site EFB-1-BIO have fluctuated from year to year since 1995 (Figure 4-4; Figure 4-5) and 2021 values were within the ranges previously sampled (within Q3; Figure 4-6; Figure 4-7). The relative weight and condition factor values of Brook Trout at Site EFB-1-BIO were both less than the optimal ranges. Determining trends in the data at Site EFB-1-BIO is less than ideal given the variable sampling years, but no increasing or decreasing trends since 2006 were observed for abundance, density, and biomass. In addition, significant trends were not present for relative weight or condition factor.

When compared to the reference site on Labrador Gulch, Brook Trout density, biomass, and relative weight were greater at Site EFB-1-BIO while condition factor was greater at Site LB-4-BIO (Table 4-9; Table 4-11), although not significantly.

4.2.7.2 Site WFB-1-BIO

Two Brook Trout were collected at Site WFB-1-BIO in August 2021 (Table 4-11). These fish were 113 to 118 mm which is within the size range for first year age class fish (Figure 4-11; Appendix A). Fish less than 150 mm have been more numerous than larger Brook Trout in most years since 2007 when sampling began (Figure 4-3; Table 4-10). In addition, the number of Brook Trout less than 150 mm collected in 2021 was less than all previous years (less than Q1) while the absence of second year-plus age class has occurred in other years since 2007 when sampling began at this site (within Q1).

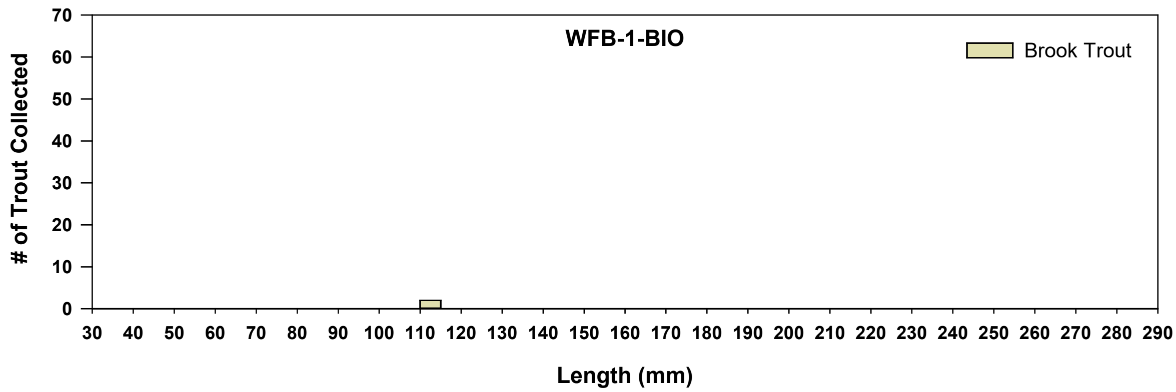


Figure 4-11: Length-frequency distribution for fish collected at Site WFB-1-BIO in False Bottom Creek, August 2021.

Distribution of Brook Trout size classes has varied greatly over time. Only in 2015 were each of the three size classes represented by more than five fish in one year. In 2007, 2008, and 2020, YOY and first year age class fish were present while second year-plus fish were absent or present in very low numbers. In addition, few YOY were sampled in 2014 while first and second year-plus age class fish were found in greater numbers. From 2011 to 2013 and 2017 to 2019 first year fish were plentiful while YOY and second year-plus age class fish were absent or present in very low numbers. Lastly, in 2016 and 2021, no YOY were collected, second year-plus age class fish were absent or low in numbers, and first year fish were sampled infrequently. This variation in size distribution observed at this site indicates that successful recruitment may be limited in some years, and fish may migrate into or out of Site WFB-1-BIO depending on habitat suitability.

Population density and biomass values have also fluctuated since 2007 (Figure 4-4; Figure 4-5) with the 2021 values being less than all previous years (less than Q1; Figure 4-6; Figure 4-7). The values observed in 2021 are similar to that observed in 2016 (Figure 4-4; Figure 4-5). Density and biomass at Site WFB-1-BIO have typically been lower than at Site EFB-1-BIO, with maximum values comparable to the median density and biomass values at Site EFB-1-BIO.

No Brook Trout greater than 120 mm were collected in 2021. However, mean relative weight at this site has slightly decreased between years since 2014 to a low of 86.2 in 2020. The mean Brook Trout condition factor was the highest of any site but still below 1.00. No increasing or decreasing trends since 2007 were observed for density or biomass and no significant trend was found for condition factor.

The suitability of habitat on the West Fork of False Bottom Creek may be strongly influenced by streamflow in a given year. For instance, low Brook Trout abundance, densities, and biomasses in 2011, 2016, and 2021 coincided with dry years (Figure 4-1). In addition, the percent surface fines has historically been moderately poor in Site WFB-1-BIO which reduces habitat suitability for fish. Iron deposits are also often visible on the substrate within WFB-1-BIO, indicating water quality issues that may impact the fish population during some years.

When compared to the reference site on Labrador Gulch, the West Fork revealed much lower density and biomass estimates (Table 4-9; Table 4-11). The condition factor values were very similar between the sites and not significantly different.

4.2.8 McKinley Gulch

McKinley Gulch was dry during August 2021 and was not sampled. Electrofishing has never been performed at this site since its inclusion in the Wharf monitoring program in 2006 due to no stream flow (GEI 2007a, 2008a, 2008c, 2009a, 2010a, 2011a, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021).

4.2.9 Cleopatra Creek

In 2021, residual pockets of water were present at Site CC-1A-BIO, but no surface flow occurred over the riffles and most pools were dry. Therefore, the decision was made to not sample (Table 4-11), the same as in 2016, 2017, 2019 and 2020 when no flowing water was present at the site (GEI 2017a, 2018a, 2020, and 2021). In years when flowing water was present (2006, when sampling began, through 2015 and 2018), no fish were collected at Site CC-1A-BIO (GEI 2007a, 2008a, 2008c, 2009a, 2010a, 2011a, 2012, 2013, 2014, 2015, 2016, and 2019). Historically, Brook Trout density and biomass had been high at the former Cleopatra Creek site, CC-1-BIO, which is located further downstream near the confluence with the East Fork Cleopatra Creek (CEC 2006a). However, the current site location is in the headwaters of Cleopatra Creek, where the lack of perennial flows is not suitable for fish.

4.2.10 Fantail Creek

Sampling in 2021 at Site FC-1-BIO on Fantail Creek produced no fish (Table 4-12). No fish have been collected during sampling in Fantail Creek except for six Brook Trout sampled in 1998 (GRMC 1987 and 1992; CEC 1998b, 1999b, 2002b, 2005b, and 2006b; GEI 2009b, 2011b, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). The presence of fish in 1998 was probably due to higher than normal summer flows, which allowed fish to move upstream from Whitetail Creek during the late summer sampling period (CEC 1999b). Usually, the small stream size and low flows make this site unsuitable for fish.

Table 4-12: Fish population metrics for Fantail Creek, Nevada Gulch, and Stewart Gulch, August 2021.

Site/Species	Number Collected	Mean Length (mm)	Mean Weight (g)	Density #/ha ± 95% C.I.	Biomass (kg/ha)	Relative Weight (W_r)	Condition (K)
FC-1-BIO	No Fish	--	--	--	--	--	--
NG-2-BIO	No Fish	--	--	--	--	--	--
SG-1-BIO							
Brook Trout	148	115.3	18.3	8,444 ± 333	154.53	83.4	0.86
Brown Trout	4	168.0	48.3	222 ± 111	10.72	89.0	0.99

4.2.11 Nevada Gulch

No fish were found during the 2021 sampling event at NG-2-BIO (Table 4-12; Appendix A). Similar to the Fantail Creek site, the only year in which fish were present in lower Nevada Gulch since sampling began in 1989 was in 1998, when a single Brook Trout was collected at Site NG-2-BIO (GRMC 1987 and 1992; CEC 1998b, 1999b, 2002b, 2005b, and 2006b; GEI 2009b, 2011b, 2012, 2013, 2014, 2015, 2016, 2017a, 2018a, 2019, 2020, and 2021). The higher flows in that year likely allowed this fish to move upstream from Whitetail Creek into lower Nevada Gulch (CEC 1999b). During most years, the small stream size and low flows make this site unsuitable for fish.

4.2.12 Stewart Gulch

One hundred and forty-eight Brook Trout were collected from Stewart Gulch in August 2021 (Table 4-12). The Brook Trout ranged in size from 52 to 212 mm, indicating an abundant number of YOY, first year, and second year-plus age class Brook Trout were collected (Figure 4-12; Appendix A). The distribution of age classes in 2021 is similar to most recent years where the population consists of all age classes and YOY was dominant. In addition, the abundance of Brook Trout less than and greater than 150 mm in 2021 were similar to that sampled in previous years (within Q4 and Q3, respectively). These data indicate that conditions at the Stewart Gulch site support successful spawning and rearing and that habitat and water quality in Stewart Gulch have sustained fish populations. The abundant macrophyte beds in portions of this site likely act as favorable rearing habitat for YOY Brook Trout and help to protect them from predation by larger fish, birds, or mammals.

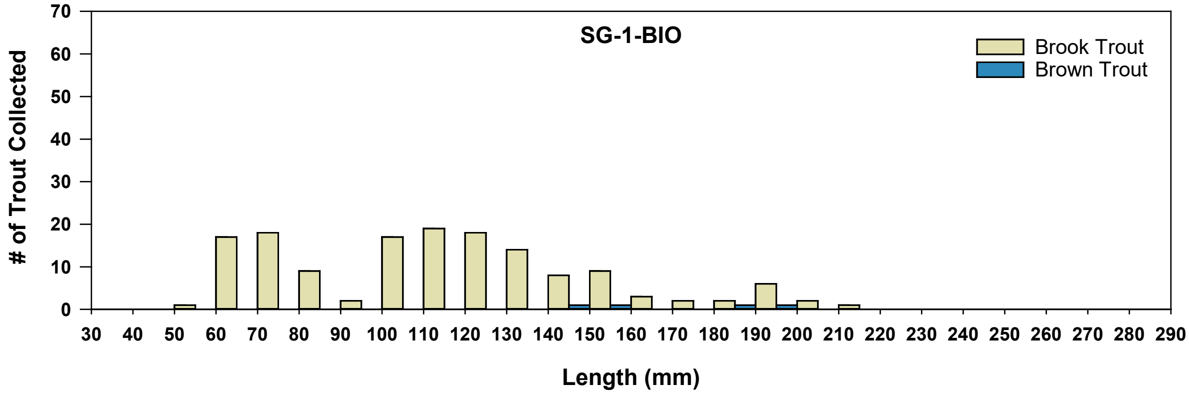


Figure 4-12: Length-frequency distribution for Brook Trout collected in Stewart Gulch, August 2021.

In 2021, two first year and two second year-plus age class Brown Trout were collected at Site SG-1-BIO. Brown Trout have been absent during all other years of sampling in Stewart Gulch except for in 2012 and 2017 when two and one Brown Trout, respectively, were collected (GEI 2012; GEI 2018a). These individuals likely moved upstream from the downstream reaches of Whitetail Creek or from Whitewood Creek.

Brook Trout density and biomass values have fluctuated since 1990 (Figure 4-4; Figure 4-5) and 2021 values were within the range previously sampled (within Q4 and Q3, respectively; Figure 4-6; Figure 4-7). The highest density of fish since 2006 was observed in 2014 (GEI 2015), and the highest observed biomass occurred in 2015 (Figure 4-4; Figure 4-5).

The average relative weight and condition factor values for Brook Trout were less than optimal ranges and both have significantly decreased since 2006 ($p = 0.016$ and 0.014, respectively, and slope = -0.869 and -0.010 Wr/year, respectively; Table 4-12). No increasing or decreasing trends since 2006 were observed for abundance, density, or biomass.

Density and biomass of Brook Trout at Site SG-1-BIO were both greater, although not significantly different from the reference site, Site LB-4-BIO, while relative weight and condition factor were lower (Table 4-9; Table 4-12). In addition, relative weight and condition factor at both sites were below the optimal range of 95.0 to 105.0 and average value of 1.00, respectively. Generally healthy populations were found at both sites, even though some adult fish may be slightly below optimal weight. More robust fish condition values are often measured at the Stewart Gulch site. This is likely partially due to inputs of nitrogen from a spring upstream of the site, which facilitates the growth of watercress, creating both favorable habitat and enriching the lower levels of the food web (GEI 2015). Stewart Gulch also contains multiple deep pools and abundant aquatic vegetation, which act as favorable habitat for adult and juvenile Brook Trout, respectively.

4.2.13 Fish Tissue Selenium

Five Brook Trout were collected at Site EFB-1-BIO in 2021 for selenium whole-body tissue analysis (Table 4-13; Appendix B). Fish were similar in size, ranging from 126 to 156 mm. Brook Trout of this size have likely spent most of their life near the vicinity of the study site and have tissues concentrations characteristic of resident fish. All replicate dry weight selenium concentrations in 2021 were less than the EPA whole-body fish tissue criterion of 8.5 µg/g dw for aquatic life and less than the whole- body selenium genus mean chronic value (GMCV) of 34.9 µg/g dw for *Salvelinus*.

Five Brown Trout were also collected from Site AC-3-BIO during the qualitative electrofishing in accessible portions of the creek. These fish were similar in size, ranging from 155 to 191 mm in length and were likely also resident fish. Historically, only Brook Trout have been sampled from Site AC-3-BIO, but given the post-disturbance conditions in 2021, only Brown Trout were present. Selenium dry weight concentrations of all but one fish were less than the EPA whole-body fish tissue criterion while all fish were less than the whole-body selenium GMCV for the most sensitive trout genus—*Oncorhynchus* (11.6 µg/g dw).

Table 4-13: Percent solids and selenium (Se) concentrations in whole-body trout at East False Bottom and Annie Creek sites, August 2021.

Site	Species	Replicate	Percent Solids	Wet Weight Se (µg/g)	Dry Weight Se (µg/g)
EFB-1-BIO	Brook Trout	1	24.1	0.86	3.56
		2	23.0	0.91	3.96
		3	21.6	0.97	4.48
		4	22.5	0.94	4.18
		5	23.6	1.13	4.79
Geometric mean			22.9	0.96	4.17
AC-3-BIO	Brown Trout	1	25.3	1.79	7.08
		2	24.7	1.96	7.94
		3	23.1	1.81	7.84
		4	24.2	1.75	7.23
		5	24.7	2.13	8.62
Geometric mean			24.4	1.88	7.72

4.3 Benthic Macroinvertebrate Populations

4.3.1 Labrador Gulch

4.3.1.1 2021 Data

In August 2021, richness, composition, tolerance, and trophic habit metric values were predominantly mildly to very favorable at Site LB-4-BIO (Table 4-14; Table 4-15; Appendix

C). Specifically, Density, Number of Taxa, and Number of EPT Taxa values were all good when compared to historical data in the Black Hills and very high compared to other sites in 2021; Diversity indicated a healthy invertebrate community; the HBI score was "Very Good"; and trophic habit metrics indicated a variety of feeding types. Mayflies and caddisflies were abundant, however, 55% of the Ephemeroptera were *Baetis tricaudatus cx.,* the dominant species at the site, which indicate that most mayflies were relatively tolerant of environmental stressors. This dominance resulted in a fair EPT Index and Percent Sensitive EPT Taxa values. In addition, life history metrics indicated that few taxa were long-lived. Overall, metric values for this site indicate that stream conditions (biotic and abiotic) in 2021 supported a rich and diverse benthic macroinvertebrate community, including numerous sensitive species.

Table 4-14: Macroinvertebrate Density (number of organisms/sample) at the reference sites on Labrador Gulch and Reno Creek, August 2021.

Taxa	LB-4-BIO	RC-1-BIO
INSECTA		
Collembola (springtails)	10	5
Ephemeroptera (mayflies)	1,425	510
Plecoptera (stoneflies)	455	685
Megaloptera (alderflies)	20	35
Coleoptera (beetles)	190	170
Trichoptera (caddisflies)	1,071	295
Diptera (true flies)	1,791	1,990
HYDRACARINA (water mites)	35	25
CRUSTACEA		
Amphipoda (Scuds)	--	380
ANNELIDA (segmented worms)		
Oligochaeta (worms)	45	120
MOLLUSCA		
Pelecypoda (clams)	40	5

Table 4-15: Macroinvertebrate population metrics at the reference sites on Labrador Gulch and Reno Creek, August 2021. NA = Not applicable.

Metric	LB-4-BIO	RC-1-BIO
RICHNESS METRICS		
Density (#/sample)	5,082	4,220
Number of Taxa	55	46
Number of EPT Taxa	18	15
COMPOSITION METRICS		
Shannon-Weaver Diversity Index	4.51	4.52
Percent Sensitive EPT Taxa	23.6%	28.3%
EPT Index	32.7%	32.6%

Metric	LB-4-BIO	RC-1-BIO
Percent *Baetis sp.*	55.4%	19.6%
Number of non-*Baetis* Ephemeroptera	635	410
Percent Chironomidae	31.6%	44.5%
Number of Plecoptera Taxa	4	6
Percent Abundance of Oligochaetes & Hirudinea	0.9%	2.8%
Dominant Taxon (Tolerance value)	*Baetis tricaudatus cx.*	*Micropsectra sp.*
Percent Dominant Taxon	15.5%	16.0%
Number of Common Taxa	NA	NA
Community Loss Index	NA	NA
TOLERANCE METRICS		
Hilsenhoff Biotic Index	4.08	4.61
Percent Tolerant Taxa	25.5%	30.4%
Percent Intolerant Taxa	50.9%	47.8%
Number of Intolerant Taxa	28	22
TROPHIC HABIT METRICS		
Number of Predator Taxa	16	12
Percent Collector-Gatherers	51.9%	56.0%
Number of Shredder Taxa	8	7
LIFE HISTORY METRICS		
Number of Univoltine Taxa	27	22
Number of Merovoltine Taxa	4	2
Percent Semivoltine Taxa	1.8%	2.2%
Number of Semivoltine Taxa	1	1

4.3.1.2 Historic Data

The macroinvertebrate community at Site LB-4-BIO changed moderately from 2006 to 2021. Richness metric have changed over time with Density and Number of Taxa significantly increasing (Table 4-16; Figure 4-13; Appendix C) and Number of EPT Taxa being greater in 2021 than previous years (previous maximums of 17 in 2006; Figure 4-14). For composition metrics, Diversity has never been less than 2.5, indicating a history of a healthy invertebrate community, and was healthier in 2021 than other years (previous maximums of 4.40 in 2006). The Ephemeroptera assemblage was comprised mostly by *Baetis spp.*, a common and relatively tolerant mayfly genus, and the Percent Sensitive EPT Taxa was poorer than in previous years (previous minimums of 23.8 in 2013). However, Percent Dominant Taxon also improved over other years (previous minimums of 17% in 2017). The only composition metrics with significant trends were Number of non-*Baetis* Ephemeroptera which has improved over time and Percent Chironomidae which has worsened since 2006 (Table 4-16).

Tolerance, trophic habit, and life history metric values have also shown few trends over time with the exception of HBI which has significantly worsened, ranging from “Excellent” (2.5 in 2007) to “Fair” (5.7 in 2019). The increasing HBI score indicates a greater abundance of more tolerant species such as the Chironomidae in recent years. Also, the Number of

Univoltine Taxa in 2021 was poorer than any other year (previous maximums of 26 in 2019). In contrast, Number of Predator and Merovoltine Taxa have significantly improved while Number of Intolerant Taxa in 2021 were better than any other year (previous maximums of 27 in 2019). Overall, most metrics have not significantly improved or worsened over time and most of the 2021 metrics where within the range observed in recent years.

Table 4-16: Slope of significant trends ($p < 0.05$) for benthic macroinvertebrate population metrics at the reference sites on Labrador Gulch and Reno Creek, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant. NA = Not applicable.

Taxa	Change Expected Following Environmental Disturbance	LB-4-BIO	RC-1-BIO
RICHNESS METRICS			
Density (#/sample)	Decrease	+ 253.91	--
Number of Taxa	Usually Decrease	+ 0.87	+ 4.10
Number of EPT Taxa	Decrease	--	+ 1.90
COMPOSITION METRICS			
Shannon-Weaver Diversity Index	Decrease	--	+ 0.24
Percent Sensitive EPT Taxa	Decrease	--	+ 0.02
EPT Index	Decrease	--	--
Percent *Baetis sp.*	Increase	--	--
Number of non-*Baetis* Ephemeroptera	Decrease	+ 35.99	--
Percent Chironomidae	Increase	+ 0.02	--
Number of Plecoptera Taxa	Decrease	--	--
Percent Abundance of Oligochaetes & Hirudinea	Variable	--	--
Percent Dominant Taxon	Increase	--	- 0.05
Number of Common Taxa	Decrease	NA	NA
Community Loss Index	Increase	NA	NA
TOLERANCE METRICS			
Hilsenhoff Biotic Index	Increase	+ 0.08	--
Percent Tolerant Taxa	Increase	--	--
Percent Intolerant Taxa	Decrease	--	--
Number of Intolerant Taxa	Decrease	--	+ 1.90
TROPHIC HABIT METRICS			
Number of Predator Taxa	Decrease	+ 0.45	--
Percent Collector-Gatherers	Variable	--	--
Number of Shredder Taxa	Decrease	--	+ 1.20
LIFE HISTORY METRICS			
Number of Univoltine Taxa	Increase	--	+ 1.80
Number of Merovoltine Taxa	Decrease	+ 0.17	+ 0.50
Number of Semivoltine Taxa	Decrease	--	--
Percent Semivoltine Taxa	Decrease	--	--

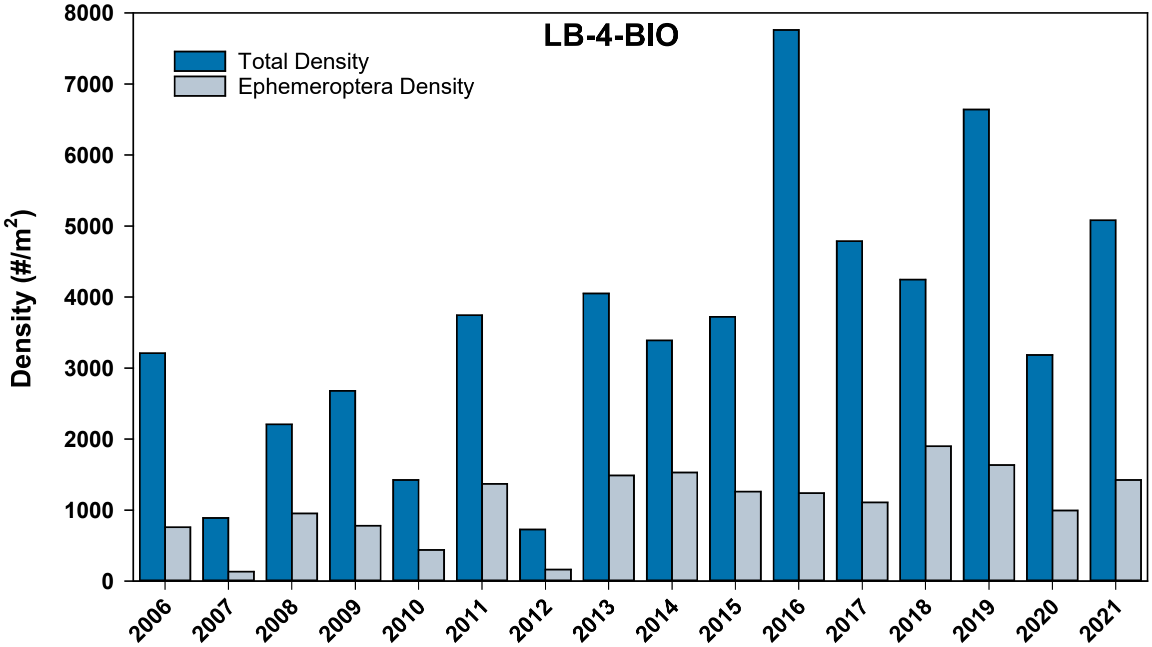


Figure 4-13: Macroinvertebrate density metrics for Site LB-4-BIO on Labrador Gulch, 2006 - 2021.

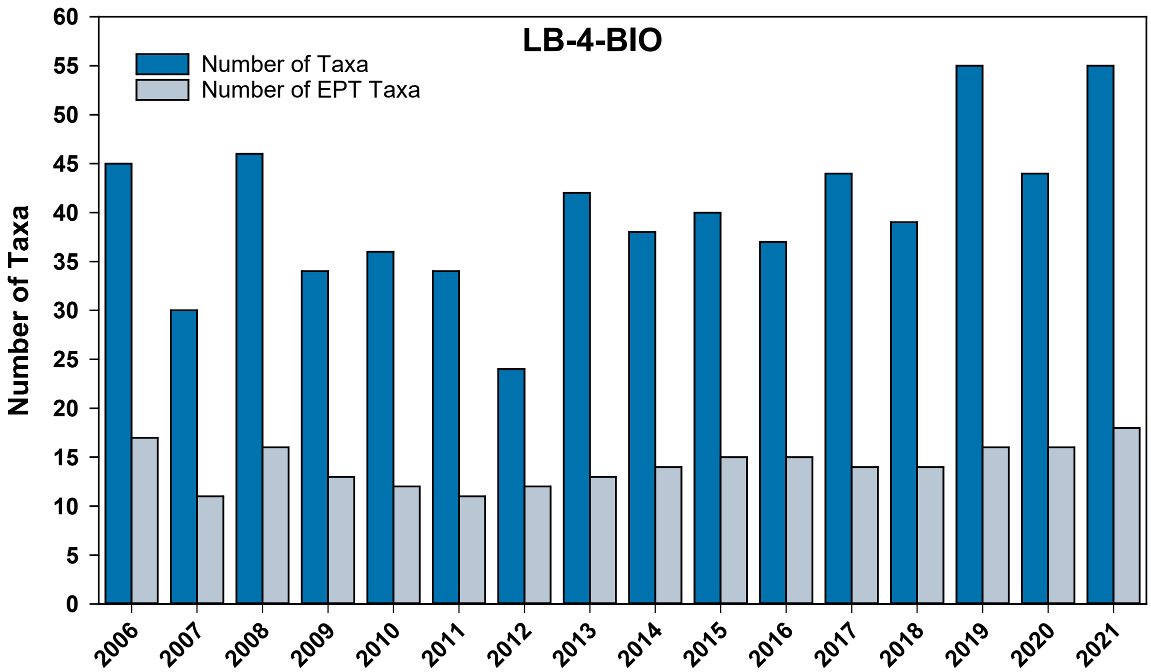


Figure 4-14: Macroinvertebrate taxa richness metrics for Site LB-4-BIO on Labrador Gulch, 2006 - 2021.

4.3.2 Reno Creek

4.3.2.1 2021 Data

Richness, composition, tolerance, trophic habit, and life history metric values were mostly mildly to very favorable at Site RC-1-BIO in August 2021 (Table 4-14; Table 4-15; Appendix C). Specifically, Density, Number of Taxa, Number of EPT Taxa, and EPT Index values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a healthy invertebrate community; the HBI indicated a "Good" benthic macroinvertebrate community including intolerant species; and a variety of feeding groups were present. *Micropsectra sp.*, a tolerant Diptera, was the most dominant taxon resulting in poorer metric values for Percent Chironomidae, Tolerant Taxa, and Collector-Gatherers values being poor. Overall, despite the abundance of *Micropsectra sp.*, stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community, including numerous sensitive species.

4.3.2.2 Historic Data

From 2017, when sampling began at Site RC-1-BIO, to 2021, the macroinvertebrate community has changed moderately. In the five years of sampling, the richness metrics, Number of Taxa and Number of EPT Taxa have significantly increased (Table 4-16; Figure 4-16; Appendix C) and were greater in 2021 than previous years (previous maximums of 3,855 in 2019 and 45 in 2020, respectively). For composition metrics, Diversity significantly improved from 2017, was healthier in 2021 than previous years (previous maximum of 4.18 in 2020), and has been greater than 2.5 in all years of sampling, indicating a healthy invertebrate community. In addition, Percent Sensitive EPT Taxa has significantly improved and was greater in 2021 than other years (previous maximum of 26.7% in 2020) and Number of Plecoptera Taxa and Percent Abundance of Oligochaetes and Hirudinea were also greater (previous maximums of 5 and 1.6%, respectively, in 2019). Percent Dominant Taxon has significantly improved and was lower in 2021 than prior years (previous minimum of 20.5% in 2020). *Baetis spp.*, a common and relatively tolerant mayfly genus, has typically been the dominant taxon at this site. The tolerance metric, HBI has ranged from "Fair" (5.64 in 2017) to "Very Good" (3.65 in 2020), and Number of Intolerant Taxa has significantly improved. Lastly, Number of Shredder Taxa, a trophic habit metric, has significantly improved over the years while the life history metrics, Number of Univoltine and Merovoltine Taxa, have significantly worsened and improved, respectively. Number of Merovoltine Taxa also were greater in 2021 than previous years (previous maximum of 1 in 2019 and 2020). Overall, the macroinvertebrate community appears to be consistently healthy and generally improving in health. However, trends should be carefully interpreted at this site given the limited number of years this site has been sampled.

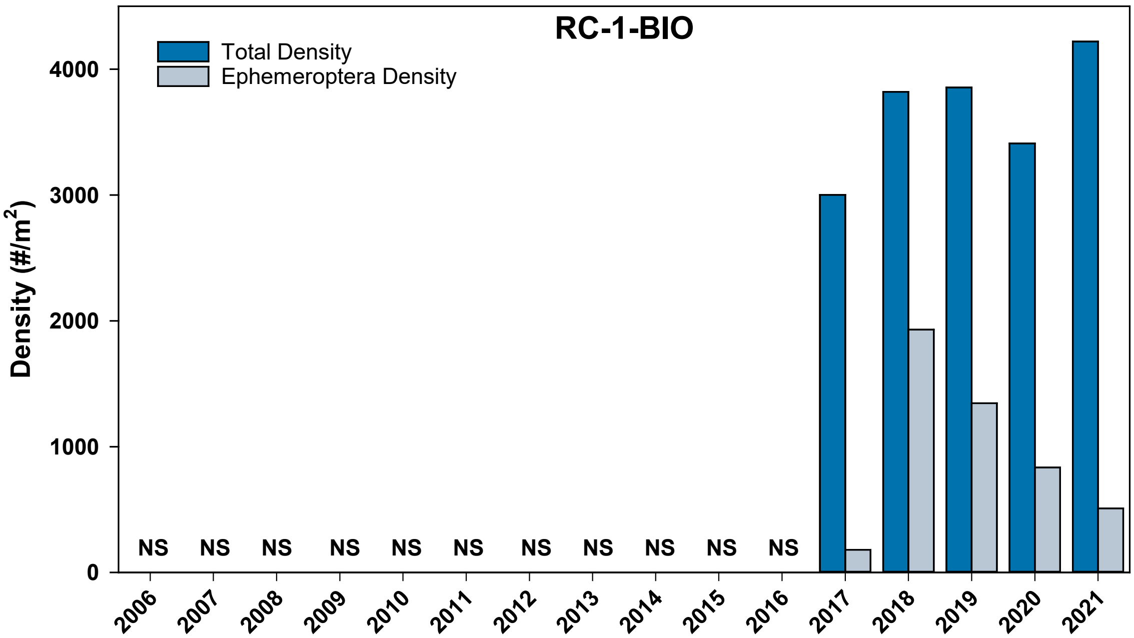


Figure 4-15: Macroinvertebrate density metrics at Site RC-1-BIO on Reno Creek, 2006 - 2021. NS = Not Sampled.

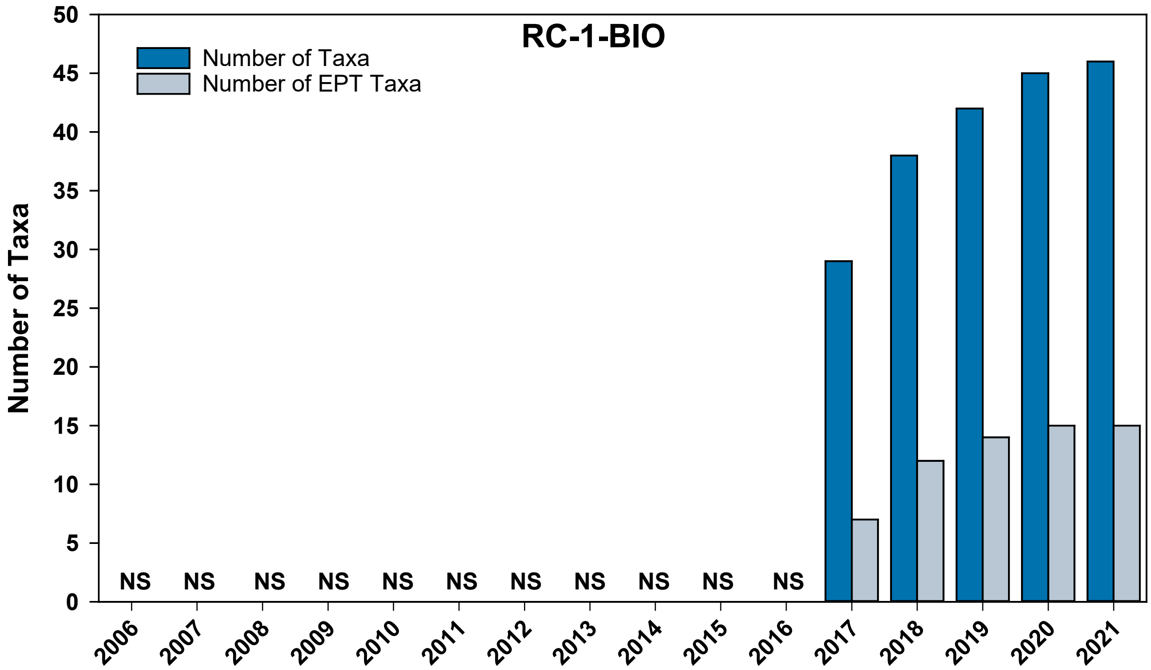


Figure 4-16: Macroinvertebrate taxa richness metrics for Site RC-1-BIO on Reno Creek, 2006 - 2021. NS = Not Sampled.

4.3.3 Annie Creek

4.3.3.1 Site AC-1-BIO

4.3.3.1.1 2021 Data

In August 2021, richness, composition, tolerance, and trophic habit metric values were largely mildly to very favorable at Site AC-1-BIO (Table 4-17; Table 4-18; Appendix C). For example, Density, Number of Taxa, Number of EPT Taxa, and EPT Index values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a balanced invertebrate community; the HBI indicated a "Very Good" benthic macroinvertebrate community including intolerant species; a diversity of feeding groups were present; and the dominant taxon, *Lepidostoma sp.,* is a sensitive caddisfly. Only life history metrics were poor indicating a dominance of short life cycle taxa. Overall, stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community, including numerous sensitive species.

Table 4-17: Macroinvertebrate density (number of organisms/sample) at Annie Creek, August 2021.

Taxa	AC-1-BIO	AC-2-BIO
INSECTA		
Ephemeroptera (mayflies)	345	805
Plecoptera (stoneflies)	960	410
Coleoptera (beetles)	840	755
Trichoptera (caddisflies)	1,750	345
Diptera (true flies)	1,780	1,030
HYDRACARINA (water mites)	85	25
TURBELLARIA (flatworms)	65	15
NEMATODA (round worms)	--	5
ANNELIDA (segmented worms)		
Oligochaeta (worms)	315	30
MOLLUSCA		
Gastropoda (snails)	--	5
Pelecypoda (clams)	20	5

Benthic macroinvertebrate richness, composition, tolerance, trophic habit, and life history metric values at Site AC-1-BIO were comparable to those at the reference site, Site RC-1-BIO. Both sites contained favorable or fair values for Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI. However, Percent *Baetis sp.,* Number of non-*Baetis* Ephemeroptera, and Number of Plecoptera Taxa were more favorable at the reference site than at Site AC-1-BIO while Percent Chironomidae, HBI, and Percent Collector-Gatherers was less favorable at the reference site. The Community Loss Index indicates that approximately half of the taxa present at Site AC-1-BIO were also present at the reference site. Overall, despite the differences, the similarities between the site were numerous and both sites contained a healthy benthic macroinvertebrate community in August 2021.

Table 4-18: Macroinvertebrate population metrics at Annie Creek, August 2021.

Metric	AC-1-BIO	AC-2-BIO
RICHNESS METRICS		
Density (#/sample)	6,160	3,425
Number of Taxa	43	52
Number of EPT Taxa	12	18
COMPOSITION METRICS		
Shannon-Weaver Diversity Index	4.13	4.56
Percent Sensitive EPT Taxa	20.9%	28.8%
EPT Index	27.9%	34.6%
Percent *Baetis sp.*	44.9%	52.2%
Number of non-*Baetis* Ephemeroptera	190	385
Percent Chironomidae	20.9%	22.2%
Number of Plecoptera Taxa	3	5
Percent Abundance of Oligochaetes & Hirudinea	5.1%	0.9%
Dominant Taxon (Tolerance value)	*Lepidostoma sp.*	*Heterlimnius corpulentus*
Percent Dominant Taxon	16.1%	13.0%
Number of Common Taxa	25	36
Community Loss Index	48.8%	36.5%
TOLERANCE METRICS		
Hilsenhoff Biotic Index	3.62	4.20
Percent Tolerant Taxa	27.9%	21.2%
Percent Intolerant Taxa	46.5%	61.5%
Number of Intolerant Taxa	20	32
TROPHIC HABIT METRICS		
Number of Predator Taxa	11	13
Percent Collector-Gatherers	32.8%	51.5%
Number of Shredder Taxa	8	8
LIFE HISTORY METRICS		
Number of Univoltine Taxa	19	26
Number of Merovoltine Taxa	1	1
Percent Semivoltine Taxa	0.0%	1.9%
Number of Semivoltine Taxa	0	1

4.3.3.1.2 Historic Data

The macroinvertebrate community at Site AC-1-BIO has changed substantially from 2006 to 2021. The high concentrations of BOD, ammonia, and cyanide and the disturbance caused by removal of excess organic matter from Annie Creek in 2007 resulted in most metric values being particularly poor in that and following years. Since then, richness metric values have significantly improved except for Density which has varied widely (1,247 in 2012 to 14,054 in 2017; Figure 4-17; Figure 4-18; Appendix C). The composition metric values, Diversity, EPT Index, Number of non-*Baetis* Ephemeroptera, Percent Dominant Taxon, Number of Common Taxa, and Community Loss Index have all significantly improved since 2006 (Table 4-19). The trend for Number of non-*Baetis* Ephemeroptera is the result of this metric being consistently zero prior to 2016. Diversity has been greater than 2.5 in all but two, non-recent years (2007 and 2012), indicating a more recent history of a healthy invertebrate community. Number of Common Taxa was also greater in 2021 than in previous years (previous maximum of 21 in 2020). The HBI tolerance metric has not significantly trended over time, ranging from “Fairly Poor” (6.79 in 2008) to “Excellent” (2.68 in 2012). The tolerance metrics, Percent and Number of Intolerant Taxa, and trophic habit metrics, Number of Predator and Shredder Taxa, also significantly improved since 2006. The only metric to decline in quality from 2006 to 2021 was the life history metric Number of Univoltine Taxa.

Overall, data in 2021 showed negligible change and metric values were similar to previous years. Almost half of the metrics have improved from 2006 to 2021, indicating that the macroinvertebrate community has become healthier over time, particularly since the disturbances in 2007. As Site AC-1-BIO represents the headwaters of the drainage, limited populations of invertebrates were present in the immediate vicinity to repopulate the area after the disturbance. Upstream colonization (i.e., adult insects flying upstream to lay eggs near Site AC-1-BIO) by insects from areas downstream was responsible for slowly returning populations at Site AC-1-BIO to similar conditions observed prior to the 2007 disturbances (Williams and Hynes 1976; Williams 1980; Hawkins and Sedell 1990; Johnson and Vaughn 1995). In addition, historic median benthic macroinvertebrate community metric values (subset of Abundance, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, HBI described in Section 3.6) between Site AC-1-BIO and its reference sites, Site RC-1-BIO, are not significantly different ($p > 0.05$). Overall, Site AC-1-BIO currently contains a rich and diverse community, with numerous intolerant taxa, indicative of healthy stream conditions.

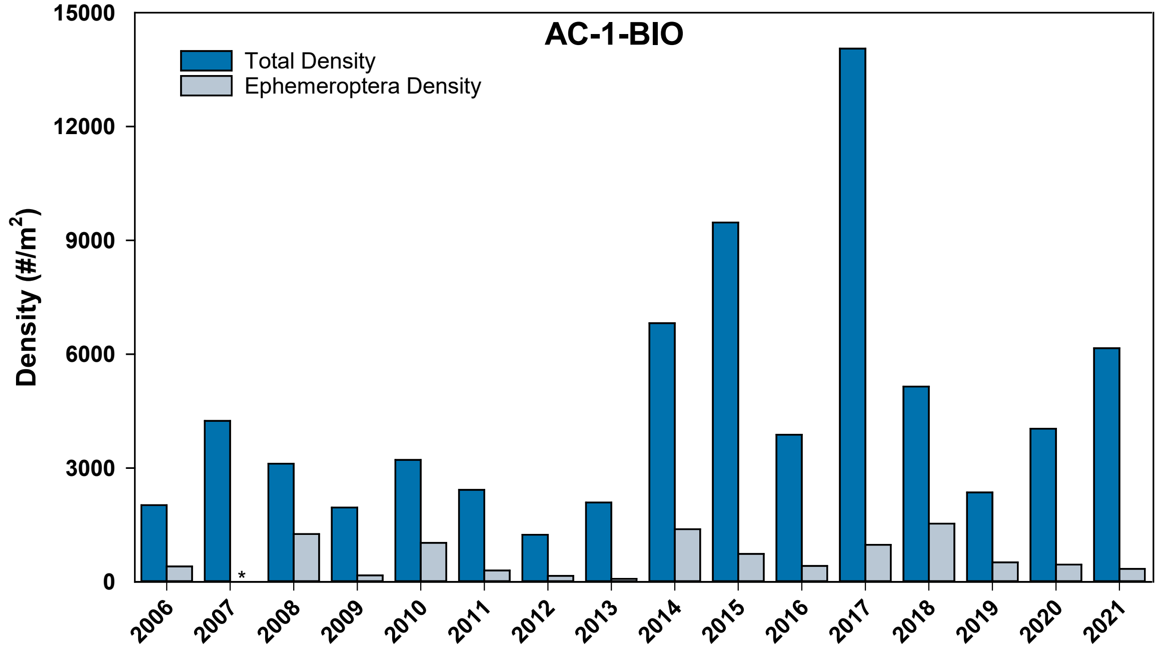


Figure 4-17: Macroinvertebrate density metrics for Site AC-1-BIO on Annie Creek, 2006 - 2021. * = Mayflies were present at low density.

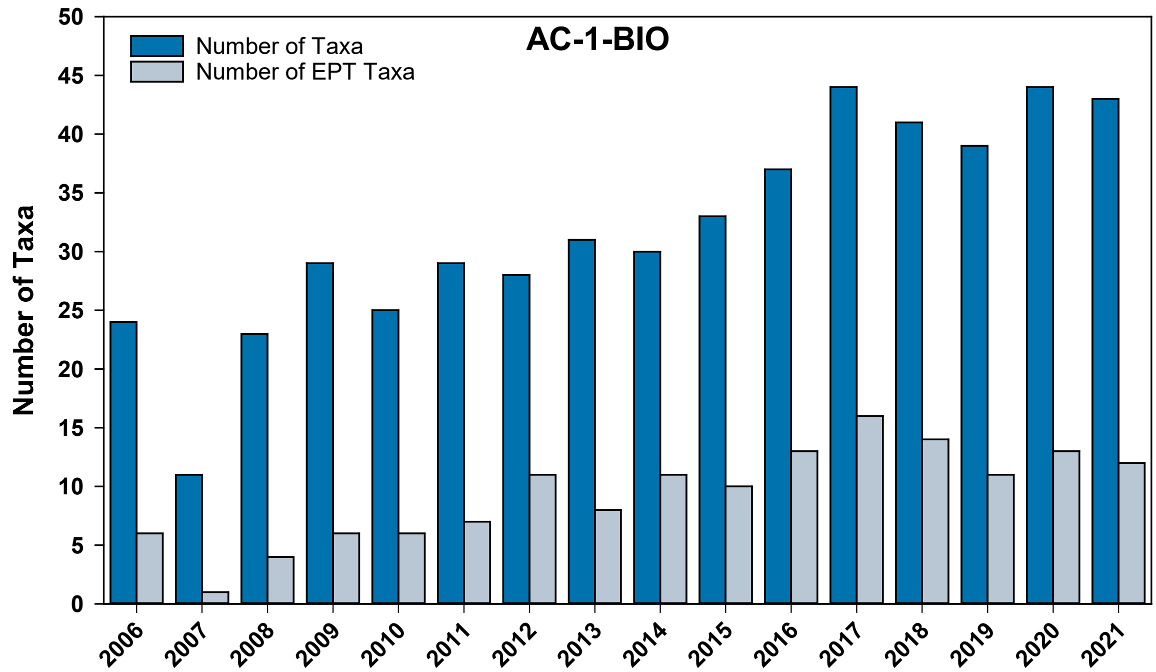


Figure 4-18: Macroinvertebrate taxa richness metrics for Site AC-1-BIO on Annie Creek, 2006 - 2021.

Table 4-19: Slopes of significant trends ($p < 0.05$) for macroinvertebrate population metrics at Annie Creek, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant.

Metric	Change Expected Following Environmental Disturbance	AC-1-BIO	AC-2-BIO	AC-3-BIO*
RICHNESS METRICS				
Density (#/sample)	Decrease	--	--	--
Number of Taxa	Usually Decrease	+ 1.74	--	--
Number of EPT Taxa	Decrease	+ 0.73	--	- 0.38
COMPOSITION METRICS				
Shannon-Weaver Diversity Index	Decrease	+ 0.11	--	--
Percent Sensitive EPT Taxa	Decrease	--	--	- 0.01
EPT Index	Decrease	0.01	--	- 0.01
Percent *Baetis sp.*	Increase	--	--	--
Number of non-*Baetis* Ephemeroptera	Decrease	+ 18.53	--	--
Percent Chironomidae	Increase	--	--	--
Number of Plecoptera Taxa	Decrease	--	--	- 0.16
Percent Abundance of Oligochaetes & Hirudinea	Variable	--	--	--
Percent Dominant Taxon	Increase	- 0.02	--	--
Number of Common Taxa	Decrease	+ 1.35	+ 0.94	--
Community Loss Index	Increase	- 0.09	--	--
TOLERANCE METRICS				
Hilsenhoff Biotic Index	Increase	--	--	+ 0.13
Percent Tolerant Taxa	Increase	--	--	+ 0.01
Percent Intolerant Taxa	Decrease	+ 0.01	--	- 0.01
Number of Intolerant Taxa	Decrease	+ 1.00	--	--
TROPHIC HABIT METRICS				
Number of Predator Taxa	Decrease	+ 0.58	--	--
Percent Collector-Gatherers	Variable	--	--	--
Number of Shredder Taxa	Decrease	+ 0.34	+ 0.25	+ 0.19
LIFE HISTORY METRICS				
Number of Univoltine Taxa	Increase	+ 0.92	--	--
Number of Merovoltine Taxa	Decrease	--	--	--
Number of Semivoltine Taxa	Decrease	--	--	--
Percent Semivoltine Taxa	Decrease	--	--	--

* AC-3-BIO was not sampled in 2020 and 2021 and trends are for 2006-2019 data.

4.3.3.2 Site AC-2-BIO

4.3.3.2.1 2021 Data

In August 2021, all metric values except for life history metrics were mainly mildly to very favorable at Site AC-2-BIO (Table 4-17; Table 4-18; Appendix C). Density, Number of Taxa, Number of EPT Taxa, and EPT Index values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a balanced invertebrate community and was more favorable than any other site; the HBI indicated a "Very Good" benthic macroinvertebrate community including intolerant species; and a diversity of feeding groups were present. Metric values associated with the EPT community were far better than in recent years with Percent Sensitive EPT Taxa and EPT Index greater than any other site, and *Baetis tricaudatus cx.* no longer the dominant Ephemeroptera. In addition, Percent and Number of Intolerant Taxa were greater than any other sites indicating that many mayfly species were relatively intolerant of environmental stressors. Only the life history metrics were poorer indicating that most taxa at the site have a short life cycle. Despite these metric values, overall, metrics for this site indicated that the stream conditions in 2021 supported a relatively abundant and diverse benthic macroinvertebrate community including intolerant species.

Benthic macroinvertebrate richness, composition, tolerance, trophic habit, and life history metric values at Site AC-2-BIO were comparable to those at the reference site, Site LB-4-BIO. Both sites contained favorable or fair values for Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI. Abundance and Number of non-*Baetis* Ephemeroptera were greater at the reference site while Percent Intolerant Taxa was more favorable at Site AC-2-BIO. The Community Loss Index and Number of Common Taxa were the best of all sites monitored in 2021, as was the case in 2020, and indicated that the majority of the taxa present at Site AC-2-BIO were also present at the reference site. Overall, the benthic macroinvertebrate communities were very similar at the two sites in August 2021 and were rich, and diverse, with numerous intolerant taxa, indicative of good stream conditions.

4.3.3.2.2 Historic Data

From 2006 to 2021 at Site AC-2-BIO, the macroinvertebrate community exhibited a variable pattern in Density and taxa richness metrics, with the Number of Taxa and Number of EPT Taxa reaching a peak every four to five years, including in 2021. The water quality disturbance in Annie Creek in 2007 also affected Site AC-2-BIO but not to the same extent as Site AC-1-BIO. Many metrics (Density, Number of Taxa, Percent *Baetis sp*., Number of non-Baetis Ephemeroptera, Percent Chironomidae, Number of Plecoptera Taxa, Percent Tolerant Taxa, Community Loss Index, HBI, Number of Intolerant Taxa, and Number of Predator Taxa) were poorer in 2007 than most other years but rebounded in one year or slowly over the years with much variably (Figure 4-19; Figure 4-20; Appendix C).

The variability in metric data resulted in only a few significant trends in metric data (Table 4-19). The Composition metric Diversity has been greater than 2.5 in all years of sampling and greater in 2021 than prior years (previous maximum of 4.28 in 2012) indicating a diverse invertebrate community. The only composition metric with a significant trend was Number of Common Taxa which improved from 2006 to 2021 and, along with Community Loss Index, were better in 2021 than all other years (previous maximum of 26 and previous minimum of 38.0%, respectively, in 2020).

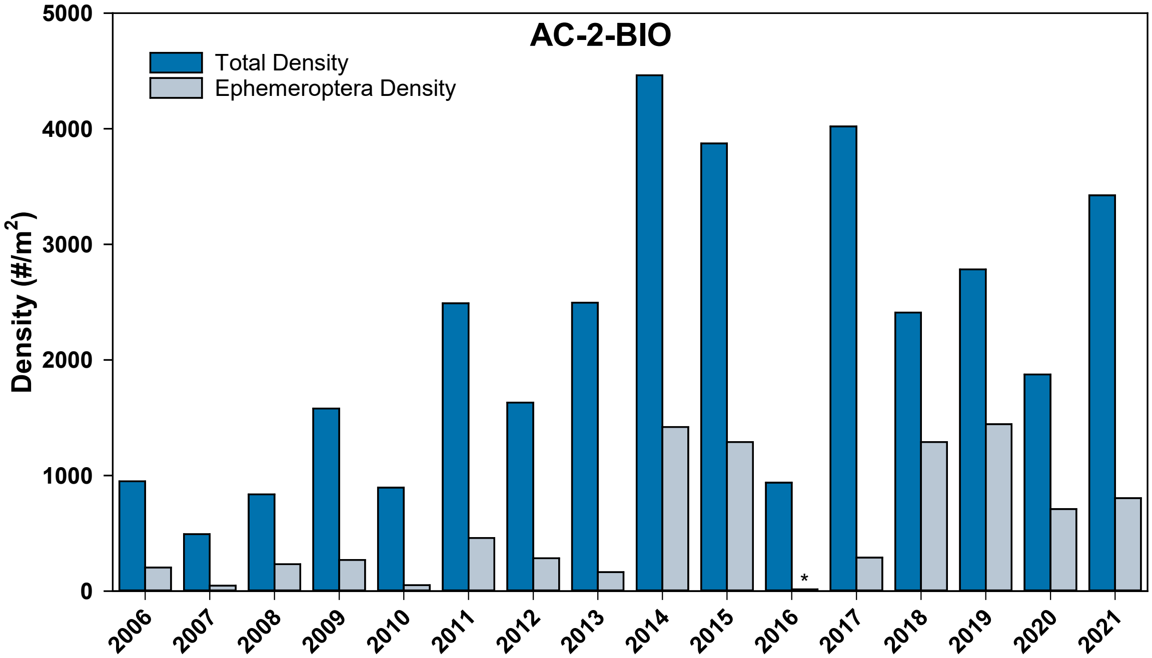


Figure 4-19: Macroinvertebrate density metrics for Site AC-2-BIO on Annie Creek, 2006 - 2021. * = Mayflies were present at low density.

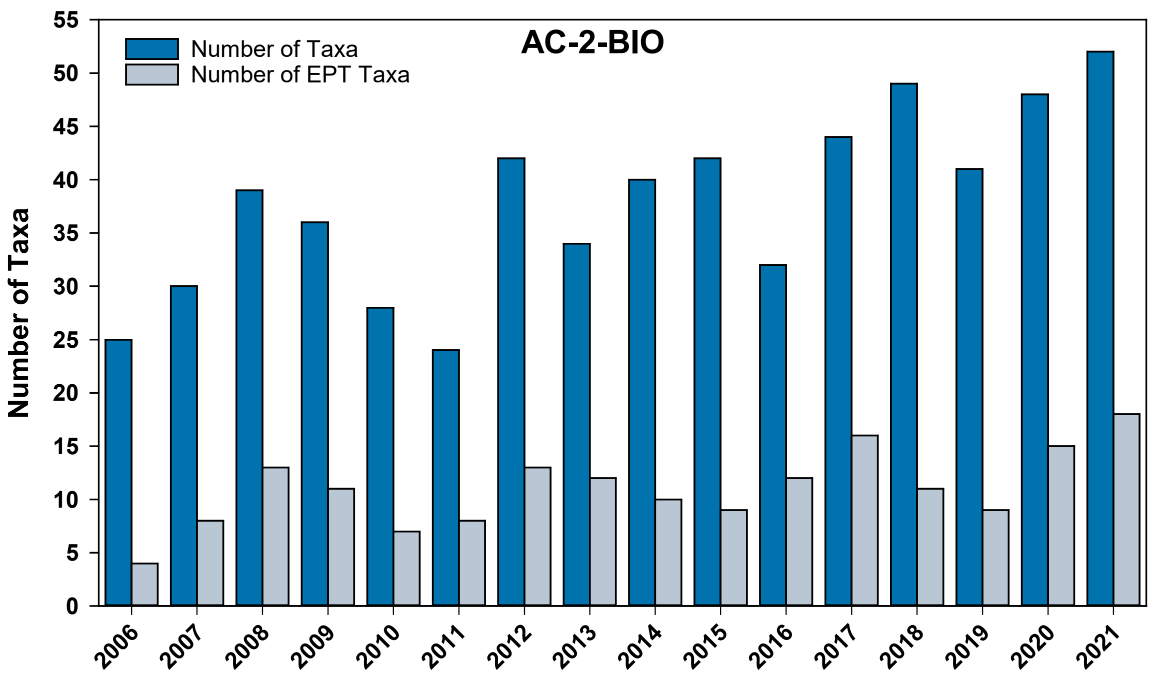


Figure 4-20: Macroinvertebrate taxa richness metrics for Site AC-2-BIO on Annie Creek, 2006 - 2021.

Tolerance, trophic habit, and life history metric values generally showed no trends over time. HBI has ranged from “Good” (5.4 in 2010) to “Very Good” (3.6 in 2009). The only trophic habit metric with a significant trend was the Number of Shredder Taxa which improved from 2006 to 2021. Number of Intolerant Taxa was greater in 2021 than past years (previous maximum of 29 in 2008) but did not significantly trend. Overall, data in 2021 showed negligible change with metric values being similar to values observed in recent years.

Few median metric values were significantly different between Site AC-2-BIO and the reference site, Site LB-4-BIO, with median Number of EPT Taxa, EPT Index, and HBI values being significantly poorer at Site AC-2-BIO than the reference site from 2006 to 2021 ($p = 0.021$, $p = 0.003$, and $p = 0.002$, respectively). EPT taxa are sensitive to sedimentation, and sediments from erosion of the nearby dirt road in recent years have negatively impacted the suitability of this site for some EPT taxa.

4.3.3.3 Site AC-3-BIO

4.3.3.3.1 2021 Data

The majority of Annie Creek at Site AC-3-BIO was not accessible for macroinvertebrate sampling in 2021 due to extensive deadfall from July 2020 tornado disturbance, and samples were not collected (Photo 4-3).

4.3.3.3.2 Historic Data

The macroinvertebrate community at Site AC-3-BIO moderately changed from 2006 to 2019. However, these changes did not appear to be related to the water quality disturbance in 2007. Richness metric values largely stayed consistent with the Number of EPT Taxa significantly decreasing from 2006 to 2019 (Figure 4-21; Figure 4-22; Appendix C). For composition metrics, Diversity has never been less than 2.5 indicating a history of a healthy invertebrate community. Percent Sensitive EPT Taxa, EPT Index, and Number of Plecoptera values also significantly decreased from 2006 to 2019.

The tolerance metric HBI has ranged from “Excellent” (2.78 in 2007) to “Fair” (5.73 in 2003) but has significantly improved over time indicating an increase in more tolerant species. Percent Tolerant Taxa and Percent Intolerant Taxa have also both slightly but significantly declined in quality from 2006 to 2019. The only metric to improve at Site AC-3-BIO from 2006 to 2019 was the trophic habit metric Number of Shredder Taxa.

Overall, the EPT based-metrics have declined from 2006 to 2019, indicating that these assemblages have been stressed over this period. The reason for this decline is unknown at this time but the changes are specific to Site AC-3-BIO and are not representative of regional changes. Only the HBI metric trended in the same direction for Site AC-3-BIO and its reference site, Site LB-4-BIO. In fact, median values for Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI were not significantly different between the two sites ($p > 0.05$) indicating that the data sets were similar. Overall, historic benthic macroinvertebrate community metric data between the two sites are not different, despite the worsening in EPT based-metric values for Site AC-3-BIO. Site AC-3-BIO has maintained a rich and diverse community with numerous intolerant taxa, indicative of healthy stream conditions.

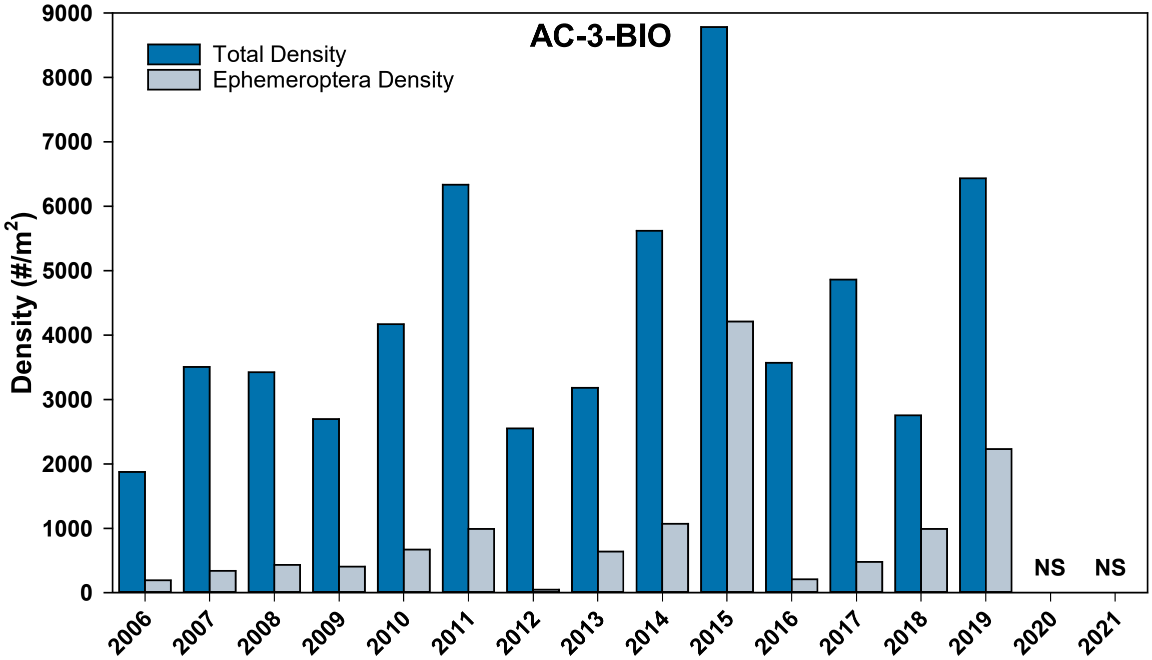


Figure 4-21: Macroinvertebrate density metrics for Site AC-3-BIO on Annie Creek, 2006 - 2021. NS = Not Sampled.

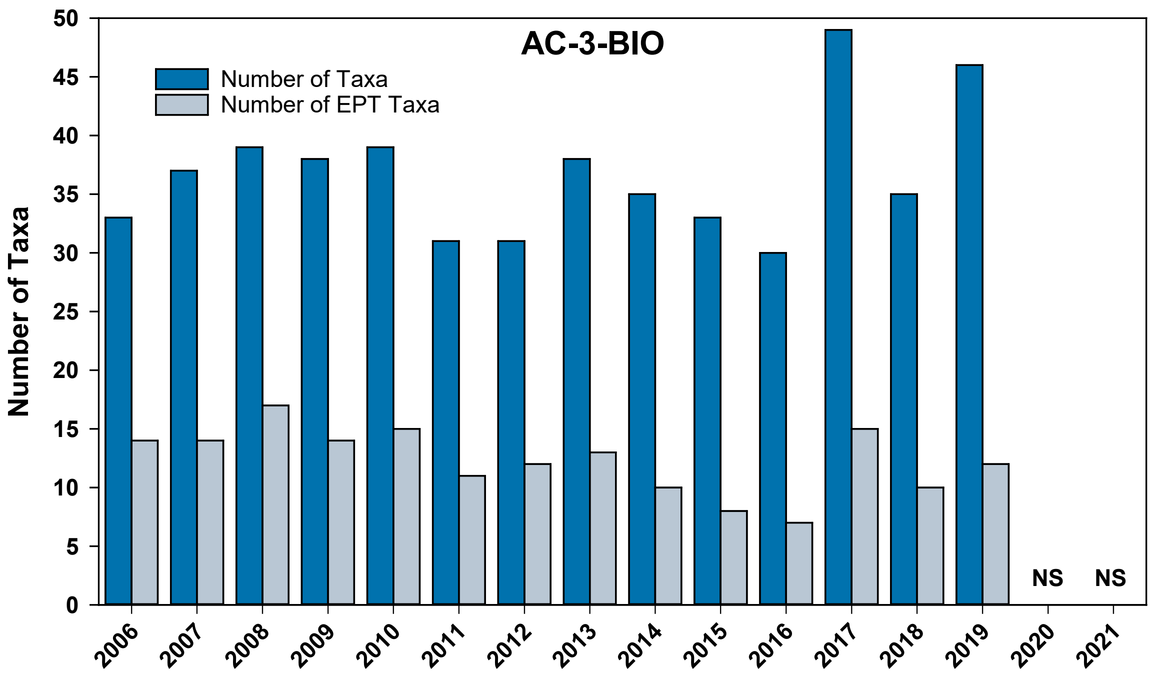


Figure 4-22: Macroinvertebrate taxa richness metrics for Site AC-3-BIO on Annie Creek, 2006 - 2021. NS = Not Sampled.

4.3.3.4 Site Comparisons

4.3.3.4.1 2021 Data

The metric results at the two Annie Creek sites sampled in 2021 were overall favorable and similar between the sites with some distinct exceptions (Table 4-17; Table 4-18; Appendix C). Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI were fair or good at both sites when compared to historic metric data in the Black Hills. Both sites contained macroinvertebrate community with a diversity of feeding groups that was dominated by short life cycle lengths. For the richness metrics, Density at Site AC-1-BIO was, however, roughly two times greater than at Site AC-2-BIO while Number of Taxa and EPT Taxa were greater at Site AC-2-BIO. The composition metric, Number of non-*Baetis* Ephemeroptera, and tolerance metrics, Percent and Number Intolerant Taxa, were better at Site AC-2-BIO while the trophic habit metric, Percent Collector-Gatherers, and the life history metric, Number of Univoltine Taxa, were better at Site AC-1-BIO. The differences in metrics between sites are likely the result of different stream sizes, the presence of tributaries, and the filling of potholes in the road near Site AC-2-BIO with sand and gravel.

4.3.3.4.2 Historic Data

Even though differences in the macroinvertebrate communities along Annie Creek appear to be influenced by stream size and sedimentation issues, the long-term median values for Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI values are not significantly different among the three sites. Only Density was significantly greater at sites AC-1-BIO and AC-3-BIO than at Site AC-2-BIO ($p = 0.02$) which, again, may be impacted by fixing of roadway erosion.

4.3.4 Ross Valley

4.3.4.1 2021 Data

Richness, composition, tolerance, and trophic habit metric values were predominantly mildly to very favorable at Site RV-2-BIO in August 2021 (Table 4-20; Table 4-21; Appendix C). Specifically, Density, Number of Taxa, Number of EPT Taxa, and EPT Index values were all fair or good when compared to historical data in the Black Hills; Diversity was indicative of a healthy invertebrate community; and the HBI indicated an “Excellent” benthic macroinvertebrate community including intolerant species, specifically *Polycelis coronata*, which was the dominant taxon. Exceptions to the favorable values were the poor Percent Sensitive EPT Taxa result and the lowest Percent Tolerant Taxa of any site. In addition, life history metrics indicated that most taxa at the site have a short life cycle. Overall, stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community (Appendix B) containing intolerant taxa.

Benthic macroinvertebrate richness, composition, tolerance, trophic habit, and life history metric values at Site RV-2-BIO were comparable to those at the reference site, Site RC-1-BIO. Both sites revealed favorable or fair values for richness metrics, Diversity, EPT Index, and HBI. The Number of Taxa and EPT Taxa, Diversity, Percent Sensitive EPT Taxa, and Percent Dominant Taxon were slightly better at the reference site than at Site RV-2-BIO indicating a slightly more favorable EPT community. However, Percent Chironomidae, HBI, Number of non-*Baetis* Ephemeroptera, Percent Collector-Gatherers, and Number of Univoltine Taxa were more favorable at Site RV-2-BIO. These differences between the sites resulted in a Community Loss Index value of 69%, indicating that the majority of taxon were not found at both sites. Overall, despite the differences, both sites containing a healthy benthic macroinvertebrate community, including intolerant species in August 2021.

Table 4-20: Macroinvertebrate density (number of organisms/sample) at Ross Valley, Lost Camp Gulch, Deadwood Creek, and East and West Fork False Bottom Creek, August 2021.

Taxa	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO
INSECTA					
Collembola (springtails)	5	--	4	--	--
Ephemeroptera (mayflies)	900	20	164	445	--
Plecoptera (stoneflies)	130	--	220	1,080	6
Megaloptera (alderflies)	--	30	28	5	--
Coleoptera (beetles)	195	365	12	120	17
Trichoptera (caddisflies)	150	180	88	80	29
Diptera (true flies)	1,180	1,500	1,665	1,365	926
NEMATODA (round worms)	--	--	8	--	
HYDRACARINA (water mites)	35	5	--	10	--
CRUSTACEA					
Amphipoda (Scuds)	--	--	--	--	--
TURBELLARIA (flatworms)	1,610	5	--	--	--
ANNELIDA (segmented worms)					
Oligochaeta (worms)	10	40	16	55	6
MOLLUSCA					
Pelecypoda (clams)	115	--	20	95	--

Table 4-21: Macroinvertebrate population metrics at Ross Valley, Lost Camp Gulch, Deadwood Creek, and East and West Fork False Bottom Creek, August 2021.

Metric	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO
RICHNESS METRICS					
Density (#/sample)	4,330	2,145	2,225	3,255	984
Number of Taxa	36	28	39	42	23
Number of EPT Taxa	9	5	12	14	5
COMPOSITION METRICS					
Shannon-Weaver Diversity Index	3.21	3.34	3.61	3.73	3.21
Percent Sensitive EPT Taxa	16.7%	10.7%	25.6%	28.6%	13.0%
EPT Index	25.0%	17.9%	30.8%	33.3%	21.7%
Percent *Baetis sp.*	10.6%	0.0%	24.4%	82.0%	0.0%
Number of non-*Baetis* Ephemeroptera	805	20	124	80	0
Percent Chironomidae	11.7%	68.5%	71.7%	33.9%	90.9%
Number of Plecoptera Taxa	3	0	4	6	2
Percent Abundance of Oligochaetes & Hirudinea	0.2%	1.9%	0.7%	1.7%	0.6%
Dominant Taxon (Tolerance value)	*Polycelis coronata*	*Polypedilum sp.*	*Trissopelopia ogemawi*	*Zapada cinctipes*	*Conchapelopia /Thienemanni myia gr.*
Percent Dominant Taxon	37.2%	32.6%	36.3%	28.1%	21.2%
Number of Common Taxa	21	17	23	27	11
Community Loss Index	69.4%	103.6%	82.1%	66.7%	191.3%
TOLERANCE METRICS					
Hilsenhoff Biotic Index	3.40	5.62	5.39	4.08	6.12
Percent Tolerant Taxa	30.6%	25.0%	25.6%	16.7%	21.7%
Percent Intolerant Taxa	50.0%	42.9%	41.0%	52.4%	34.8%
Number of Intolerant Taxa	18	12	16	22	8
TROPHIC HABIT METRICS					
Number of Predator Taxa	10	7	10	14	6
Percent Collector-Gatherers	38.7%	36.8%	37.2%	47.6%	56.9%
Number of Shredder Taxa	7	5	7	8	5
LIFE HISTORY METRICS					
Number of Univoltine Taxa	16	17	20	19	13
Number of Merovoltine Taxa	2	2	2	2	1
Percent Semivoltine Taxa	0.0%	0.0%	0.0%	2.4%	0.0%
Number of Semivoltine Taxa	0	0	0	1	0

4.3.4.2 Historic Data

From 2006 to 2021, the macroinvertebrate community at Site RV-2-BIO changed very little except for Density which has been variable over time. Richness, composition, tolerance, trophic habit, and life history metric values have largely stayed consistent over these years (Figure 4-23; Figure 4-24; Appendix C) and not trended except for Number of Common Taxa and Percent Intolerant Taxa which both improved significantly (Table 4-22). In addition, the 2021 Number of non-*Baetis* Ephemeroptera was better and Percent Dominant Taxon values were poorer than in past years (previous maximums of 315 in 2008 and 35.4% in 2012, respectively). Diversity has been greater than 2.5 in all years of sampling, indicating a healthy invertebrate community. In addition, the HBI metric fluctuated from "Good" (5.47 in 2011) to "Excellent" (2.77 in 2014). The subset of historic benthic macroinvertebrate community metric values (see Section 3.6) were not substantially different between Site RV-2-BIO and its reference site, Site RC-1-BIO. Overall, data in 2021 indicated negligible changes with the metric values similar to those in previous years.

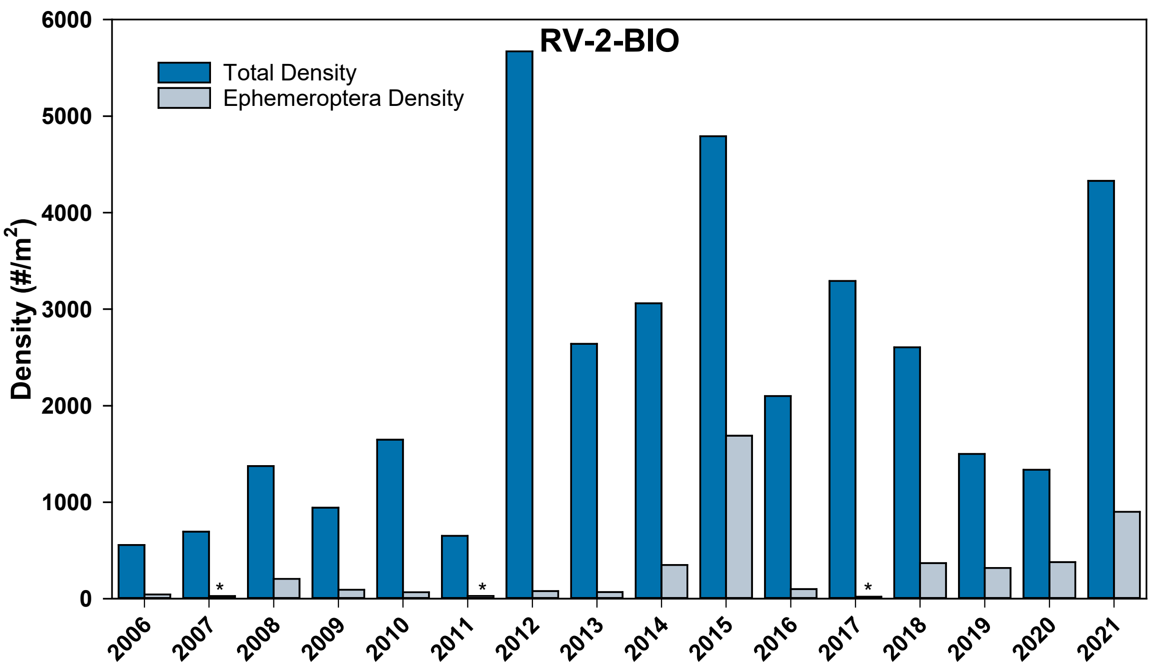


Figure 4-23: Macroinvertebrate density metrics for Site RV-2-BIO on Ross Valley, 2006 - 2021. * = Mayflies were present at low density.

Table 4-22: Slopes of significant trends ($p < 0.05$) for macroinvertebrate population metrics at Ross Valley, Lost Camp Gulch, Deadwood Creek, East Fork False Bottom Creek, and West Fork False Bottom Creek, 2006 - 2021. + = positive slope. - = negative slope. -- = not significant.

Metric	Change Expected Following Environmental Disturbance	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO
RICHNESS METRICS						
Density (#/sample)	Decrease	--	--	--	--	--
Number of Taxa	Usually Decrease	--	--	+ 1.02	--	--
Number of EPT Taxa	Decrease	--	- 0.60	--	--	- 0.33
COMPOSITION METRICS						
Shannon-Weaver Diversity Index	Decrease	--	- 0.10	--	--	- 0.06
Percent Sensitive EPT Taxa	Decrease	--	- 0.02	--	--	- 0.01
EPT Index	Decrease	--	- 0.02	- 0.01	--	--
Percent *Baetis sp.*	Increase	--	--	- 0.05	--	--
Number of non-*Baetis* Ephemeroptera	Decrease	--	--	--	--	--
Percent Chironomidae	Increase	--	--	--	--	--
Number of Plecoptera Taxa	Decrease	--	- 0.26	--	--	- 0.22
Percent Abundance of Oligochaetes & Hirudinea	Variable	--	--	--	--	--
Percent Dominant Taxon	Increase	--	--	--	--	--
Number of Common Taxa	Decrease	+ 0.68	--	+ 0.61	--	--
Community Loss Index	Increase	--	--	--	--	0.05
TOLERANCE METRICS						
Hilsenhoff Biotic Index	Increase	--	--	--	--	--
Percent Tolerant Taxa	Increase	--	--	--	--	--
Percent Intolerant Taxa	Decrease	+ 0.01	--	--	--	--
Number of Intolerant Taxa	Decrease	--	--	--	--	- 0.39
TROPHIC HABIT METRICS						
Number of Predator Taxa	Decrease	--	--	--	+ 0.42	- 0.26
Percent Collector-Gatherers	Variable	--	--	--	--	--
Number of Shredder Taxa	Decrease	--	--	+ 0.34	--	--
LIFE HISTORY METRICS						
Number of Univoltine Taxa	Increase	--	--	+ 0.49	--	- 0.50
Number of Merovoltine Taxa	Decrease	--	--	--	--	--
Number of Semivoltine Taxa	Decrease	--	--	--	+ 0.06	--
Percent Semivoltine Taxa	Decrease	--	--	--	--	--

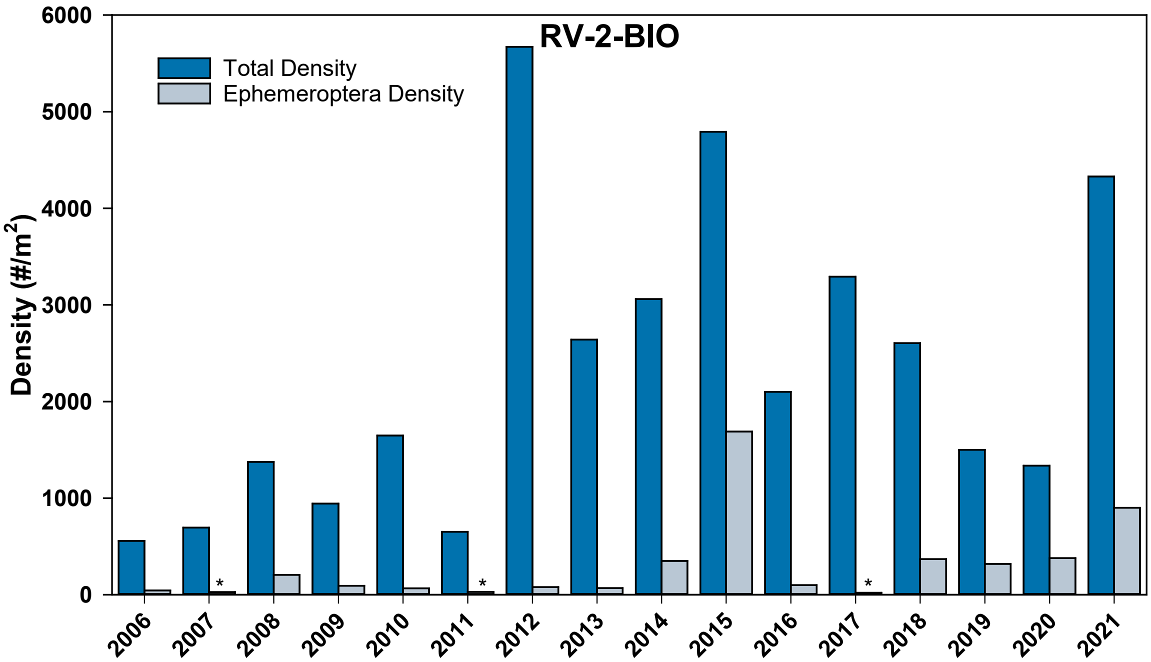


Figure 4-24: Macroinvertebrate taxa richness metrics for Site RV-2-BIO on Ross Valley, 2006 - 2021.

4.3.5 Lost Camp Gulch

4.3.5.1 2021 Data

In August 2021, richness, tolerance, and trophic metric values were predominantly mildly to very favorable at Site LC-1-BIO (Table 4-20; Table 4-21; Appendix C). Density, Number of Taxa, and Number of EPT Taxa values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a balanced invertebrate community; and the HBI indicated a "Good" benthic macroinvertebrate community including intolerant species. However, very poor metric values for EPT Index, Percent Sensitive EPT Taxa, Number of non-*Baetis* Ephemeroptera, and Number of Plecoptera Taxa in combination with no *Baetis sp.* indicated a very limited EPT community. Instead, the macroinvertebrate community at Site LC-1-BIO consisted primarily of Chironomidae. Lastly, life history metrics indicated the community was dominated by short lived taxa. These deficiencies are the result of the low flows and resulting increased sedimentation observed in the past few years, which is strongly linked to the dirt road adjacent to Lost Camp Gulch. Despite the unfavorable EPT and related metric values, overall, metrics for this site indicated that the stream conditions in 2021 supported a relatively abundant and diverse benthic macroinvertebrate community including intolerant species.

Benthic macroinvertebrate richness, composition, tolerance, and trophic habit metric values at Site LC-1-BIO were mostly not comparable to those at the reference site, Site RC-1-BIO. This difference is largely because the reference site contained a more diverse and higher quality EPT assemblage than Site LC-1-BIO and Site LC-1-BIO contained a more robust

Chironomid assemblage. Overall, only 17 taxa were shared between the sites with the reference site containing 19 taxa not found at Site LC-1-BIO (Appendix B). As a result, Density, Number of Taxa and EPT Taxa, Diversity, Percent Sensitive EPT Taxa, EPT index, Number of non-*Baetis* Ephemeroptera, Percent Chironomidae, Number of Plecoptera Taxa, HBI, Number of Intolerant Taxa, and Number of Predator Taxa were all more favorable at the reference site. However, many of these metrics were still considered good at Site LC-1-BIO, including Diversity and HBI calculations, which are not solely dependent on EPT taxa, indicated a healthy benthic macroinvertebrate community.

These results indicated that the EPT community at Site LC-1-BIO was poorer compared to the reference site. The community at Site LC-1-BIO is more limited by the lack of flushing flows and increased sedimentation than some other Wharf study sites in 2021. The site was affected by the low flow conditions in 2016, 2017, 2020 and 2021 which also limited aquatic habitat available to benthic macroinvertebrates. None-the-less, the benthic macroinvertebrate community at Site LC-1-BIO in August 2021 was diverse and included intolerant taxa which is indicative of good water quality.

4.3.5.2 Historic Data

The macroinvertebrate community at Site LC-1-BIO has slightly changed from 2010, when sampling began, to 2021. Richness, tolerance, trophic habit, and life history metric values have largely stayed consistent over these years (Figure 4-25; Figure 4-26; Appendix C) while many composition metrics have trended. Density has fluctuated over time, often due to highly variable numbers of *Baetis* or *Fallceon* mayflies from year to year. These genera are widespread and moderately tolerant of water quality conditions. Number of EPT Taxa was the only richness metric to significantly decrease from 2010 to 2021 (Table 4-22). For composition metric values from 2010 to 2021, Diversity, Percent Sensitive EPT Taxa, EPT Index values, and Number of Plecoptera Taxa significantly declined in quality over time. However, Diversity has been less than 2.5 only once in recent years indicating a history of a diverse invertebrate community. The 2021 HBI tolerance metric was within the range of previously recorded values, ranging from “Fair” (5.9 in 2014) to “Very Good” (3.7 in 2010).

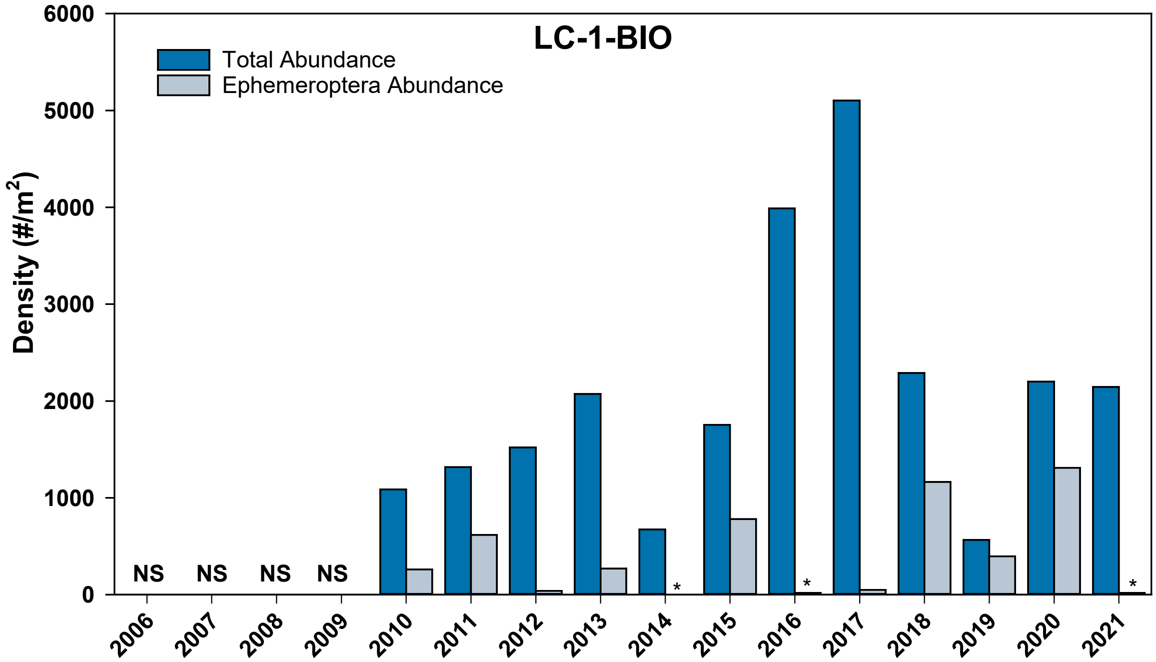


Figure 4-25: Macroinvertebrate density metrics for Site LC-1-BIO on Lost Camp Gulch, 2006 - 2021. NS = Not sampled. * = Mayflies absent or present at low density.

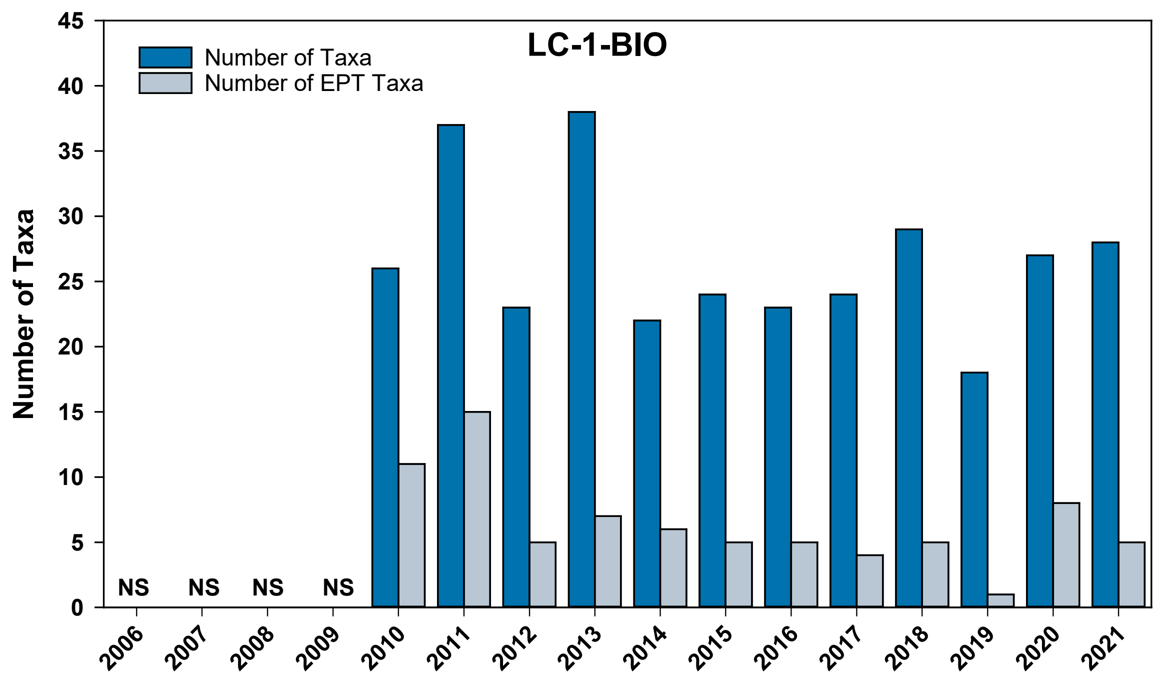


Figure 4-26: Macroinvertebrate taxa richness metrics for Site LC-1-BIO on Lost Camp Gulch, 2006 - 2021. NS = Not sampled.

Overall, four of the 11 composition metrics have declined in quality from 2010 to 2021 indicating that the macroinvertebrate community, specifically EPT taxa, has become stressed over that period. In addition, historic median Density, Number of Taxa and EPT Taxa, and Diversity data were significantly better at the reference site, Site RC-1-BIO, than at Site

LC-1-BIO (p = 0.027, p = 0.004, p = 0.012, and p = 0.020, respectively). Periodic low flows at this site negatively impact the benthic macroinvertebrate community and are responsible for changes in metrics. In 2016 and 2017, portions of the site exhibited little to no flow, which likely limited available habitat for more sensitive benthic macroinvertebrates. Sedimentation from the road adjacent to Site LC-1-BIO may also be negatively impacting the benthic macroinvertebrate community in some years, as the percent surface fines were relatively poor in most habitat units at this site when compared with surface fines values at other biomonitoring sites in 2017. The low flows during 2016 and 2017 limited the potential to scour and remove fine sediments that have settled in the substrate. Precipitation from 2018 to 2021 was higher (Figure 4-1), and the percentage of surface fines were low at this site, but an appreciable improvement in metrics from 2017 to 2021 was not detected. With higher perennial flows at this site, the benthic macroinvertebrate community may improve during future sampling events.

4.3.6 Deadwood Creek

4.3.6.1 2021 Data

In August 2021, richness, composition, tolerance, and trophic habit metric values were predominantly mildly to very favorable at Site DC-2-BIO (Table 4-20; Table 4-21; Appendix C). Specifically, Density, Number of Taxa and EPT Taxa, and EPT Index values were fair or good when compared to historical data in the Black Hills; Diversity indicated a healthy invertebrate community; the HBI indicated a “Good” benthic macroinvertebrate community including intolerant species; and a diversity of feeding groups were present. However, this site contained a very high percentage of Chironomids (Appendix B), specifically the non-biting midge *Trissopelopia sp.* This subfamily, as well as many other Chironomid subfamilies, has a short life cycle leading to this site being dominated by univoltine taxa. Overall, despite the large midge population, stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community including intolerant species.

Benthic macroinvertebrate richness, composition, tolerance, trophic habit, and life history metric values at Site DC-2-BIO were comparable to those at the reference site, Site LB-4-BIO. Density, Number of Taxa, Number of EPT Taxa, Diversity, Number of non-*Baetis* Ephemeroptera, Percent Dominant Taxon, HBI, Number of Intolerant Taxa, and Number of Predator Taxa were more favorable at the reference site but good at both sites. Percent Chironomidae was also more favorable at the reference site compared to a very low value at Site DC-2-BIO. At the same time, Percent *Baetis sp.*, Percent Collector-Gatherers, and Number of Univoltine Taxa were more favorable at Site DC-2-BIO. These differences between the sites resulted in a Community Loss Index of 82%, indicating that the majority of the taxa present at the reference site were not present at Site DC-2-BIO. While many metric values at Site DC-2-BIO were less than the values at the reference site, Site DC-2-BIO contained an overall healthy benthic macroinvertebrate community in August 2021.

4.3.6.2 Historic Data

The macroinvertebrate community at Site DC-2-BIO moderately changed from 2010 to 2021. Richness metric values stayed consistent and did not trend except for Number of Taxa which significantly increased (Table 4-22; Figure 4-27; Figure 4-28; Appendix C). Number of EPT Taxa exhibit a cyclical pattern where every three to four years the number of taxa peak and then decline. Diversity has been above 2.5 in all years indicating a history of a diverse invertebrate community. The EPT Index has slightly, but significantly, decreased in quality while Percent *Baetis sp*. and Number of Common Taxa have significantly improved since 2010 (Table 4-21). Community Loss Index was greater in 2021 than previous years (previous maximum of 77.8% in 2016).

The 2021 HBI value was poorer than previously recorded at this site (previous maximum of 4.94 in 2020) and has ranged from “Good” (5.4 in 2021) to “Excellent” (3.2 in 2010). Trophic habit metrics, Number of Shredder Taxa significantly improved from 2010 to 2021 while the life history metric Number of Univoltine Taxa significantly worsened over time.

Few median metric values were significantly different between Site DC-2-BIO and the reference site, Site LB-4-BIO, with only the median Number of EPT Taxa being significantly higher at the reference site than at Site DC-2-BIO from 2010 to 2021 ($p = 0.003$). Historic benthic macroinvertebrate community metric data (i.e., long-term median values) between the two sites are not different and both have been rich and diverse, with numerous intolerant taxa, indicative of good historic stream conditions.

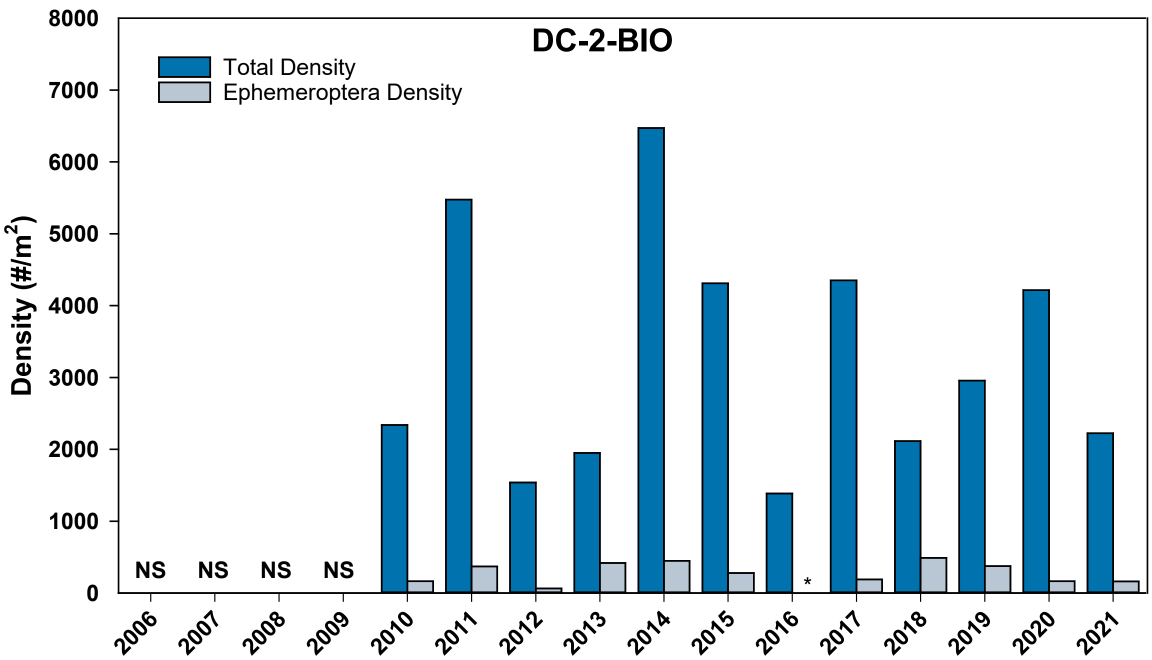


Figure 4-27: Macroinvertebrate density metrics for Site DC-2-BIO on Deadwood Creek, 2006 - 2021. NS = Not sampled. * = Mayflies not present.

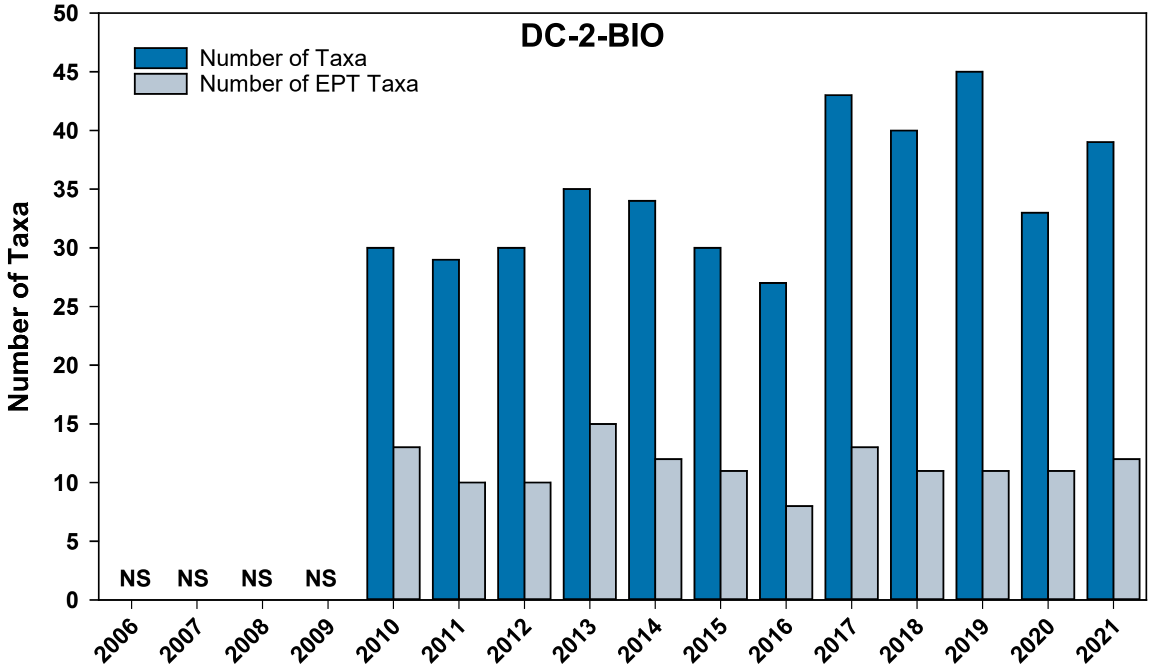


Figure 4-28: Macroinvertebrate taxa richness metrics for Site DC-2-BIO on Deadwood Creek, 2006 - 2021.

4.3.7 False Bottom Creek

4.3.7.1 Site EFB-1-BIO

4.3.7.1.1 2021 Data

Metric values were predominantly mildly to very favorable at Site EFB-1-BIO in August 2021 (Table 4-20; Table 4-21; Appendix C). Specifically, Density, Number of Taxa, Number of EPT Taxa, and EPT Index values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a healthy invertebrate community; the HBI indicated a “Very Good” benthic macroinvertebrate community that included intolerant species, diversity of feeding groups, and a variety of life cycle lengths. Site EFB-1-BIO had a lower percentage of tolerant species than all other sites. An exception was the high percentage of *Baetis tricaudatus cx.* at 82% (Appendix B) of the mayflies population indicating that the majority of Ephemeroptera were relatively tolerant of environmental stressors. Overall, stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community including sensitive taxa.

Benthic macroinvertebrate richness, composition, tolerance, trophic habit, and life history metric values at Site EFB-1-BIO were comparable to those at the reference site, Site LB-4-BIO. Both sites contained favorable or fair values for Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI metric values. The Ephemeroptera population at both sites was dominated by *Baetis tricaudatus cx.,* however this metric and the Number of non-*Baetis* Ephemeroptera were, again, better at the reference site. In addition, Density, Number of Taxa, Diversity, Percent Dominant Taxa, Number of Intolerant Taxa, and Number of Semivoltine Taxa were better at the reference site while Number of Univoltine Taxa were more favorable at the Site EFB-1-BIO. These differences between the sites resulted in the Community Loss Index of 67%, indicating that the majority of taxa present at the reference site were not present at Site EFB-1-BIO. Overall, both sites contained a healthier macroinvertebrate community while the community at the reference site was slightly more robust in August 2021.

4.3.7.1.2 Historic Data

From 2006 to 2021 at Site EFB-1-BIO, the macroinvertebrate community richness, composition, tolerance, trophic habit, and life history metric values were relatively consistent (Figure 4-29; Figure 4-30; Appendix C) and few significant trends observed over time (Table 4-22). However, only seven of the last 16 years were sampled, so trends should be cautiously interpreted. Diversity has been greater than 2.5 in all years of sampling indicating a healthy invertebrate community. In addition, HBI has fluctuated from “Good” (4.6 in 2009) to “Excellent” (3.5 in 2019). The 2021 Number of Taxa, Number of EPT Taxa, Number of Plecoptera Taxa, Number of Intolerant Taxa, Number of Predator Taxa, and Number of Shredder Taxa metric data were all improved over previous years (previous maximums of 41 in 2018, 12 in 2010, 4 in most years, 18 in 2018, 13 in 2018, 5 in 2009 and 2019, respectively) while Percent Chironomidae was poorer (previous maximum of 33% in 2009). However, the only metrics to significantly trend over these years were the trophic habit metric, Number of Predator Taxa, and the life history metric, Number of Semivoltine Taxa, which both significantly improved. Overall, macroinvertebrate data in 2021 showed negligible change in the metric values and were similar to those in previous years.

When compared to the reference site, Site LB-4-BIO, the long-term median Number of EPT Taxa and Diversity at Site EFB-1-BIO were significantly poorer ($p = 0.005$ and $p = 0.004$), although the low number of sampling events at EFB-1-BIO, and differences between sample size, influences the robustness of these comparisons. There were no significant differences observed among Density, Number of Taxa, EPT Index, and HBI metrics for the two sites.

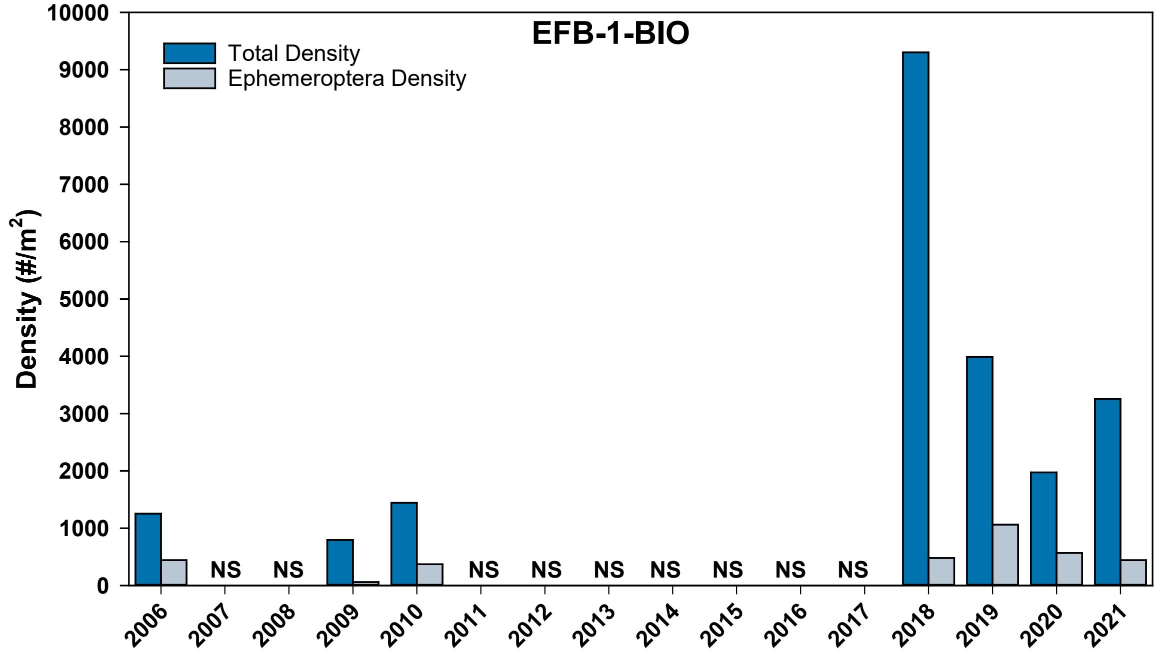


Figure 4-29: Macroinvertebrate density metrics for Site EFB-1-BIO in False Bottom Creek, 2006 - 2021. NS = Not Sampled.

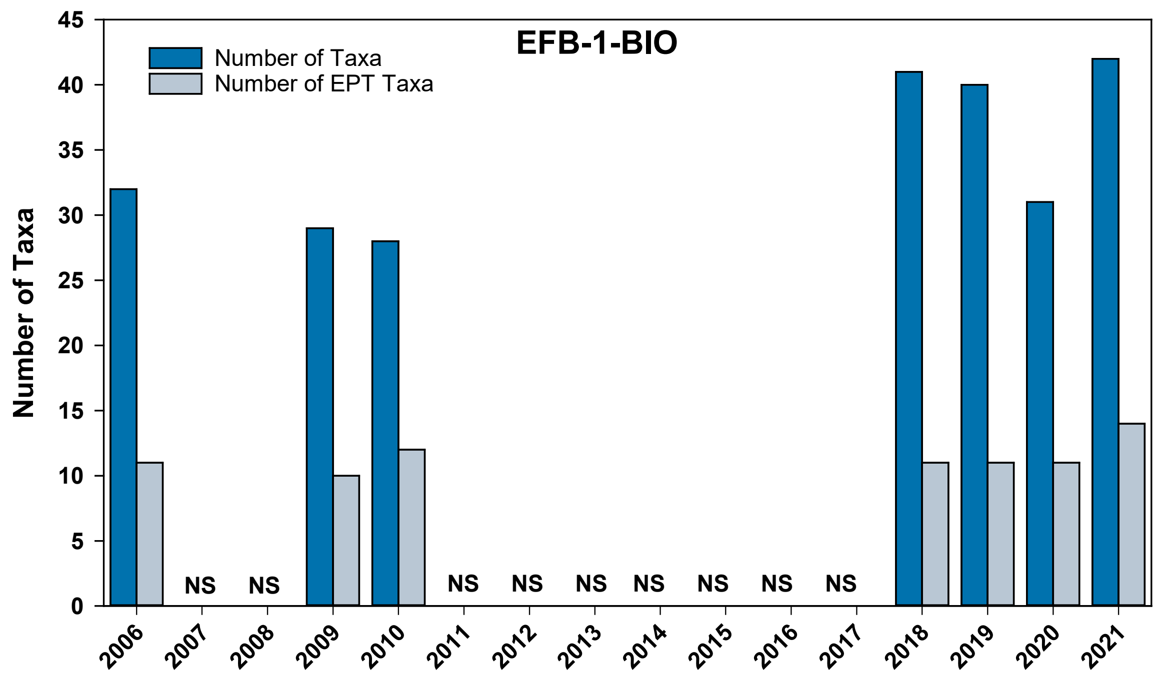


Figure 4-30: Macroinvertebrate taxa richness metrics for Site EFB-1-BIO on False Bottom Creek, 2006 - 2021. NS = Not Sampled.

4.3.7.2 Site WFB-1-BIO

4.3.7.2.1 2021 Data

No Ephemeroptera and low numbers of Plecoptera (six organisms) and Trichoptera (29 organisms) were collected at Site WFB-1-BIO in 2021 resulting in a poor EPT community and mostly unfavorable metric values. Density was very low, the lowest of any site, and Number of EPT Taxa was low, lower than most other sites. Number of Taxa was good when compared to historical data in the Black Hills but was the lowest of any site observed in 2021 sampling (Table 4-20; Table 4-21; Appendix C). Composition metrics were mostly poor, even though the Diversity value was 3.2. Percent Sensitive EPT Taxa, EPT Index, Number of non-*Baetis* Ephemeroptera, Percent Chironomidae, and Ephemeroptera Density were also poor and close to, or the poorest, of any Wharf site.

In addition, the tolerance metrics were fair with HBI and Percent and Number of Intolerant Taxa values being the lowest of any site. Life history metrics indicated a dominance of short-lived taxa. Despite these low metric scores, the HBI tolerance-based metric scored as "Good," and the trophic habit metrics indicated a diversity of feeding types, although with a relatively low Number of Predator and Shredder Taxa. The majority of data indicated that the stream conditions resulted in a fair to poor benthic macroinvertebrate community in 2021.

Benthic macroinvertebrate richness metric values at Site WFB-1-BIO were all relatively poor when compared to the reference site, Site LB-4-BIO. Similarly, most composition metrics, specifically EPT related metrics, were poorer than the reference site metrics. Tolerance metrics, including HBI, and trophic habit metrics were also generally less favorable at Site WFB-1-BIO than the reference site. These differences between the sites were reflected in the Community Loss Index of 191%, which showed only 11 taxa common to both sites and 44 reference site taxa not found at Site WFB-1-BIO. In general, the metrics indicate that invertebrate communities at these sites are different, and the poor stream conditions at Site WFB-1-BIO, linked to the iron oxide deposition have negatively affected the macroinvertebrate assemblage.

4.3.7.2.2 Historic Data

The macroinvertebrate community at Site WFB-1-BIO has somewhat changed from 2007 to 2021. Density in 2021 was greater than in 2020 when the lowest value was observed; however, this value is still low when compared to all other years. The Number of EPT Taxa, Diversity, Percent Sensitive EPT Taxa, Number of Plecoptera Taxa, Community Loss Index, Number of Intolerant Taxa, and Number of Predator Taxa have all significantly declined in quality over time (Table 4-22; Figure 4-31; Figure 4-32; Appendix C). Ephemeroptera density continues to be poor, and the mayfly population has either been absent or present in very low numbers during all years of sampling at Site WFB-1-BIO. HBI, Number of Predator Taxa, and Percent Collector-Gatherers at Site WFB-1-BIO were of poorer quality in 2021 than in prior years (previous maximum of 5.97 in 2012, minimum of 8 in 2018, and maximum of 49.5% in 2017, respectively). The Number of Univoltine Taxa is the only metric to have significantly improve over the years.

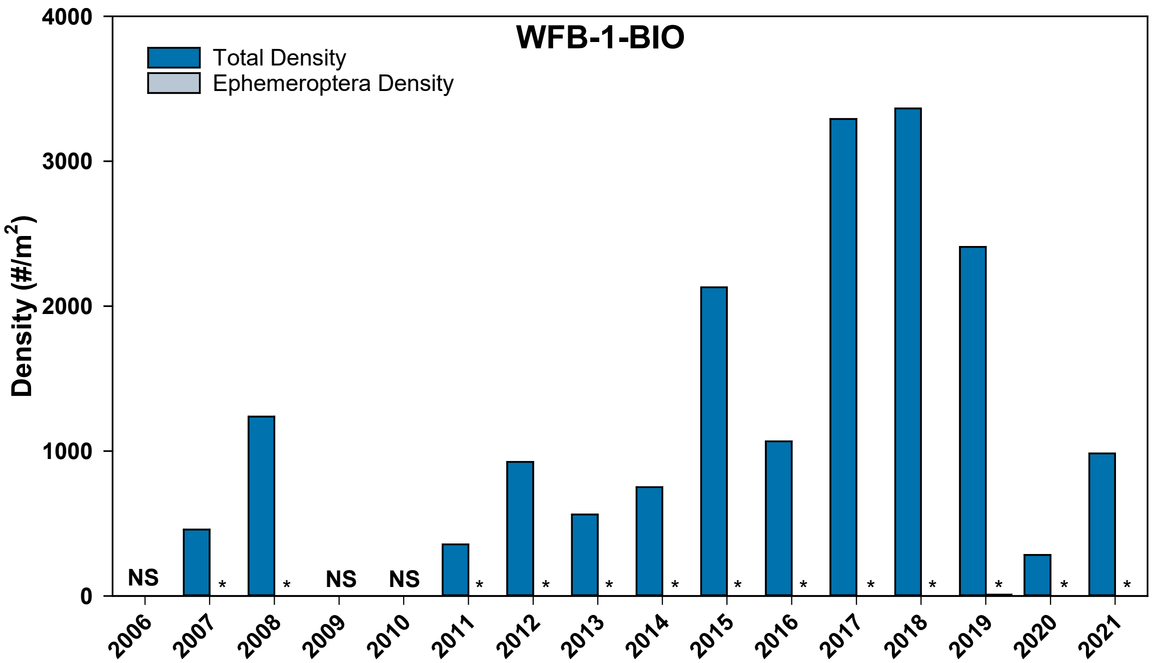


Figure 4-31: Macroinvertebrate density metrics for Site WFB-1-BIO in False Bottom Creek, 2006 - 2021. NS = Not Sampled. * = Mayflies absent or present at low density.

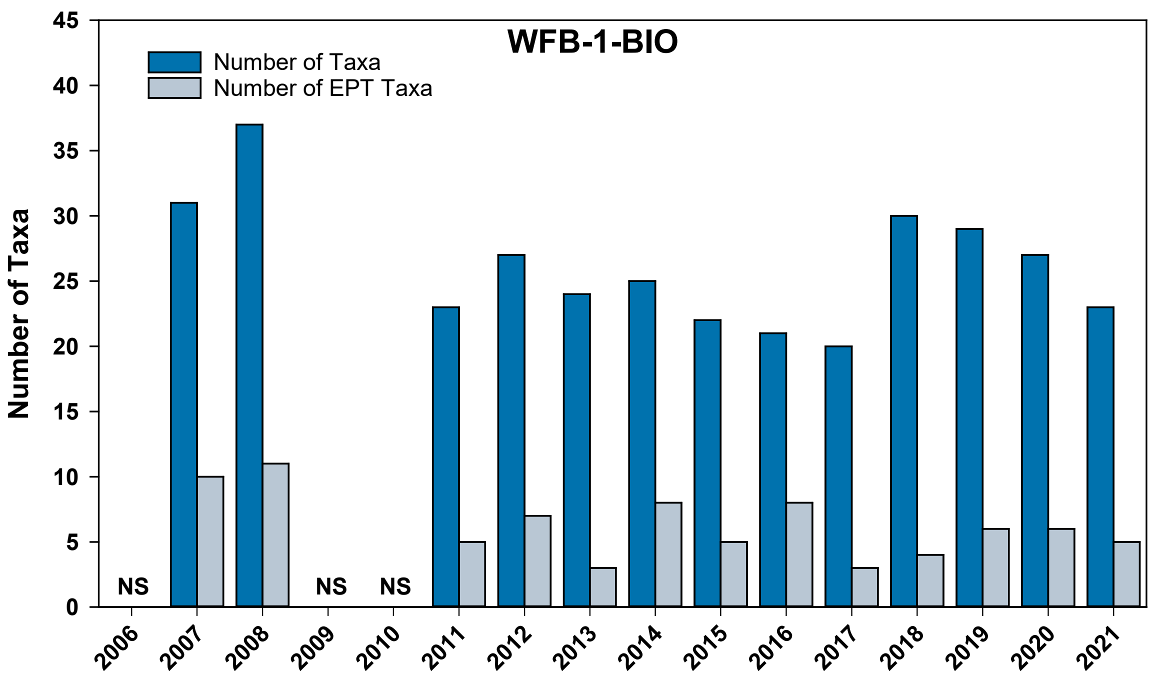


Figure 4-32: Macroinvertebrate taxa richness metrics for Site WFB-1-BIO on False Bottom Creek, 2006 - 2021. NS = Not Sampled.

When Site WFB-1-BIO is compared to the reference site, Site LB-4-BIO, all six selected metrics tested are significantly worse than the reference site metrics (Density: $p = 0.001$, Number of Taxa: $p < 0.001$, Number of EPT Taxa: $p < 0.001$, Diversity: $p = 0.005$, EPT Index: $p < 0.001$, and HBI: $p < 0.001$) indicating a poorer macroinvertebrate community at Site WFB-1-BIO. Overall, data in 2021 were not remarkable and the metric values were similar to those in previous years. A moderate number of metrics have declined in quality since 2007, indicating that the health of the macroinvertebrate community has become poorer over time and is stressed due to low flows and iron oxide deposition that negatively affects habitat quality.

4.3.8 McKinley Gulch

McKinley Gulch was dry during August 2021 and was not sampled. Macroinvertebrates have never been sampled at this site due to no stream flow.

4.3.9 Cleopatra Creek

4.3.9.1 2021 Data

Residual pockets of water were present, but water was not flowing through the riffles at Site CC-1A-BIO in 2019 to 2021. Therefore, macroinvertebrates were not sampled.

4.3.9.2 Historic Data

The macroinvertebrate community at Site CC-1A-BIO changed slightly from 2006 to 2018. Richness metrics, Density and Number of Taxa, the composition metric, Community Loss Index, and the tolerance metric, Number of Intolerant Taxa, significantly improved over the years the site was sampled (Table 4-23; Figure 4-33; Figure 4-34; Appendix C). In addition, Univoltine Taxa have significantly decreased in quality over time. Historic median Number of Taxa and EPT Taxa, Diversity, and HBI are also poorer at Site CC-1A-BIO ($P = 0.003$, $P = 0.006$, $P = 0.002$, and $p = 0.015$, respectively). These changes are largely attributed to a few dominant taxa collected in 2015 and 2018 that influenced the relationship.

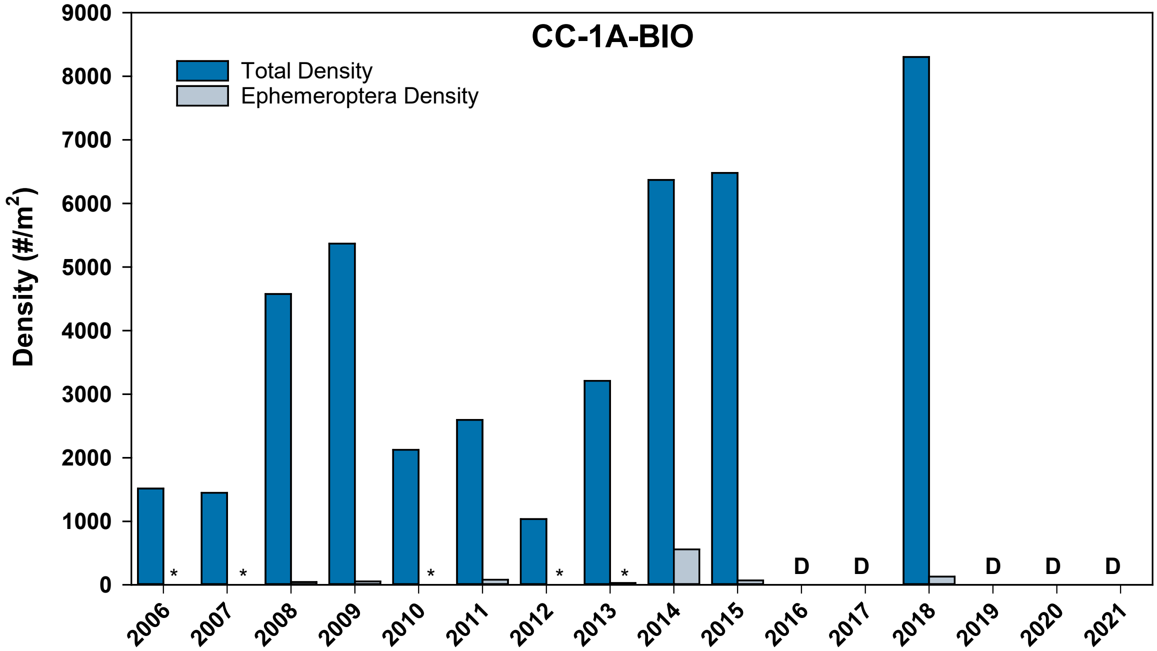


Figure 4-33: Macroinvertebrate density metrics for Site CC-1A-BIO on Cleopatra Creek, 2006 - 2021. * = Mayflies absent or present at low density. D = Dry.

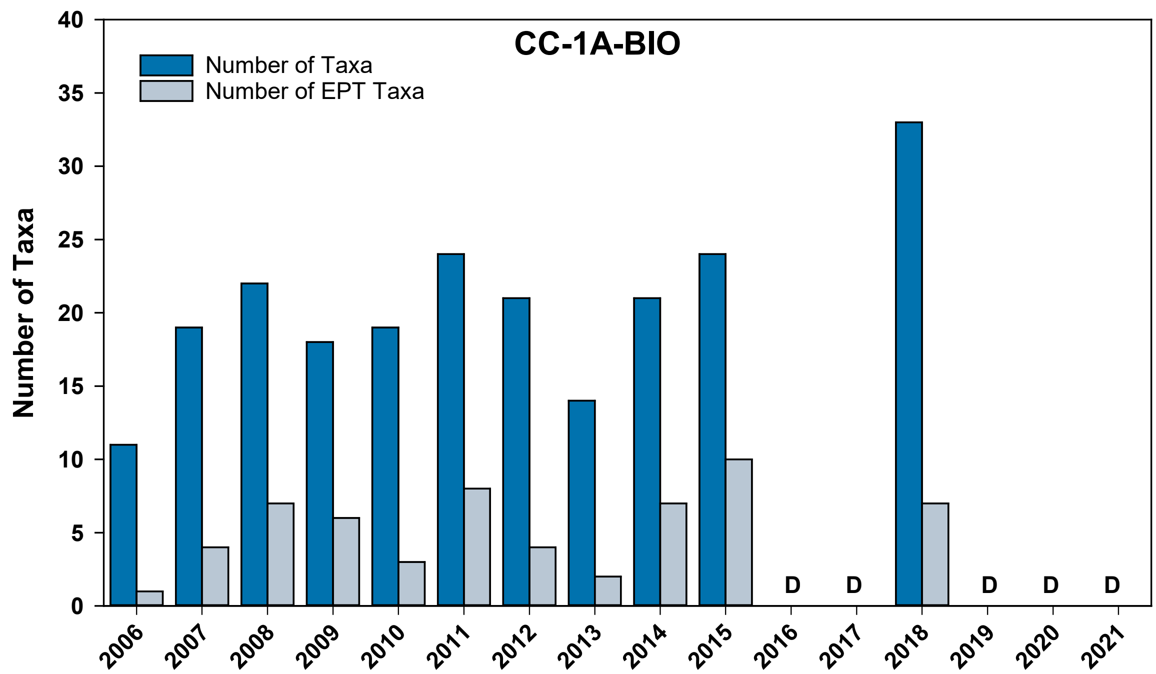


Figure 4-34: Macroinvertebrate taxa richness metrics for Site CC-1A-BIO on Cleopatra Creek, 2006 - 2021. NS = Not Sampled. D = Dry.

Table 4-23: Slopes of significant trends ($p < 0.05$) for macroinvertebrate population metrics at Cleopatra Creek, Fantail Creek, Nevada Gulch, and Stewart Gulch, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant.

Metric	Change Expected Following Environmental Disturbance	CC-1A-BIO	FC-1-BIO	NG-2-BIO	SG-1-BIO
RICHNESS METRICS					
Density (#/sample)	Decrease	+ 470.15	--	--	+ 311.24
Number of Taxa	Usually Decrease	+ 1.09	--	--	+ 0.59
Number of EPT Taxa	Decrease	--	--	--	--
COMPOSITION METRICS					
Shannon-Weaver Diversity Index	Decrease	--	--	- 0.10	--
Percent Sensitive EPT Taxa	Decrease	--	- 0.01	--	--
EPT Index	Decrease	--	- 0.01	--	--
Percent *Baetis sp.*	Increase	--	--	--	--
Number of non-*Baetis* Ephemeroptera	Decrease	--	--	- 12.07	--
Percent Chironomidae	Increase	--	--	- 0.04	--
Number of Plecoptera Taxa	Decrease	--	--	--	--
Percent Abundance of Oligochaetes & Hirudinea	Variable	--	--	0.00	--
Percent Dominant Taxon	Increase	--	--	+ 0.02	--
Number of Common Taxa	Decrease	--	--	--	--
Community Loss Index	Increase	- 0.09	--	--	--
TOLERANCE METRICS					
Hilsenhoff Biotic Index	Increase	--	--	- 0.06	--
Percent Tolerant Taxa	Increase	--	--	--	--
Percent Intolerant Taxa	Decrease	--	--	--	--
Number of Intolerant Taxa	Decrease	+ 0.60	--	--	--
TROPHIC HABIT METRICS			--		
Number of Predator Taxa	Decrease	--	--	--	+ 0.27
Percent Collector-Gatherers	Variable	--	--	- 0.02	--
Number of Shredder Taxa	Decrease	--	--	--	--
LIFE HISTORY METRICS					
Number of Univoltine Taxa	Increase	+ 0.68	--	--	--
Number of Merovoltine Taxa	Decrease	--	--	--	--
Number of Semivoltine Taxa	Decrease	--	--	--	--
Percent Semivoltine Taxa	Decrease	--	--	--	--

Note: CC-1A-BIO was sampled in 2006-2015 and 2018.

4.3.10 Fantail Creek

4.3.10.1 2021 Data

At Site FC-1-BIO in August 2021, richness, tolerance, and trophic habit metric values were all mildly to very favorable (Table 4-24; Table 4-25; Appendix C). Specifically, Density, Number of Taxa, and Number of EPT Taxa values were all good when compared to historical data in the Black Hills; the HBI indicated a "Very Good" benthic macroinvertebrate community including intolerant species; and diversity of feeding types were present.

Composition metric values were not consistently favorable. Diversity indicated a healthy invertebrate community. However, EPT related metric values indicated a fair EPT assemblage that was dominated by *Baetis tricaudatus cx.* (96%) mayflies. This percentage resulted in a fair EPT Index and poor metric values for Percent Sensitive EPT Taxa and Number of non-*Baetis* Ephemeroptera. Life history metric values indicated that most taxa at the site have short life cycles. This section of the stream exhibits low flow, and the retention pond limits the drift of benthic invertebrates from upstream. Despite the poor Ephemeroptera community (Appendix B), metrics for this site indicate that the stream conditions in 2021 supported a diverse benthic macroinvertebrate community that included intolerant species.

Table 4-24: Macroinvertebrate density (number of organisms/sample) at Fantail Creek, Nevada Gulch, Stewart Gulch, August 2021.

Taxa	FC-1-BIO	NG-2-BIO	SG-1-BIO
INSECTA			
Collembola (springtails)	--	4	--
Ephemeroptera (mayflies)	66	16	1,295
Plecoptera (stoneflies)	115	44	760
Hemiptera (true bugs)	--	4	--
Megaloptera (alderflies)	--	4	--
Coleoptera (beetles)	151	1,040	1,900
Trichoptera (caddisflies)	57	17	1,910
Diptera (true flies)	479	412	600
HYDRACARINA (water mites)	3	--	30
TURBELLARIA (flatworms)	160	4	5
ANNELIDA (segmented worms)			
Oligochaeta (worms)	19	76	30
MOLLUSCA			
Pelecypoda (clams)	3	--	--

Benthic macroinvertebrate richness, composition, tolerance, and trophic habit metric values at Site FC-1-BIO were somewhat comparable to those at the reference site, Site RC-1-BIO. Both sites contained a favorable Number of Taxa, Number of EPT Taxa, Diversity, and HBI and fair EPT Index metric values. However, all values but the HBI were more favorable at the reference site. The Percent *Baetis sp.* and Number of non-*Baetis* Ephemeroptera metrics were more favorable at the reference site. Alternatively, Percent Chironomidae, HBI, and trophic habit metrics were generally better at Site FC-1-BIO. The Community Loss Index indicated that approximately 55% of the taxa present at the reference site were also found at Site FC-1-BIO. Generally, the benthic macroinvertebrate community in Fantail Creek was less healthy than the Reno Creek site but differences were minimal and both sites displayed a healthy macroinvertebrate community.

Table 4-25: Macroinvertebrate population metrics at Fantail Creek, Nevada Gulch, Stewart Gulch, August 2021.

Metric	FC-1-BIO	NG-2-BIO	SG-1-BIO
RICHNESS METRICS			
Density (#/sample)	1,053	1,621	6,530
Number of Taxa	40	37	38
Number of EPT Taxa	11	8	12
COMPOSITION METRICS			
Shannon-Weaver Diversity Index	4.32	3.04	3.20
Percent Sensitive EPT Taxa	17.5%	16.2%	26.3%
EPT Index	27.5%	21.6%	31.6%
Percent *Baetis sp.*	95.5%	25.0%	97.3%
Number of non-*Baetis* Ephemeroptera	3	12	35
Percent Chironomidae	33.2%	23.9%	6.8%
Number of Plecoptera Taxa	3	4	3
Percent Abundance of Oligochaetes & Hirudinea	1.8%	4.7%	0.5%
Dominant Taxon (Tolerance value)	*Polycelis coronata*	*Optioservus divergens*	*Heterlimnius corpulentus*
Percent Dominant Taxon	15.2%	44.7%	28.3%
Number of Common Taxa	19	19	23
Community Loss Index	67.5%	73.0%	84.2%
TOLERANCE METRICS			
Hilsenhoff Biotic Index	3.85	4.54	3.13
Percent Tolerant Taxa	17.5%	27.0%	23.7%
Percent Intolerant Taxa	45.0%	43.2%	44.7%
Number of Intolerant Taxa	18	16	17
TROPHIC HABIT METRICS			
Number of Predator Taxa	12	10	9
Percent Collector-Gatherers	30.3%	17.5%	55.2%
Number of Shredder Taxa	8	6	7
LIFE HISTORY METRICS			
Number of Univoltine Taxa	20	17	16
Number of Merovoltine Taxa	2	3	1
Percent Semivoltine Taxa	0.0%	2.7%	0.0%
Number of Semivoltine Taxa	0	1	0

4.3.10.2 Historic Data

The macroinvertebrate community at Site FC-1-BIO has changed very little between 2006 to 2021. The 2021 richness metrics did not trend but Density starting in 2013 was generally greater than 2006 to 2012 (Table 4-23; Figure 4-35; Figure 4-36; Appendix C). Composition metric values largely stayed consistent with only Percent Sensitive EPT Taxa and EPT Index significantly decreasing in quality since 2006, indicating poorer conditions for EPT taxa in recent years. The 2021 HBI tolerance metric was better than most recent years with data ranging from "Good" (5.4 in 2012) to "Excellent" (3.2 in 2020). In addition, the Percent Tolerant Taxa was better than previously recorded (previous minimum of 17.9% in 2009 and 2010). Historic median data for the select benthic macroinvertebrate community metrics were not significantly different between Site FC-1-BIO and its reference site, Site RC-1-BIO.

The combination of low flow and increased amount of fine-grained sediment in recent years have likely influenced the macroinvertebrate community at Site FC-1-BIO. However, historical data exhibited negligible change with many metric values being similar to those in previous years and not different than the reference site.

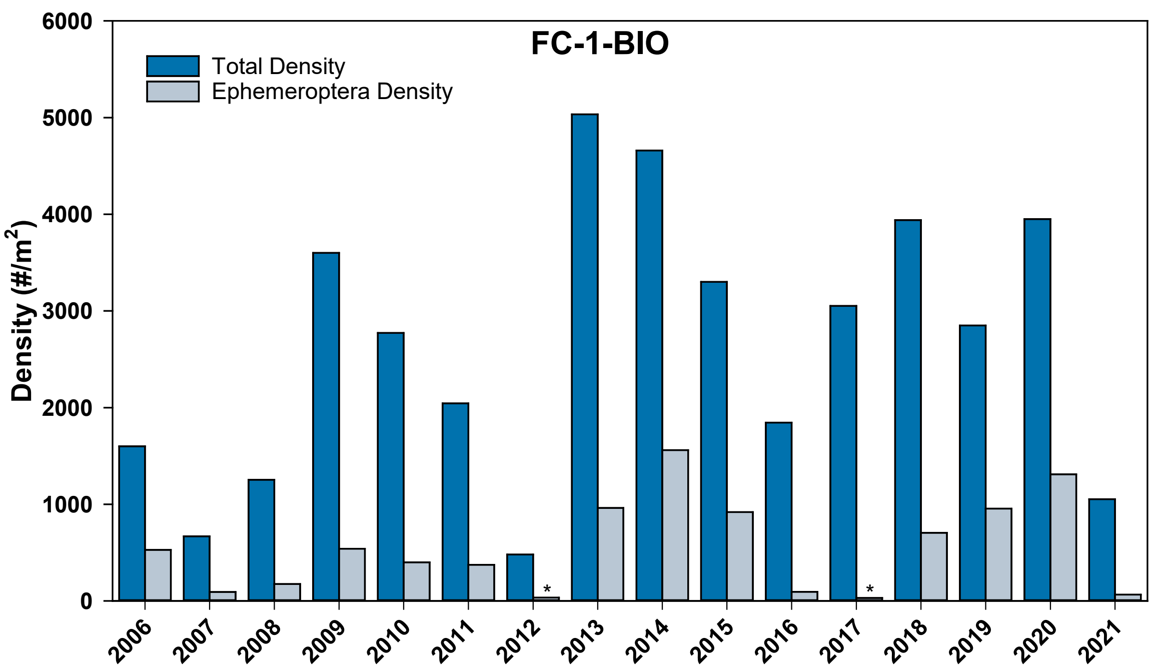


Figure 4-35: Macroinvertebrate density metrics for Site FC-1-BIO on Fantail Creek, 2006 - 2021. * = Mayflies present at low density.

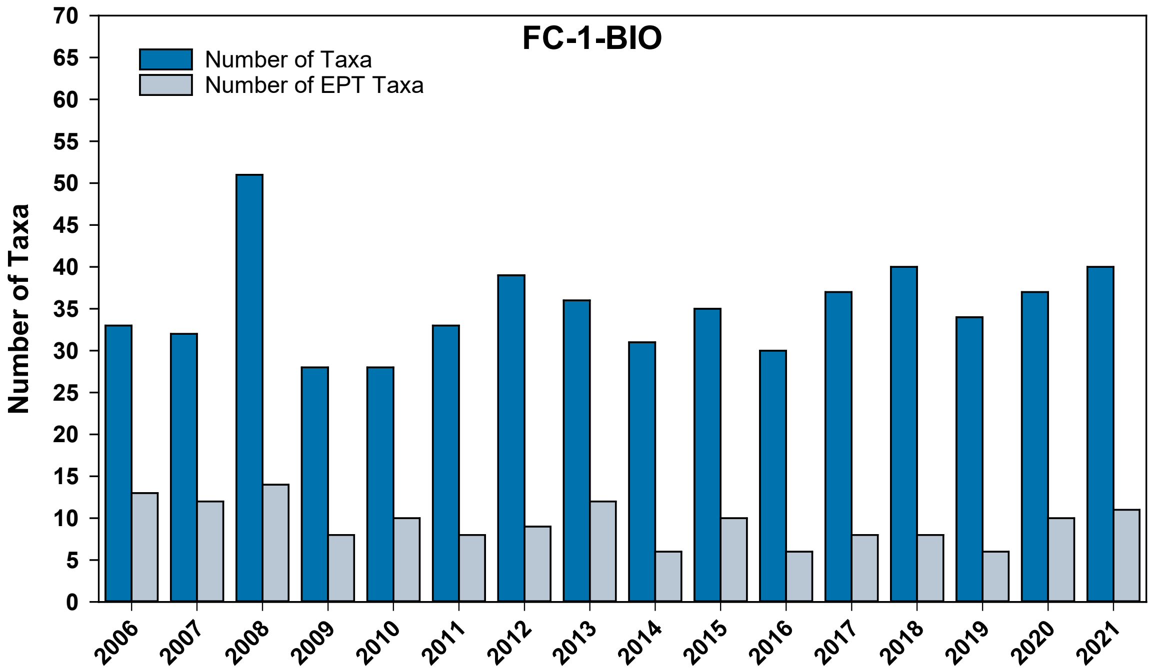


Figure 4-36: Macroinvertebrate taxa richness metrics for Site FC-1-BIO on Fantail Creek, 2006 - 2021.

4.3.11 Nevada Gulch

4.3.11.1 2021 Data

In August 2021, metric values were predominantly mildly to very favorable at Site NG-2-BIO (Table 4-24; Table 4-25; Appendix C). Density, Number of Taxa, and Number of EPT Taxa values were all fair or good when compared to historical data in the Black Hills; Diversity indicated a balanced invertebrate community; the HBI indicated a "Very Good" benthic macroinvertebrate community including intolerant species; and a diversity of feeding groups and life cycle lengths were present. However, the diversity value was poorer than all other Wharf sample sites and poor metric values for Percent Sensitive EPT Taxa, EPT Index, Number of non-*Baetis* Ephemeroptera indicated a limited EPT community. The macroinvertebrate community at Site NG-2-BIO consisted primarily of Coleoptera, specifically *Optioservus divergens* and *O. seriatus*, moderately sensitive species, resulting in a poorer Percent Dominant Taxon value than at all other sites. These deficiencies are the result of the low flows and resulting increased sedimentation observed in the past few years, which is strongly linked to the salting and sanding and general run-off from Nevada Gulch Road. However, the dominance by the moderately sensitive riffle beetle species is an improvement over the previous year. Despite the small EPT community, overall, metrics for this site indicated that the stream conditions in 2021 supported a moderately healthy benthic macroinvertebrate community (Appendix B).

Benthic macroinvertebrate richness and composition metric values were worse at Site NG-2-BIO than at the reference site, Site RC-1-BIO, with the exception of Percent

Chironomidae. Tolerance, trophic habit, and life history metrics were generally similar between the two sites except for Number of Intolerant Taxa which was poorer at Site NG-2-BIO. These differences between the sites resulted in a Community Loss Index of 73%, indicating that three-quarters of macroinvertebrate taxon present at Site NG-2-BIO were not present at the reference site. The poorer condition of Site NG-2-BIO is likely the result of salting and sanding and general run-off from Nevada Gulch Road. Despite these differences, the Number of Taxa and EPT Taxa, Diversity, and HBI were good and intolerant species were present at both sites. Generally, the benthic macroinvertebrate community in Nevada Gulch was less healthy than the reference site but both sites displayed a healthy macroinvertebrate community.

4.3.11.2 Historic Data

The macroinvertebrate community at Site NG-2-BIO has moderately changed from 2006 to 2021. Richness metric values have not trended in this time (Figure 4-37; Figure 4-38; Table 4-23; Appendix C) while the 2021 composition metrics Diversity, Number of non-*Baetis* Ephemeroptera, and Percent Dominant Taxa have significantly worsened since 2006 and Percent Chironomidae has significantly improved. The HBI tolerance metric also significantly improved over this time. Diversity has only been less than 2.5 in two of last 16 years of sampling and HBI has ranged from “Fair” (5.68 in 2013) to “Very Good” (3.75 in 2020) indicating a healthy invertebrate community was present in most years. In addition, the trophic habit metric, Percent Collector-Gatherers has significantly decreased over time. The 2006 to 2021 median richness metrics were significantly worse at Site NG-2-BIO than at the reference site (Density: $p = 0.010$, Number of Taxa: $p = 0.018$, and Number of EPT Taxa: $p = 0.028$). Overall, historic benthic macroinvertebrate community metric data at Site NG-2-BIO has decreased in quality and is slightly poorer than at the reference site which is likely due to the salting and sanding and general run-off from Nevada Gulch Road.

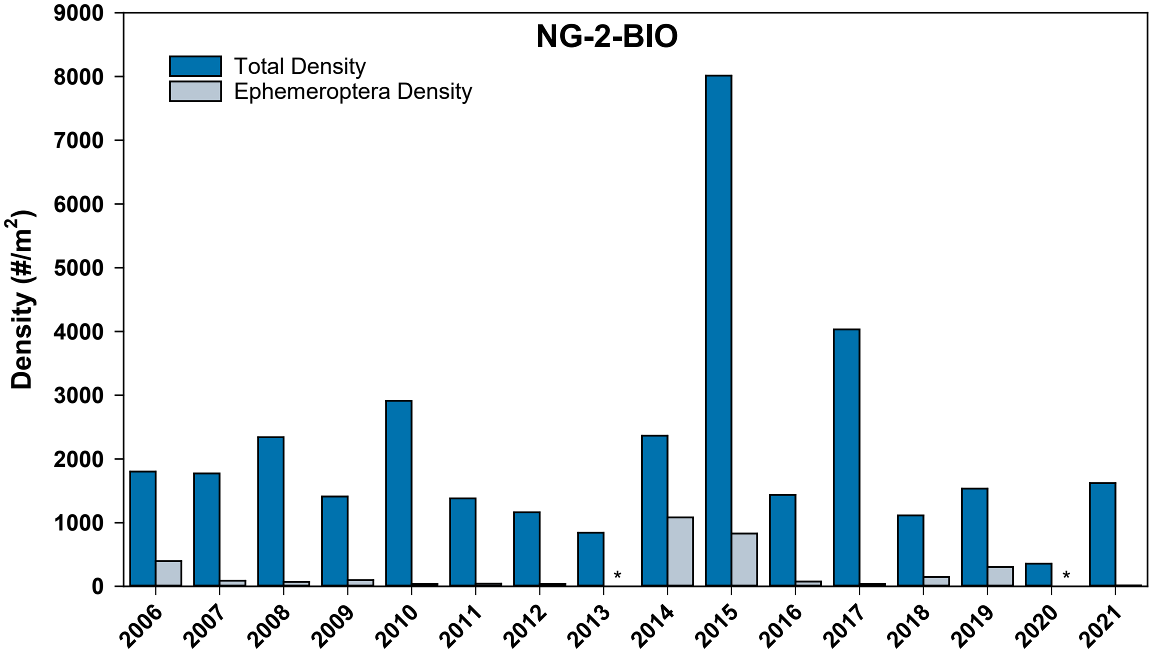


Figure 4-37: Macroinvertebrate density metrics for Site NG-2-BIO on Nevada Gulch, 2006 - 2021. * = Mayflies present at low density. D= Dry.

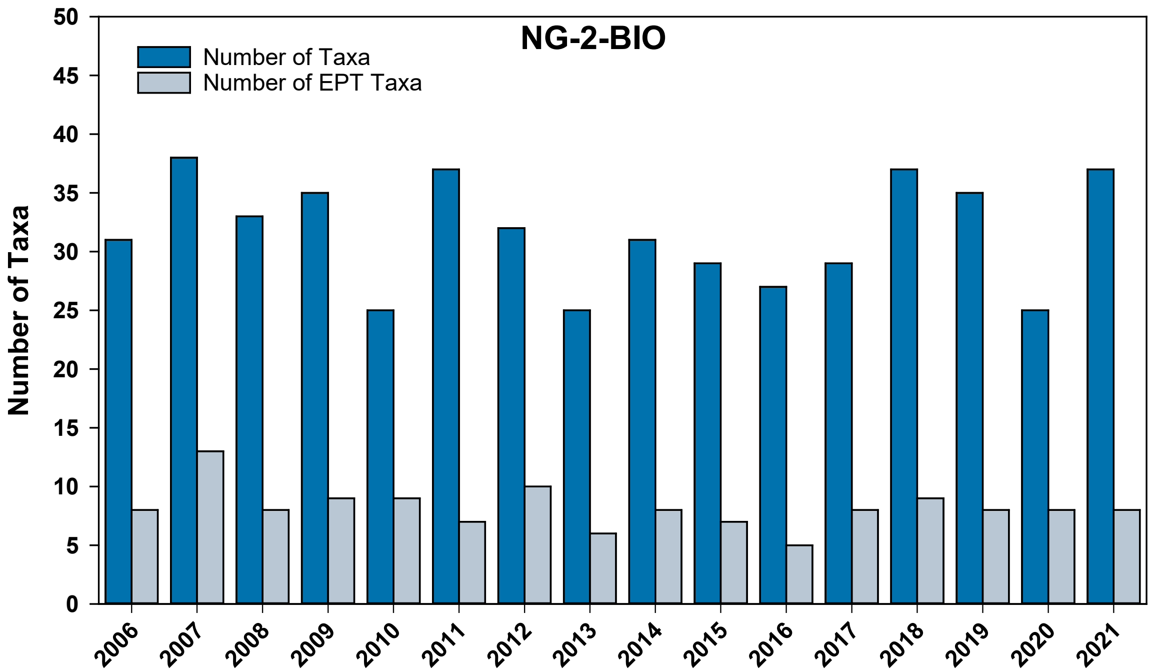


Figure 4-38: Macroinvertebrate taxa richness metrics for Site NG-2-BIO on Nevada Gulch, 2006 - 2021. NS = Not sampled. D= Dry.

4.3.12 Stewart Gulch

4.3.12.1 2021 Data

In August 2021, richness, composition, tolerance, and trophic habit metric values were mostly mildly to very favorable at Site SG-1-BIO (Table 4-24; Table 4-25; Appendix C). Specifically, Density was good and the highest of any site; Number of Taxa, Number of EPT Taxa, and EPT Index values were fair or good when compared to historical data in the Black Hills; Diversity indicated a healthy macroinvertebrate community; and the HBI was better than all other sites and indicated an "Excellent" benthic macroinvertebrate community including intolerant species. However, the Percent *Baetis sp.* metric was the poorest of all sites and indicated that the Ephemeroptera assemblage was comprised almost entirely of tolerant mayflies and that Number of non-*Baetis* Ephemeroptera was very low. In addition, the life history metric values indicated that most taxa at the site have short life cycles. These poor metric values are likely the result of elevated nitrate concentrations from the spring upstream of the site. Overall, despite a few poor metric values, the stream conditions in 2021 supported a rich and diverse benthic macroinvertebrate community (Appendix B) including many intolerant taxa.

Benthic macroinvertebrate richness, tolerance, and trophic habit metric values at Site SG-1-BIO were similar to those at the reference site, Site LB-4-BIO. Both sites revealed favorable or fair metric values for Density, Number of Taxa, Number of EPT Taxa, Diversity, EPT Index, and HBI. Of these, Number of EPT Taxa and Diversity were greater at the reference site while HBI was better at Site SG-1-BIO. In addition, Number of non-*Baetis* Ephemeroptera, Percent Dominant Taxa, Number of Intolerant Taxa, and Number of Predator Taxa values were more favorable at the reference site than at Site SG-1-BIO. Percent Chironomidae and Number of Univoltine Taxa, however, were better at Site SG-1-BIO. Despite these differences between the sites, Site SG-1-BIO contained a healthy benthic macroinvertebrate community in August 2021 that was comparable to the community sampled at the reference site.

4.3.12.2 Historic Data

From 2006 to 2021 at Site SG-1-BIO composition, tolerance, trophic habit, and life history metric values of the macroinvertebrate community remained relatively consistent (Figure 4-39; Figure 4-40; Appendix C). Only Density, Number of Taxa, and Number or Predator Taxa have significantly increased from 2006 to 2021 with no other metric revealing any trend (Table 4-23). Diversity has been greater than 2.5 in all years of sampling indicating a healthy invertebrate community and HBI has fluctuated from the poorest rating of "Good" (4.71) in 2017 to the highest rating of "Excellent" (3.13) in 2021. Community Loss Index was also greatest in 2021 (previous maximum of 76% in 2013).

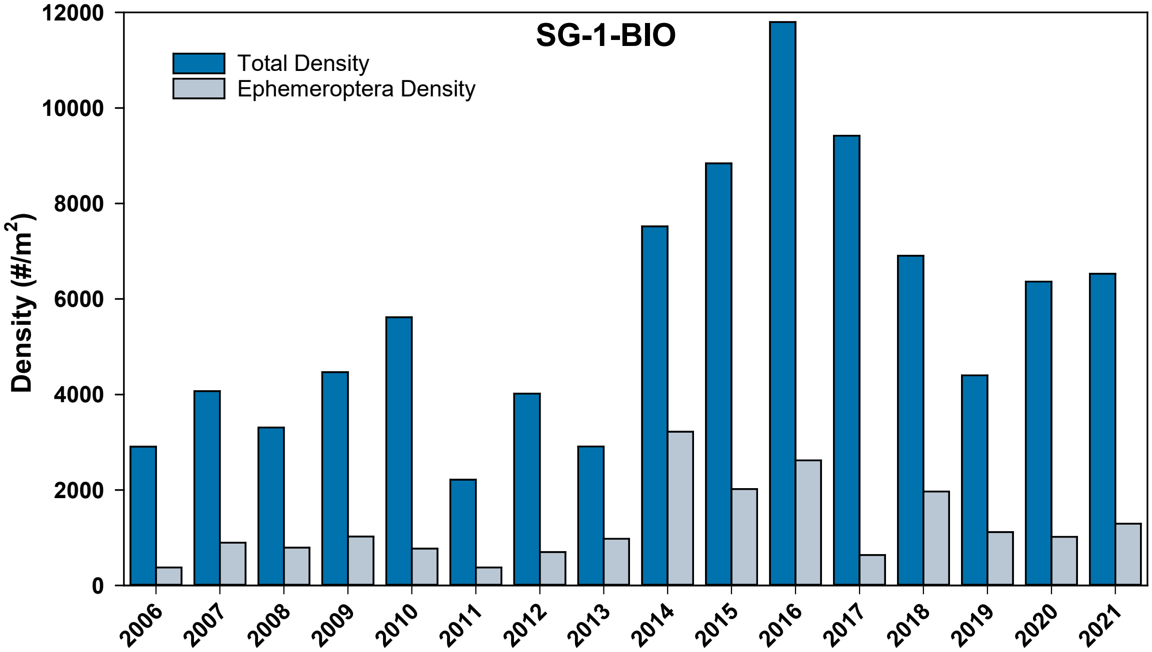


Figure 4-39: Macroinvertebrate density metrics for Site SG-1-BIO on Stewart Gulch, 2006 - 2021.

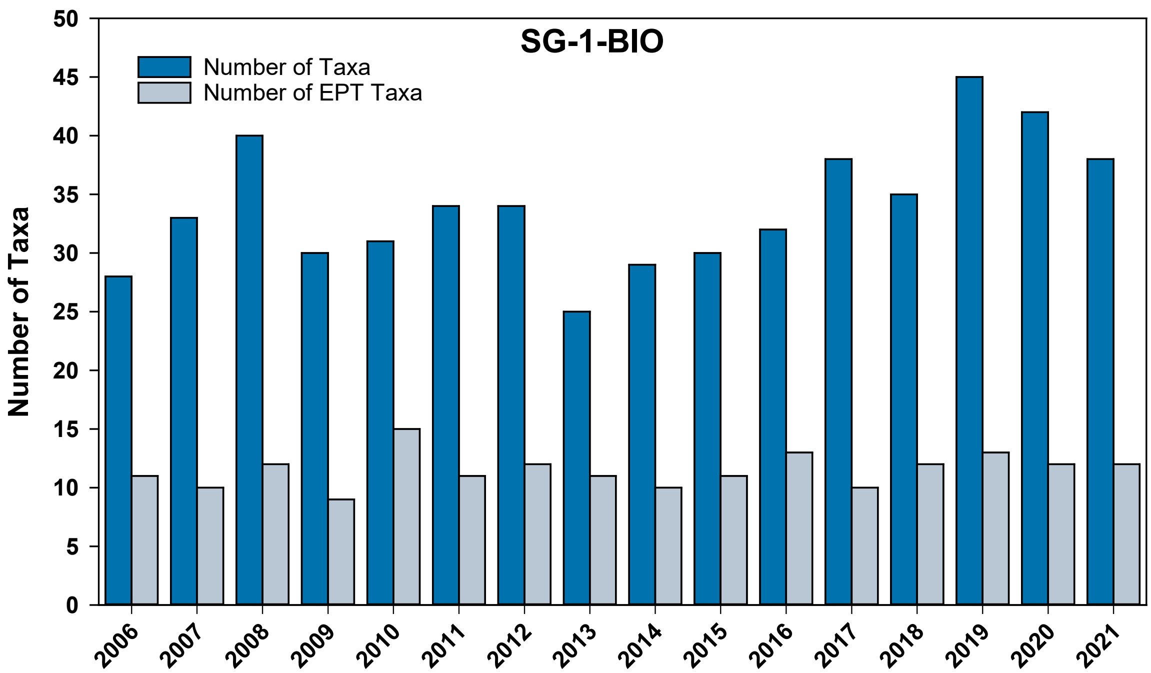


Figure 4-40: Macroinvertebrate taxa richness metrics for Site SG-1-BIO on Stewart Gulch, 2006 - 2021. NS = Not Sampled. D= Dry.

Overall, the 2021 data showed little change with metric values being similar to those in previous years. The long-term median Density was, however, greater at Site SG-1-BIO than at the reference site ($p = 0.029$) while the Number of Taxa and EPT Taxa and Diversity were significantly better at the reference site from 2006 to 2021 ($p = 0.\ 018$, $p = 0.001$, and p = 0.016, respectively). The benthic macroinvertebrate community at Site SG-1-BIO is influenced by elevated nitrate concentrations from the spring upstream of the site causing the increased growth of watercress since 2011. The increased habitat provided by the macrophyte growth may also have contributed to the increased benthic macroinvertebrate density over time. Generally, historic benthic macroinvertebrate community metric data at Site SG-1-BIO reveals rich and diverse macroinvertebrate communities, with numerous intolerant taxa.

4.4 Periphyton Populations

4.4.1 Labrador Gulch

4.4.1.1 2021 Data

In August 2021, the periphyton community at Site LB-4-BIO consisted completely of Pennate diatoms (Table 4-26; Appendix D). Diatoms were more diverse than any other site and the Diversity value for diatoms was well above the 2.5 threshold, indicating a diverse community (Table 4-27). The Autotrophic Index was slightly above 400 and indicated that organic matter may be influencing the site. Pollution tolerant diatoms were present at a small percentage of relative abundance but still more so than at any other site, and motile diatoms were present indicating some organic matter deposition and siltation occurs in this reach. However, the Pollution Index value also showed that a relatively large proportion of the periphyton community was comprised of sensitive diatoms and that minor organic enrichment occurred.

Table 4-26: Relative periphyton density (%) and biomass estimates the reference sites on Labrador Gulch and Reno Creek, August 2021.

Taxa/Metric	LB-4-BIO	RC-1-BIO
BACILLARIOPHYTA		
Pennales (Pennate diatoms)	100.00	100.00
BIOMASS		
AFDM (mg/m^2)	5,372	4,408
Chlorophyll-a (mg/m^2)	10.4	10.8

Table 4-27: Periphyton population metrics for sites at the reference sites on Labrador Gulch and Reno Creek, August 2021.

Metric	LB-4-BIO	RC-1-BIO
RICHNESS		
Density (cells/cm^2)	183,479	313,358
Relative Diatom Density	100%	100%
Number of Taxa (species)	22	14
Number of Diatom Taxa	22	14
Number of Periphyton Divisions	1	1
Number of Periphyton Genera	10	7

Metric	LB-4-BIO	RC-1-BIO
COMPOSITION		
Shannon-Weaver Diversity Index for Diatoms	3.92	2.74
Bahls Similarity Index	--	--
Autotrophic Index	519	410
TOLERANCE		
Percent Tolerant Diatoms	12.5%	3.3%
Lange-Bertalot Pollution Index	2.48	2.83
TROPHIC HABIT		
Percent Eutrophic Diatoms	50.0%	64.3%
Percent Acidiphilic Diatoms	0.0%	0.0%
Percent Alkaliphilic Diatoms	50.0%	50.0%
Percent Nitrogen Heterotrophs	9.1%	28.6%
Percent High Oxygen Diatoms	40.9%	28.6%
Percent Motile Diatoms	54.5%	64.3%
Percent Saprobic Diatoms	9.1%	28.6%

In headwaters streams, such as the ones surveyed for the Wharf Biomonitoring project, organic inputs are often influenced by allochthonous (i.e., outside of the stream) sources, particularly leaf litter from within the watershed. This detritus can then settle to the substrate, be collected as part of the periphyton sample, and potentially skew the Autotrophic Index value by inflating the AFDM measurement. This scenario likely leads to the disagreement between the Pollution Index and Autotrophic Index metrics observed at this and other Wharf sites. Site LB-4-BIO, specifically, contains a large amount of low, overhanging vegetation, log jams, and leaf litter which may have contributed to the poor Autotrophic Index. In 2021, the chlorophyll-a concentration was also high resulting in the Autotrophic Index not being as poor as calculated in previous years (maximum of 1,534 in 2016). Taking this into account, this site appeared to have a relatively healthy periphyton community in 2021.

4.4.1.2 Historic Data

The periphyton community at Site LB-4-BIO changed moderately from 2006 to 2021. Pennate diatoms have been the dominant group for every year, with centric diatoms and cyanobacteria also occasionally found at this site. Richness metric values for Numbers of Taxa significantly increased while the Number of Periphyton Divisions decreased over the time period (Figure 4-41; Table 4-28; Appendix E). For the composition metrics, Diversity significantly improved from 2006 to 2021 and Site LB-4-BIO was more diverse in 2021 than previously recorded (previous maximum of 3.87 in 2017). The Autotrophic Index has not significantly trended, but this reference site has been affected by organic matter deposition in most years. The tolerance metric, Percent Tolerant Diatoms, and the habit metric, Percent Motile Diatoms, significantly worsened from 2006 to 2021, although the magnitude was small. Lastly, the Pollution Index has not trended. Overall, despite these few poorer trends, data in 2021 revealed negligible changes with metric values being similar to those observed in recent years.

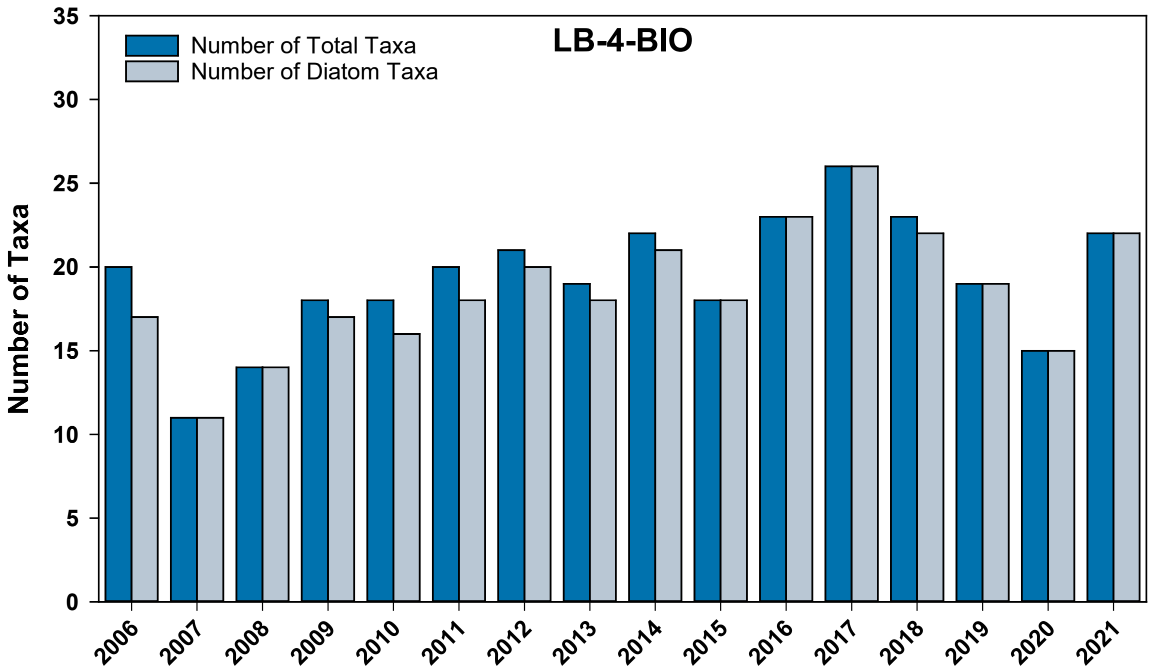


Figure 4-41: Periphyton taxa richness metrics at Site LB-4-BIO on Labrador Gulch, 2006 – 2021.

Table 4-28: Slopes of significant trends ($p < 0.05$) for periphyton population parameters at the reference sites on Labrador Gulch and Reno Creek, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant. NA = Not applicable.

Metric	Change Expected Following Environmental Disturbance	LB-4-BIO	RC-1-BIO
RICHNESS			
Density (cells/cm^2)	Variable	--	--
Relative Diatom Density	Variable	--	--
Number of Taxa (species)	Decrease	--	--
Number of Diatom Taxa	Decrease	+ 0.47	--
Number of Periphyton Divisions	Variable	- 0.20	--
Number of Periphyton Genera	Variable	--	--
COMPOSITION			
Shannon-Weaver Diversity Index for Diatoms	Decrease	+ 0.06	--
Bahls Similarity Index	Variable	NA	NA
Autotrophic Index	Increase	--	--
TOLERANCE			
Percent Tolerant Diatoms Density	Increase	+ 0.01	--
Lange-Bertalot Pollution Index	Decrease	--	--

Metric	Change Expected Following Environmental Disturbance	LB-4-BIO	RC-1-BIO
TROPHIC HABIT			
Percent Eutrophic Diatoms	Variable	--	--
Percent Acidiphilic Diatoms	Variable	--	--
Percent Alkaliphilic Diatoms	Variable	--	--
Percent Nitrogen Heterotrophs	Variable	--	--
Percent High Oxygen Diatoms	Variable	--	
Percent Motile Diatoms	Increase	+ 0.01	+ 0.06
Percent Saprobic Diatoms	Variable	--	--

4.4.2 Reno Creek

4.4.2.1 2021 Data

Pennate diatoms comprised 100% of the periphyton community at Site RC-1-BIO in 2021 (Table 4-26; Appendix D). The Diversity value for diatoms was above the threshold of 2.5 (Table 4-27), indicating a diverse community. The Autotrophic Index was roughly a third of that in 2020 but still slightly above 400, and indicated that organic matter deposition was affecting the site. The improvement in Autotrophic Index resulted from the AFDM returning to 2017 and 2018 concentrations while the chlorophyll-a content increased substantially in 2021. As a headwater stream, there appears to have been some allochthonous loading to the reach which inflated the Autotrophic Index as is the case with Site LB-4-BIO. Pollution tolerant diatoms were present and Percent Motile Diatoms was poor indicating siltation. In addition, this site had some of the poorest richness and trophic habitat metric values of any site in the Wharf study. However, the Pollution Index value also showed that a relatively large proportion of the periphyton community was comprised of sensitive diatoms and that no organic matter pollution occurred. As with Site LB-4-BIO, allochthonous plant matter likely affected this site resulting in the contradicting results. Overall, this site appeared to have a healthy periphyton community in 2021.

4.4.2.2 Historic Data

The periphyton community at Site RC-1-BIO changed very little since 2017. Pennate diatoms have been the dominant group for every year. Richness, composition, and tolerance metric values have not trended (Table 4-28). The Autotrophic Index reveals that this reference site has been affected by organic matter deposition in most years while Diversity and Pollution Index values have been variable. The only metric to trend was the trophic habit metric, Percent Motile Diatoms, which significantly improved from 2017 to 2021. In addition, two metrics were more favorable while six were less favorable in 2021 than previous years. However, these extremes are not surprising as only five years of data have been collected. Overall, the presence/absence of trends at Site RC-1-BIO should be cautiously interpreted given the limited years of data collection.

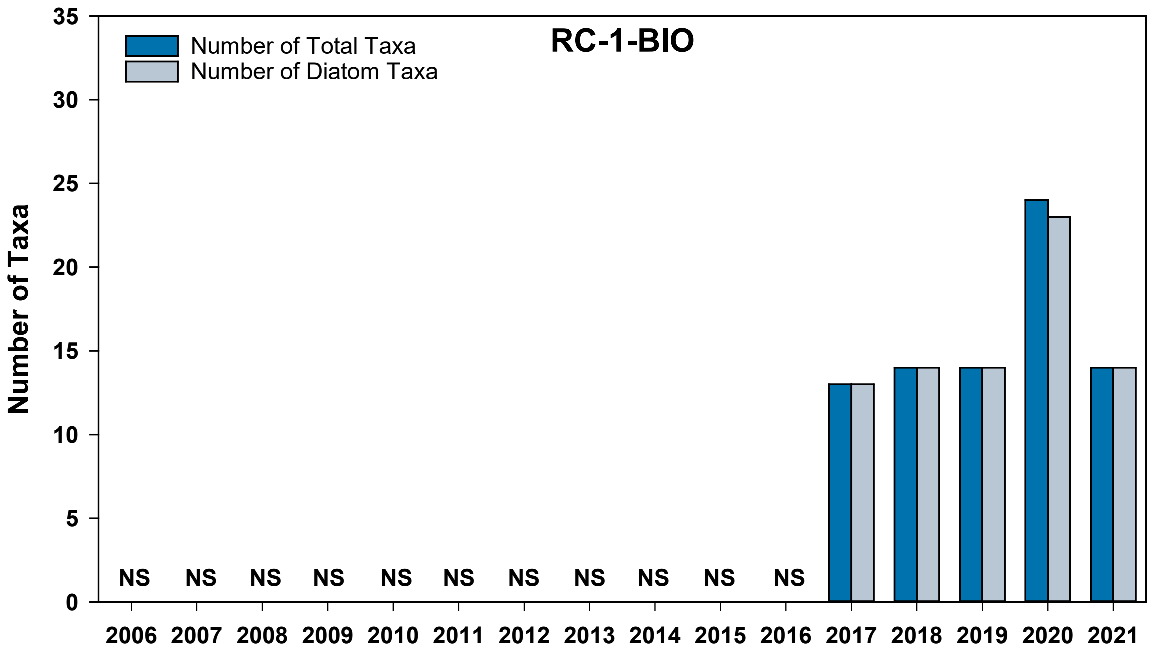


Figure 4-42: Periphyton taxa richness metrics at Site RC-1-BIO on Labrador Gulch, 2006 – 2021. NS = not sampled.

4.4.3 Annie Creek

4.4.3.1 Site AC-1-BIO

4.4.3.1.1 2021 Data

In August 2021 at Site AC-1-BIO, the periphyton assemblage consisted of pennate diatoms, green algae, and cyanobacteria (Table 4-29; Appendix D). The Diversity value for diatoms was above the 2.5 threshold, indicating a diverse community (Table 4-30). This site was less affected by organic pollution than any other site, but the Autotrophic Index was still fair and indicated that organic enrichment likely occurred. Pollution tolerant and motile diatoms were present and indicated some organic matter deposition occurs in this reach. Despite having a Pollution Index value poorer than all other sites, the metric indicated that only minor organic enrichment has occurred and that the periphyton community contained some sensitive diatoms.

Table 4-29: Relative periphyton Density (%) and biomass estimates for sites on Annie Creek, August 2021.

Taxa/Metric	AC-1-BIO	AC-2-BIO
BACILLARIOPHYTA		
Pennales (Pennate diatoms)	66.93	98.58
CHLOROPHYTA (Green algae)	1.58	0.71
CYANOPHYTA (Cyanobacteria)	31.50	0.71
BIOMASS		
AFDM (mg/m^2)	11,570	6,061
Chlorophyll-a (mg/m^2)	43.3	21.7

Table 4-30: Periphyton population metrics for sites on Annie Creek, August 2021.

Metric	AC-1-BIO	AC-2-BIO
RICHNESS		
Density ($cells/cm^2$)	535,963	452,432
Relative Diatom Density	67%	99%
Number of Taxa (species)	25	21
Number of Diatom Taxa	21	19
Number of Periphyton Divisions	3	3
Number of Periphyton Genera	15	12
COMPOSITION		
Shannon-Weaver Diversity Index for Diatoms	3.08	3.20
Bahls Similarity Index	13.6%	61.1%
Autotrophic Index	268	279
TOLERANCE		
Percent Tolerant Diatoms Density	4.7%	0.0%
Lange-Bertalot Pollution Index	2.20	2.81
TROPHIC HABIT		
Percent Eutrophic Diatoms	47.6%	42.1%
Percent Acidiphilic Diatoms	0.0%	0.0%
Percent Alkaliphilic Diatoms	38.1%	31.6%
Percent Nitrogen Heterotrophs	14.3%	0.0%
Percent High Oxygen Diatoms	33.3%	42.1%
Percent Motile Diatoms	57.1%	42.1%
Percent Saprobic Diatoms	14.3%	5.3%

Periphyton richness, composition, tolerance, and trophic habit metric values at Site AC-1-BIO were mostly dissimilar to those at the reference site, Site RC-1-BIO, in 2021. The richness metrics Density, Number of Taxa, Number of Diatom Taxa, and Number of Periphyton Taxa were all greater at Site AC-1-BIO. In addition, the composition metrics Diversity and Autotrophic Index and more than half of the Trophic Habit metrics were more favorable at Site AC-1-BIO. Only the tolerance metric Pollution Index, was more favorable at the reference site. Lastly, Bahls Similarity Index revealed a “Very Dissimilar” assemblage between Site AC-1-BIO and the reference site. However, Diversity was good at both sites, Autotrophic index showed both sites are affected by some organic pollution, the Pollution index revealed the sites to be affected by none or minor organic enrichment, and both sites contained a similar percentage of motile diatoms. Overall, both sites appeared to have a healthy periphyton community in 2021 despite the metrics at Site AC-1-BIO being more favorable than at the reference site.

4.4.3.1.2 Historic Data

The periphyton community at Site AC-1-BIO has been variable over the years with most metrics indicating no significant trends from 2006 to 2021. Pennate diatoms have been the dominant group in most years, although green algae were dominant in 2010 and 2011. Cyanobacteria and cryptomonads have also been present in small numbers in some years. Richness, composition, and tolerance metric values have not significantly trended from 2006 to 2021 (Figure 4-43; Appendix E). Diversity at Site AC-1-BIO has been good in most years while Autotrophic Index and Pollution Index values have been variable. Two trophic habit metrics, Percent Eutrophic and Saprobic Diatoms, significantly improved, albeit slightly from 2006 to 2021. When compared to the reference site, Site RC-1-BIO, only long-term Autotrophic Index data, of the eight metrics assessed, were significantly different ($p = 0.017$) with Site AC-1-BIO being more favorable. Overall, Site AC-1-BIO has supported a healthy periphyton assemblage since 2006 similar to the reference site conditions.

Table 4-31: Slopes of significant trends ($p < 0.05$) for periphyton population parameters at Annie Creek, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant.

Metric	Change Expected Following Environmental Disturbance	AC-1-BIO	AC-2-BIO	AC-3-BIO*
RICHNESS				
Density (cells/cm^2)	Variable	--	--	--
Relative Diatom Density	Variable	--	--	--
Number of Taxa (species)	Decrease	--	--	--
Number of Diatom Taxa	Decrease	--	--	--
Number of Periphyton Divisions	Variable	--	--	--
Number of Periphyton Genera	Variable	--	--	--
COMPOSITION				
Shannon-Weaver Diversity Index for Diatoms	Decrease	--	--	--
Bahls Similarity Index	Variable	--	--	--
Autotrophic Index	Increase	--	--	--
TOLERANCE				
Percent Tolerant Diatoms Density	Increase	--	--	--
Lange-Bertalot Pollution Index	Decrease	--	--	--
TROPHIC HABIT				
Percent Eutrophic Diatoms	Variable	- 0.01	--	--
Percent Acidiphilic Diatoms	Variable	--	--	--

Metric	Change Expected Following Environmental Disturbance	AC-1-BIO	AC-2-BIO	AC-3-BIO*
Percent Alkaliphilic Diatoms	Variable	--	- 0.01	--
Percent Nitrogen Heterotrophs	Variable	--	--	--
Percent High Oxygen Diatoms	Variable	--	--	--
Percent Motile Diatoms	Increase	--	--	--
Percent Saprobic Diatoms	Variable	- 0.01	--	--

* AC-3-BIO was not sampled in 2020 and 2021 and trends are for 2006-2019 data.

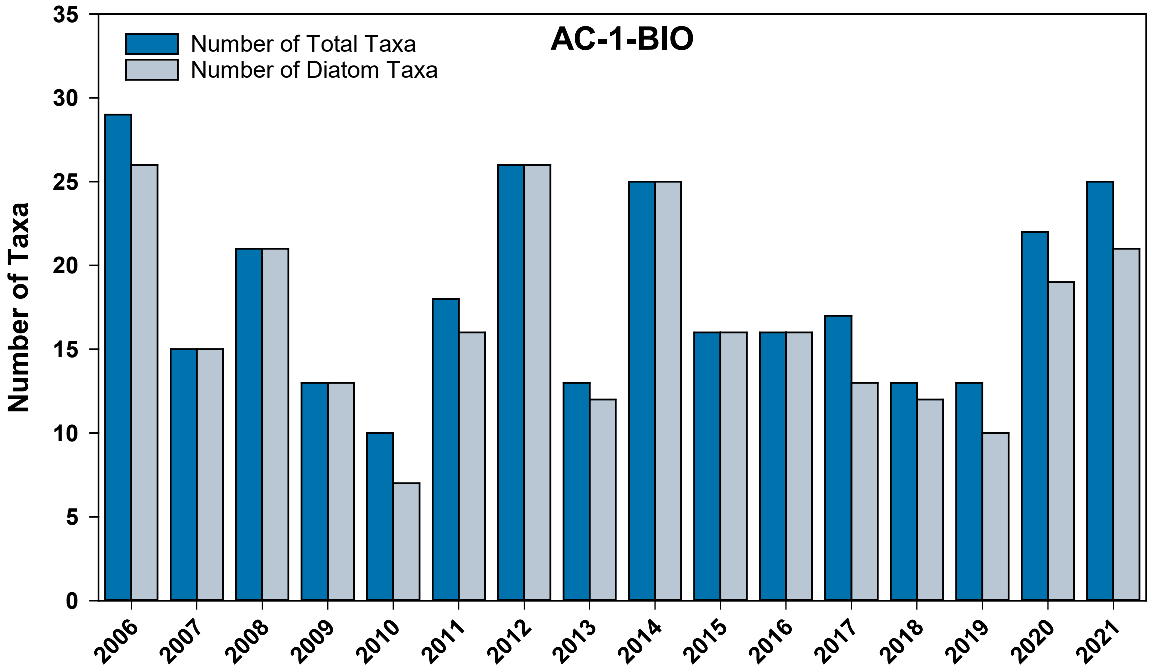


Figure 4-43: Periphyton taxa richness metrics at Site AC-1-BIO on Annie Creek, 2006 – 2021.

4.4.3.2 Site AC-2-BIO

4.4.3.2.1 2021 Data

The periphyton assemblage consisted of pennate diatoms, green algae, and cyanobacteria at Site AC-2-BIO in August 2021 (Table 4-29; Appendix D). The Diversity value for diatoms was well above the 2.5 threshold, indicating a diverse community (Table 4-30). This site was less affected by organic pollution than most other sites, but the Autotrophic Index was still fair and indicated that some organic enrichment likely occurred. Motile diatoms were present and indicated some siltation occurs in this reach. However, no pollution tolerant diatoms were present, and the Pollution Index value indicated that a relatively large proportion of the periphyton community was comprised of sensitive diatoms. Again, the disparity between these metrics indicates that some organic matter deposition is likely from the heavily vegetated stream banks.

In 2021 metric values at Site AC-2-BIO were comparable to those at the reference site, Site LB-4-BIO. Richness, composition, tolerance, and trophic habit metrics were similar between the sites. Density, Autotrophic Index, Percent Tolerant Diatoms, and Pollution Index were slightly more favorable at Site AC-2-BIO than the reference site while diversity was slightly better at the reference site. Despite these small differences, the Bahls Similarity Index indicated "Very Similar" taxa between Site AC-2-BIO and the reference Site LB-4-BIO. Overall, both sites support healthy periphyton communities with Site AC-2-BIO influenced by the increased siltation in recent years.

4.4.3.2.2 Historic Data

The periphyton community at Site AC-2-BIO changed little from 2006 to 2021. Pennate diatoms have been the dominant group in all years while green algae, cyanobacteria, and cryptomonads were present in small numbers in some years. Significant trends were not observed for richness, composition, and tolerance metrics (Table 4-30). Only the trophic habit metric, Percent Alkaliphilic Diatoms, significantly improved from 2006 to 2021, albeit slightly, and was more favorable in 2021 than previous years (previous minimum of 43.8% in 2020; Appendix E). In addition, six other metrics were also more favorable in 2021 than previous years. Diversity at Site AC-2-BIO has not trended but has been good in most years while Autotrophic Index and Pollution Index values have been variable. When Site AC-2-BIO metrics were compared to the reference Site LB-4-BIO metrics, only one of the eight tests indicated a significant difference between the long-term median values. The Number of Taxa was significantly lower ($p = 0.027$) at Site AC-2-BIO than at the reference site, while all other metrics showed no differences. Metric data in 2021 revealed negligible annual changes with many of the metric values being similar to those in previous years.

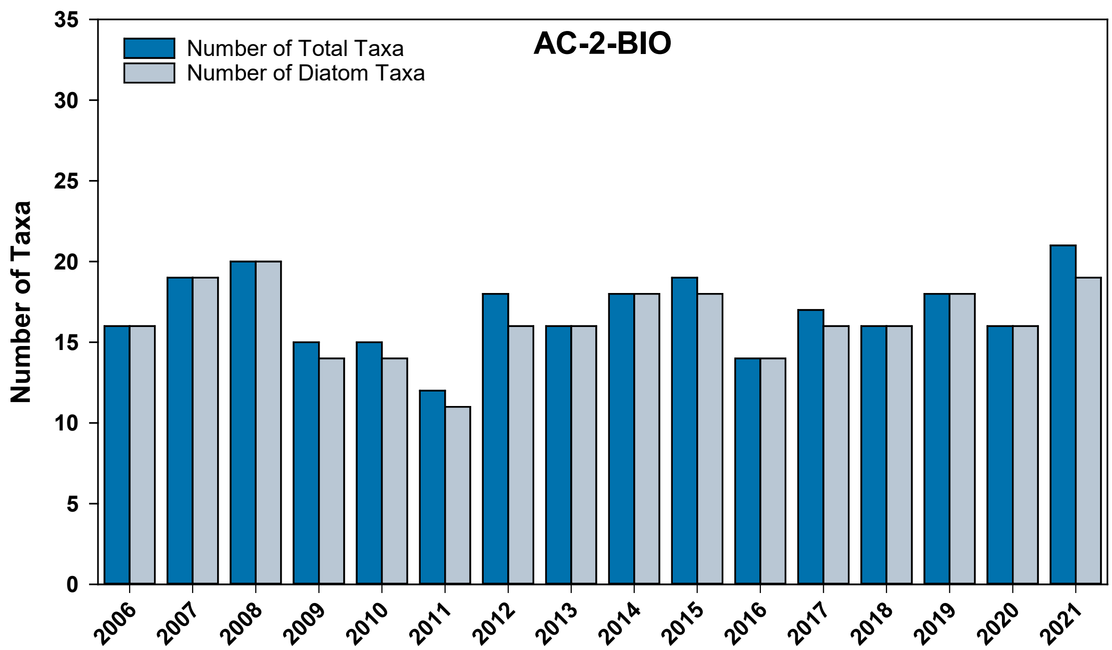


Figure 4-44: Periphyton taxa richness metrics at Site AC-2-BIO on Annie Creek, 2006 - 2021.

4.4.3.3 Site AC-3-BIO

4.4.3.3.1 2021 Data

The majority of Annie Creek at Site AC-3-BIO was not accessible for periphyton monitoring in 2021 due to extensive deadfall from tornado activity in July 2020 and samples were not collected (Photo 4-3).

4.4.3.3.2 Historic Data

From 2006 to 2019, the periphyton community at Site AC-3-BIO was variable with no significant trends (Table 4-30; Appendix E). Pennate diatoms have been the dominant group in all years with green algae and cyanobacteria were present in small numbers in some years. The long-term Autotrophic Index values indicate that this site has been affected by minimal organic matter deposition in most years and Diversity values indicate good diversity in most years. Pollution Index values have been variable. When Site AC-3-BIO metrics were compared to the reference Site LB-4-BIO metrics, only two of the eight tests indicated significant differences between the long-term median values. Density was significantly higher and Autotrophic Index was significantly better ($p = 0.031$ and $p = 0.003$, respectively) at Site AC-3-BIO than at the reference site. Overall, periphyton communities at both sites have had good diversity with little organic enrichment from 2006 to 2019.

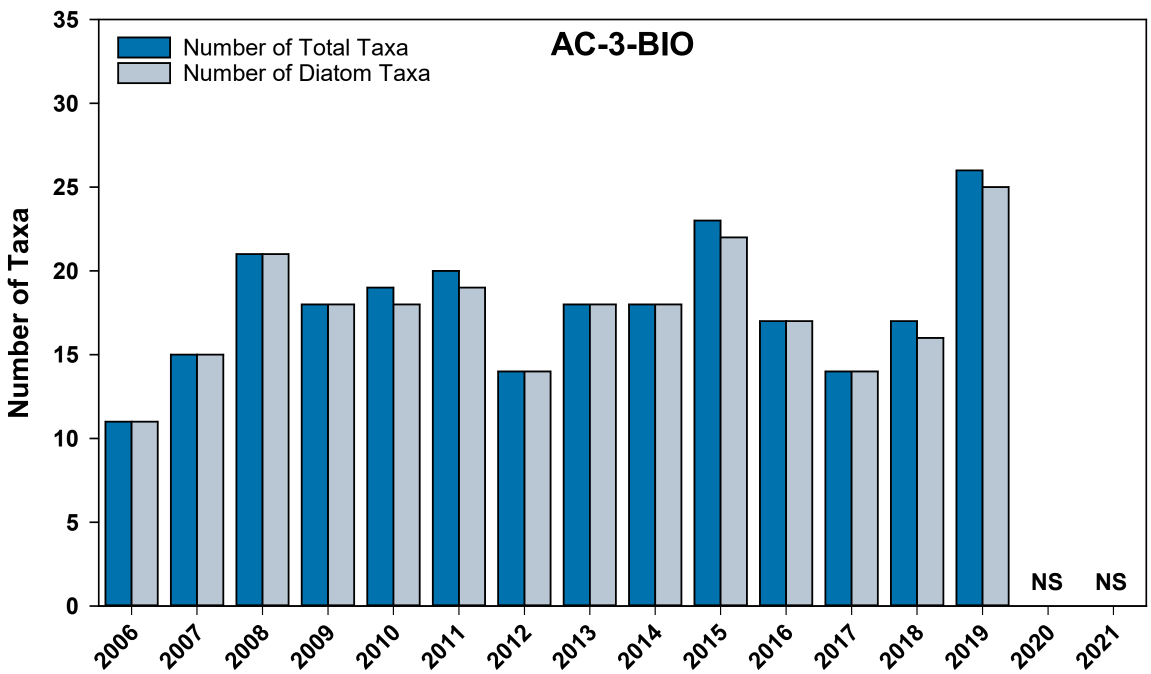


Figure 4-45: Periphyton taxa richness metrics at Site AC-3-BIO on Annie Creek, 2006 – 2021. NS = not sampled.

4.4.3.4 Site Comparisons

4.4.3.4.1 2021 Data

Periphyton metric values were similar between the two Annie Creek sites in 2021. Diversity, Autotrophic Index, and Pollution index were good or fair at both sites; tolerant diatoms were scarce or non-existent, and motile diatoms were present at both sites. The only major difference between these sites was that Site AC-1-BIO was “Very Dissimilar” from its reference site while Site AC-2-BIO was “Very Similar”.

4.4.3.4.2 Historic Data

Median 2006 to 2021 metric values were similar between the three Annie Creek sites. No metrics were significantly different between the sites ($p > 0.05$).

4.4.4 Ross Valley

4.4.4.1 2021 Data

In August 2021, the periphyton assemblage consisted of 99% pennate diatoms, and 1% cyanobacteria at Site RV-2-BIO (Table 4-32; Appendix D). The Diversity value for diatoms was above the 2.5 threshold, indicating a diverse community (Table 4-33), while the Pollution Index was good and Percent Tolerant Diatoms poor, indicated that the periphyton community contained sensitive diatoms and no organic enrichment was occurring. However, the Autotrophic Index metric was just above 400 and Percent Motile Diatom metric was worse than all other sites in 2021 indicating allochthonous inputs, increased algal growth, and siltation. Again, the disparity between these metrics suggests that some organic matter deposition occurs in the stream, likely caused by the large amount of overhanging vegetation and woody debris at the site.

Table 4-32: Relative periphyton Density (%) and biomass estimates for sites on Ross Valley, Lost Camp Gulch, Deadwood Creek, False Bottom Creek, and Cleopatra Creek, August 2021.

Taxa/Metric	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO	WFB-1-BIO
BACILLARIOPHYTA					
Pennales (Pennate diatoms)	98.67	100.00	99.15	--	86.66
CHLOROPHYTA (Green algae)	--	--	0.85	--	10.00
CRYPTOPHYTA (Cryptomonads)	--	--	--	--	3.34
CYANOPHYTA (Cyanobacteria)	1.33	--	--	--	--
BIOMASS					
AFDM (mg/m^2)	8,953	2,893	17,355	2,342	3,857
Chlorophyll-a (mg/m^2)	21.5	7.9	17.3	1.2	3.7

Periphyton richness, composition, tolerance, and trophic habit metric values at Site RV-2-BIO were comparable to those at the reference site, Site RC-1-BIO, in 2021 (Table 4-27, Table 4-33). Density was greater at the reference site than Site RV-2-BIO. However, composition metrics indicated that Diversity was better at Site RV-2-BIO than the reference site. Tolerance metrics were also similar between the two sites while trophic habit metrics were slightly better at Site RV-2-BIO. With these minor differences between the sites, Diversity and Pollution index metrics were good, the Autotrophic Index metrics were equally poor, and the Bahls Similarity Index indicated "Very Similar" taxa between Site RV-2-BIO and the reference site. Overall, both sites appeared to have healthy periphyton communities in 2021.

Table 4-33: Periphyton population metrics for sites on Ross Valley, Lost Camp Gulch, Deadwood Creek, False Bottom Creek, and Cleopatra Creek, August 2021.

Metric	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO*	WFB-1-BIO
RICHNESS					
Density (cells/cm^2)	136,844	178,236	1,122,898	<415	17,920
Relative Diatom Density	99%	100%	99%	--	87%
Number of Taxa (species)	18	18	20	--	12
Number of Diatom Taxa	17	18	19	--	9
Number of Periphyton Divisions	2	1	2	--	3
Number of Periphyton Genera	8	8	15	--	11
COMPOSITION					
Shannon-Weaver Diversity Index for Diatoms	3.48	3.00	2.68	--	1.82
Bahls Similarity Index	68.0%	48.9%	41.0%	--	10.1%
Autotrophic Index	416	365	1,006	--	1,055
TOLERANCE					
Percent Tolerant Diatoms Density	5.4%	4.5%	0.0%	--	0.0%
Lange-Bertalot Pollution Index	2.70	2.78	2.66	--	2.83
TROPHIC HABIT					
Percent Eutrophic Diatoms	52.9%	44.4%	21.1%	--	22.2%
Percent Acidiphilic Diatoms	0.0%	0.0%	10.5%	--	22.2%
Percent Alkaliphilic Diatoms	47.1%	50.0%	26.3%	--	33.3%
Percent Nitrogen Heterotrophs	11.8%	22.2%	5.3%	--	11.1%
Percent High Oxygen Diatoms	23.5%	22.2%	31.6%	--	66.7%
Percent Motile Diatoms	70.6%	50.0%	42.1%	--	33.3%
Percent Saprobic Diatoms	17.6%	11.1%	0.0%	--	0.0%

*Metrics were not calculated

4.4.4.2 Historic Data

The periphyton community at Site RV-2-BIO has been variable over the years, and as a result, few significant trends were observed from 2006 to 2021. Pennate diatoms have been the dominant group in all years while green algae, cyanobacteria, cryptomonads, and golden algae were present in small numbers in some years. The 2021 metric values were largely within the range sampled in previous years except Number of Periphyton Genera which was lower than in previous years (previous minimum of 9 in 2015; Table 4-34; Figure 4-46; Appendix E). Diversity has been good in most years while Autotrophic Index has shown that the site was affected by organic matter deposition in most years. Pollution Index values indicated that sensitive species have primarily inhabited this site over the years and has slightly but significantly improved from 2006 to 2021. However, Percent Motile Diatoms has slightly but significantly worsened indicating siltation. This metric was also worse in 2021 than other years (previous maximum of 70% in 2011). Only two of the eight tests comparing median metric values were significantly different between Site RV-2-BIO and the reference Site RC-1-BIO. Diversity was significantly better (p = 0.013) and Percent Motile Diatoms was significantly worse (p = 0.023) at Site RV-2-BIO than at the reference site. Despite some poor metrics as a result of allochthonous inputs and siltation, the 2021 metric values were similar to those in previous years and to the reference site. The periphyton community has remained relatively consistent in Ross Valley with good diversity with little organic enrichment.

Table 4-34: Slopes of significant trends (p < 0.05) for periphyton population metrics at Ross Valley, Lost Camp Gulch, Deadwood Creek, East Fork False Bottom Creek, and West Fork False Bottom Creek, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant. NA = Not assessed, to many results of zero to run analysis.

Metric	Change Expected Following Environmental Disturbance	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO*	WFB-1-BIO
RICHNESS						
Density (cells/cm^2)	Variable	--	--	--	--	--
Relative Diatom Density	Variable	--	--	--	--	--
Number of Taxa (species)	Decrease	--	--	--	--	--
Number of Diatom Taxa	Decrease	--	--	--	--	--
Number of Periphyton Divisions	Variable	--	--	--	--	--
Number of Periphyton Genera	Variable	--	--	--	--	
COMPOSITION						
Shannon-Weaver Diversity Index for Diatoms	Decrease	--	--	--	--	--
Bahls Similarity Index	Variable	--	--	--	--	--
Autotrophic Index	Increase	--	--	--	--	--
TOLERANCE						
Percent Tolerant Diatoms Density	Increase	--	+ 0.01	- 0.01	--	NA
Lange-Bertalot Pollution Index	Decrease	+ 0.01	--	--	- 0.02	--
TROPHIC HABIT						
Percent Eutrophic Diatoms	Variable	--	--	--	--	- 0.02

Metric	Change Expected Following Environmental Disturbance	RV-2-BIO	LC-1-BIO	DC-2-BIO	EFB-1-BIO*	WFB-1-BIO
Percent Acidiphilic Diatoms	Variable	--	--	--	--	--
Percent Alkaliphilic Diatoms	Variable	--	--	--	--	--
Percent Nitrogen Heterotrophs	Variable	--	--	--	--	- 0.01
Percent High Oxygen Diatoms	Variable	--	- 0.02	--	--	+ 0.02
Percent Motile Diatoms	Increase	+ 0.01	+ 0.01	--	--	--
Percent Saprobic Diatoms	Variable	--	--	--	--	- 0.01

*No 2021 data

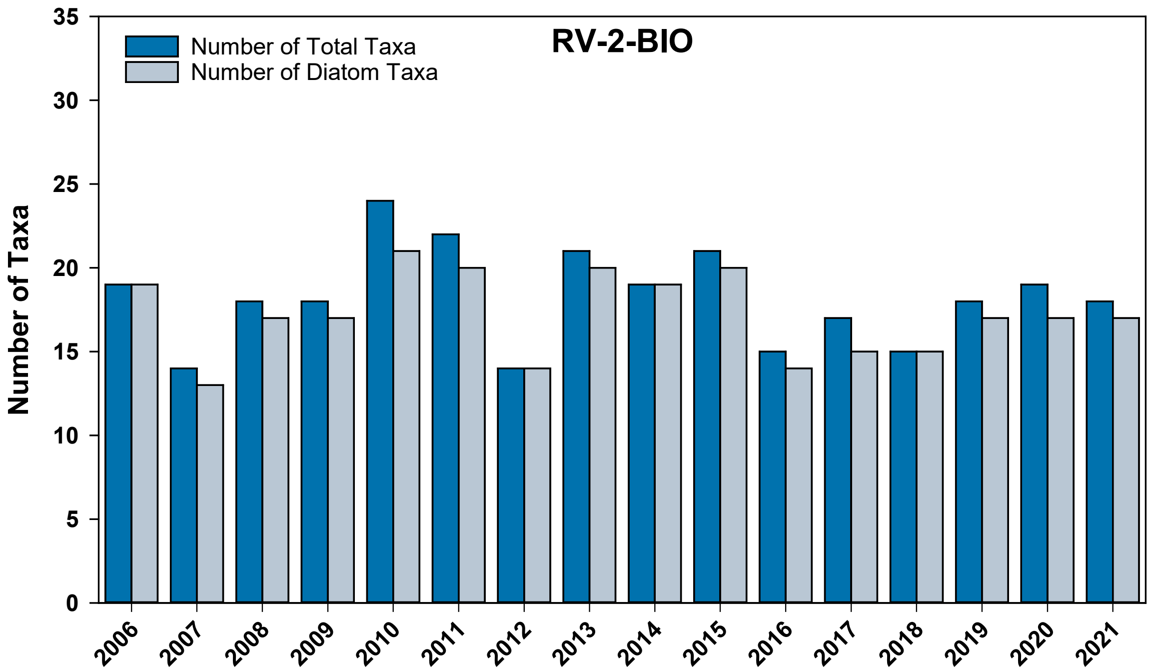


Figure 4-46: Periphyton taxa richness metrics at Site RV-2-BIO on Ross Valley, 2006 – 2021.

4.4.5 Lost Camp Gulch

4.4.5.1 2021 Data

The periphyton assemblage at Site LC-1-BIO in August 2021 consisted of 100% pennate diatoms (Table 4-32; Appendix D). The Diatom Diversity value was above the 2.5 threshold, indicating a balanced periphyton community (Table 4-33) and the Pollution Index was indicative of no organic enrichment. The Autotrophic Index indicated that the stream was enriched with some nutrients and shows a potential for increased periphyton growth. Again, the disparity between these metrics suggests that some organic matter deposition occurs in the stream, likely caused by the large amount of overhanging vegetation and woody debris at the site. A small percentage of tolerant diatoms were present and half of the assemblage at this site were motile diatoms, indicating that siltation was affecting the assemblage. The adjacent dirt roadway acts as a source of sediment for this site.

In 2021, metric values at Site LC-1-BIO were comparable to those at the reference site, Site RC-1-BIO. Density was greater at the reference site, Diversity was good and comparable at both sites, and the Autotrophic Index indicated less organic pollution at Site LC-1-BIO. Tolerance and trophic habit metrics were similar at the sites and the Pollution Index values indicated no organic enrichment. These minor differences between the sites resulted in a Bahls Similarity Index of "Somewhat Similar" taxa between Site LC-1-BIO and the reference site. Overall, both sites appeared to have healthy periphyton communities in 2021 while Site LC-1-BIO is influenced by low flows and siltation.

4.4.5.2 Historic Data

The periphyton community at Site LC-1-BIO changed little since 2010. Pennate diatoms have been the dominant group in all years with green algae, cyanobacteria, and cryptomonads present in small numbers in some years. Richness and composition metric values have not significantly trended over the years (Table 4-34; Figure 4-47; Appendix E). Diversity at Site AC-1-BIO has been good in most years while Autotrophic Index and Pollution Index values have been variable. The tolerance metric, Percent Tolerant Diatoms, and the trophic habit metrics, Percent High Oxygen Diatoms and Motile Diatoms, have slightly but significantly declined in quality since 2010. Also, Percent Nitrogen Heterotrophic Diatom Taxa was poorer in 2021 than any other year (previous maximum of 20% in 2015). The median Autotrophic Index from 2010 to 2021 was significantly better ($p = 0.027$) at Site LC-1-BIO than at the reference site while no other long-term metrics were different between the sites. Overall, the metrics indicate that periphyton conditions have remained relatively favorable at the Lost Camp Gulch site for the duration of the study period, although periodic low flows, sedimentation, and organic enrichment influence the periphyton community.

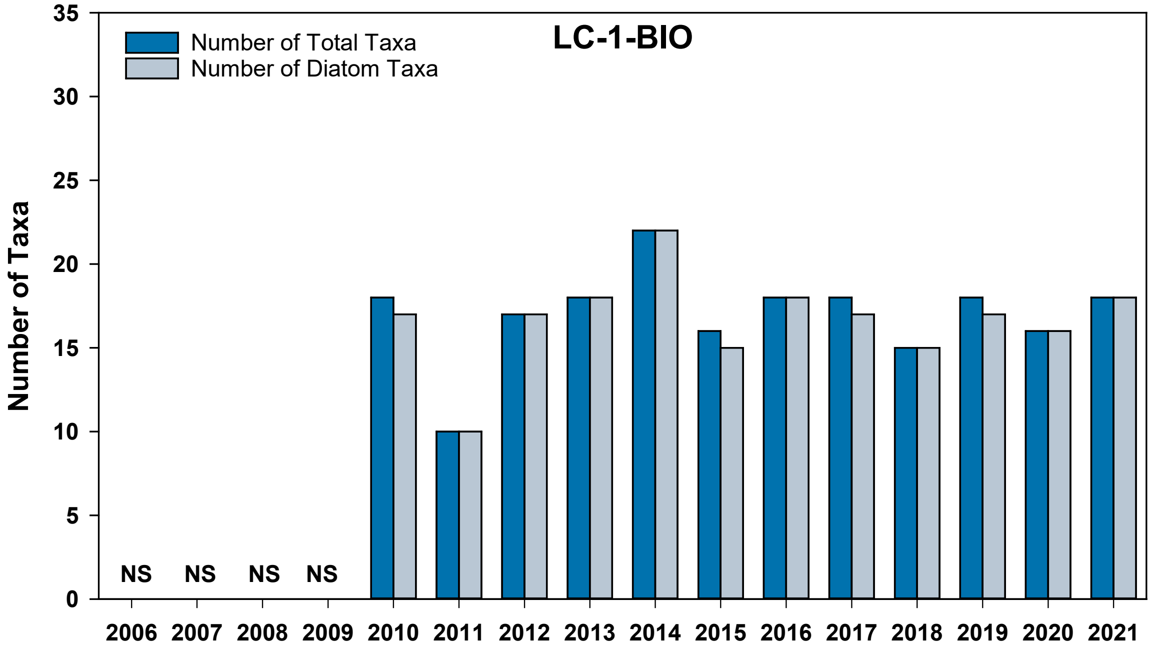


Figure 4-47: Periphyton taxa richness metrics at Site LC-1-BIO on Lost Camp Gulch, 2006 – 2021. NS = Not Sampled.

4.4.6 Deadwood Creek

4.4.6.1 2021 Data

The periphyton assemblage consisted of 99% pennate diatoms with the remainder being green algae at Site DC-2-BIO in August 2021 (Table 4-32; Appendix D). The Diversity value for diatoms was above the 2.5 threshold, indicating a diverse periphyton community (Table 4-33). The Autotrophic Index value was poor as a result of the high AFDM, indicating that the community is being affected by organic matter inputs. However, the Pollution Index value indicated that no organic enrichment occurred. Again, the disparity between these metrics suggests that some organic matter deposition occurs in the stream, likely caused by the large amount of overhanging vegetation and woody debris at the site. Percent Tolerant Diatoms and most trophic habit metrics were favorable indicating little siltation or pollution.

In 2021, metric values at Site DC-2-BIO were comparable to those at the reference site, Site LB-4-BIO. For richness metrics, Density at Site DC-2-BIO was roughly six times as large as at the reference site. Also, the Number of Periphyton Genera was greater at Site DC-2-BIO. Both composite metrics were more favorable at the reference site while the tolerance metric, Percent Tolerant Diatoms, was better at Site DC-2-BIO. These differences resulted in the Bahls Similarity Index indicating a “Somewhat Dissimilar” community at Site DC-2-BIO. Despite these differences, Diversity, Autotrophic Index, Pollution Index, and Percent Motile Diatoms values were similar between the sites and, overall, the metrics for Site DC-2-BIO indicates that this reach supports an abundant and relatively diverse periphyton community.

4.4.6.2 Historic Data

The periphyton community at Site DC-2-BIO has remained relatively stable since 2010. Pennate diatoms have been the dominant periphyton group in all years with green algae present in small numbers in some years. Richness, composition, and trophic habit metric values have not significantly trended over the years (Figure 4-48; Table 4-34; Appendix E). Diversity has been high in most years and the Autotrophic Index has been affected by organic matter deposition from 2010 to 2021. Percent Eutrophic Diatom Taxa was more favorable than those previously observed (previous minimum of 24% in 2012). The Pollution Index has been variable over time and, lastly, the tolerance metric, Percent Tolerant Diatoms, has significantly improved.

When Site DC-2-BIO metrics were compared to the metrics for the reference site (LB-4-BIO), only three of the eight tests indicated a significant difference between the long-term median values. Density has been significantly less ($p < 0.001$) and Percent Tolerant and Motile Diatom metrics have been significantly better ($p = 0.020$, $p < 0.001$, respectively) at Site DC-2-BIO. Overall, periphyton communities at both sites have shown good diversity with few differences between the assemblages.

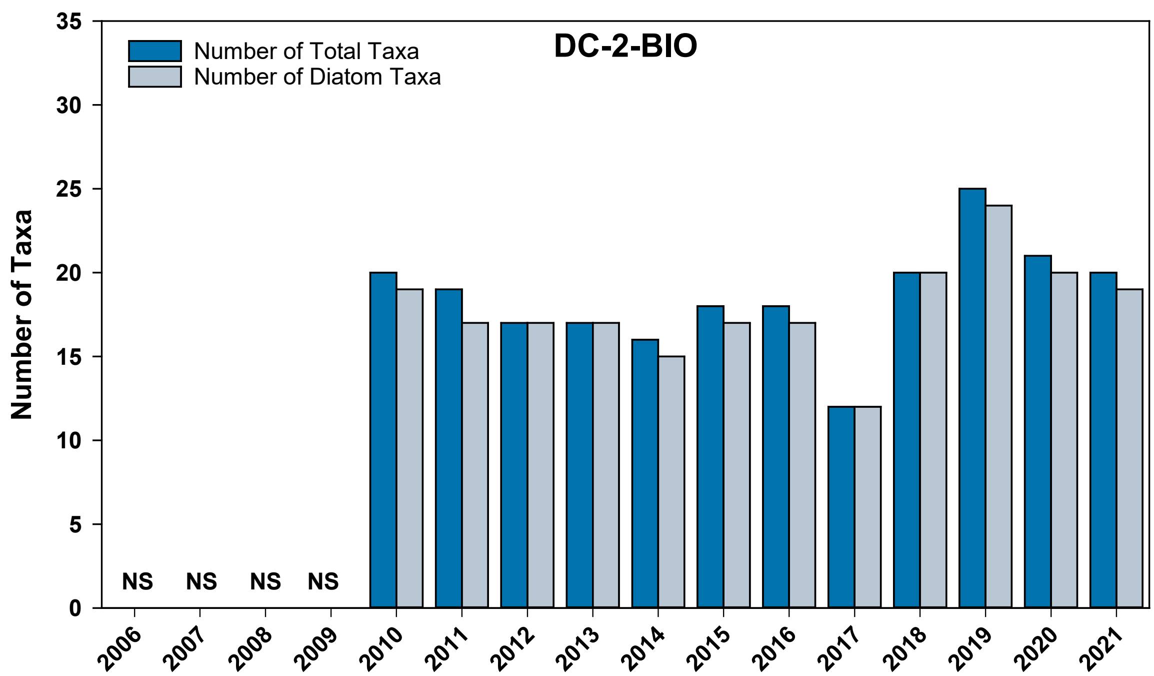


Figure 4-48: Periphyton taxa richness metrics at Site DC-2-BIO on Deadwood Creek, 2006 – 2021. NS = Not Sampled.

4.4.7 False Bottom Creek

4.4.7.1 Site EFB-1-BIO

4.4.7.1.1 2021 Data

Essentially, no algal cells were observed in the sample collected at Site EFB-1-BIO, because the results were reported as less than 415 periphyton cells/cm^2. In addition, a measurable amount of chlorophyll-a was obtained for this site, albeit the lowest value observed in 2021. The lack of periphyton cells at this site during 2021 is questionable, because historically periphyton have been abundant at this site. However, the results for the fish and macroinvertebrates communities and water quality are comparable to past years. Upon GEI's request, the periphyton lab analyzed the sample again and obtained the same result. The field notes were reviewed, and in fact, the field crew leader reviewed the sampling protocols with the team members to ensure the field data were collected according to the sampling and analysis plan. In addition, the appropriate field methodology and sample volumes were recorded in the field book. It's unclear whether the periphyton community was naturally very limited this past year or whether there was a sampling handling error somewhere along the process.

4.4.7.1.2 Historic Data

From 2006 to 2020, the periphyton community at Site EFB-1-BIO has revealed some variable patterns in the metrics due to limited sampling (n = 6) which has limited the value of trend analyses. Pennate diatoms have been the dominant group in all years with green algae, cyanobacteria, and cryptomonads being present in small numbers in some years. Only one significant trend, a decrease in the Pollution Index, was observed for the periphyton metrics due to the sporadic sampling events. However, Number of Taxa and Diatom Taxa; Diversity; and Percent Tolerant, Eutrophic, Nitrogen Heterotrophic, Motile, and Saprobic Diatoms were all greatest in 2020 (Table 4-33; Figure 4-49; Appendix E).

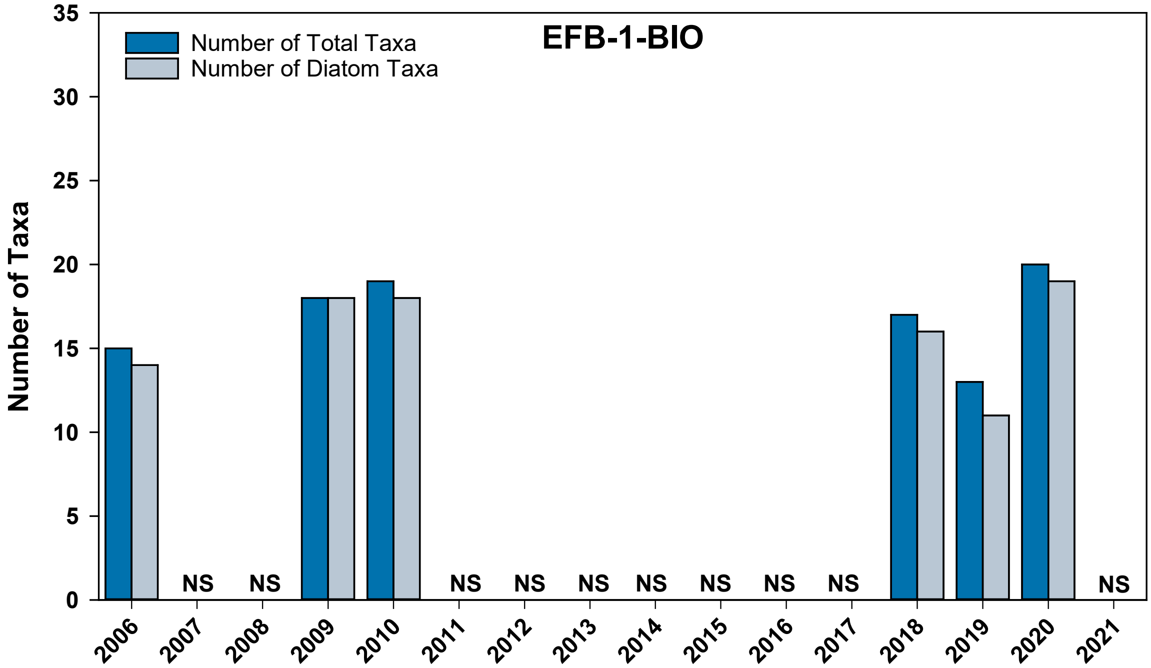


Figure 4-49: Periphyton taxa richness metrics at Site EFB-1-BIO on East Fork False Bottom Creek, 2006-2020. NS = Not Sampled.

When Site EFB-1-BIO metrics were compared to the reference Site LB-4-BIO metrics, three of the eight tests indicated significant differences between the long-term median values. However, these tests were similarly limited by the fewer sampling events at Site EFB-1-BIO. Nonetheless, Autotrophic Index was significantly worse and the metrics for Percent Tolerant and Motile Diatoms were significantly better ($p = 0.010$, $p = 0.010$ and $p = 0.002$, respectively) at Site EFB-1-BIO than at the reference site. Overall, periphyton communities at both sites have shown good diversity with few differences between the assemblages.

4.4.7.2 Site WFB-1-BIO

4.4.7.2.1 2021 Data

The periphyton assemblage consisted of 87% pennate diatoms, 10% green algae, and 3% cryptomonads at Site WFB-1-BIO in August 2021 (Table 4-32; Appendix D). The Diversity value for diatoms was well below the 2.5 threshold and the lowest of all sites, indicating an unbalanced community (Table 4-33). In addition, the Autotrophic Index was poor, indicating organic matter deposition. However, Percent Tolerant and Motile Diatoms were favorable, and the Pollution Index indicated no organic enrichment. This mix of favorable and unfavorable metric values included a very high Percent of Acidiphilic and High Oxygen Diatoms at Site WFB-1-BIO.

In 2021, metric values at Site WFB-1-BIO were different than those at the reference site, Site LB-4-BIO. Number of Taxa and Diatom Taxa were greater, and Diversity and composition metrics were more favorable at the reference site than at Site WFB-1-BIO.

Trophic habit metrics were more favorable at Site WFB-1-BIO when compared to the reference site except for Percent Acidiphilic Diatoms which was very high and not found at most other sites. These differences between these two sites resulted in a Bahls Similarity Index of a “Very Dissimilar” community between the two sites. Overall, Site WFB-1-BIO was very different from the reference site and contained a limited periphyton community that was considered in poor health for 2021. This site continued to be influenced by iron oxide deposition which impacts the periphyton community.

4.4.7.2.2 Historic Data

The periphyton community at Site WFB-1-BIO has changed moderately over the years with some metrics revealing significant trends. Pennate diatoms have been the dominant group for all years except 2018 when green algae were dominant. Richness, composition, and tolerance metric values have remained relatively consistent (Table 4-34; Figure 4-50; Appendix E) since 2007 and density was lower in 2021 than all other years (previous minimum of 36,272 in 2011). The Autotrophic Index indicates that Site WFB-1-BIO has been affected by organic pollution in all sample years while Diversity and Pollution Index values have been variable. Multiple trophic habit metrics (Percent Eutrophic, Nitrogen Heterotrophs, High Oxygen, and Saprobic Diatoms) have slightly, but significantly, improved in recent years with Percent High Oxygen Diatom Taxa being more favorable in 2021 than other years (previous maximum of 64% in 2020). Yet, these metrics remain in relatively poor condition due to the iron oxide deposition.

When Site WFB-1-BIO metrics were compared to the reference Site LB-4-BIO metrics, six of the eight tests indicated a significant difference between the sites. The Number of Taxa and Diatom Taxa, Diversity, and Autotrophic Index values were all significantly worse ($p < 0.001$, $p < 0.001$, $p = 0.001$, and $p = 0.020$, respectively) at Site WFB-1-BIO than compared to the reference site. Only the metrics for Percent Tolerant and Motile Diatoms were significantly better ($p < 0.001$ and $p < 0.001$, respectively) at Site WFB-1-BIO than the reference site. These trends and comparisons should be cautiously interpreted because of the poor periphyton assemblage at the site. Conditions at Site WFB-1-BIO are less favorable to support a diverse diatom community in recent years than they were during 2007 and 2008. A combination of very low flows and iron oxide precipitates contribute to the poor periphyton metrics observed in recent years.

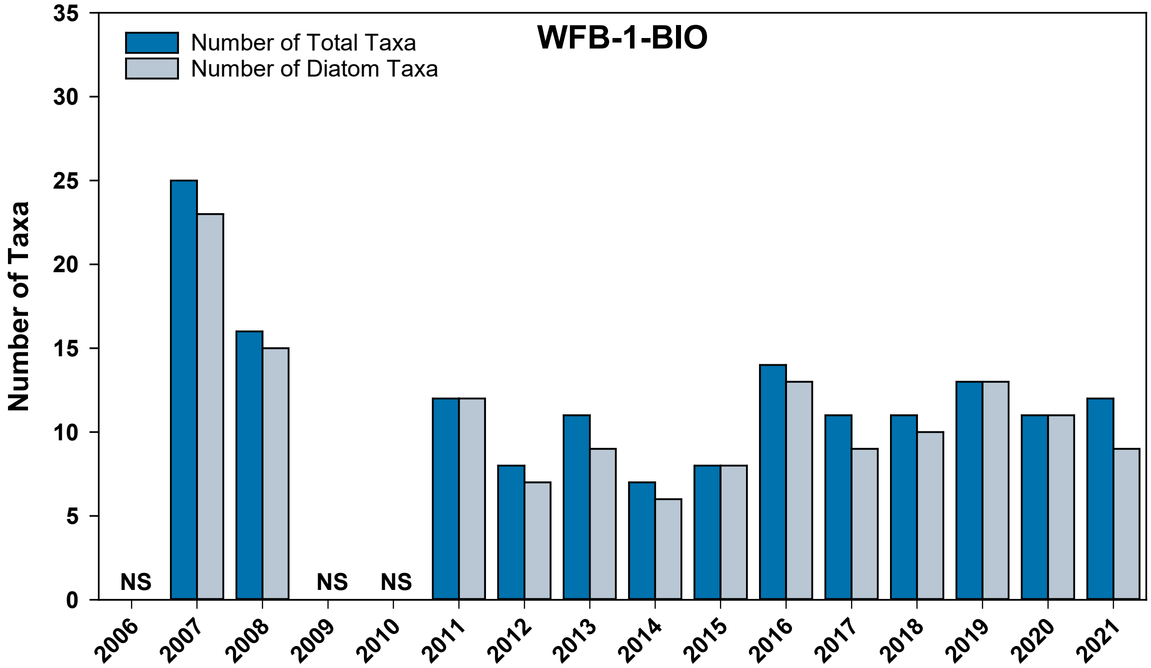


Figure 4-50: Periphyton taxa richness metrics at Site WFB-1-BIO on West Fork False Bottom Creek, 2006 – 2021. NS = Not Sampled.

4.4.8 McKinley Gulch

McKinley Gulch was dry during August 2021 and was not sampled. Periphyton has never been sampled at this site due to no stream flow.

4.4.9 Cleopatra Creek

4.4.9.1 2021 Data

The residual pockets of water and no flowing water precluded periphyton sampling at Site CC-1A-BIO in 2019 to 2021.

4.4.9.2 Historic Data

The periphyton community at Site CC-1A-BIO was highly variable from 2006 to 2018. Pennate diatoms were the dominant group in all years with cyanobacteria present in small numbers in some years. Richness, composition, and tolerance metric values have not significantly trended over time, partially due to the high variability observed in the metrics (Table 4-35; Figure 4-51; Appendix E). The Percent Nitrogen Heterotrophic Diatoms was the only trophic habit metric to reveal a significant worsening trend over time (Table 4-35).

Table 4-35: Slopes of significant trends ($p < 0.05$) for periphyton population metrics at Cleopatra Creek, Fantail Creek, Nevada Gulch, and Stewart Gulch, 2006 - 2021. + = Positive slope. - = Negative slope. -- = Not significant.

Metric	Change Expected Following Environmental Disturbance	CC-1A-BIO	FC-1-BIO	NG-2-BIO	SG-1-BIO
RICHNESS					
Density (cells/cm^2)	Variable	--	--	--	--
Relative Diatom Density	Variable	--	--	--	--
Number of Taxa (species)	Decrease	--	--	--	--
Number of Diatom Taxa	Decrease	--	--	--	--
Number of Periphyton Divisions	Variable	--	--	--	--
Number of Periphyton Genera	Variable	--	--	- 0.49	--
COMPOSITION					
Shannon-Weaver Diversity Index for Diatoms	Decrease	--	--	--	--
Bahls Similarity Index	Variable	--	- 0.02	--	--
Autotrophic Index	Increase	--	--	--	--
TOLERANCE					
Percent Tolerant Diatoms Density	Increase	--	< 0.01	--	--
Lange-Bertalot Pollution Index	Decrease	--	--	--	--
TROPHIC HABIT					
Percent Eutrophic Diatoms	Variable	--	--	--	--
Percent Acidiphilic Diatoms	Variable	--	--	--	--
Percent Alkaliphilic Diatoms	Variable	--	--	--	--
Percent Nitrogen Heterotrophs	Variable	+ 0.01	--	--	--
Percent High Oxygen Diatoms	Variable	--	--	--	--
Percent Motile Diatoms	Increase	--	--	--	--
Percent Saprobic Diatoms	Variable	--	--	--	--

Note: CC-1A-BIO was sampled in 2006-2015 and 2018.

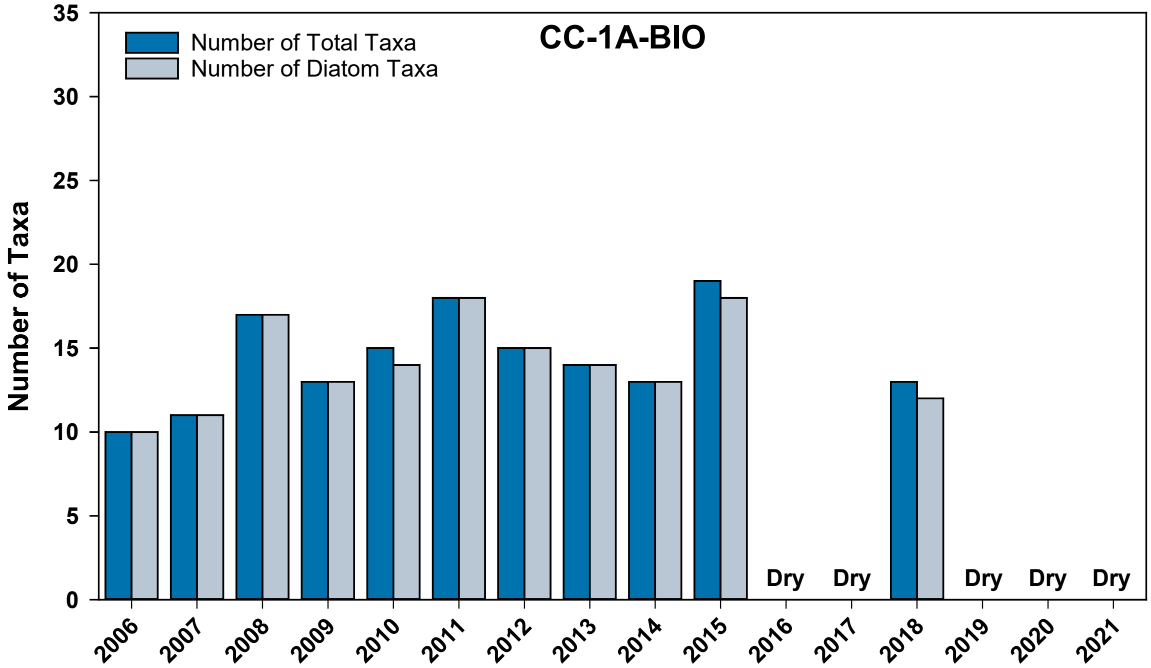


Figure 4-51: Periphyton taxa richness metrics at Site CC-1A-BIO on Cleopatra Creek, 2006-2021.

4.4.10 Fantail Creek

4.4.10.1 2021 Data

The periphyton assemblage consisted of 100% pennate diatoms at Site FC-1-BIO in August 2021 (Table 4-36; Appendix D). Periphyton density was extremely low and much lower than all other sites. The Diatom Diversity value was greater than the 2.5 threshold, indicating a balanced periphyton community (Table 4-37). The Autotrophic Index was poor and indicated that the site was affected by organic pollution. However, the Percent Tolerant and Motile Diatoms were relatively favorable in 2021 indicating organic pollution content and low siltation in this site. The Pollution Index also indicated that a majority of the periphyton community was indifferent to organic enrichment and that minor organic enrichment influenced the site. In 2013, vegetation and trees were cut down as part of the powerline maintenance along Fantail Creek, and the organic debris was left in place likely inflating the Autotrophic Index. Over the years, the natural decay of organic matter has likely influenced the organic inputs to this stream. High sedimentation from the adjacent dirt road was also noted at this site since 2018.

In 2021, periphyton richness, composition, tolerance, and trophic habit, metric values at Site FC-1-BIO were comparable to those at the reference site, Site RC-1-BIO. Total Density was 58 times greater at the reference site, but other richness metrics were similar. Composition metrics indicated that Diversity was slightly more favorable at Site FC-1-BIO in 2021 while Autotrophic Index was more favorable at the reference site. Trophic habit metrics were not consistently better at either site. The Bahls Similarity Index indicated a

"Somewhat Dissimilar" community between Site FC-1-BIO and the reference site. Despite these differences, Diversity and Pollution Index were good while the Autotrophic index was poor at both sites. Overall, the metrics for Site FC-1-BIO indicate that this reach supports a high number of taxa with a relatively diverse periphyton assemblage.

Table 4-36: Relative periphyton density (%) and biomass estimates for sites on Fantail Creek, Nevada Gulch, and Stewart Gulch, August 2021.

Taxa/Metric	FC-1-BIO	NG-2-BIO	SG-1-BIO
BACILLARIOPHYTA			
Pennales (Pennate diatoms)	100.00	99.13	100.00
CRYPTOPHYTA (Cryptomonads)	--	0.87	--
BIOMASS			
AFDM (mg/m^2)	3,444	8,540	4,132
Chlorophyll-a (mg/m^2)	4.5	22.3	12.4

Table 4-37: Periphyton population metrics for sites on Fantail Creek, Nevada Gulch, and Stewart Gulch, August 2021.

Metric	FC-1-BIO	NG-2-BIO	SG-1-BIO
RICHNESS			
Density (cells/cm^2)	5,388	1,653,691	219,091
Relative Diatom Density	100%	99%	100%
Number of Taxa (species)	16	17	16
Number of Diatom Taxa	16	16	16
Number of Periphyton Divisions	1	2	1
Number of Periphyton Genera	11	10	8
COMPOSITION			
Shannon-Weaver Diversity Index for Diatoms	3.11	2.60	2.15
Bahls Similarity Index	38.2%	29.7%	33.9%
Autotrophic Index	771	382	334
TOLERANCE			
Percent Tolerant Diatoms Density	2.2%	4.4%	5.7%
Lange-Bertalot Pollution Index	2.65	2.59	2.61
TROPHIC HABIT			
Percent Eutrophic Diatoms	56.3%	50.0%	56.3%
Percent Acidiphilic Diatoms	0.0%	0.0%	0.0%
Percent Alkaliphilic Diatoms	50.0%	37.5%	56.3%
Percent Nitrogen Heterotrophs	6.3%	18.8%	18.8%
Percent High Oxygen Diatoms	37.5%	43.8%	37.5%
Percent Motile Diatoms	43.8%	56.3%	56.3%
Percent Saprobic Diatoms	6.3%	6.3%	12.5%

4.4.10.2 Historic Data

The periphyton community metrics for Site FC-1-BIO have been variable from 2006 to 2021. Pennate diatoms have been the dominant group in all years with green and golden algae present in small numbers in some years. No richness or trophic habit metrics trended over time while the composition metric, Bahls Similarity Index, and tolerance metric, Percent Tolerant Diatoms (Table 4-35; Figure 4-52; Appendix E) have worsened over time. The Autotrophic Index indicates that Site FC -1-BIO has been affected by organic pollution in all sample years while Diversity and Pollution Index values have been variable. Two median periphyton metrics from 2006 to 2021 were significantly different between Site FC-1-BIO and the refence site. Density was significantly poorer and Percent Tolerant Diatoms significantly better at Site FC-1-BIO than at the reference site. While Site FC-1-BIO may experience some organic enrichment and siltation, the data indicate that the site has supported a diverse assemblage and is similar to the refence site.

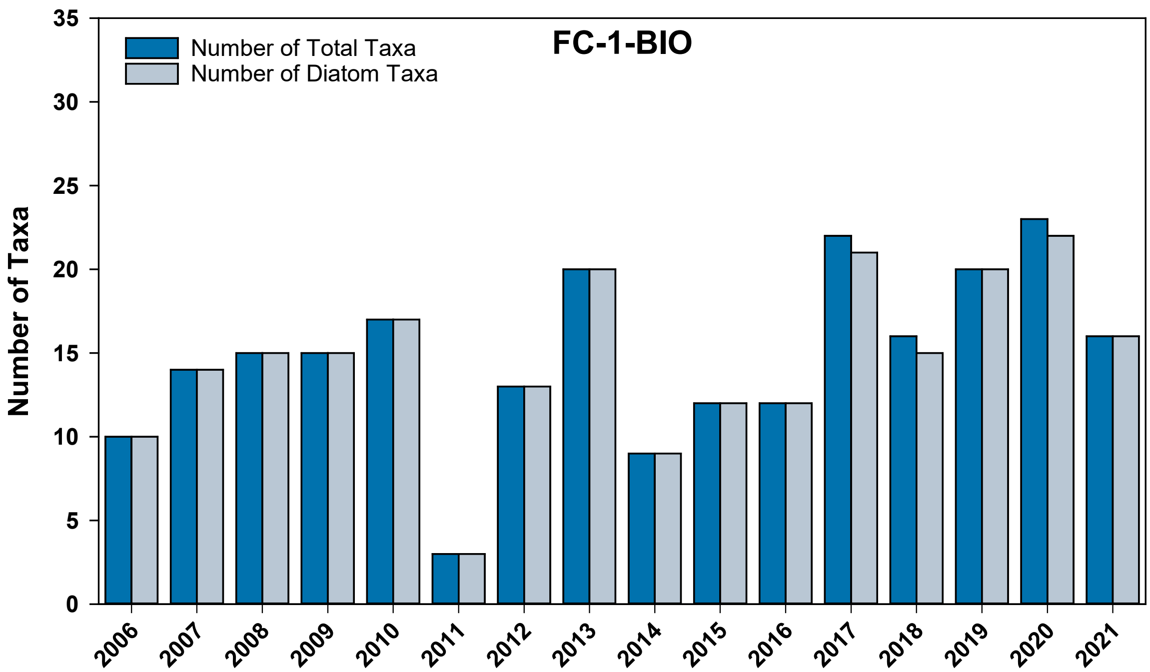


Figure 4-52: Periphyton taxa richness metrics at Site FC-1-BIO on Fantail Creek, 2006 - 2021.

4.4.11 Nevada Gulch

4.4.11.1 2021 Data

In August 2021, the periphyton assemblage at Site NG-2-BIO consisted of 99% pennate diatoms and 1% cryptomonads (Table 4-36; Appendix D). The density at this site was the highest of any site and the diatom Diversity value was above the 2.5 threshold, indicating an abundant and balanced community (Table 4-37). The Autotrophic Index was fair indicating some enrichment with organic matter and a potential for increased algal growth. Pollution tolerant species were present in low numbers, and the Pollution Index indicated that a relatively larger proportion of the periphyton community was comprised of sensitive diatoms.

In 2021, periphyton richness, composition, and tolerance metric values at Site NG-2-BIO were comparable to those at the reference site, Site RC-1-BIO. Density was five times greater at Site NG-2-BIO while the other richness metrics were similar between the two sites. Diversity, Autotrophic Index, Pollution Index, and Percent Motile Diatoms metrics were also similar between the two sites. Most tolerance metrics were, however, slightly more favorable at Site NG-2-BIO. Overall, both sites appeared to have healthy periphyton communities in 2021.

4.4.11.2 Historic Data

The periphyton community metrics for Site NG-2-BIO have been variable over the years. Pennate diatoms have been the dominant group in all years with green algae and cyanobacteria present in small numbers in some years. No significant trends for periphyton metric were observed from 2006 to 2021 except for a significant decrease in the Number of Periphyton Genera (Table 4-35). Diversity has not trended and has been good in all years while Autotrophic and Pollution Indexes values have been variable. Data in 2021 revealed few changes, with many of the metric values being similar to those observed in the past few years. This site continues to support a diverse periphyton assemblage.

When Site NG-2-BIO metrics were compared to the reference Site RC-1-BIO metrics, four of the eight tests indicated significant differences between the median values. Density, Diversity, and Autotrophic Index have been significantly better ($p = 0.004$, $p = 0.010$, $p = 0.013$, respectively) at Site NG-2-BIO, while the Pollution Index has been significantly worse ($p = 0.005$) at Site NG-2-BIO when compared to the reference site conditions. While differences exist, the periphyton communities at both sites have shown good diversity with few differences between the assemblages.

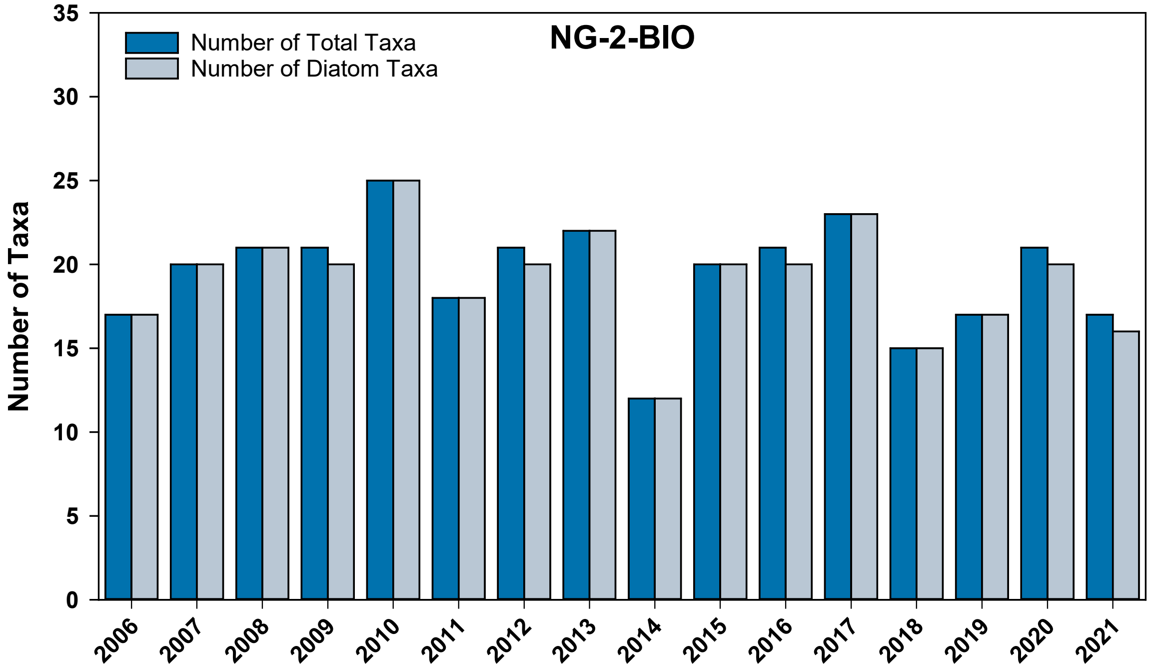


Figure 4-53: Periphyton taxa richness metrics at Site NG-2-BIO on Nevada Gulch, 2006 – 2021.

4.4.12 Stewart Gulch

4.4.12.1 2021 Data

The periphyton assemblage at Site SG-1-BIO in August 2021 consisted of all pennate diatoms (Table 4-36; Appendix D) and the diatom Diversity value was less than the 2.5 threshold, indicating an unbalance assemblage (Table 4-37). The Autotrophic Index showed that the stream was enriched with nutrients and shows a potential for increased periphyton growth. The pollution tolerant and motile diatoms were present in low numbers, and the Pollution Index value indicated that a relatively larger proportion of the periphyton community was comprised of sensitive diatoms. This site continues to be influenced by the small spring immediately upstream and its higher nitrogen content, even though the growth and expanse of the watercress was less in 2021 than observed in recent years.

In 2021, periphyton richness, tolerance, and trophic habit metric values at Site SG-1-BIO were comparable to those at the reference site, Site LB-4-BIO. Number of Taxa and Diatom Taxa were greater at Site SG-1-BIO, but all other richness metric values were similar between the sites. Composition and tolerance metrics indicated that Diversity was better at the reference site and the Autotrophic and Pollution indices were better at Site SG-1-BIO. Trophic habit metrics were similar between the sites. These differences between the sites resulted in a Bahls Similarity Index of a "Somewhat Dissimilar" community between Site SG-1-BIO and the reference site. Overall, Site SG-1-BIO in 2021 appeared to have a different but no less robust periphyton community than the reference site.

4.4.12.2 Historic Data

The periphyton community metrics for Site SG-1-BIO have been highly variable over the years with no significant trends being observed for 2006 to 2021. Pennate diatoms have been the dominant group in all years while green algae and cyanobacteria were present in small numbers in some years. Diversity has been low in most years, Autotrophic Index has been variable, and Pollution Index indicated no organic enrichment in most years despite the nitrogen enriched ground water entering the site. All 2021 metrics were within the range of those previously observed for this site (Table 4-37; Figure 4-54; Appendix E) except for Percent Eutrophic and Alkaliphilic Diatom Taxa metrics which were poorer in 2021 than other years (previous minimum of 53.3% in 2016 and 50% in 2007, 2011, and 2012, respectively). The 2021 data revealed small changes compared to the recent years data and continues to support a limited diatom assemblage that is influenced by the emergent vegetation that covers the margins of the stream yet allows for an open thalweg (Photo 4-6).

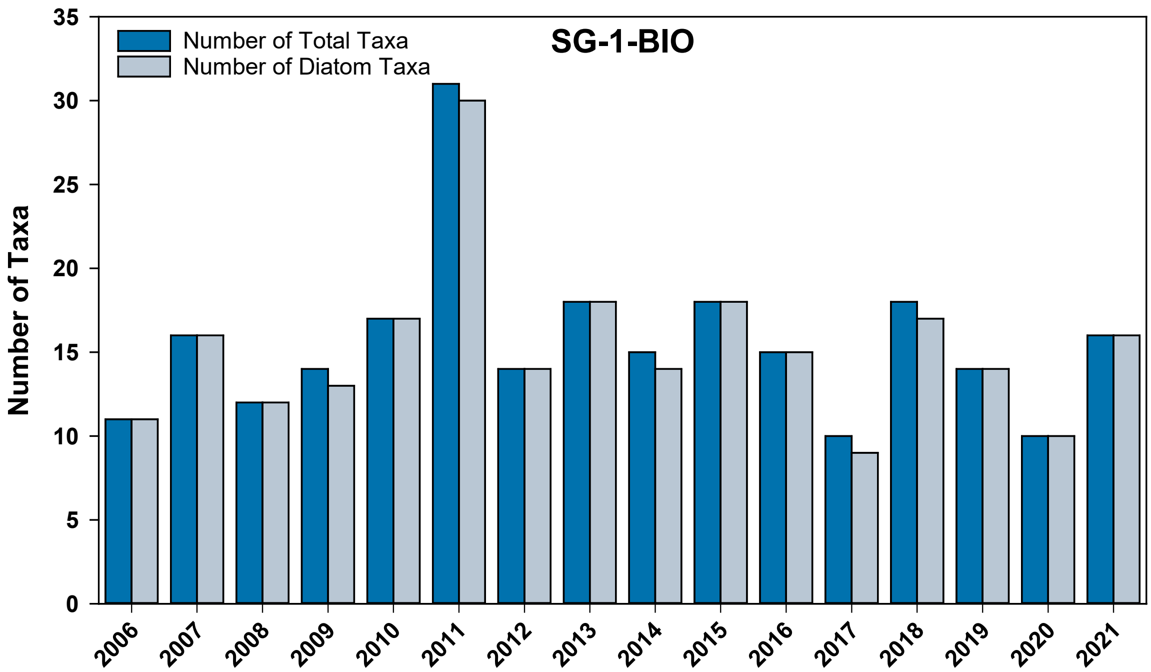


Figure 4-54: Periphyton taxa richness metrics at Site SG-1-BIO on Stewart Gulch, 2006 - 2021.

When Site SG-1-BIO metrics were compared to the reference Site LB-4-BIO metrics, four of the eight tests indicated significant differences between the sites. Density values have been significantly greater ($p = 0.011$) at Site SG-1-BIO, while the Number of Taxa and Diatom Taxa and Diversity have all been significantly less ($p = 0.003$, $p = 0.008$, and $p = 0.001$, respectively) at Site SG-1-BIO when compared to the reference site (Table 4-35). Despite the differences in richness, composition, and tolerance metrics, Site SG-1-BIO continues to support an assemblage with sensitive diatoms.

4.5 Water Quality

4.5.1 Biomonitoring

Water quality samples were collected by Wharf personnel from August 20 to 23 and results were obtained for sites LB-4-BIO, RC-1-BIO, AC-1-BIO (NPDES Compliance Point 001), AC-2-BIO (NPDES Compliance Point 005), AC-3-BIO, RV-2-BIO (NPDES Compliance Point 002), LC-1-BIO, DC-2-BIO, EFB-1-BIO, WFB-1-BIO, FC-1-BIO, NG-2-BIO, and SG-1-BIO.

Total recoverable concentrations for cadmium, copper, mercury, silver, and weak acid dissociable cyanide were less than their respective detection limits at all of the biological monitoring sites, while chromium, lead, selenium, and zinc concentrations were less than their respective detection limits at most sites (Appendix F). Measurable concentrations of arsenic, iron, and nickel were observed for most biological monitoring sites while concentrations of calcium and magnesium were observed at all sites, although the magnitude of concentrations were similar to the respective reference sites. For arsenic and nickel, concentrations were considerably less than the continuous criterion concentration (CCC) for aquatic life use (ARSD 2019 §74:51:01:55).

Nutrient analyses indicated that nitrate was not detected at sites LB-4-BIO and WFB-1-BIO (Appendix F). However, the concentrations of nitrate were similar to those at the reference sites except for sites AC-3-BIO, EFB-1-BIO, and SG-1-BIO which were greater. The nitrogen-rich groundwater flowing into Stewart Gulch (4.5 mg/L) could be influenced by the historic Golden Reward mine, but the source of the groundwater is difficult to trace. Explosives containing nitrate are often used to break up bedrock during mining operations, and remnants of these charges can leach into groundwater. Similar concentrations of nitrate-nitrogen were observed in Annie Creek (4.8 mg/L) and East Fork False Bottom Creek (6.0 mg/L), but all concentrations were less than the numerical ground water standard of 10 mg/L and the surface water standard for fish and wildlife propagation, recreation, and stock watering uses (ARSD 2021 §74:54:01:04 and §74:51:01:52).

In addition, dissolved phosphorous was above the detection limit at most sites, except for sites WFB-1-BIO, EFB-1-BIO, FC-1-BIO, and NG-2-BIO where it was not detected. Measurable concentrations of phosphorus originate in Lost Camp Gulch and influence the downstream waters in Annie Creek. Overall, the data indicate no patterns in water chemistry relative to Wharf Mine permitted outfalls and few localized patterns such as the phosphorus in the Annie Creek basin and nickel in the Deadwood Creek, East and West False Bottom Creek, and Stewart Gulch.

4.5.1.1 Selenium

Annie Creek and East False Bottom Creek have been routinely sampled over the years for selenium water quality while selenium fish tissues were only sampled from Annie Creek. In 2021, fish tissue sampling began in East False Bottom Creek to supplement the selenium data for this stream. South Dakota's chronic selenium water quality criteria is 5.0 µg/L (total recoverable) and is assessed as the median concentration over a three-year period. In addition, the monthly chronic limit for Wharf compliance points is 4.6 ug/L. However, the EPA's 2016 Aquatic Life Ambient Water Quality Criterion for Selenium provides guidance on the most scientifically up-to-date criterion which prioritizes the use of fish tissue concentrations over water quality concentrations (EPA 2016). While the biomonitoring water quality data at sites AC-3-BIO and EFB-1-BIO are only a subset of the water quality data collected by Wharf each year, comparisons of the biomonitoring data provide some context to the selenium water quality conditions in each stream.

From 2019 to 2021, the median total recoverable selenium concentration at Site AC-3-BIO was < 5 µg/L and did not exceed South Dakota's chronic criteria. Furthermore, over the ten years of collecting fish tissues (n = 45 Brook Trout and 5 Brown Trout) from Annie Creek, only one individual Brown Trout (8.6 µg/g) collected in 2021 exceeded the EPA tissue criterion (8.5 µg/g). The median total recoverable selenium concentration measured at Site EFB-1-BIO during the 2019-2021 biomonitoring events is 9 µg/L which exceeds South Dakota's chronic criterion and the monthly chronic limit for compliance points. However, the selenium concentrations in the five Brook Trout collected from East False Bottom Creek in 2021 are considerably less than the EPA 2016 selenium whole-body tissue criterion (Section 3.2.2).

This page intentionally left blank

5. Conclusions

Fish, benthic macroinvertebrate, and periphyton populations were sampled and habitat was evaluated at sites near the Wharf Mine on Labrador Gulch, Reno Creek, Annie Creek, Ross Valley, Lost Camp Gulch, Deadwood Creek, East and West Fork False Bottom Creek, Fantail Creek, Nevada Gulch, and Stewart Gulch in August 2021. Cleopatra Creek and McKinley Gulch remained dry. Data from these sites were analyzed to evaluate current conditions of aquatic biological populations in the streams and compared to reference site conditions. In addition, these data were compared to data from previous years to evaluate relationships between the aquatic populations and mining activities over time. The site on Labrador Gulch (LB-4-BIO) was used as the reference site for the sites of AC-2-BIO, DC-2-BIO, EFB-1-BIO, WFB-1-BIO, and SG-1-BIO which either support or have historically supported the biological target of fish, benthic macroinvertebrates, and periphyton. In most years, Site LB-4-BIO is also the reference site for Site AC-3-BIO but this site on lower Annie Creek was not sampled in 2020 and 2021 due to the extensive deadfalls and disturbance resulting from the July 8, 2020 tornado activity. The site on Reno Creek (RC-1-BIO) was used as the reference site for the sites of AC-1-BIO, RV-2-BIO, LC-1-BIO, CC-1A-BIO, FC-1-BIO, and NG-2-BIO which have supported the biological target of benthic macroinvertebrates and periphyton. These reference sites were selected based on their similar characteristics to the mining activity sites with respect to stream size and order, flow regime, ecoregion, elevation, geology, and biological populations and represent a comparative system influenced by local climatic conditions rather than mining related influences.

5.1 Habitat

Site LB-4-BIO is a high-gradient site that flows through a steep, bedrock canyon while Site RC-1-BIO is lower gradient stream, in a more vegetated and forested valley, with ample sources of fine sediments. The majority of both sites were comprised of fast water habitat, but each site also contained ample slow-water pool habitat. Substrate compositions varied between sites, with Site LB-4-BIO being dominated by boulders, bedrock, and rubble and substrate being mainly comprised of gravel at Site RC-1-BIO.

The two sites on Annie Creek and sites RV-2-BIO and LC-1-BIO sampled in 2021 had a diversity of habitat types, with abundant riffles and pools. Boulders and bedrock were the dominant substrate size classes at Site AC-1-BIO, fine sediment at Site RV-2-BIO, and rubble at Site LC-1-BIO. Substrate at Site AC-2-BIO was not dominated by any particular substrate size. Percent surface fines were fair at all sites except at Site RV-2-BIO where fines were much higher. Storm runoff and recreational usage of the dirt road adjacent to sites AC-2-BIO and LC-1-BIO likely contribute to the surface fines at these sites and not mining activities. The higher percentages of fine substrates at the Ross Valley site are likely due to the low discharge typically found at this site and the accumulation of organic matter from the surrounding forest combined with a lack of flushing flows.

The EFB-1-BIO and WFB-1-BIO sites were dominated by riffle habitat in 2021, as in previous years, while Site DC-2-BIO was dominated by pools. All sites included at least one pool while substrate composition varied between sites. The general increase in stream flows due to wet-year type conditions have reduced surface fine percentages in recent years in False Bottom Creek. Eroding banks have been observed at sites DC-2-BIO and WFB-1-BIO in past years leading to the elevated surface fines in 2021. Habitat conditions in 2021 were generally comparable to past years. Fine iron oxide precipitates were again observed at Site WFB-1-BIO, and the data indicate that this condition is negatively affecting the benthic macroinvertebrate and periphyton communities.

Sites FC-1-BIO, NG-2-BIO, and SG-1-BIO were predominantly composed of riffle habitat, but all sites also contained some pool habitat formed by various elements, such as logs or boulders. Stream widths and average water depths were greatest in Stewart Gulch, while the remaining stream sites were generally narrower and shallower. Gravel was the most abundant substrate size class at sites FC-1-BIO and NG-2-BIO while course gravel was most abundant at Site SG-1-BIO. Surface fines were slightly greater at Fantail Creek, which may be from the nearby adjacent road and nearby residential construction activities. The upstream location of the Fantail Gulch site limits the occurrence of high flow events which would scour fine sediments from the site. All sites contained a variety of substrate sizes. A shift in groundwater patterns upstream of the Stewart Gulch site continues to provide nitrogen-rich groundwater that promotes the growth of watercress, albeit to a lesser degree in 2021, in Stewart Gulch.

In general, habitat characteristics at most sites have shown some variability over time, but no substantial, long term patterns have been observed. The study sites contained a diverse range of habitats in most years of sampling. Flows in 2018 and 2019 were high due to increased precipitation in the Black Hills and fine sediment percentages have decreased at some sites in 2018 through 2021. Most sites did not differ greatly from their respective reference sites.

5.2 Fish

Site LB-4-BIO contained a small population of Brook Trout that included YOY, first year age class, and second year-plus age class individuals. First year age class have been collected in previous years indicating that this site supports all life stages of Brook Trout. The mean length and weight for trout were greater at Site LB-4-BIO than the comparative mining activity sites. However, the mean condition factor and relative weight values were lower than the optimal ranges for trout at Site LB-4-BIO. In fact, the relative weight of Brook Trout has significantly decreased over time at the reference site. These fish may be stressed due to natural factors such as the generally lower stream flows and higher water temperature in recent years.

Eleven Brook and Brown Trout were collected at the reference site on Reno Creek which is similar to the number collected from 2017 to 2019. All fish were second year-plus. As with Labrador Gulch, mean condition factor and relative weight values at Site RC-1-BIO were less than the optimal but similar to other sites sampled in 2021. Site RC-1-BIO is likely near the upstream extent of suitable fish habitat that provides spawning habitat for trout. Brook Trout have been the dominant fish observed in Reno Creek in all years, except for 2019 when Brown Trout were more abundant. Fish usage of this site may be seasonal or variable depending on flows within the drainage, and trout may be expanding their spawning habitat up from Whitewood Creek into Reno Creek. With only four years of data, future sampling at this site will better illustrate the patterns in fish use over time.

Two of the three Annie Creek biomonitoring sites were surveyed for fish in 2021. The extensive habitat disturbance in the lower portion of the basin precluded sampling at Site AC-3-BIO. No fish were collected in the upper portion of the basin at either AC-1-BIO or AC-2-BIO. Historically, Mountain Suckers were common at Site AC-2-BIO but have been absent at this site since 2011. This absence is attributed to the water quality disturbances in 2007. There are no upstream sources of fish, and the movement of fish into this site from downstream is prevented by Annie Creek Falls. Based on the data collected, Mountain Suckers are now absent from Annie Creek upstream of Annie Creek Falls.

Despite not sampling Site AC-3-BIO in 2020 and 2021, past data show that the Brook Trout population, specifically YOY and first year age class fish, has been trending downwards in recent years and that the Brown Trout population has been increasing. This shift in species dominance has been a common occurrence in many Rocky Mountain streams where the transition of colder to cool water habitat has shifted downstream due to increasing ambient air temperature and water temperature. In these streams, Brown Trout populations are expanding upstream into habitat typically dominated by Brook Trout and are beginning to competitively exclude Brook Trout from their traditional habitat. The larger numbers of second year-plus age class of Brown Trout began showing up in 2014, and 2019 is the first year when a larger number of YOY and first year age class Brown Trout were observed for this site. The limited survey in 2021 found that only second-and third-year Brown Trout were present in lower Annie Creek. Overall, both Brook Trout and Brown Trout have historically maintained resident populations of all age classes at this site, and the data do not indicate that mining activities upstream of the compliance points in the Annie Creek basin have adversely affected the fish assemblage at Site AC-3-BIO.

Site DC-2-BIO maintains perennial flows and supports a naturally reproducing Brook Trout population, with multiple size classes present during most years. The density and biomass of Brook Trout in 2021 were both within the range of historical conditions. Over the years, fewer second year-plus age class sized fish have been observed compared to YOY and first year age class fish at this site, indicating this section of stream may serve primarily as a spawning and rearing area. Deep water habitat is also minimal at this site, limiting suitable habitat for larger, second year-plus age class trout.

Each site on the East Fork and West Fork False Bottom Creek has contained a Brook Trout population during the years sampled. The 2021 trout population at Site EFB-1-BIO was comprised mainly of YOY age class Brook Trout, indicating that spawning was taking place. The population at Site WFB-1-BIO consisted of only two, first-year age class Brook Trout. Abundance, density, and biomass values at Site WFB-1-BIO have been relatively lower when compared to Site EFB-1-BIO over time, but the sampling frequency has varied at these sites. The smaller population, density, and biomass at Site WFB-1-BIO is likely related to low flows and the poorer water quality conditions. Iron oxide precipitates have been present at this site in recent years which have reduced its habitat suitability. Deep water habitat at this site, in the form of pools, is also very limited, likely decreasing usage by second year-plus age class fish in some years, particularly when flows are low. Brook Trout density and biomass estimates for the East Fork were similar to the values observed for the reference site on Labrador Gulch. Overall, the data shows that East Fork False Bottom maintains suitable spawning and rearing habitat and supports multiple age classes of fish each year, whereas as the habitat in West Fork only supports transitory first-year age class of fish.

The Brook Trout population in Stewart Gulch revealed density and biomass estimates greater than the long-term median conditions for Site SG-1-BIO. The presence of a high nutrient content spring immediately upstream of Site SG-1-BIO facilitates plant growth and provides habitat for benthic macroinvertebrates and YOY Brook Trout. The presence of a stable Brook Trout population, with all age classes, indicates that the habitat and water quality conditions in Stewart Gulch sustains a naturally reproducing fishery. Historically, few Brown Trout have been observed in Stewart Gulch.

No fish have been collected from sites AC-1-BIO, RV-2-BIO, LC-1-BIO, and CC-1A-BIO since monitoring at these sites began. These stream reaches are small and narrow, and the sites are in the headwaters, upstream of natural fish barriers and suitable fish habitat. Because no fish have been found upstream of Annie Creek Falls since 2010, the movement of fish into the upper reaches of Annie Creek, Lost Camp Gulch, and Ross Valley are unlikely. In addition, no fish have been present at sites sampled on Nevada Gulch and Fantail Creek since 1998.

5.3 Benthic Macroinvertebrates

In 2021, the reference sites LB-4-BIO and RC-1-BIO both supported a rich and diverse benthic macroinvertebrate community including numerous sensitive species and a variety of feeding types. In fact, these sites had two of the highest diversity values of all sites. In addition, Site LB-4-BIO contained a higher Number of Taxa than any other Wharf site. *Micropsectra sp.*, a tolerant Diptera, was the most dominant taxon at Site RC-1-BIO resulting in Percent Chironomidae, Tolerant Taxa, and Collector-Gatherers values being relatively poorer, despite the site's robust diversity. Overall, data in 2021 showed negligible differences with metric values being similar to those in previous years, indicating the macroinvertebrate community has changed little over time. Both sites exhibit a healthy macroinvertebrate community, with little to no influence from anthropogenic sources.

The Annie Creek sites, AC-1-BIO and AC-2-BIO, supported a rich and diverse benthic macroinvertebrate community in 2021, including numerous sensitive species, and both sites were comparable to their respective reference site community. Site AC-1-BIO was dominated by a sensitive taxon and metric values at Site AC-2-BIO associated with the EPT community were far better than in recent years with Percent Sensitive EPT Taxa and EPT Index greater than any other site. Twelve of the twenty-five macroinvertebrate metrics for Site AC-1-BIO significantly improved from 2006 to 2021 showing complete recovery from the water quality disturbances in 2007. Site AC-2-BIO was much less affected by this disturbance with few trending metrics. This site has historically been impacted by poorer sediment conditions caused by the recreational use of the dirt trail adjacent to Lost Camp Gulch.

Each site on Ross Valley, Site RV-2-BIO, and Deadwood Creek, Site DC-2-BIO, supports rich and diverse benthic macroinvertebrate communities, including numerous sensitive species, and were comparable to their respective reference site community. Exceptions to the favorable values were the poor Percent Sensitive EPT Taxa and Percent Tolerant Taxa at Site RV-2-BIO and high percentage of Chironomids at Site DC-2-BIO. At Site RV-2-BIO, only two macroinvertebrate metrics significantly trended over time while both improving and declining metrics were revealed at Site DC-2-BIO; however, overall, the communities at both sites have been relatively stable over the years.

The site on Lost Camp Gulch supported few sensitive EPT taxa in 2021; although it scored well for diversity and tolerance metrics. When compared to the reference site on Reno Creek, the metrics for Lost Camp generally indicated a poorer community but still moderately healthy. Five of the twenty-five metrics, mostly EPT related metrics, significantly declined over time. The combination of low flows and the influence of poorer sediment conditions have negatively affected the macroinvertebrate community at Site LC-1-BIO.

The East Fork and West Fork False Bottom sites continued to show a disparity in many 2021 macroinvertebrate metrics. Despite a high percentage of *Baetis tricaudatus cx.*, a tolerant mayfly, Site EFB-1-BIO supported a rich and diverse benthic macroinvertebrate community, including numerous intolerant species, with many metrics comparable to the metrics for Labrador Gulch. Site WFB-1-BIO supported a limited community that scored poorly when compared to the reference site conditions on Labrador Gulch. On the West Fork, eight of the twenty-five metrics significantly trended with seven of the metrics indicating a decline in quality over time. When compared to the reference site conditions, each selected metric was significantly worse at Site WFB-1-BIO. The iron oxide and poorer flow conditions in the

West Fork has decreased habitat suitability and negatively affected the macroinvertebrate assemblage in recent years.

Despite a poor Ephemeroptera assemblage at Fantail Creek, a small EPT community at Nevada Gulch, and poor percentage of *Baetis sp.* at Stewart Gulch, these sites supported a taxonomically rich and diverse macroinvertebrate community, including intolerant species and were relatively comparable to their respective reference site condition. Sites FC-1-BIO and NG-2-BIO revealed two and seven significant trends, respectively, with all but two of the trends revealing declining conditions over time. Many of the declining metrics were related to community composition. Both of these sites are influenced by nearby road conditions and typically receive a greater amount of sediment that decreases habitat suitability. In contrast, the Stewart Gulch site showed improvement for three of the twenty-five metrics, indicating that the macroinvertebrate community has remained stable, if not slightly improved over the years. The higher productivity at Site SG-1-BIO likely benefits the macroinvertebrate community.

Overall, the macroinvertebrate community has changed at multiple sites from 2006 to 2021. Historic metric data at Site AC-1-BIO indicate an increased community quality since 2006, and this site has become more similar to the reference site. The community was particularly poor at Site AC-1-BIO in 2007 following multiple disturbances but has recovered well. Conversely, the quality of the macroinvertebrate community at sites AC-3-BIO, LC-1-BIO, and WFB-1-BIO have slightly worsened over time and long term data at Site WFB-1-BIO were significantly poorer when compared to the Labrador Gulch reference site. Periodic low flows and sedimentation from adjacent roads have influence the habitat in Lost Camp Gulch and middle Annie Creek, while the iron oxide and low flow conditions have led to the poor habitat and biological conditions on West Fork False Bottom Creek.

5.4 Periphyton

In 2021, the reference sites LB-4-BIO and RC-1-BIO both supported healthy periphyton communities. Diatom diversity at Site LB-4-BIO was very high. The Autotrophic Index at both sites indicated that allochthonous organic matter may be influencing the periphyton community. However, the Pollution Index indicated no or minor organic enrichment. Overall, data in 2021 showed negligible changes for these two sites, with metric values being similar to those in previous years. Only five and one of the eighteen metrics significantly trended for Labrador Gulch and Reno Creek, respectively, with a mixture of improving and declining periphyton metrics over time.

The Annie Creek sites, AC-1-BIO and AC-2-BIO, each exhibited variable periphyton conditions in 2021. However, both sites exhibited a fair Autotrophic Index and Diversity above the 2.5 threshold indicating that balanced diatom assemblages were present. Both sites also revealed minimal significant trends over time. When compared to their respective reference sites, both Annie Creek sites revealed few differences, indicating that the periphyton communities were relatively stable and healthy at these sites.

The sites on Ross Valley, RV-2-BIO, Lost Camp Gulch, LC-1-BIO, and Deadwood Creek, DC-2-BIO contained a diverse diatom community that exhibited similar periphyton metrics with their respective reference sites in 2021. The Pollution Index was relatively good and Autotrophic Index was relatively poor or fair at these sites indicating that allochthonous organic matter was likely influencing these sites. Also, the Percent Tolerant Diatoms was poor at Site LC-1-BIO indicating that siltation from adjacent dirt roadway was affecting the assemblage. Few periphyton metrics trended over time at these sites.

The East Fork False Bottom periphyton sample essentially contained no periphyton in 2021. Historically, Site EFB-1-BIO contained a diverse diatom community, with many motile diatoms, and exhibited similar periphyton metrics with its reference site that mostly have not trended over time. The macroinvertebrate community at Site WFB-1-BIO is poorer than at its reference site due to the community being dominated by acidiphilic and high oxygen diatoms. As a result, diversity and other metrics were poor. Site WFB-1-BIO revealed four significant worsening trends over time for the autecological metrics which are largely tied to the decrease in taxa. This site continued to be influenced by iron oxide deposition that negatively affects the periphyton community.

The sites on Fantail Creek, Nevada Gulch, and Stewart Gulch supported a taxonomically rich and diverse diatom community in 2021. Sites FC-1-BIO and NG-2-BIO were similar to their refence site and the poor Autotrophic Index at Site FC-1-BIO suggesting allochthonous inputs. Diversity was low at Site SG-1-BIO resulting in a different but no less robust periphyton community when compared to the reference site. Two periphyton metrics trended over time for Site FC-1-BIO, while only one of the eighteen periphyton metrics revealed significant trends for Site NG-2-BIO. At Site SG-1-BIO, many of the periphyton metrics have been highly variable over the years with no significant trends observed over time. The extensive macrophyte growth at this site has somewhat limited the periphyton community in recent years.

5.5 Overview

Aquatic biological data collected in 2021 largely indicated the presence of abundant and healthy communities of aquatic organisms near the Wharf mine, while the long-term data indicated maintenance of healthy communities over time. The sites on Labrador Gulch and Reno Creek revealed quality habitat and contained healthy fish, macroinvertebrate, and periphyton populations. These sites continue to be appropriate reference sites that show little to no influence by anthropogenic activity.

The benthic macroinvertebrate population in upper Annie Creek has fully recovered from the water quality disturbances and the disturbance caused by removal of excess biomass at Site

AC-1-BIO. Mountain Suckers continue to be absent from the Site AC-2-BIO, and likely no longer inhabit any portion of the stream upstream of the falls. The macroinvertebrate community at Site AC-2-BIO did appear to be affected by fine sediments and increased siltation observed in past years. In lower Annie Creek, healthy trout populations have historically been present, although 2019 represented the first year when Brown Trout dominated the fish assemblage. Site AC-3-BIO was not sampled in 2020 and 2021 due to the tornado activity on July 8, 2020. Overall, the sites on Annie Creek contained quality habitat and healthy fish (where present), and the macroinvertebrate and periphyton populations do not appear to be affected by mining activity.

Healthy trout populations were also present in Deadwood Creek, East Fork False Bottom Creek, and Stewart Gulch. These sites also supported a rich and diverse macroinvertebrate community that was comparable to the reference site on Labrador Gulch. Deadwood Creek also contained a rich and diverse periphyton community comparable to the reference site while the Stewart Gulch periphyton assemblages was slightly different compared to the reference site assemblage. Trout were present in West Fork False Bottom Creek as well, but successful recruitment is limited due to the low flow and poorer water quality conditions in this stream. The macroinvertebrate and periphyton communities are also limited by the iron oxide deposition and habitat at this site.

The macroinvertebrate and periphyton assemblages on Ross Valley, Lost Camp Gulch, Fantail Creek, and Nevada Gulch were slightly limited by periods of low or no flow, siltation, or organic matter deposition that resulted in minor changes from 2006 to 2021. Although many of the macroinvertebrate and periphyton metrics indicated that these sites were similar to slightly better than their respective reference site on Reno Creek.

The majority of the biological metrics and habitat measurements do not indicate direct impacts from active mining in 2021. However, past mining activities may indirectly affect Stewart Gulch in terms of increasing productivity and biomass due to nitrogen inputs, while the iron oxide deposition in West Fork False Bottom is affecting the overall health of the biological assemblages. The lack of perennial or low flows in Cleopatra Creek and Lost Camp Gulch, respectively, affect the overall health of the periphyton and macroinvertebrate assemblages over the long-term, and influence the sediment conditions in Lost Camp Gulch and Annie Creek.

6. References

Anderson, R. O. 1980. Proportional stock density (PSD) and relative weight (W_r): Interpretive indices for fish populations and communities. Pp. 27-33 *in* Gloss, S., and B. Shupp (eds.). Practical Fisheries Management: More With Less in the 1980's. New York Chapter American Fisheries Society, Ithaca, NY.

Anderson, R. O., and R. M. Neumann. 1996. Length, weight, and associated structural indices. Pp. 447-482 *in* Murphy, B. R., and D. W. Willis (eds). Fisheries Techniques, Second Edition. American Fisheries Society, Bethesda, MD.

APHA (American Public Health Association). 2005. Standard Methods for the Examination of Water and Wastewater. 21st Edition, American Public Health Association/American Water Works Association/Water Environment Federation, Washington DC.

Bahls, L. L. 1993. Periphyton Bioassessment Methods for Montana Streams. Department of Health and Environmental Science, Helena, MT.

Barbour, M. T., J. Gerritsen, B. D. Snyder, and J. B. Stribling. 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates and Fish, 2nd Edition. EPA 841-B-99-002. U.S. Environmental Protection Agency, Washington, DC.

Biggs, B. J. F. 1996. Patterns in benthic algae of streams. In: Algal Ecology. Stevenson, J., M. L. Bothwell, and R. L. Lowe (eds.). Academic Press, San Diego, CA. pp. 31-51.

Bowman, M. F., and K. M. Somers. 2005. Considerations when using the Reference Condition Approach for bioassessment of freshwater ecosystems. Water Quality Research Journal of Canada 40:347-360.

Carlander, K. D. 1969. *Handbook of Freshwater Fishery Biology*, Vol. 1. Iowa State University Press, Ames, IA.

Carter, J. L., and V. H. Resh. 2001. After site selection and before data analysis: Sampling, sorting, and laboratory procedures used in stream benthic macroinvertebrate monitoring programs by U.S.A. state agencies. Journal of the North American Benthological Society 20:658-682.

Chadwick & Associates, Inc. (C&A). 1993. Aquatic Biology Assessment, Annie Creek. Draft report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick & Associates, Inc. (C&A). 1994a. Annie Creek Biological Monitoring. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick & Associates, Inc. (C&A). 1994b. Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick & Associates, Inc. (C&A). 1995a. Annie Creek Biological Monitoring. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick & Associates, Inc. 1995b. (C&A). 1994 Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1996a. Baseline Aquatic Biology of Upper Deadwood Creek and False Bottom Creek, South Dakota, 1995. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1996b. Biological Monitoring of Annie Creek, South Dakota, 1995. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1996c. Response of the Fish, Benthic Macroinvertebrate, and Periphyton Communities of Annie Creek, South Dakota, to an Accidental Release of Ammonia and Cyanide. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1996d. The 1995 Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1997a. 1996 Aquatic Biological Monitoring of Annie Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1997b. Baseline Aquatic Biology of Upper Deadwood Creek and False Bottom Creek, South Dakota, 1996. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1997c. The 1996 Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1998a. 1997 Aquatic Biological Monitoring of Annie Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1998b. 1997 Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1998c. Aquatic Biology Baseline Monitoring of Upper Deadwood Creek and False Bottom Creek, South Dakota, 1997. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1999a. 1998 Aquatic Biological Monitoring of Annie Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1999b. Aquatic Biology Assessment of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for the Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 1999c. Aquatic Biology Baseline Monitoring of Upper Deadwood Creek and False Bottom Creek, South Dakota, 1998. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2000a. 1999 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, and False Bottom Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2000b. 1999 Aquatic Biology Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2000c. Aquatic Biological Monitoring Plan for Annie Creek, Deadwood Creek, False Bottom Creek, McKinley Gulch, and Squaw Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2001a. 2000 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, and McKinley Gulch, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2001b. 2000 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2002a. 2001 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, and McKinley Gulch, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2002b. 2001 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2003a. 2002 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, and McKinley Gulch, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2003b. 2002 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2004a. 2003 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2004b. 2003 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2005a. 2004 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2005b. 2004 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Report prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2006a. 2005 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2006b. 2005 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

Chadwick Ecological Consultants, Inc. (CEC). 2006d. Aquatic Biological Monitoring Plan for Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

Everhart, W.H., and W.D. Youngs. 1981. Principles of Fishery Science, 2nd Edition. Cornell University Press, Ithaca, NY.

Fore, L. and C. Grafe. 2002. Using diatoms to assess the biological condition of large rivers in Idaho (U.S.A.). Freshwater Biology 47:2015-2037.Wege, G. J., and R. O. Anderson. 1978. Relative Weight (W_r): A new index of condition for largemouth bass. Pp. 79-91 *in* Novinger, G.D., and J.G. Dillard (eds.). New Approaches to the Management of Small Impoundments. Special Publication 5. North Central Division, American Fisheries Society.

GEI Consultants, Inc. (GEI). 2007a. 2006 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2007b. 2006 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

GEI Consultants, Inc. (GEI). 2008a. 2007 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2008b. Fish, benthic macroinvertebrate, and periphyton evaluation, June 2008 sampling. Memo prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2008c. 2007 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

GEI Consultants, Inc. (GEI). 2009a. 2008 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2009b. 2008 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

GEI Consultants, Inc. (GEI). 2010a. 2009 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2010b. 2009 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

GEI Consultants, Inc. (GEI). 2011a. 2010 Aquatic Biological Monitoring of Annie Creek, Upper Deadwood Creek, False Bottom Creek, McKinley Gulch, and Cleopatra Creek, South Dakota. Report prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2011b. 2010 Aquatic Biological Monitoring of Streams in the Vicinity of the Golden Reward Mine, Lawrence County, South Dakota. Prepared for Golden Reward Mining Company, Lead, SD.

GEI Consultants, Inc. (GEI). 2012. 2011 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf and Golden Reward Mines, Lawrence County, South Dakota. Prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2013. 2012 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf and Golden Reward Mines, Lawrence County, South Dakota. Prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2014. 2013 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf and Golden Reward Mines, Lawrence County, South Dakota. Prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2015. 2014 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf and Golden Reward Mines, Lawrence County, South Dakota. Prepared for Wharf Resources (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2016. 2015 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf and Golden Reward Mines, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2017a. 2016 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2017b. Root Cause Analysis: False Bottom Creek Sampling Location. Memorandum prepared on March 19, 2017 for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2018a. 2017 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2018b. 2018 Aquatic Biological Sampling and Analysis Plan for Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD,.

GEI Consultants, Inc. (GEI). 2019. 2018 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2020. 2019 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

GEI Consultants, Inc. (GEI). 2021. 2020 Aquatic Biological Monitoring of Streams in the Vicinity of the Wharf Mine, Lawrence County, South Dakota. Prepared for Coeur Mining (U.S.A.), Inc., Lead, SD.

Golden Reward Mining Company (GRMC). 1987. Golden Reward Mine: Aquatic Biology and Habitat Baseline Investigations. Report prepared by OEA Research, Inc.

Golden Reward Mining Company (GRMC). 1990. Golden Reward Mine: 1989 Aquatic Habitat, Macroinvertebrate, and Periphyton Surveys. Report prepared by OEA Research, Inc.

Golden Reward Mining Company (GRMC). 1992. Aquatic Biology and Habitat Survey, 1991, Golden Reward Mine. Report prepared by OEA Research, Inc.

Golden Reward Mining Company (GRMC). 1993. Aquatic Macroinvertebrate and Periphyton Survey, 1992, Golden Reward Mine. Report prepared by OEA Research, Inc.

Grafe, C.S. (ed.). 2002. Idaho Small Stream Ecological Assessment Framework: An Integrated Approach. Idaho Department of Environmental Quality, Boise, ID.

Hargett, E. G. 2011. The Wyoming Stream Integrity Index (WSII)- Multimetric Indices for Assessment of Wadeable Streams and Large Rivers in Wyoming. Document #11-0787, Water Quality Division, Cheyenne, Wyoming.

Hawkins, C. P., and J. R. Sedell. 1990. The role of refugia in the recolonization of streams devastated by the 1980 eruption of Mount St. Helens. Northwest Science 64:271-274.

Hill, B.H., A.T. Herlihy, P.R. Kaufmann, R.J. Stevenson, F.H. McCormick, and C.B. Johnson. 2000. Use of periphyton assemblage data as an Index of Biotic Integrity. Journal of North American Benthological Society 19(1):50-67.

Hilsenhoff, W. L. 1987. An improved biotic index of organic stream pollution. The Great Lakes Entomologist 20:31-39.

Hintze, J. L. 2004. NCSS and PASS. Number Cruncher Statistical Systems, Kaysville, UT.

Hynes, H. B. N. 1970. The Ecology of Running Waters. University of Toronto Press, Toronto, Canada.

Johnson, S. L., and C. C. Vaughn. 1995. A hierarchical study of macroinvertebrate recolonization of disturbed patches along a longitudinal gradient in a prairie river. Freshwater Biology 34:531-540.

Klemm, D. J., P. A. Lewis, F. Fulk, and J. M. Lazorchak. 1990. Macroinvertebrate Field and Laboratory Methods for Evaluating the Biological Integrity of Surface Waters. EPA/600/4-90/030. U.S. Environmental Protection Agency.

Knudson, K. 2003. An Evaluation of the Biological Communities of Squaw Creek, Labrador & Rubicon Gulches, Lawrence County near Lead, South Dakota. Report prepared for LAC Minerals (U.S.A.) LLC, Lead, SD.

Lange-Bertalot, H. 1979. Pollution tolerance of diatoms as a criterion for water quality estimation. Nova Hedwigia 64:285-304.

Lemly, D. A. 1997. Ecosystem recovery following selenium contamination in a freshwater reservoir. Ecotoxicology and Environmental Safety 36:275-281.

Lowe, R. L. 1974. Environmental Requirements and Pollution Tolerance of Freshwater Diatoms. EPA 670/4-74-005. U.S. Environmental Protection Agency, Cincinnati, OH.

Mariah Associates, Inc. 1990. Aquatic Report for the Annie Creek-Spearfish Creek (Reliance Waste Dump) and Squaw Creek (Foley Ridge Waste Dump) Drainage Areas. Report prepared for Wharf Resources, (U.S.A.), Inc., Deadwood, SD.

Mariah Associates, Inc. 1992a. Results of Aquatic Sampling on Annie Creek, Lawrence County, South Dakota, 1991. Report prepared for Wharf Resources (U.S.A.), Inc., Deadwood, SD.

Mariah Associates, Inc. 1992b. Results of Aquatic Sampling on Annie Creek, Lawrence County, South Dakota, 1992. Report prepared for Wharf Resources (U.S.A.), Inc., Deadwood, SD.

Merritt, R. W., K. W. Cummins, and M. B. Berg. 2008. An Introduction to the Aquatic Insects of North America, Fourth Edition. Kendall/Hunt Publishing Company, Dubuque, IA.

Murphy, B. R., and D. W. Willis. 1991. Application of relative weight (W_r) to western warmwater fisheries. Pp. 243-248 *in* Cooper, J.L., and R.H. Hamre (eds.). Warmwater Fisheries symposium I. General Technical Report RM-207. U.S. Forest Service, Fort Collins, CO.

National Oceanic and Atmospheric Administration (NOAA). 2020. National Centers for Environmental Information: Daily Summaries Station Details. Available: https://www.ncdc.noaa.gov/cdo-web/datasets/GHCND/stations/GHCND:USC00394834/detail. (December 2020).

Neumann, R.M., C.S. Guy, and D.W. Willis. 2012. Length, weight, and associated indices. Pp. 637-676 *in* Zale, A. V., D.L. Parrish, and T. M. Sutton (eds). Fisheries Techniques, Third Edition. American Fisheries Society, Bethesda, MD.

Omernik, J. M. 1987. Ecoregions of the Conterminous United States. Annals of the Association of American Geographers 77:118-125.

Omernik, J. M., and A. L. Gallant. 1987. Ecoregions of the West Central United States. EPA/600/D-87/317. U.S. Environmental Protection Agency, Corvallis, OR.

Overton, C. K., S. P. Wollrab, B. C. Roberts, and M. A. Radko. 1997. R1/R4 (Northern/Intermountain Regions) Fish and Fish Habitat Standard Inventory Procedures Handbook. General Technical Report INT-GTR-346. U.S. Department of Agriculture Forest Service Intermountain Research Station, Ogden, UT.

Patrick, R., and C. W. Reimer. 1966. The Diatoms of the United States, Exclusive of Alaska and Hawaii. Monograph 13. Academy of Natural Sciences, Philadelphia, PA.

Patrick, R., and C. W. Reimer. 1975. The Diatoms of the United States, Volume 2, Part 1, Monograph 13. Academy of Natural Sciences, Philadelphia, PA.

Plafkin, J.L., M.T. Barbour, K.D. Porter, S.K. Gross, and R.M. Hughes. 1989. Rapid bioassessment protocols for use in streams and rivers: Benthic macroinvertebrates and fish. U.S. Environmental Protection Agency, Office of Water Regulations and Standards, Washington, D.C. EPA 440-4-89-001.

Platts, W. S., W. F. Megahan, and G. W. Minshall. 1983. Methods for Evaluating Stream, Riparian, and Biotic Conditions. General Technical Report INT-138. U.S. Department of Agriculture Forest Service Intermountain Research Station, Ogden, UT.

Porter, S.D. 2008. Algal Attributes: An autecological classification of algal taxa collected by the National Water-Quality Assessment Program. U.S. Geological Survey Data Series 329, https://pubs.usgs.gov/ds/ds329/pdf/ds329.pdf.

Presser T.S., M. Hardy, M.A. Huebner, and P.J. Lamothe. 2004. Selenium loading through the Blackfoot River watershed: linking sources to ecosystems. In: J.R. Hein, editor. Life cycle of the phosphoria formation: From deposition to post-mining environment. New York (NY): Elsevier. p 299–319.

South Dakota Department of Agriculture and Natural Resources (SDDANR). 2021. South Dakota Administrative Rules for Surface Water Quality Uses Assigned to Streams. Chapter 74:51:03. Sections 1 and 10. Available: http://legis.sd.gov/rules/DisplayRule.aspx?Rule=74:51:03. (December 2021).

South Dakota Department of Environment and Natural Resources (SDDENR). 2017. Standard Operating Procedures for Field Samplers, Volume II: Biological and Habitat Related Techniques. Revision 3.1, Watershed Protection Team, January 2017.

Stribling, J. B., S. R. Moulton, II, and G. T. Lester. 2003. Determining the quality of taxonomic data. Journal of the North American Benthological Society 22:621-631.

United States Environmental Protection Agency (EPA). 2016. Aquatic life ambient water quality criterion for selenium – freshwater 2016. Office of Water, Office of Science and Technology, Washington D.C. EPA 822-R-16-006, June 2016.

Van Deventer, J. S., and W. S. Platts. 1983. Sampling and estimating fish populations for streams. Transactions of the North American Wildlife and Natural Resource Conference 48:349-354.

Van Deventer, J. S., and W. S. Platts. 1986. MicroFish Interactive Program, Version 3.0.

Van Dam, H., A. Mertens, and J. Sinkeldam. 1994. A Coded Check List and Ecological Indicator Values of Freshwater Diatoms and Ecological Indicator Values of Freshwater Diatoms from Netherland. Netherlands Journal of Aquatic Ecology, 28, 117-133. http://dx.doi.org/10.1007/BF02334251

Vinson, M. R., and C. P. Hawkins. 1996. Effects of sampling area and subsampling procedure on comparisons of taxa richness among streams. Journal of the North American Benthological Society 15:392-399.

Waters, T. F. 1995. *Sediment in Streams: Sources, Biological Effects, and Control.* Monograph 7, American Fisheries Society, Bethesda, MD.

Watson, V. J., and B. Gestring. 1996. Monitoring algae levels in the Clark Fork River. Intermountain J. Sci. 2:17-26.

Wehr, J. D., and R. G. Sheath (eds.). 2003. Freshwater Algae of North America. Academic Press, San Diego, CA.

Whittaker, R. H. 1975. Communities and Ecosystems, 2nd edition. Macmillan Publishing Co., New York, NY.

Wiederholm, T. 1984. Responses of aquatic insects to environmental pollution. Pp. 508-557 *in* Resh, V.H., and D.M. Rosenberg (eds.). The Ecology of Aquatic Insects. Praeger Scientific, New York, NY.

Wilhm, J. L. 1970. Range of diversity index in benthic macroinvertebrate populations. Journal of the Water Pollution Control Federation 42:R227-R234.

Williams, D. D. 1980. Temporal patterns in recolonization of stream benthos. Archive für Hydrobiologia 90:56-74.

Williams, D. D., and H. B. N. Hynes. 1976. The recolonization mechanisms of stream benthos. Oikos 27:265-272.

This page intentionally left blank

Appendix A 2021 Fish Data

This page intentionally left blank

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/27/2021
SITE: **LABRADOR GULCH, LB-4-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	200	80	1.00	90.0	88.9
1	BRK	189	66	0.98	75.5	87.4
1	BRK	175	54	1.01	59.4	90.8
1	BRK	171	51	1.02	55.3	92.2
1	BRK	160	33	0.81	45.0	73.3
1	BRK	150	33	0.98	36.8	89.6
1	BRK	149	32	0.97	36.1	88.7
1	BRK	148	33	1.02	35.3	93.4
1	BRK	147	31	0.98	34.6	89.6
1	BRK	144	29	0.97	32.5	89.3
1	BRK	139	25	0.93	29.1	85.9
1	BRK	136	24	0.95	27.2	88.3
1	BRK	136	21	0.83	27.2	77.2
1	BRK	135	22	0.89	26.6	82.8
1	BRK	132	21	0.91	24.8	84.7
1	BRK	130	22	1.00	23.6	93.1
1	BRK	128	17	0.81	22.5	75.5
1	BRK	127	17	0.83	22.0	77.3
1	BRK	126	17	0.85	21.5	79.2
1	BRK	121	17	0.96	18.9	89.9
1	BRK	121	16	0.90	18.9	84.6
1	BRK	111	12	0.88		
1	BRK	95	7.6	0.89		
1	BRK	89	7.1	1.01		
1	BRK	86	6.6	1.04		
2	BRK	192	67	0.95	79.3	84.5
2	BRK	149	33	1.00	36.1	91.4
2	BRK	126	17	0.85	21.5	79.2
2	BRK	91	7.2	0.96		
2	BRK	85	6.0	0.98		
3	BRK	136	25	0.99	27.2	92.0
3	BRK	93	8.9	1.11		
3	BRK	88	6.7	0.98		

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	33	33	33	25
	MIN:	85	6.0	0.81	73.3
	MAX:	200	80	1.11	93.4
	MEAN:	133.5	26.2	0.95	86.0

	1st Pass	2nd Pass	3rd Pass	Pop Est	95% CI	Site Area (acre)	Density (#/acre)	95% CI	Biomass (lbs/acre)
BRK	25	5	3	33	± 2	0.039	846	± 51	48.87

	1st Pass	2nd Pass	3rd Pass	Pop Est	95% CI	Site Area (ha)	Density (#/ha)	95% CI	Biomass (kg/ha)
BRK	25	5	3	33	± 2	0.016	2063	± 125	54.05

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/25/2021
SITE: **RENO CREEK, RC-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	173	41	0.79	57.4	71.5
1	BRK	152	27	0.77	38.4	70.3
1	BRK	144	29	0.97	32.5	89.3
1	BRK	143	26	0.89	31.8	81.8
1	BRK	138	22	0.84	28.4	77.3
1	BRK	128	24	1.14	22.5	106.5
1	BRK	95	8.3	0.97		
1	LOC	181	59	0.99	65.4	90.2
1	LOC	176	61	1.12	60.2	101.3
1	LOC	160	46	1.12	45.4	101.3
2	BRK	144	28	0.94	32.5	86.3
3		Pass 3 - No Fish				

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	8	8	8	7
	MIN:	95	8.3	0.77	70.3
	MAX:	173	41	1.14	106.5
	MEAN:	139.6	25.7	0.91	83.3

LOC		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	3	3	3	3
	MIN:	160	46	0.99	90.2
	MAX:	181	61	1.12	101.3
	MEAN:	172.3	55.3	1.08	97.6

	1st Pass	2nd Pass	3rd Pass	Pop Est	95% CI		Site Area (acre)	Density (#/acre)	95% CI		Biomass (lbs/acre)
BRK	7	1	0	8	±	0	0.029	276	±	0	15.64
LOC	3	0	0	3	±	0	0.029	103	±	0	12.56

	1st Pass	2nd Pass	3rd Pass	Pop Est	95% CI		Site Area (ha)	Density (#/ha)	95% CI		Biomass (kg/ha)
BRK	7	1	0	8	±	0	0.012	667	±	0	17.14
LOC	3	0	0	3	±	0	0.012	250	±	0	13.83

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/26/2021
SITE: **ANNIE CREEK, AC-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/26/2021
SITE: **ANNIE CREEK, AC-2-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/25/2021
SITE: **ANNIE CREEK, AC-3-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	LOC	204	102	1.20	93.2	109.4
1	LOC	191	84	1.21	76.7	109.5
1	LOC	191	68	0.98	76.7	88.6
1	LOC	172	52	1.02	56.3	92.4
1	LOC	170	54	1.10	54.3	99.4
1	LOC	155	39	1.05	41.3	94.3
1	LOC	125	21	1.08		

Only targeted electrofishing occurred to collect fish for tissue analysis.

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/26/2021
SITE: **ROSS VALLEY CREEK, RV-2-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/26/2021
SITE: **LOST CAMP GULCH, LC-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **DEADWOOD CREEK, DC-2-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	175	50	0.93	59.4	84.1
1	BRK	139	24	0.89	29.1	82.5
1	BRK	136	22	0.87	27.2	80.9
1	BRK	127	17	0.83	22.0	77.3
1	BRK	126	19	0.95	21.5	88.6
1	BRK	125	17	0.87	20.9	81.2
1	BRK	119	16	0.95		
1	BRK	119	16	0.95		
1	BRK	118	16	0.97		
1	BRK	117	14	0.87		
1	BRK	116	14	0.90		
1	BRK	116	14	0.90		
1	BRK	115	14	0.92		
1	BRK	115	13	0.85		
1	BRK	114	12	0.81		
1	BRK	111	12	0.88		
1	BRK	111	11	0.80		
1	BRK	110	11	0.83		
1	BRK	109	10	0.77		
1	BRK	107	11	0.90		
1	BRK	106	11	0.92		
1	BRK	106	11	0.92		
1	BRK	106	8.4	0.71		
1	BRK	105	10	0.86		
1	BRK	105	10	0.86		
1	BRK	104	10	0.89		
1	BRK	104	9.6	0.85		
1	BRK	104	9.3	0.83		
1	BRK	104	9.0	0.80		
1	BRK	104	8.8	0.78		
1	BRK	103	8.4	0.77		
1	BRK	102	9.0	0.85		
1	BRK	100	9.7	0.97		
1	BRK	100	8.8	0.88		
1	BRK	100	8.3	0.83		
1	BRK	100	8.1	0.81		
1	BRK	99	8.5	0.88		
1	BRK	98	8.2	0.87		
1	BRK	98	6.8	0.72		
1	BRK	95	7.4	0.86		
1	BRK	95	6.4	0.75		
1	BRK	94	7.1	0.85		
1	BRK	94	7.0	0.84		
1	BRK	94	6.7	0.81		
1	BRK	94	6.6	0.79		
1	BRK	94	6.4	0.77		
1	BRK	93	7.1	0.88		
1	BRK	93	6.9	0.86		
1	BRK	93	6.1	0.76		
1	BRK	92	7.1	0.91		

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **DEADWOOD CREEK, DC-2-BIO**

1	BRK	92	7.0	0.90
1	BRK	92	6.1	0.78
1	BRK	91	6.2	0.82
1	BRK	91	6.1	0.81
1	BRK	90	6.1	0.84
1	BRK	90	6.0	0.82
1	BRK	89	6.1	0.87
1	BRK	89	5.9	0.84
1	BRK	89	5.6	0.79
1	BRK	88	5.5	0.81
1	BRK	88	5.5	0.81
1	BRK	87	5.3	0.80
1	BRK	86	5.9	0.93
1	BRK	85	5.4	0.88
1	BRK	84	4.9	0.83
1	BRK	84	4.6	0.78
1	BRK	84	4.4	0.74
1	BRK	83	5.1	0.89
1	BRK	81	4.4	0.83
1	BRK	80	4.7	0.92
1	BRK	74	2.9	0.72
1	BRK	70	3.1	0.90
1	BRK	69	3.1	0.94
1	BRK	69	1.7	0.52
1	BRK	68	3.0	0.95
1	BRK	68	3.0	0.95
1	BRK	68	2.8	0.89
1	BRK	66	2.5	0.87
1	BRK	66	2.4	0.83
1	BRK	66	2.4	0.83
1	BRK	66	2.3	0.80
1	BRK	66	2.3	0.80
1	BRK	65	2.4	0.87
1	BRK	65	2.4	0.87
1	BRK	65	2.4	0.87
1	BRK	65	2.4	0.87
1	BRK	64	2.1	0.80
1	BRK	63	2.6	1.04
1	BRK	63	2.3	0.92
1	BRK	63	1.9	0.76
1	BRK	62	2.3	0.97
1	BRK	62	1.9	0.80
1	BRK	62	1.8	0.76
1	BRK	61	2.1	0.93
1	BRK	61	1.9	0.84
1	BRK	60	2.2	1.02
1	BRK	60	2.1	0.97
1	BRK	60	2.0	0.93
1	BRK	60	1.8	0.83
1	BRK	59	2.0	0.97
1	BRK	59	1.8	0.88
1	BRK	59	1.8	0.88

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **DEADWOOD CREEK, DC-2-BIO**

1	BRK	59	1.7	0.83
1	BRK	59	1.5	0.73
1	BRK	58	1.9	0.97
1	BRK	58	1.9	0.97
1	BRK	58	1.9	0.97
1	BRK	58	1.7	0.87
1	BRK	58	1.6	0.82
1	BRK	58	1.5	0.77
1	BRK	58	1.4	0.72
1	BRK	57	1.8	0.97
1	BRK	57	1.8	0.97
1	BRK	57	1.7	0.92
1	BRK	57	1.6	0.86
1	BRK	57	1.6	0.86
1	BRK	57	1.6	0.86
1	BRK	57	1.6	0.86
1	BRK	57	1.5	0.81
1	BRK	56	1.8	1.02
1	BRK	56	1.8	1.02
1	BRK	56	1.6	0.91
1	BRK	56	1.4	0.80
1	BRK	56	1.4	0.80
1	BRK	55	1.5	0.90
1	BRK	55	1.5	0.90
1	BRK	55	1.4	0.84
1	BRK	55	1.2	0.72
1	BRK	54	1.5	0.95
1	BRK	54	1.3	0.83
1	BRK	54	1.3	0.83
1	BRK	54	1.2	0.76
1	BRK	54	1.2	0.76
1	BRK	54	1.1	0.70
1	BRK	53	1.4	0.94
1	BRK	53	1.3	0.87
1	BRK	53	1.3	0.87
1	BRK	53	1.1	0.74
1	BRK	52	1.3	0.92
1	BRK	52	1.1	0.78
1	BRK	52	1.1	0.78
1	BRK	52	0.8	0.57
1	BRK	51	1.3	0.98
1	BRK	51	1.2	0.90
1	BRK	50	1.1	0.88
1	BRK	50	1.1	0.88
1	BRK	50	0.9	0.72
1	BRK	49	1.1	0.93
1	BRK	49	0.9	0.76
1	BRK	48	1.2	1.09
1	BRK	48	1.1	0.99
1	BRK	48	0.7	0.63
1	BRK	48	0.7	0.63
1	BRK	48	0.6	0.54

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **DEADWOOD CREEK, DC-2-BIO**

1	BRK	45	0.6	0.66		
2	BRK	154	36	0.99	40.0	90.0
2	BRK	137	24	0.93	27.8	86.3
2	BRK	133	20	0.85	25.4	78.8
2	BRK	130	26	1.18	23.6	110.0
2	BRK	127	23	1.12	22.0	104.6
2	BRK	123	17	0.91	19.9	85.4
2	BRK	121	14	0.79	18.9	74.0
2	BRK	120	15	0.87	18.4	81.4
2	BRK	118	15	0.91		
2	BRK	113	13	0.90		
2	BRK	107	8.9	0.73		
2	BRK	106	11	0.92		
2	BRK	105	10	0.86		
2	BRK	103	9.4	0.86		
2	BRK	100	8.6	0.86		
2	BRK	99	6.8	0.70		
2	BRK	98	7.4	0.79		
2	BRK	93	6.9	0.86		
2	BRK	92	6.2	0.80		
2	BRK	90	6.4	0.88		
2	BRK	89	5.1	0.72		
2	BRK	87	6.0	0.91		
2	BRK	86	5.2	0.82		
2	BRK	65	2.6	0.95		
2	BRK	64	2.0	0.76		
2	BRK	63	2.1	0.84		
2	BRK	63	1.9	0.76		
2	BRK	61	2.2	0.97		
2	BRK	61	1.7	0.75		
2	BRK	60	1.8	0.83		
2	BRK	59	2.0	0.97		
2	BRK	59	1.7	0.83		
2	BRK	59	1.5	0.73		
2	BRK	59	1.4	0.68		
2	BRK	58	2.3	1.18		
2	BRK	57	2.1	1.13		
2	BRK	57	1.5	0.81		
2	BRK	55	1.9	1.14		
2	BRK	55	1.6	0.96		
2	BRK	55	1.4	0.84		
2	BRK	54	1.2	0.76		
3	BRK	119	14	0.83		
3	BRK	110	11	0.83		
3	BRK	109	11	0.85		
3	BRK	109	10	0.77		
3	BRK	107	9.7	0.79		
3	BRK	104	9.1	0.81		
3	BRK	96	7.0	0.79		
3	BRK	80	4.0	0.78		
3	BRK	71	2.7	0.75		
3	BRK	64	2.2	0.84		

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **DEADWOOD CREEK, DC-2-BIO**

3	BRK	63	2.3	0.92
3	BRK	59	1.5	0.73
3	BRK	58	2.0	1.03
3	BRK	56	1.3	0.74
3	BRK	55	1.4	0.84
3	BRK	54	1.5	0.95
3	BRK	51	0.8	0.60
3	BRK	49	0.6	0.51

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	214	214	214	14
	MIN:	45	0.6	0.51	74.0
	MAX:	175	50	1.18	110.0
	MEAN:	79.8	5.9	0.85	86.1

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (acre)	Density (#/acre)		95% CI	Biomass (lbs/acre)
BRK	155	41	18	220	±	7	0.035	6286	±	200	81.76

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (ha)	Density (#/ha)		95% CI	Biomass (kg/ha)
BRK	155	41	18	220	±	7	0.014	15714	±	500	92.71

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/23/2021
SITE: **EAST FORK FALSE BOTTOM CREEK, EFB-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	156	39	1.03	41.6	93.7
1	BRK	140	27	0.98	29.7	90.8
1	BRK	136	26	1.03	27.2	95.6
1	BRK	131	23	1.02	24.2	95.0
1	BRK	126	20	1.00	21.5	93.2
1	BRK	124	17	0.89	20.4	83.3
1	BRK	113	14	0.97		
1	BRK	113	13	0.90		
1	BRK	106	10	0.84		
1	BRK	104	11	0.98		
1	BRK	104	11	0.98		
1	BRK	102	11	1.04		
1	BRK	101	9.6	0.93		
1	BRK	100	11	1.10		
1	BRK	100	9.5	0.95		
1	BRK	100	8.3	0.83		
1	BRK	98	10	1.06		
1	BRK	96	7.4	0.84		
1	BRK	92	7.0	0.90		
1	BRK	91	6.1	0.81		
1	BRK	90	8.5	1.17		
1	BRK	90	6.6	0.91		
1	BRK	87	5.7	0.87		
1	BRK	74	3.7	0.91		
1	BRK	72	3.2	0.86		
1	BRK	70	3.0	0.87		
1	BRK	70	2.8	0.82		
1	BRK	68	2.5	0.80		
1	BRK	66	2.2	0.77		
1	BRK	63	2.2	0.88		
2	BRK	144	29	0.97	32.5	89.3
2	BRK	117	16	1.00		
2	BRK	112	17	1.21		
2	BRK	109	13	1.00		
2	BRK	109	11	0.85		
2	BRK	107	11	0.90		
2	BRK	105	10	0.86		
2	BRK	103	9.6	0.88		
2	BRK	100	9.6	0.96		
2	BRK	97	7.7	0.84		
2	BRK	93	6.2	0.77		
2	BRK	89	7.9	1.12		
2	BRK	71	3.1	0.87		
2	BRK	68	2.6	0.83		
2	BRK	68	2.6	0.83		
2	BRK	63	2.4	0.96		
3	BRK	117	17	1.06		
3	BRK	104	11	0.98		
3	BRK	101	9.2	0.89		
3	BRK	96	7.6	0.86		

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/23/2021
SITE: **EAST FORK FALSE BOTTOM CREEK, EFB-1-BIO**

3	BRK	94	7.1	0.85
3	BRK	93	7.2	0.90

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	52	52	52	7
	MIN:	63	2.2	0.77	83.3
	MAX:	156	39	1.21	95.6
	MEAN:	98.9	10.6	0.93	91.6

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (acre)	Density (#/acre)		95% CI	Biomass (lbs/acre)
BRK	30	16	6	56	±	8	0.022	2545	±	364	59.47

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (ha)	Density (#/ha)		95% CI	Biomass (kg/ha)
BRK	30	16	6	56	±	8	0.009	6222	±	889	65.95

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/23/2021
SITE: **WEST FORK FALSE BOTTOM CREEK, WFB-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	118	16	0.97		
1	BRK	113	14	0.97		

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K
	N:	2	2	2
	MIN:	113	14	0.97
	MAX:	118	16	0.97
	MEAN:	115.5	15.0	0.97

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (acre)	Density (#/acre)		95% CI	Biomass (lbs/acre)
BRK	2	0	0	2	±	0	0.021	95	±	0	3.14

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (ha)	Density (#/ha)		95% CI	Biomass (kg/ha)
BRK	2	0	0	2	±	0	0.008	250	±	0	3.75

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/25/2021
SITE: **FANTAIL CREEK, FC-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **NEVADA GULCH, NG-2-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1			NO FISH			

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **STEWART GULCH, SG-1-BIO**

PASS	SPECIES	LENGTH (mm)	WEIGHT (g)	K	Ws	Wr
1	BRK	206	81	0.93	98.6	82.1
1	BRK	205	80	0.93	97.1	82.4
1	BRK	195	68	0.92	83.2	81.8
1	BRK	193	72	1.00	80.6	89.4
1	BRK	192	69	0.97	79.3	87.0
1	BRK	191	61	0.88	78.0	78.2
1	BRK	190	61	0.89	76.7	79.5
1	BRK	188	57	0.86	74.3	76.8
1	BRK	177	46	0.83	61.6	74.7
1	BRK	170	43	0.88	54.3	79.1
1	BRK	165	43	0.96	49.5	86.8
1	BRK	163	43	0.99	47.7	90.2
1	BRK	162	35	0.82	46.8	74.8
1	BRK	159	37	0.92	44.2	83.8
1	BRK	156	36	0.95	41.6	86.5
1	BRK	155	32	0.86	40.8	78.4
1	BRK	154	36	0.99	40.0	90.0
1	BRK	154	33	0.90	40.0	82.5
1	BRK	153	34	0.95	39.2	86.8
1	BRK	153	31	0.87	39.2	79.1
1	BRK	152	33	0.94	38.4	86.0
1	BRK	148	29	0.89	35.3	82.0
1	BRK	146	27	0.87	33.9	79.7
1	BRK	145	27	0.89	33.2	81.4
1	BRK	144	26	0.87	32.5	80.1
1	BRK	142	23	0.80	31.1	74.0
1	BRK	140	29	1.06	29.7	97.5
1	BRK	138	26	0.99	28.4	91.4
1	BRK	138	24	0.91	28.4	84.4
1	BRK	137	22	0.86	27.8	79.1
1	BRK	135	21	0.85	26.6	79.0
1	BRK	134	20	0.83	26.0	77.0
1	BRK	133	23	0.98	25.4	90.7
1	BRK	131	22	0.98	24.2	90.9
1	BRK	128	20	0.95	22.5	88.8
1	BRK	128	20	0.95	22.5	88.8
1	BRK	128	19	0.91	22.5	84.3
1	BRK	127	17	0.83	22.0	77.3
1	BRK	126	20	1.00	21.5	93.2
1	BRK	126	18	0.90	21.5	83.9
1	BRK	125	19	0.97	20.9	90.8
1	BRK	125	18	0.92	20.9	86.0
1	BRK	125	17	0.87	20.9	81.2
1	BRK	124	16	0.84	20.4	78.4
1	BRK	121	17	0.96	18.9	89.9
1	BRK	120	16	0.93	18.4	86.8
1	BRK	120	15	0.87	18.4	81.4
1	BRK	120	14	0.81	18.4	75.9
1	BRK	118	17	1.03		
1	BRK	118	15	0.91		

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **STEWART GULCH, SG-1-BIO**

1	BRK	118	14	0.85
1	BRK	118	14	0.85
1	BRK	117	15	0.94
1	BRK	116	15	0.96
1	BRK	116	15	0.96
1	BRK	115	13	0.85
1	BRK	114	13	0.88
1	BRK	110	12	0.90
1	BRK	109	12	0.93
1	BRK	108	12	0.95
1	BRK	108	12	0.95
1	BRK	107	11	0.90
1	BRK	107	11	0.90
1	BRK	106	11	0.92
1	BRK	105	13	1.12
1	BRK	105	11	0.95
1	BRK	105	11	0.95
1	BRK	105	10	0.86
1	BRK	105	10	0.86
1	BRK	104	9.5	0.84
1	BRK	100	9.4	0.94
1	BRK	100	8.9	0.89
1	BRK	97	7.4	0.81
1	BRK	95	6.5	0.76
1	BRK	81	4.8	0.90
1	BRK	81	2.3	0.43
1	BRK	80	3.6	0.70
1	BRK	79	4.0	0.81
1	BRK	79	3.5	0.71
1	BRK	75	3.2	0.76
1	BRK	75	2.9	0.69
1	BRK	74	3.3	0.81
1	BRK	73	2.5	0.64
1	BRK	72	2.9	0.78
1	BRK	72	2.7	0.72
1	BRK	71	2.4	0.67
1	BRK	70	2.9	0.85
1	BRK	70	2.7	0.79
1	BRK	69	3.2	0.97
1	BRK	69	2.8	0.85
1	BRK	68	2.0	0.64
1	BRK	67	2.3	0.76
1	BRK	67	2.1	0.70
1	BRK	66	2.2	0.77
1	BRK	66	2.1	0.73
1	BRK	66	2.1	0.73
1	BRK	65	1.6	0.58
1	BRK	64	2.0	0.76
1	BRK	63	2.0	0.80
1	BRK	62	2.2	0.92
1	BRK	60	1.4	0.65
1	BRK	52	0.8	0.57

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **STEWART GULCH, SG-1-BIO**

1	LOC	190	71	1.04	75.5	94.0
1	LOC	180	53	0.91	64.4	82.4
1	LOC	147	31	0.98	35.3	87.7
2	BRK	212	89	0.93	107.8	82.6
2	BRK	195	72	0.97	83.2	86.6
2	BRK	185	60	0.95	70.6	84.9
2	BRK	153	31	0.87	39.2	79.1
2	BRK	146	28	0.90	33.9	82.6
2	BRK	141	25	0.89	30.4	82.2
2	BRK	138	24	0.91	28.4	84.4
2	BRK	137	22	0.86	27.8	79.1
2	BRK	136	24	0.95	27.2	88.3
2	BRK	133	22	0.94	25.4	86.7
2	BRK	132	21	0.91	24.8	84.7
2	BRK	130	20	0.91	23.6	84.6
2	BRK	127	19	0.93	22.0	86.4
2	BRK	127	17	0.83	22.0	77.3
2	BRK	126	16	0.80	21.5	74.6
2	BRK	122	15	0.83	19.4	77.3
2	BRK	116	12	0.77		
2	BRK	115	13	0.85		
2	BRK	115	13	0.85		
2	BRK	114	13	0.88		
2	BRK	112	13	0.93		
2	BRK	110	13	0.98		
2	BRK	110	11	0.83		
2	BRK	109	12	0.93		
2	BRK	106	12	1.01		
2	BRK	88	4.8	0.70		
2	BRK	86	5.3	0.83		
2	BRK	85	5.2	0.85		
2	BRK	80	4.3	0.84		
2	BRK	78	3.6	0.76		
2	BRK	75	3.0	0.71		
2	BRK	73	2.5	0.64		
2	BRK	70	2.7	0.79		
2	BRK	68	2.3	0.73		
2	BRK	68	2.2	0.70		
2	BRK	67	2.2	0.73		
3	BRK	133	23	0.98	25.4	90.7
3	BRK	112	12	0.85		
3	BRK	110	12	0.90		
3	BRK	106	10	0.84		
3	BRK	84	4.3	0.73		
3	BRK	82	3.7	0.67		
3	BRK	74	3.2	0.79		
3	BRK	74	2.5	0.62		
3	BRK	70	2.5	0.73		
3	BRK	69	2.3	0.70		
3	LOC	155	38	1.02	41.3	91.9

DATA: FISH
CLIENT: WHARF
SAMPLED: 08/24/2021
SITE: **STEWART GULCH, SG-1-BIO**

SUMMARY:

BRK		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	148	148	148	65
	MIN:	52	0.8	0.43	74.0
	MAX:	212	89	1.12	97.5
	MEAN:	115.3	18.3	0.86	83.4

LOC		LENGTH (mm)	WEIGHT (g)	K	Wr
	N:	4	4	4	4
	MIN:	147	31	0.91	82.4
	MAX:	190	71	1.04	94.0
	MEAN:	168.0	48.3	0.99	89.0

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (acre)	Density (#/acre)		95% CI	Biomass (lbs/acre)
BRK	102	36	10	152	±	6	0.045	3378	±	133	136.28
LOC	3	0	1	4	±	2	0.045	89	±	44	9.48

	1st Pass	2nd Pass	3rd Pass	Pop Est		95% CI	Site Area (ha)	Density (#/ha)		95% CI	Biomass (kg/ha)
BRK	102	36	10	152	±	6	0.018	8444	±	333	154.53
LOC	3	0	1	4	±	2	0.018	222	±	111	10.72

This page intentionally left blank

Appendix B 2021 Benthic Macroinvertebrate Data

This page intentionally left blank

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/27/2021
Site: LABRADOR GULCH, LB-4-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
COLLEMBOLA	**10**	
Unid. Collembola	10	0.2
EPHEMEROPTERA	**1,425**	
Ameletus sp.	45	0.9
Baetis tricaudatus cx.	790	15.5
Diphetor hageni	465	9.1
Leptophlebiidae	125	2.5
PLECOPTERA	**455**	
Hesperoperla pacifica	100	2.0
Skwala americana	10	0.2
Sweltsa sp.	30	0.6
Zapada cinctipes	315	6.2
MEGALOPTERA	**20**	
Sialis sp.	20	0.4
COLEOPTERA	**190**	
Cleptelmis addenda	5	0.1
Heterlimnius corpulentus	135	2.7
Optioservus seriatus	45	0.9
Zaitzevia parvula	5	0.1
TRICHOPTERA	**1,071**	
Dolophilodes distinctus	5	0.1
Hesperophylax sp.	25	0.5
Hydropsyche sp.	45	0.9
Lepidostoma sp.	75	1.5
Micrasema bactro	705	13.9
Oecetis sp.	5	0.1
Oligophlebodes minutus	130	2.6
Polycentropus sp.	1	<0.1
Rhyacophila brunnea/vao	50	1.0
Rhyacophila sp.	30	0.6
DIPTERA	**1,791**	
Antocha monticola	85	1.7
Conchapelopia/Thienemannimyia gr.	25	0.5
Corynoneura sp.	25	0.5
Cricotopus (Nostococladius) nostocicola	80	1.6
Dicranota sp.	10	0.2
Dixa sp.	55	1.1

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/27/2021
Site: LABRADOR GULCH, LB-4-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
Eukiefferiella sp.	25	0.5
DIPTERA (cont.)		
Meringodixa chalonensis	5	0.1
Micropsectra sp.	190	3.7
Neoplasta sp.	5	0.1
Odontomesa sp.	165	3.2
Orthocladius/Cricotopus gr.	245	4.8
Pagastia sp.	25	0.5
Pentaneura sp.	55	1.1
Pericoma sp.	5	0.1
Phaenopsectra sp.	25	0.5
Polypedilum sp.	55	1.1
Prodiamesa sp.	55	1.1
Radotanypus sp.	215	4.2
Rheocricotopus sp.	340	6.7
Simulium sp.	15	0.3
Synorthocladius sp.	25	0.5
Thienemanniella sp.	55	1.1
Tipula sp.	1	<0.1
Trichoclinocera sp.	5	0.1
HYDRACARINA	**35**	
Atractides sp.	15	0.3
Protzia sp.	15	0.3
Sperchon sp.	5	0.1
ANNELIDA		
OLIGOCHAETA	**45**	
Eiseniella tetraedra	15	0.3
Unid. Immature Tubificidae w/o Capilliform Chaetae	30	0.6
MOLLUSCA		
PELECYPODA	**40**	
Pisidium sp.	40	0.8
TOTAL (#/sample)	5,082	
NUMBER OF TAXA	55	
SHANNON-WEAVER (H')	4.51	
TOTAL EPT TAXA	18	
EPT INDEX (% of Total Taxa)	33	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	28	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/25/2021
Site: RENO CREEK, RC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
COLLEMBOLA	**5**	
Unid. Collembola	5	0.1
EPHEMEROPTERA	**510**	
Baetis tricaudatus cx.	100	2.4
Diphetor hageni	330	7.8
Leptophlebiidae	80	1.9
PLECOPTERA	**685**	
Amphinemura banksi	5	0.1
Capniidae	5	0.1
Hesperoperla pacifica	210	5.0
Isoperla sp.	5	0.1
Sweltsa sp.	145	3.4
Zapada cinctipes	315	7.5
MEGALOPTERA	**35**	
Sialis sp.	35	0.8
COLEOPTERA	**170**	
Heterlimnius corpulentus	120	2.8
Optioservus seriatus	50	1.2
TRICHOPTERA	**295**	
Hydropsyche sp.	5	0.1
Lepidostoma sp.	195	4.6
Micrasema bactro	5	0.1
Oligophlebodes minutus	50	1.2
Rhyacophila brunnea/vao	35	0.8
Rhyacophila sp.	5	0.1
DIPTERA	**1,990**	
Ceratopogoninae	65	1.5
Conchapelopia/Thienemannimyia gr.	30	0.7
Corynoneura sp.	60	1.4
Dicranota sp.	10	0.2
Eukiefferiella sp.	30	0.7
Hydrobaenus sp.	30	0.7
Limnophyes sp.	60	1.4
Meringodixa chalonensis	30	0.7
Micropsectra sp.	675	16.0
Odontomesa sp.	90	2.1
Orthocladius/Cricotopus gr.	120	2.8

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/25/2021
Site: RENO CREEK, RC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
DIPTERA (cont.)		
Pagastia sp.	60	1.4
Parakiefferiella sp.	60	1.4
Polypedilum sp.	60	1.4
Radotanypus sp.	60	1.4
Rheocricotopus sp.	365	8.6
Simulium sp.	5	0.1
Synorthocladius sp.	30	0.7
Tvetenia sp.	150	3.6
HYDRACARINA	**25**	
Lebertia sp.	5	0.1
Protzia sp.	20	0.5
CRUSTACEA		
AMPHIPODA	**380**	
Gammarus sp.	380	9.0
ANNELIDA		
OLIGOCHAETA	**120**	
Eiseniella tetraedra	30	0.7
Limnodrilus sp.	5	0.1
Unid. Immature Tubificidae w/ Capilliform Chaetae	35	0.8
Unid. Immature Tubificidae w/o Capilliform Chaetae	50	1.2
MOLLUSCA		
PELECYPODA	**5**	
Pisidium sp.	5	0.1

TOTAL (#/sample)	4,220
NUMBER OF TAXA	46
SHANNON-WEAVER (H')	4.52
TOTAL EPT TAXA	15
EPT INDEX (% of Total Taxa)	33
EPHEMEROPTERA ABUNDANCE (% of Total Number)	12

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**345**	
Baetis tricaudatus cx.	155	2.5
Diphetor hageni	190	3.1
PLECOPTERA	**960**	
Hesperoperla pacifica	325	5.3
Sweltsa sp.	30	0.5
Zapada cinctipes	605	9.8
COLEOPTERA	**840**	
Helophorus sp.	5	0.1
Heterlimnius corpulentus	725	11.8
Narpus concolor	10	0.2
Optioservus divergens	100	1.6
TRICHOPTERA	**1,750**	
Anagapetus debilis	5	0.1
Hesperophylax sp.	60	1.0
Lepidostoma sp.	990	16.1
Micrasema bactro	305	5.0
Oligophlebodes minutus	340	5.5
Rhyacophila brunnea gr.	45	0.7
Rhyacophila sp.	5	0.1
DIPTERA	**1,780**	
Antocha monticola	5	0.1
Brillia sp.	25	0.4
Conchapelopia/Thienemannimyia gr.	25	0.4
Corynoneura sp.	50	0.8
Dicranota sp.	150	2.4
Ephydridae	5	0.1
Eukiefferiella sp.	95	1.5
Meringodixa chalonensis	5	0.1
Micropsectra sp.	25	0.4
Neoplasta sp.	5	0.1
Orthocladius/Cricotopus gr.	825	13.4
Pagastia sp.	170	2.8
Pericoma sp.	190	3.1
Prodiamesa sp.	25	0.4
Pseudodiamesa sp.	25	0.4
Simulium sp.	5	0.1
Tipula sp.	125	2.0
Tvetenia sp.	25	0.4

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
HYDRACARINA	**85**	
Hygrobates sp.	5	0.1
Lebertia sp.	30	0.5
Protzia sp.	15	0.2
Sperchon sp.	15	0.2
Thyopsis sp.	20	0.3
TURBELLARIA	**65**	
Polycelis coronata	65	1.1
ANNELIDA		
OLIGOCHAETA	**315**	
Unid. Immature Tubificidae w/ Capilliform Chaetae	295	4.8
Unid. Immature Tubificidae w/o Capilliform Chaetae	20	0.3
MOLLUSCA		
PELECYPODA	**20**	
Pisidium sp.	20	0.3
TOTAL (#/sample)	6,160	
NUMBER OF TAXA	43	
SHANNON-WEAVER (H')	4.13	
TOTAL EPT TAXA	12	
EPT INDEX (% of Total Taxa)	28	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	6	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**805**	
Ameletus sp.	25	0.7
Baetis tricaudatus cx.	420	12.3
Diphetor hageni	355	10.4
Leptophlebiidae	5	0.1
PLECOPTERA	**410**	
Amphinemura banksi	5	0.1
Hesperoperla pacifica	105	3.1
Skwala americana	10	0.3
Sweltsa sp.	160	4.7
Zapada cinctipes	130	3.8
COLEOPTERA	**755**	
Heterlimnius corpulentus	445	13.0
Narpus concolor	20	0.6
Optioservus divergens	180	5.3
Optioservus seriatus	85	2.5
Zaitzevia parvula	25	0.7
TRICHOPTERA	**345**	
Amphicosmoecus canax	5	0.1
Glossosoma ventrale	50	1.5
Hesperophylax sp.	20	0.6
Hydropsyche sp.	45	1.3
Lepidostoma sp.	60	1.8
Micrasema bactro	5	0.1
Oligophlebodes minutus	95	2.8
Rhyacophila brunnea gr.	60	1.8
Rhyacophila sp.	5	0.1
DIPTERA	**1,030**	
Antocha monticola	5	0.1
Clinocera sp.	10	0.3
Conchapelopia/Thienemannimyia gr.	15	0.4
Corynoneura sp.	30	0.9
Diamesa sp.	15	0.4
Dicranota sp.	10	0.3
Dixa sp.	50	1.5
Eukiefferiella sp.	30	0.9
Hexatoma sp.	10	0.3
Limnophila sp.	5	0.1
Meringodixa chalonensis	15	0.4
Micropsectra sp.	135	3.9
Odontomesa sp.	30	0.9
Orthocladius/Cricotopus gr.	265	7.7

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
DIPTERA (cont.)		
Pagastia sp.	15	0.4
Pericoma sp.	95	2.8
Radotanypus sp.	180	5.3
Simulium sp.	45	1.3
Synorthocladius sp.	15	0.4
Tipula sp.	25	0.7
Tvetenia sp.	30	0.9
HYDRACARINA	**25**	
Hygrobates sp.	10	0.3
Lebertia sp.	15	0.4
TURBELLARIA	**15**	
Polycelis coronata	15	0.4
ANNELIDA		
OLIGOCHAETA	**30**	
Eiseniella tetraedra	15	0.4
Lumbriculidae	5	0.1
Unid. Immature Tubificidae w/o Capilliform Chaetae	10	0.3
MOLLUSCA		
GASTROPODA	**5**	
Physa sp.	5	0.1
PELECYPODA	**5**	
Pisidium sp.	5	0.1
TOTAL (#/sample)	3,425	
NUMBER OF TAXA	52	
SHANNON-WEAVER (H')	4.56	
TOTAL EPT TAXA	18	
EPT INDEX (% of Total Taxa)	35	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	24	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ROSS VALLEY, RV-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
COLLEMBOLA	**5**	
Unid. Collembola	5	0.1
EPHEMEROPTERA	**900**	
Baetis tricaudatus cx.	95	2.2
Diphetor hageni	805	18.6
PLECOPTERA	**130**	
Perlidae	20	0.5
Sweltsa sp.	15	0.3
Zapada cinctipes	95	2.2
COLEOPTERA	**195**	
Heterlimnius corpulentus	45	1.0
Microcylloepus pusillus	5	0.1
Narpus concolor	5	0.1
Optioservus divergens	140	3.2
TRICHOPTERA	**150**	
Hesperophylax sp.	15	0.3
Lepidostoma sp.	65	1.5
Micrasema bactro	25	0.6
Rhyacophila brunnea gr.	45	1.0
DIPTERA	**1,180**	
Caloparyphus/Euparyphus sp.	15	0.3
Ceratopogoninae	30	0.7
Conchapelopia/Thienemannimyia gr.	10	0.2
Corynoneura sp.	25	0.6
Dicranota sp.	30	0.7
Dixa sp.	10	0.2
Eukiefferiella sp.	10	0.2
Meringodixa chalonensis	40	0.9
Micropsectra sp.	60	1.4
Orthocladius/Cricotopus gr.	25	0.6
Pericoma sp.	525	12.1
Polypedilum sp.	10	0.2
Prodiamesa sp.	10	0.2
Radotanypus sp.	330	7.6
Synorthocladius sp.	10	0.2
Tipula sp.	25	0.6
Zavrelimyia (Paramerina)	15	0.3

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: ROSS VALLEY, RV-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
HYDRACARINA	**35**	
Arrenurus sp.	30	0.7
Sperchon sp.	5	0.1
TURBELLARIA	**1,610**	
Polycelis coronata	1,610	37.2
ANNELIDA		
OLIGOCHAETA	**10**	
Unid. Immature Tubificidae w/ Capilliform Chaetae	10	0.2
MOLLUSCA		
PELECYPODA	**115**	
Pisidium sp.	115	2.7
TOTAL (#/sample)	4,330	
NUMBER OF TAXA	36	
SHANNON-WEAVER (H')	3.21	
TOTAL EPT TAXA	9	
EPT INDEX (% of Total Taxa)	25	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	21	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: LOST CAMP GULCH, LC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**20**	
Leptophlebiidae	20	0.9
MEGALOPTERA	**30**	
Sialis sp.	30	1.4
COLEOPTERA	**365**	
Heterlimnius corpulentus	225	10.5
Optioservus divergens	125	5.8
Paracymus sp.	5	0.2
Sanfilippodytes vilis	10	0.5
TRICHOPTERA	**180**	
Hesperophylax sp.	130	6.1
Lepidostoma sp.	5	0.2
Limnephilus sp.	30	1.4
Oligophlebodes minutus	15	0.7
DIPTERA	**1,500**	
Brillia sp.	25	1.2
Conchapelopia/Thienemannimyia gr.	25	1.2
Dicranota sp.	20	0.9
Eukiefferiella sp.	25	1.2
Meringodixa chalonensis	10	0.5
Micropsectra sp.	370	17.2
Odontomesa sp.	25	1.2
Pagastia sp.	25	1.2
Phaenopsectra sp.	25	1.2
Polypedilum sp.	700	32.6
Prodiamesa sp.	25	1.2
Pseudodiamesa sp.	25	1.2
Radotanypus sp.	200	9.3
HYDRACARINA	**5**	
Hygrobates sp.	5	0.2
TURBELLARIA	**5**	
Polycelis coronata	5	0.2
ANNELIDA		

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/26/2021
Site: LOST CAMP GULCH, LC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
OLIGOCHAETA	**40**	
Eiseniella tetraedra	25	1.2
Unid. Immature Tubificidae w/ Capilliform Chaetae	5	0.2
Unid. Immature Tubificidae w/o Capilliform Chaetae	10	0.5
TOTAL (#/sample)	2,145	
NUMBER OF TAXA	28	
SHANNON-WEAVER (H')	3.34	
TOTAL EPT TAXA	5	
EPT INDEX (% of Total Taxa)	18	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	1	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: DEADWOOD CREEK, DC-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
COLLEMBOLA	**4**	
Unid. Collembola	4	0.2
EPHEMEROPTERA	**164**	
Baetis tricaudatus cx.	40	1.8
Diphetor hageni	52	2.3
Leptophlebiidae	72	3.2
PLECOPTERA	**220**	
Amphinemura banksi	4	0.2
Hesperoperla pacifica	92	4.1
Sweltsa sp.	16	0.7
Zapada cinctipes	108	4.9
MEGALOPTERA	**28**	
Sialis sp.	28	1.3
COLEOPTERA	**12**	
Heterlimnius corpulentus	8	0.4
Zaitzevia parvula	4	0.2
TRICHOPTERA	**88**	
Hydropsychidae	8	0.4
Lepidostoma sp.	36	1.6
Micrasema bactro	36	1.6
Ochrotrichia sp.	4	0.2
Rhyacophila brunnea gr.	4	0.2
DIPTERA	**1,665**	
Ceratopogoninae	44	2.0
Conchapelopia/Thienemannimyia gr.	76	3.4
Corynoneura sp.	24	1.1
Dicranota sp.	4	0.2
Limnophyes sp.	24	1.1
Meringodixa chalonensis	4	0.2
Micropsectra sp.	336	15.1
Neoplasta sp.	4	0.2
Oreogeton sp.	8	0.4
Orthocladius/Cricotopus gr.	76	3.4

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: DEADWOOD CREEK, DC-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
DIPTERA (cont.)		
Parakiefferiella sp.	52	2.3
Parametriocnemus sp.	24	1.1
Polypedilum sp.	24	1.1
Rheocricotopus sp.	128	5.8
Tabanus gr.	4	0.2
Tipula sp.	1	<0.1
Trissopelopia ogemawi	808	36.3
Tvetenia sp.	24	1.1
NEMATODA	**8**	
Unid. Nematoda	8	0.4
ANNELIDA		
OLIGOCHAETA	**16**	
Lumbriculidae	4	0.2
Nais sp.	4	0.2
Unid. Immature Tubificidae w/ Capilliform Chaetae	8	0.4
MOLLUSCA		
PELECYPODA	**20**	
Pisidium sp.	20	0.9
TOTAL (#/sample)	2,225	
NUMBER OF TAXA	39	
SHANNON-WEAVER (H')	3.61	
TOTAL EPT TAXA	12	
EPT INDEX (% of Total Taxa)	31	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	7	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/23/2021
Site: EAST FORK FALSE BOTTOM CREEK, EFB-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**445**	
Baetis tricaudatus cx.	365	11.2
Diphetor hageni	75	2.3
Leptophlebiidae	5	0.2
PLECOPTERA	**1,080**	
Amphinemura banksi	5	0.2
Capniidae	5	0.2
Hesperoperla pacifica	25	0.8
Skwala americana	45	1.4
Sweltsa sp.	85	2.6
Zapada cinctipes	915	28.1
MEGALOPTERA	**5**	
Sialis sp.	5	0.2
COLEOPTERA	**120**	
Helichus striatus	5	0.2
Heterlimnius corpulentus	70	2.2
Narpus concolor	25	0.8
Optioservus divergens	5	0.2
Zaitzevia parvula	15	0.5
TRICHOPTERA	**80**	
Dolophilodes distinctus	15	0.5
Glossosoma ventrale	5	0.2
Hydropsychidae	5	0.2
Lepidostoma sp.	20	0.6
Rhyacophila brunnea gr.	35	1.1
DIPTERA	**1,365**	
Bilyjomyia algens	60	1.8
Brillia sp.	20	0.6
Ceratopogoninae	90	2.8
Conchapelopia/Thienemannimyia gr.	20	0.6
Dicranota sp.	80	2.5
Dixa sp.	25	0.8
Meringodixa chalonensis	5	0.2
Micropsectra sp.	640	19.7
Oreogeton sp.	10	0.3
Orthocladius/Cricotopus gr.	20	0.6

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/23/2021
Site: EAST FORK FALSE BOTTOM CREEK, EFB-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
DIPTERA (cont.)		
Pagastia sp.	185	5.7
Polypedilum sp.	40	1.2
Radotanypus sp.	40	1.2
Simulium sp.	35	1.1
Tabanidae	5	0.2
Tipula sp.	5	0.2
Trichoclinocera sp.	5	0.2
Tvetenia sp.	80	2.5
HYDRACARINA	**10**	
Hygrobates sp.	5	0.2
Protzia sp.	5	0.2
ANNELIDA		
OLIGOCHAETA	**55**	
Eiseniella tetraedra	55	1.7
MOLLUSCA		
PELECYPODA	**95**	
Pisidium sp.	95	2.9
TOTAL (#/sample)	3,255	
NUMBER OF TAXA	42	
SHANNON-WEAVER (H')	3.73	
TOTAL EPT TAXA	14	
EPT INDEX (% of Total Taxa)	33	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	14	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/23/2021
Site: WEST FORK FALSE BOTTOM CREEK, WFB-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
PLECOPTERA	**6**	
Leuctridae	3	0.3
Sweltsa sp.	3	0.3
COLEOPTERA	**17**	
Heterlimnius corpulentus	3	0.3
Hydroporinae	3	0.3
Narpus concolor	11	1.1
TRICHOPTERA	**29**	
Lepidostomatidae	3	0.3
Limnephilus sp.	6	0.6
Polycentropus sp.	20	2.0
DIPTERA	**926**	
Ceratopogoninae	20	2.0
Conchapelopia/Thienemannimyia gr.	209	21.2
Corynoneura sp.	209	21.2
Heterotrissocladius sp.	63	6.4
Limnophyes sp.	14	1.4
Meringodixa chalonensis	9	0.9
Oreogeton sp.	3	0.3
Orthocladius/Cricotopus gr.	14	1.4
Pagastia sp.	46	4.7
Parametriocnemus sp.	14	1.4
Polypedilum sp.	140	14.2
Psectrocladius sp.	171	17.4
Thienemanniella sp.	14	1.4
ANNELIDA		
OLIGOCHAETA	**6**	
Eiseniella tetraedra	3	0.3
Unid. Immature Tubificidae w/ Capilliform Chaetae	3	0.3
TOTAL (#/sample)	984	
NUMBER OF TAXA	23	
SHANNON-WEAVER (H')	3.21	
TOTAL EPT TAXA	5	
EPT INDEX (% of Total Taxa)	22	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	0	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/25/2021
Site: FANTAIL CREEK, FC-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**66**	
Baetis tricaudatus cx.	63	6.0
Diphetor hageni	3	0.3
PLECOPTERA	**115**	
Amphinemura banksi	50	4.7
Sweltsa sp.	10	0.9
Zapada cinctipes	55	5.2
COLEOPTERA	**151**	
Agabus cx.	5	0.5
Enochrus sp.	3	0.3
Optioservus divergens	128	12.2
Paracymus sp.	10	0.9
Sanfilippodytes vilis	5	0.5
TRICHOPTERA	**57**	
Glossosoma ventrale	5	0.5
Hesperophylax sp.	10	0.9
Lepidostoma sp.	23	2.2
Limnephilus sp.	8	0.8
Oligophlebodes minutus	3	0.3
Rhyacophila brunnea gr.	8	0.8
DIPTERA	**479**	
Brillia sp.	8	0.8
Ceratopogoninae	23	2.2
Dicranota sp.	13	1.2
Dixa sp.	13	1.2
Empididae	3	0.3
Heleniella sp.	13	1.2
Hexatoma sp.	15	1.4
Limnophila sp.	10	0.9
Meringodixa chalonensis	18	1.7
Micropsectra sp.	63	6.0
Ormosia sp.	3	0.3
Pagastia sp.	25	2.4
Polypedilum sp.	20	1.9
Prodiamesa sp.	88	8.4
Psilometriocnemus sp.	8	0.8
Radotanypus sp.	125	11.9
Simulium sp.	13	1.2
Tipula sp.	18	1.7
HYDRACARINA	**3**	
Sperchon sp.	3	0.3

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/25/2021
Site: FANTAIL CREEK, FC-1-BIO

TAXA		REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
TURBELLARIA		**160**	
	Polycelis coronata	160	15.2
ANNELIDA			
OLIGOCHAETA		**19**	
	Eiseniella tetraedra	3	0.3
	Limnodrilus sp.	3	0.3
	Unid. Immature Tubificidae w/o Capilliform Chaetae	13	1.2
MOLLUSCA			
PELECYPODA		**3**	
	Pisidium sp.	3	0.3
TOTAL (#/sample)		1,053	
NUMBER OF TAXA		40	
SHANNON-WEAVER (H')		4.32	
TOTAL EPT TAXA		11	
EPT INDEX (% of Total Taxa)		28	
EPHEMEROPTERA ABUNDANCE (% of Total Number)		6	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: NEVADA GULCH, NG-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
COLLEMBOLA	**4**	
Unid. Collembola	4	0.2
EPHEMEROPTERA	**16**	
Baetis tricaudatus cx.	4	0.2
Siphlonurus sp.	12	0.7
PLECOPTERA	**44**	
Amphinemura banksi	12	0.7
Hesperoperla pacifica	4	0.2
Sweltsa sp.	24	1.5
Zapada cinctipes	4	0.2
HEMIPTERA	**4**	
Aquarius sp.	4	0.2
MEGALOPTERA	**4**	
Sialis sp.	4	0.2
COLEOPTERA	**1,040**	
Agabus cx.	4	0.2
Boreonectes striatellus	4	0.2
Cleptelmis addenda	4	0.2
Narpus concolor	8	0.5
Optioservus divergens	724	44.7
Optioservus seriatus	284	17.5
Zaitzevia parvula	12	0.7
TRICHOPTERA	**17**	
Glossosoma ventrale	16	1.0
Rhyacophila brunnea gr.	1	0.1
DIPTERA	**412**	
Chironomus sp.	16	1.0
Conchapelopia/Thienemannimyia gr.	8	0.5
Cricotopus sp.	8	0.5
Dixidae	4	0.2
Eukiefferiella sp.	8	0.5
Orthocladius/Cricotopus gr.	16	1.0
Pedicia sp.	4	0.2
Polypedilum sp.	40	2.5
Prodiamesa sp.	44	2.7
Radotanypus sp.	156	9.6
Rheocricotopus sp.	8	0.5

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: NEVADA GULCH, NG-2-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
DIPTERA (cont.)		
Simulium sp.	4	0.2
Thienemanniella sp.	24	1.5
Tipula sp.	12	0.7
Tvetenia sp.	60	3.7
TURBELLARIA	**4**	
Polycelis coronata	4	0.2
ANNELIDA		
OLIGOCHAETA	**76**	
Eiseniella tetraedra	64	3.9
Limnodrilus sp.	4	0.2
Unid. Immature Tubificidae w/o Capilliform Chaetae	8	0.5
TOTAL (#/sample)	1,621	
NUMBER OF TAXA	37	
SHANNON-WEAVER (H')	3.04	
TOTAL EPT TAXA	8	
EPT INDEX (% of Total Taxa)	22	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	1	

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: STEWART GULCH, SG-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
INSECTA		
EPHEMEROPTERA	**1,295**	
Baetis tricaudatus cx.	1,260	19.3
Diphetor hageni	35	0.5
PLECOPTERA	**760**	
Hesperoperla pacifica	280	4.3
Sweltsa sp.	180	2.8
Zapada cinctipes	300	4.6
COLEOPTERA	**1,900**	
Heterlimnius corpulentus	1,845	28.3
Optioservus divergens	55	0.8
TRICHOPTERA	**1,910**	
Glossosoma ventrale	55	0.8
Lepidostoma sp.	155	2.4
Limnephilidae	5	0.1
Micrasema bactro	1,560	23.9
Oligophlebodes minutus	100	1.5
Rhyacophila brunnea gr.	25	0.4
Rhyacophila sp.	10	0.2
DIPTERA	**600**	
Brillia sp.	45	0.7
Chironomus sp.	10	0.2
Corynoneura sp.	25	0.4
Diamesa sp.	15	0.2
Dicranomyia sp.	10	0.2
Dicranota sp.	30	0.5
Dixa sp.	35	0.5
Eukiefferiella sp.	30	0.5
Limnophyes sp.	10	0.2
Meringodixa chalonensis	10	0.2
Metriocnemus sp.	55	0.8
Micropsectra sp.	100	1.5
Oreogeton sp.	30	0.5
Orthocladius/Cricotopus gr.	10	0.2
Pagastia sp.	75	1.1
Rheocricotopus sp.	15	0.2
Simulium sp.	40	0.6
Thienemanniella sp.	30	0.5
Tvetenia sp.	25	0.4

DATA: MACROINVERTEBRATE DENSITY
Client: WHARF
Sampled: 8/24/2021
Site: STEWART GULCH, SG-1-BIO

TAXA	REACH WIDE COMPOSITE (#/SAMPLE)	% OF TOTAL
HYDRACARINA	**30**	
Lebertia sp.	5	0.1
Protzia sp.	20	0.3
Thyopsis sp.	5	0.1
TURBELLARIA	**5**	
Polycelis coronata	5	0.1
ANNELIDA		
OLIGOCHAETA	**30**	
Eiseniella tetraedra	30	0.5
TOTAL (#/sample)	6,530	
NUMBER OF TAXA	38	
SHANNON-WEAVER (H')	3.20	
TOTAL EPT TAXA	12	
EPT INDEX (% of Total Taxa)	32	
EPHEMEROPTERA ABUNDANCE (% of Total Number)	20	

This page intentionally left blank

Appendix C Historical Benthic Macroinvertebrate Data

This page intentionally left blank

Table C-1: Select benthic macroinvertebrate population metrics for Site LB-4-BIO, South Dakota, 1995 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. Data is shown for only years in which the site was sampled. NC = not calculated.

Site LB-4-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2005
Total Density	572	1,155	877	1,037	1,256	1,323	1,000	1,415	2,760	994
Number of Taxa	15	32	31	24	29	32	32	33	36	34
Shannon-Weaver Diversity (H’)	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC
Number of EPT Taxa	NC	NC	NC	14	16	16	21	18	NC	16

Table C-2: Benthic macroinvertebrate population metrics for Site LB-4-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years.

Site LB-4-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	3,210	891	2,210	2,680	1,424	3,745	729	4,051	3,390	3,722	7,760	4,788	4,245	6,640	3,185	5,082
Number of Taxa	45	30	46	34	36	34	24	42	38	40	37	44	39	55	44	55
Number of EPT Taxa	17	11	16	13	12	11	12	13	14	15	15	14	14	16	16	18
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H')	4.40	3.88	3.67	3.91	4.09	3.65	3.62	4.06	3.61	4.03	3.67	4.19	4.07	4.01	4.04	4.51
Percent Sensitive EPT Taxa	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.2	0.3	0.3	0.3	0.3	0.3	0.3	0.3	0.2
EPT Index (%)	0.4	0.4	0.3	0.4	0.3	0.3	0.5	0.3	0.4	0.4	0.4	0.3	0.4	0.3	0.4	0.3
Percent *Baetis sp.*	0.5	0.2	0.9	0.8	0.8	0.8	0.9	0.7	0.9	0.7	0.8	0.5	0.5	0.5	0.6	0.6
Number of non-*Baetis* Ephemeroptera	400	107	135	180	100	301	23	480	150	380	220	590	960	760	360	635
Percent Chironomidae	13.7	13.8	12.0	14.9	26.7	14.2	17.4	16.8	11.8	12.6	56.4	33.3	29.2	61.1	15.2	31.6
Number of Plecoptera Taxa	5	4	5	4	3	4	3	5	5	4	4	4	4	6	5	4
Percent Abundance of Oligochaetes and Hirudinea	2.2	0.0	0.9	1.5	2.0	2.5	1.8	3.0	3.8	1.6	1.0	1.3	0.5	0.5	1.7	0.9
Percent Dominant Taxon	21.5	18.7	34.2	25.4	19.7	28.6	26.5	23.9	32.4	22.3	32.9	16.9	22.1	22.6	20.9	15.5
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	3.41	2.50	3.84	3.55	4.09	3.85	3.15	3.91	3.86	3.77	5.00	4.24	4.24	5.65	3.39	4.08
Percent Tolerant Taxa	20.0	16.7	23.9	26.5	22.2	20.6	16.7	19.0	18.4	22.5	21.6	20.5	20.5	25.5	27.3	25.5
Percent Intolerant Taxa	46.7	56.7	54.3	44.1	50.0	47.1	54.2	47.6	42.1	45.0	48.6	47.7	46.2	49.1	43.2	50.9
Number of Intolerant Taxa	21	17	25	15	18	16	13	20	16	18	18	21	18	27	19	28
TROPHIC HABIT METRICS																
Number of Predator Taxa	10	6	10	6	7	11	5	11	10	9	9	10	12	17	11	16
Percent Collector-Gatherers	37.6	26.9	52.4	37.9	49.9	54.3	43.6	56.3	60.5	47.6	43.0	40.2	58.5	45.3	43.6	51.9
Number of Shredder Taxa	8	6	9	6	4	4	5	5	6	9	6	7	7	9	8	8
LIFE HISTORY METRICS																
Number of Univoltine Taxa	22	15	19	13	16	17	8	21	15	16	17	21	19	26	20	27
Number of Merovoltine Taxa	3	0	1	1	3	0	1	3	1	3	2	2	1	6	3	4
Number of Semivoltine Taxa	1	1	1	1	0	1	0	1	1	1	0	1	1	1	1	1
Percent Semivoltine taxa	2.2	3.3	2.2	2.9	0.0	2.9	0.0	2.4	2.6	2.5	0.0	2.3	2.6	1.8	2.3	1.8

Table C-3: Benthic macroinvertebrate population metrics for Site RC-1-BIO, South Dakota, 2017 - 2021. Semiquantitative sampling methods (Kick) were performed in these years.

Site RC-1-BIO	2017	2018	2019	2020	2021
RICHNESS METRICS					
Total Density	3,001	3,821	3,855	3,410	4,220
Number of Taxa	29	38	42	45	46
Number of EPT Taxa	7	12	14	15	15
COMPOSITION METRICS					
Shannon-Weaver Diversity Index (H’)	3.59	3.61	3.78	4.18	4.52
Percent Sensitive EPT Taxa	0.2	0.2	0.3	0.3	0.3
EPT Index (%)	0.2	0.3	0.3	0.3	0.3
Percent *Baetis sp.*	0.0	0.3	0.7	0.3	0.2
Number of non-*Baetis* Ephemeroptera	180	1,380	395	590	410
Percent Chironomidae	64.3	11.6	9.5	16.6	44.5
Number of Plecoptera Taxa	4	3	5	4	6
Percent Abundance of Oligochaetes and Hirudinea	0.3	1.2	1.6	0.6	2.8
Percent Dominant Taxon	34.0	32.6	28.7	20.5	16.0
TOLERANCE METRICS					
Hilsenhoff Biotic Index (HBI)	5.64	4.24	3.79	3.65	4.61
Percent Tolerant Taxa	31.0	15.8	23.8	20.0	30.4
Percent Intolerant Taxa	51.7	44.7	40.5	48.9	47.8
Number of Intolerant Taxa	15	17	17	22	22
TROPHIC HABIT METRICS					
Number of Predator Taxa	10	11	13	10	12
Percent Collector-Gatherers	60.6	63.2	45.3	44.1	56.0
Number of Shredder Taxa	3	4	6	8	7
LIFE HISTORY METRICS					
Number of Univoltine Taxa	16	17	19	23	22
Number of Merovoltine Taxa	0	0	1	1	2
Number of Semivoltine Taxa	0	0	1	0	1
Percent Semivoltine taxa	0.0	0.0	2.4	0.0	2.2

Table C-4: Select benthic macroinvertebrate population metrics for Site AC-1-BIO, South Dakota, 1992 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. Data is shown for only years in which the site was sampled.

Site AC-1-BIO	1992[a]	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Total Density	3,932	4,508	8,696	10,397	4,909	1,841	819	4,775	39,612	38,461	7,641	1,977
Number of Taxa	33	28	27	24	26	19	29	34	21	27	29	29
Shannon-Weaver Diversity (H’)	3.71	3.2	3.16	3.11	3.17	3.09	3.84	3.18	2.49	3.18	3.03	2.93
Number of EPT Taxa	12	12	9	7	10	7	9	10	3	3	4	9

[a] Data from Site 1-A of 1992 Aquatic Biological Assessment, just downstream of Site AC-1 (C&A 1993).

Table C-5: Benthic macroinvertebrate population metrics for Site AC-1-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. N/A = no reference site established.

Site AC-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	2,026	4,243	3,311	1,960	3,216	2,429	1,247	2,093	6,820	9,473	3,881	14,054	5,150	2,360	4,040	6,160
Number of Taxa	24	11	40	29	25	29	28	31	30	33	37	44	41	39	44	43
Number of EPT Taxa	6	1	12	6	6	7	11	8	11	10	13	16	14	11	13	12
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H’)	2.52	1.75	3.99	4.08	3.36	3.85	2.39	3.87	3.83	3.82	4.31	3.80	3.83	3.97	4.17	4.13
Percent Sensitive EPT Taxa	0.2	0.1	0.3	0.1	0.1	0.2	0.4	0.2	0.3	0.2	0.2	0.2	0.2	0.2	0.2	0.2
EPT Index (%)	0.3	0.1	0.3	0.2	0.2	0.2	0.4	0.3	0.4	0.3	0.4	0.4	0.3	0.3	0.3	0.3
Percent *Baetis sp.*	1.0	0.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	0.5	0.4	0.8	1.0	0.7	0.4
Number of non-*Baetis* Ephemeroptera	0	0	1	0	0	0	0	0	0	0	210	571	350	5	115	190
Percent Chironomidae	4.9	99.0	23.6	27.6	27.1	38.2	6.7	58.5	59.5	60.9	41.0	35.3	9.2	12.9	17.3	20.9
Number of Plecoptera Taxa	3	0	3	3	2	4	4	2	4	3	3	5	4	3	3	3
Percent Abundance of Oligochaetes & Hirudinea	0.2	0.5	0.7	34.2	1.6	3.0	0.0	0.0	0.1	0.3	2.1	2.6	3.0	0.6	2.0	5.1
Percent Dominant Taxon	54.3	60.3	22.0	15.3	28.3	25.5	58.0	23.7	20.4	19.8	17.0	18.4	23.0	21.6	23.1	16.1
Number of Common Taxa	3	1	21	10	7	16	11	11	10	N/A	N/A	24	17	20	21	25
Community Loss Index	1.2	3.4	0.6	0.9	0.7	0.7	0.8	0.5	0.7	N/A	N/A	0.3	0.4	0.5	0.6	0.5
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	3.22	6.35	3.69	6.54	4.33	4.21	2.68	4.33	5.22	5.20	4.23	4.94	3.33	3.50	3.07	3.62
Percent Tolerant Taxa	25.0	36.4	15.0	31.0	24.0	24.1	14.3	12.9	23.3	18.2	24.3	22.7	24.4	23.1	31.8	27.9
Percent Intolerant Taxa	41.7	18.2	50.0	37.9	36.0	34.5	53.6	51.6	43.3	48.5	45.9	43.2	46.3	46.2	45.5	46.5
Number of Intolerant Taxa	10	2	20	11	9	10	15	16	13	16	17	19	19	18	20	20
TROPHIC HABIT METRICS																
Number of Predator Taxa	5	2	9	5	3	7	7	8	7	8	8	11	10	9	12	11
Percent Collector-Gatherers	25.9	36.7	56.8	58.6	59.0	46.8	23.3	46.8	53.8	44.4	38.4	23.1	44.1	44.5	31.6	32.8
Number of Shredder Taxa	4	3	6	6	4	6	7	8	8	7	9	11	7	8	7	8
LIFE HISTORY METRICS																
Number of Univoltine Taxa	11	4	23	12	12	14	14	14	15	20	14	22	22	21	19	19
Number of Merovoltine Taxa	0	1	1	2	1	1	0	3	1	1	3	2	1	2	4	1
Number of Semivoltine Taxa	0	0	0	1	0	0	0	0	0	0	0	1	1	0	1	0
Percent Semivoltine taxa	0.0	0.0	0.0	3.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	2.3	2.4	0.0	2.3	0.0

Table C-6: Select benthic macroinvertebrate population metrics for Site AC-2-BIO, South Dakota, 1992 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. Data is shown for only years in which the site was sampled.

Site AC-2-BIO	1992	1995 [a]	1996 [a]	1997 [a]	1998 [a]	1999 [a]	2000	2001	2002	2003	2004	2005
Total Density	10,840	1,333	4,586	3,597	992	5,215	2,119	10,097	4,607	2,046	1,025	2,708
Number of Taxa	37	30	29	32	17	31	34	34	42	48	30	35
Shannon-Weaver Diversity (H')	2.82	3.8	2.11	3.13	2.32	3.2	3.89	2.65	3.94	4.31	3.74	3.67
Number of EPT Taxa	17	9	14	16	6	9	13	8	12	11	10	10

[a] Data from Site AC-2 of 1995-1999 Aquatic Biological Monitoring, downstream of Site AC-2-BIO (CEC 1996b, 1997a, 1998a, 1999a, 2000).

Table C-7: Benthic macroinvertebrate population metrics for Site AC-2-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years.

Site AC-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	950	493	4,937	1,580	896	2,491	1,632	2,497	4,463	3,873	939	4,021	2,411	2,783	1,875	3,425
Number of Taxa	25	30	67	36	28	24	42	34	40	42	32	44	49	41	48	52
Number of EPT Taxa	4	8	24	11	7	8	13	12	10	9	12	16	11	9	15	18
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H')	3.54	3.84	3.30	4.05	3.84	3.49	4.28	3.44	3.61	3.91	3.81	4.21	3.32	3.08	3.88	4.56
Percent Sensitive EPT Taxa	0.1	0.2	0.3	0.2	0.2	0.3	0.2	0.2	0.2	0.1	0.3	0.2	0.2	0.1	0.3	0.3
EPT Index (%)	0.2	0.3	0.4	0.3	0.3	0.3	0.3	0.4	0.3	0.2	0.4	0.4	0.2	0.2	0.3	0.3
Percent *Baetis sp.*	1.0	1.0	0.8	0.8	1.0	1.0	0.8	0.9	1.0	1.0	0.6	0.4	0.9	1.0	0.8	0.5
Number of non-*Baetis* Ephemeroptera	0	0	442	45	0	0	70	20	10	0	7	180	100	70	125	385
Percent Chironomidae	5.8	72.6	8.1	22.2	60.7	52.7	36.8	69.9	30.7	22.7	42.2	50.2	11.0	11.3	6.4	22.2
Number of Plecoptera Taxa	3	2	7	5	3	3	3	3	3	3	3	4	3	2	3	5
Percent Abundance of Oligochaetes & Hirudinea	21.1	0.0	1.0	3.2	5.4	0.4	1.8	0.4	1.6	3.4	1.4	2.0	2.5	1.6	1.6	0.9
Percent Dominant Taxon	22.1	28.8	35.5	30.4	26.5	30.0	12.6	37.8	31.6	33.3	29.5	31.6	49.4	49.4	31.2	13.0
Number of Common Taxa	12	13	23	17	13	12	16	22	17	0	2	9	16	9	26	36
Community Loss Index	0.7	0.9	0.5	0.6	0.9	0.9	0.5	0.4	0.5	0.8	1.2	0.5	0.4	0.7	0.4	0.4
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	4.77	5.20	4.73	3.57	5.43	4.81	3.97	5.36	4.74	4.59	4.30	5.04	4.92	4.97	4.13	4.20
Percent Tolerant Taxa	40.0	3.3	22.4	19.4	25.0	8.3	23.8	26.5	32.5	26.2	21.9	15.9	34.7	31.7	27.1	21.2
Percent Intolerant Taxa	36.0	43.3	43.3	50.0	42.9	62.5	52.4	44.1	32.5	45.2	50.0	50.0	38.8	43.9	43.8	61.5
Number of Intolerant Taxa	9	13	29	18	12	15	22	15	13	19	16	22	19	18	21	32
TROPHIC HABIT METRICS																
Number of Predator Taxa	6	5	14	8	7	3	10	8	8	10	7	8	10	9	13	13
Percent Collector-Gatherers	50.0	69.8	43.9	39.6	40.3	65.8	49.6	59.1	57.6	48.0	18.0	55.5	64.7	67.4	52.3	51.5
Number of Shredder Taxa	2	6	10	7	4	6	4	7	9	10	8	10	9	8	7	8
LIFE HISTORY METRICS																
Number of Univoltine Taxa	8	19	34	15	15	11	21	15	20	19	18	25	24	19	19	26
Number of Merovoltine Taxa	1	2	2	3	1	2	3	2	2	3	1	2	2	2	5	1
Number of Semivoltine Taxa	0	0	0	1	0	1	0	0	1	0	0	1	0	0	0	1
Percent Semivoltine taxa	0.0	0.0	0.0	2.8	0.0	4.2	0.0	0.0	2.5	0.0	0.0	2.3	0.0	0.0	0.0	1.9

Table C-8: Select benthic macroinvertebrate population metrics for Site AC-3-BIO, South Dakota, 1992 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. Data is shown for only years in which the site was sampled.

Site AC-3-BIO	1992 [a]	1995 [b]	1996 [b]	1997 [b]	1998 [b]	1999 [b]	2000	2001	2002	2003	2004	2005
Total Density	10,840	1,333	4,586	3,597	992	5,215	2,119	10,097	4,607	2,046	1,025	2,708
Number of Taxa	37	30	29	32	17	31	34	34	42	48	30	35
Shannon-Weaver Diversity (H’)	2.82	3.8	2.11	3.13	2.32	3.2	3.89	2.65	3.94	4.31	3.74	3.67
Number of EPT Taxa	17	9	14	16	6	9	13	8	12	11	10	10

[a] Data from Site AC-6 of 1992 Aquatic Biological Assessment (C&A 1993).

[b] Data from Site AC-6 (CEC 1996a).

Table C-9: Benthic macroinvertebrate population metrics for Site AC-3-BIO, South Dakota, 2006 - 2019. Semiquantitative sampling methods (Kick) were performed in these years. Data is shown for only years in which the site was sampled.

Site AC-3-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019
RICHNESS METRICS														
Total Density	1,875	3,505	3,424	2,697	4,170	6,335	2,551	3,181	5,620	8,783	3,570	4,862	2,755	6,436
Number of Taxa	33	37	39	38	39	31	31	38	35	33	30	49	35	46
Number of EPT Taxa	14	14	17	14	15	11	12	13	10	8	7	15	10	12
COMPOSITION METRICS														
Shannon-Weaver Diversity Index (H’)	3.92	3.64	4.12	4.21	4.22	2.94	3.64	4.37	3.85	2.89	3.40	4.19	3.77	3.56
Percent Sensitive EPT Taxa	0.3	0.3	0.4	0.3	0.3	0.3	0.3	0.3	0.2	0.2	0.1	0.2	0.2	0.2
EPT Index (%)	0.4	0.4	0.4	0.4	0.4	0.4	0.4	0.3	0.3	0.2	0.2	0.3	0.3	0.3
Percent *Baetis sp.*	0.5	0.4	0.7	0.8	0.9	0.7	0.1	0.4	1.0	1.0	1.0	0.6	0.9	1.0
Number of non-*Baetis* Ephemeroptera	94	190	120	100	80	320	40	360	20	0	0	190	120	5
Percent Chironomidae	31.5	14.6	21.0	15.2	35.0	15.5	76.6	28.3	36.1	20.2	66.7	24.9	11.1	18.2
Number of Plecoptera Taxa	5	4	5	5	5	4	5	3	4	3	3	5	2	3
Percent Abundance of Oligochaetes & Hirudinea	1.1	0.0	1.2	0.7	1.0	0.4	0.0	0.0	0.4	0.3	2.2	1.9	4.5	1.4
Percent Dominant Taxon	23.1	34.5	21.4	16.7	18.6	51.3	25.1	23.3	18.7	47.9	24.4	24.7	31.6	34.6
Number of Common Taxa	13	16	27	22	22	19	20	20	16	17	15	13	15	20
Community Loss Index	0.5	0.7	0.4	0.4	0.4	0.5	0.5	0.4	0.6	0.5	0.8	0.4	0.6	0.6
TOLERANCE METRICS														
Hilsenhoff Biotic Index (HBI)	4.18	2.78	3.38	3.04	4.12	3.17	5.73	3.31	4.58	4.72	5.49	4.02	4.60	5.02
Percent Tolerant Taxa	21.2	13.5	20.5	15.8	12.8	22.6	16.1	18.4	17.1	21.2	30.0	20.4	31.4	28.3
Percent Intolerant Taxa	57.6	62.2	51.3	47.4	53.8	51.6	61.3	55.3	45.7	39.4	36.7	53.1	42.9	45.7
Number of Intolerant Taxa	19	23	20	18	21	16	19	21	16	13	11	26	15	21
TROPHIC HABIT METRICS														
Number of Predator Taxa	10	8	8	9	12	7	10	8	7	9	8	14	7	10
Percent Collector-Gatherers	47.8	18.5	28.5	30.1	49.2	33.3	37.2	37.4	41.8	65.9	25.8	35.6	49.2	55.1
Number of Shredder Taxa	5	6	6	6	8	4	7	8	7	8	6	9	7	8
LIFE HISTORY METRICS														
Number of Univoltine Taxa	18	21	20	17	19	13	15	18	17	16	13	26	17	21
Number of Merovoltine Taxa	2	0	0	2	3	0	3	3	2	2	1	2	1	4
Number of Semivoltine Taxa	1	0	1	1	1	0	1	1	1	0	0	1	0	1
Percent Semivoltine taxa	3.0	0.0	2.6	2.6	2.6	0.0	3.2	2.6	2.9	0.0	0.0	2.0	0.0	2.2

Table C-10: Benthic macroinvertebrate population metrics for Site RV-2-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. N/A = no reference site established.

Site RV-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	558	714	3,094	944	1,648	652	5,671	2,640	3,060	4,792	2,100	3,292	2,605	1,500	1,336	4,330
Number of Taxa	25	38	62	26	33	37	23	27	28	33	36	39	38	39	26	36
Number of EPT Taxa	6	10	16	8	10	7	9	7	9	12	10	12	11	11	10	9
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H’)	3.48	4.11	4.44	3.75	3.97	3.91	2.91	3.73	3.10	3.33	3.86	4.08	4.09	3.91	3.04	3.21
Percent Sensitive EPT Taxa	0.2	0.1	0.2	0.2	0.2	0.1	0.3	0.2	0.2	0.3	0.2	0.2	0.2	0.2	0.3	0.2
EPT Index (%)	0.2	0.3	0.3	0.3	0.3	0.2	0.4	0.3	0.3	0.4	0.3	0.3	0.3	0.3	0.4	0.3
Percent *Baetis sp.*	1.0	0.9	0.7	0.8	0.5	0.3	0.0	1.0	0.8	1.0	0.3	0.5	0.3	0.9	0.5	0.1
Number of non-*Baetis* Ephemeroptera	0	2	315	24	33	21	80	0	80	10	73	11	275	25	173	805
Percent Chironomidae	5.9	26.6	7.6	2.5	25.4	23.0	21.5	18.9	11.1	10.2	6.4	14.9	9.6	7.3	1.9	11.7
Number of Plecoptera Taxa	3	3	6	3	4	2	3	1	3	4	3	4	5	4	4	3
Percent Abundance of Oligochaetes & Hirudinea	20.3	8.8	9.2	4.7	2.7	25.3	0.0	0.4	0.3	0.4	0.0	1.8	20.3	9.0	0.2	0.2
Percent Dominant Taxon	29.2	20.2	15.8	21.2	23.8	25.3	35.4	16.3	33.0	34.4	24.1	24.9	25.0	28.0	32.4	37.2
Number of Common Taxa	6	15	13	10	11	18	10	8	12	N/A	N/A	29	19	26	18	21
Community Loss Index	1.0	0.6	0.6	1.0	0.4	0.5	1.0	0.6	0.7	N/A	N/A	0.4	0.5	0.5	1.0	0.7
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	4.72	4.35	4.50	4.05	4.07	5.47	3.78	4.72	2.77	3.70	4.15	3.44	4.95	4.14	2.85	3.40
Percent Tolerant Taxa	28.0	21.1	19.4	19.2	24.2	18.9	21.7	18.5	17.9	27.3	36.1	25.6	28.9	30.8	15.4	30.6
Percent Intolerant Taxa	36.0	34.2	37.1	42.3	45.5	40.5	52.2	48.1	42.9	51.5	38.9	43.6	47.4	38.5	61.5	50.0
Number of Intolerant Taxa	9	13	23	11	15	15	12	13	12	17	14	17	18	15	16	18
TROPHIC HABIT METRICS																
Number of Predator Taxa	8	6	10	4	5	9	9	8	7	5	14	11	8	11	5	10
Percent Collector-Gatherers	31.9	48.2	51.1	22.5	27.2	51.5	56.6	37.5	22.5	49.5	38.1	20.1	46.3	37.7	41.1	38.7
Number of Shredder Taxa	5	8	11	5	6	8	6	4	5	7	8	8	8	9	6	7
LIFE HISTORY METRICS																
Number of Univoltine Taxa	14	19	28	12	14	20	11	15	15	12	16	21	18	15	10	16
Number of Merovoltine Taxa	0	1	2	1	2	2	1	1	0	1	1	1	1	1	2	2
Number of Semivoltine Taxa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent Semivoltine taxa	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table C-11: Benthic macroinvertebrate population metrics for Site LC-1-BIO, South Dakota, 2010 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. N/A = no reference site established.

Site LC-1-BIO	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS												
Total Density	1,087	1,317	1,521	2,074	675	1,755	3,990	5,103	2,290	566	2,200	2,145
Number of Taxa	26	37	23	38	22	24	22	24	29	18	27	28
Number of EPT Taxa	11	15	5	7	6	5	5	4	5	1	8	5
COMPOSITION METRICS												
Shannon-Weaver Diversity Index (H')	3.87	3.73	3.51	4.17	3.09	3.29	2.03	3.27	2.81	2.60	2.81	3.34
Percent Sensitive EPT Taxa	0.3	0.2	0.1	0.1	0.1	0.1	0.1	0.0	0.1	0.0	0.1	0.1
EPT Index (%)	0.4	0.4	0.2	0.2	0.3	0.2	0.2	0.2	0.2	0.1	0.3	0.2
Percent *Baetis sp.*	0.9	0.2	0.0	1.0	0.0	1.0	1.0	0.0	1.0	0.0	0.9	0.0
Number of non-*Baetis* Ephemeroptera	20	471	40	0	0	0	0	51	20	0	165	20
Percent Chironomidae	30.1	17.8	64.4	75.0	77.6	20.8	84.5	69.8	13.3	24.9	19.3	68.5
Number of Plecoptera Taxa	3	3	2	2	1	1	1	0	1	0	1	0
Percent Abundance of Oligochaetes & Hirudinea	1.3	0.6	1.0	0.2	3.0	4.0	2.0	2.0	0.9	0.0	0.7	1.9
Percent Dominant Taxon	21.7	34.7	23.0	16.9	32.3	36.5	67.7	23.3	50.0	38.5	52.0	32.6
Number of Common Taxa	11	21	8	16	10	N/A	N/A	24	23	24	12	17
Community Loss Index	0.5	0.4	1.0	0.2	1.0	N/A	N/A	0.8	0.8	1.8	1.2	1.0
TOLERANCE METRICS												
Hilsenhoff Biotic Index (HBI)	3.72	4.07	4.89	5.39	5.87	4.97	5.69	5.86	4.83	4.43	4.95	5.62
Percent Tolerant Taxa	15.4	18.9	30.4	28.9	36.4	25.0	31.8	37.5	27.6	27.8	29.6	25.0
Percent Intolerant Taxa	46.2	43.2	26.1	39.5	31.8	37.5	45.5	29.2	31.0	38.9	48.1	42.9
Number of Intolerant Taxa	12	16	6	15	7	9	10	7	9	7	13	12
TROPHIC HABIT METRICS												
Number of Predator Taxa	2	9	9	14	5	6	5	7	5	4	5	7
Percent Collector-Gatherers	54.1	64.3	40.8	40.5	59.9	66.1	17.3	43.9	72.5	53.4	69.1	36.8
Number of Shredder Taxa	7	6	3	6	7	4	4	3	5	2	5	5
LIFE HISTORY METRICS												
Number of Univoltine Taxa	13	21	13	19	14	8	10	12	11	9	16	17
Number of Merovoltine Taxa	1	1	1	3	2	2	3	3	1	2	1	2
Number of Semivoltine Taxa	0	1	0	0	0	0	0	1	0	0	1	0
Percent Semivoltine taxa	0.0	2.7	0.0	0.0	0.0	0.0	0.0	4.2	0.0	0.0	3.7	0.0

Table C-12: Select benthic macroinvertebrate population metrics for Site DC-2-BIO, South Dakota, 2000 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years.

Site DC-2-BIO	2000	2001	2002	2003	2004	2005
Total Density	1,320	3,292	5,160	1,877	7,865	7,951
Number of Taxa	33	27	43	31	39	41
Shannon-Weaver Diversity (H')	3.5	2.83	3.68	3.41	3.81	3.91
Number of EPT Taxa	13	5	13	12	16	14

Table C-13: Benthic macroinvertebrate population metrics for Site DC-2-BIO, South Dakota, 2010 - 2021. Semiquantitative sampling methods (Kick) were performed in these years.

Site DC-2-BIO	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS												
Total Density	2,340	5,476	1,540	1,950	6,472	4,313	1,386	4,353	2,116	2,956	4,215	2,225
Number of Taxa	30	29	30	35	34	30	27	44	40	45	33	39
Number of EPT Taxa	13	10	10	15	12	11	8	14	11	11	11	12
COMPOSITION METRICS												
Shannon-Weaver Diversity Index (H’)	3.23	3.36	3.83	4.30	2.70	3.38	3.78	4.46	4.35	4.52	3.27	3.61
Percent Sensitive EPT Taxa	0.4	0.2	0.2	0.3	0.2	0.3	0.3	0.2	0.2	0.2	0.3	0.3
EPT Index (%)	0.4	0.3	0.3	0.4	0.4	0.4	0.3	0.3	0.3	0.2	0.3	0.3
Percent *Baetis sp.*	0.8	0.7	0.5	0.8	0.8	0.7	0.0	0.1	0.3	0.6	0.2	0.2
Number of non-*Baetis* Ephemeroptera	40	110	35	105	100	90	0	170	355	155	135	124
Percent Chironomidae	31.1	58.7	79.2	45.4	36.9	36.4	77.2	67.8	37.3	49.6	79.4	71.7
Number of Plecoptera Taxa	5	3	3	4	3	3	3	3	3	3	3	4
Percent Abundance of Oligochaetes & Hirudinea	1.1	0.2	0.0	0.0	1.1	0.0	0.0	0.5	0.2	3.0	0.1	0.7
Percent Dominant Taxon	43.0	29.4	19.2	12.8	47.9	41.5	22.7	18.8	17.5	16.1	39.4	36.3
Number of Common Taxa	18	14	17	17	19	17	19	23	20	21	19	23
Community Loss Index	0.7	0.7	0.6	0.5	0.5	0.5	0.8	0.6	0.5	0.7	0.8	0.8
TOLERANCE METRICS												
Hilsenhoff Biotic Index (HBI)	3.20	4.59	4.61	4.53	3.74	3.43	4.57	4.66	3.64	4.85	4.94	5.39
Percent Tolerant Taxa	13.3	20.7	23.3	11.4	14.7	16.7	7.4	18.2	22.5	31.1	15.2	25.6
Percent Intolerant Taxa	56.7	37.9	43.3	51.4	44.1	46.7	55.6	43.2	40.0	35.6	54.5	41.0
Number of Intolerant Taxa	17	11	13	18	15	14	15	19	16	16	18	16
TROPHIC HABIT METRICS												
Number of Predator Taxa	9	11	11	11	10	11	11	12	11	16	8	10
Percent Collector-Gatherers	35.1	52.0	46.4	57.4	39.4	31.8	35.7	40.7	52.9	47.5	19.7	37.2
Number of Shredder Taxa	4	3	4	5	6	7	6	8	7	7	6	7
LIFE HISTORY METRICS												
Number of Univoltine Taxa	16	16	16	17	20	16	20	24	18	23	19	20
Number of Merovoltine Taxa	1	0	0	3	1	1	1	3	3	2	1	2
Number of Semivoltine Taxa	0	1	0	0	0	0	0	0	0	0	0	0
Percent Semivoltine taxa	0.0	3.4	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table C-14: Select benthic macroinvertebrate population metrics for Site EFB-1-BIO, South Dakota, 1995 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years.

Site EFB-1-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Total Density	3,247	619	1,451	1,313	511	3,711	2,313	3,927	2,498	2,901	7,726
Number of Taxa	24	20	27	22	23	30	36	44	39	48	33
Shannon-Weaver Diversity (H’)	2.81	3.26	3.57	3.1	3.57	3.48	3.35	3.8	3.7	4.02	3.34
Number of EPT Taxa	8	9	12	6	11	13	10	13	13	15	12

Table C-15: Benthic macroinvertebrate population metrics for Site EFB-1-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. Data is shown for only years in which the site was sampled.

Site EFB-1-BIO	2006	2009	2010	2018	2019	2020	2021
RICHNESS METRICS							
Total Density	1,255	797	1,443	9,305	3,990	1,975	3,255
Number of Taxa	32	29	28	41	40	31	42
Number of EPT Taxa	11	10	12	11	11	11	14
COMPOSITION METRICS							
Shannon-Weaver Diversity Index (H')	3.66	3.87	3.38	2.92	3.20	3.40	3.73
Percent Sensitive EPT Taxa	0.3	0.2	0.4	0.2	0.2	0.3	0.3
EPT Index (%)	0.3	0.3	0.4	0.3	0.3	0.4	0.3
Percent *Baetis sp.*	0.9	0.9	0.9	0.9	1.0	0.8	0.8
Number of non-*Baetis* Ephemeroptera	45	8	27	40	35	140	80
Percent Chironomidae	12.0	33.2	16.6	27.7	13.7	4.6	33.9
Number of Plecoptera Taxa	3	3	4	4	4	4	6
Percent Abundance of Oligochaetes & Hirudinea	4.4	29.0	1.9	0.4	1.4	4.3	1.7
Percent Dominant Taxon	31.9	27.1	33.7	45.4	39.1	32.4	28.1
Number of Common Taxa	13	16	16	17	29	15	27
Community Loss Index	0.5	0.8	0.8	0.5	0.7	0.9	0.7
TOLERANCE METRICS							
Hilsenhoff Biotic Index (HBI)	3.69	4.63	3.58	3.54	3.50	3.27	4.08
Percent Tolerant Taxa	18.8	27.6	17.9	22.0	25.0	16.1	16.7
Percent Intolerant Taxa	53.1	34.5	53.6	43.9	40.0	51.6	52.4
Number of Intolerant Taxa	17	10	15	18	16	16	22
TROPHIC HABIT METRICS							
Number of Predator Taxa	7	8	7	13	12	10	14
Percent Collector-Gatherers	53.7	60.9	47.3	31.0	40.4	44.8	47.6
Number of Shredder Taxa	4	5	4	4	5	2	8
LIFE HISTORY METRICS							
Number of Univoltine Taxa	18	11	12	21	18	12	19
Number of Merovoltine Taxa	0	2	1	0	1	3	2
Number of Semivoltine Taxa	0	0	1	1	1	1	1
Percent Semivoltine taxa	0.0	0.0	3.6	2.4	2.5	3.2	2.4

Table C-16: Benthic macroinvertebrate population metrics for Site WFB-1-BIO, South Dakota, 2007 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. Data is shown for only years in which the site was sampled.

Site WFB-1-BIO	2007	2008	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS													
Total Density	459	1,238	357	926	563	751	2,131	1,068	2,935	3,365	2,410	284	984
Number of Taxa	31	37	23	27	24	25	22	21	20	30	29	27	23
Number of EPT Taxa	10	11	5	7	3	8	5	8	3	4	6	6	5
COMPOSITION METRICS													
Shannon-Weaver Diversity Index (H’)	4.18	4.16	3.91	3.52	3.22	3.36	3.28	3.22	2.92	3.65	3.02	3.72	3.21
Percent Sensitive EPT Taxa	0.2	0.2	0.2	0.2	0.1	0.2	0.1	0.2	0.1	0.1	0.1	0.1	0.1
EPT Index (%)	0.3	0.3	0.2	0.3	0.1	0.3	0.2	0.4	0.2	0.1	0.2	0.2	0.2
Percent *Baetis sp.*	0.4	1.0	0.0	0.0	0.0	1.0	0.0	0.0	0.0	0.0	0.5	0.0	0.0
Number of non-*Baetis* Ephemeroptera	3	0	0	0	0	0	0	0	0	0	5	0	0
Percent Chironomidae	50.8	54.0	61.6	84.0	75.3	56.2	60.1	64.8	91.3	67.8	27.8	58.1	90.9
Number of Plecoptera Taxa	5	4	3	3	1	4	2	2	1	2	2	1	2
Percent Abundance of Oligochaetes & Hirudinea	2.2	3.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.9	1.2	3.2	0.6
Percent Dominant Taxon	18.5	16.2	16.2	22.9	39.4	30.4	22.2	28.1	27.3	27.2	35.3	25.7	21.2
Number of Common Taxa	9	15	8	10	7	7	7	9	13	18	26	12	11
Community Loss Index	1.0	0.8	1.1	0.9	1.2	1.2	1.1	1.5	2.0	1.0	1.5	1.2	1.9
TOLERANCE METRICS													
Hilsenhoff Biotic Index (HBI)	4.69	4.71	4.27	5.97	5.69	4.82	5.02	5.65	5.90	5.32	4.43	5.12	6.12
Percent Tolerant Taxa	19.4	13.5	17.4	14.8	29.2	8.0	22.7	19.0	30.0	26.7	27.6	22.2	21.7
Percent Intolerant Taxa	41.9	40.5	39.1	48.1	25.0	40.0	40.9	47.6	30.0	26.7	24.1	40.7	34.8
Number of Intolerant Taxa	13	15	9	13	6	10	9	10	6	8	7	11	8
TROPHIC HABIT METRICS													
Number of Predator Taxa	10	12	10	14	11	9	10	10	9	8	11	9	6
Percent Collector-Gatherers	20.7	34.5	34.2	47.7	28.8	11.5	37.0	14.2	49.5	32.8	17.0	19.4	56.9
Number of Shredder Taxa	8	6	3	4	4	8	3	5	2	6	4	5	5
LIFE HISTORY METRICS													
Number of Univoltine Taxa	20	18	16	19	15	14	12	15	12	14	13	13	13
Number of Merovoltine Taxa	1	1	0	1	0	1	1	1	1	1	0	3	1
Number of Semivoltine Taxa	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent Semivoltine taxa	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table C-17: Benthic macroinvertebrate population metrics for Site CC-1A-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. Data is shown for only years in which the site was sampled.

Site CC-1A-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2018
RICHNESS METRICS											
Total Density	1,515	1,450	4,576	5,369	2,126	2,597	1,035	3,210	6,370	6,480	8,305
Number of Taxa	11	19	22	18	19	24	21	14	21	24	33
Number of EPT Taxa	1	4	7	6	3	8	4	2	7	10	7
COMPOSITION METRICS											
Shannon-Weaver Diversity Index (H’)	1.71	3.01	3.31	2.35	3.11	3.11	2.73	2.43	3.27	2.73	3.11
Percent Sensitive EPT Taxa	0.1	0.2	0.2	0.2	0.1	0.2	0.0	0.1	0.1	0.3	0.2
EPT Index (%)	0.1	0.2	0.3	0.3	0.2	0.3	0.2	0.1	0.3	0.4	0.2
Percent *Baetis sp.*	0.0	1.0	0.4	0.2	0.0	0.0	0.0	0.0	0.1	0.7	0.1
Number of non-*Baetis* Ephemeroptera	0	0	30	52	0	78	11	30	530	20	120
Percent Chironomidae	96.7	38.6	65.8	81.7	69.8	42.6	70.5	96.3	72.7	78.7	95.3
Number of Plecoptera Taxa	0	1	2	2	2	1	0	0	0	3	2
Percent Abundance of Oligochaetes & Hirudinea	2.0	9.0	0.0	0.2	0.0	0.2	0.0	0.0	0.0	0.3	0.2
Percent Dominant Taxon	71.0	38.6	19.4	48.3	37.3	39.5	46.4	35.5	31.7	38.6	38.4
Number of Common Taxa	2	10	10	10	7	12	7	6	6	N/A	17
Community Loss Index	2.6	1.5	1.0	1.4	0.9	1.0	1.2	1.4	1.2	N/A	0.8
TOLERANCE METRICS											
Hilsenhoff Biotic Index (HBI)	6.87	4.30	4.95	6.12	5.94	4.20	5.69	6.80	5.71	6.35	6.74
Percent Tolerant Taxa	45.5	36.8	27.3	27.8	31.6	33.3	33.3	35.7	23.8	16.7	33.3
Percent Intolerant Taxa	18.2	36.8	36.4	27.8	31.6	29.2	28.6	35.7	38.1	50.0	36.4
Number of Intolerant Taxa	2	7	8	5	6	7	6	5	8	12	12
TROPHIC HABIT METRICS											
Number of Predator Taxa	1	1	4	5	4	5	8	1	3	5	5
Percent Collector-Gatherers	89.7	41.0	41.4	66.5	27.4	35.9	68.0	52.6	61.1	32.4	78.4
Number of Shredder Taxa	1	5	8	4	3	4	3	2	4	5	6
LIFE HISTORY METRICS											
Number of Univoltine Taxa	5	6	6	8	9	12	10	6	11	12	14
Number of Merovoltine Taxa	1	1	1	1	1	1	1	0	2	2	0
Number of Semivoltine Taxa	0	0	0	0	0	1	1	0	1	1	0
Percent Semivoltine taxa	0.0	0.0	0.0	0.0	0.0	4.2	4.8	0.0	4.8	4.2	0.0

Table C-18: Select benthic macroinvertebrate population metrics for Site FC-1-BIO, South Dakota, 1995 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. NC = not calculated.

Site FC-1-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Total Density	2,061	4,714	1,738	1,088	7,897	24,540	25,085	506	14,518	9,349	1,900[a]
Number of Taxa	38	35	27	30	40	49	40	41	57	41	34
Shannon-Weaver Diversity (H’)	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC
Number of EPT Taxa	11	12	7	11	13	14	10	12	14	9	11

Table C-19: Benthic macroinvertebrate population metrics for Site FC-1-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. N/A = no reference site established.

Site FC-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	1,600	669	1,254	3,600	2,772	2,045	480	5,034	4,660	3,300	1,845	3,053	3,941	2,850	3,950	1,053
Number of Taxa	33	32	51	28	28	33	39	36	31	35	30	37	40	34	37	40
Number of EPT Taxa	13	12	14	8	10	8	9	12	6	10	6	8	8	6	10	11
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H')	3.20	3.72	4.60	3.05	2.96	3.19	4.36	4.05	3.28	3.64	3.96	3.94	4.08	2.74	2.96	4.32
Percent Sensitive EPT Taxa	0.3	0.3	0.2	0.2	0.3	0.2	0.2	0.2	0.1	0.2	0.1	0.2	0.1	0.1	0.2	0.2
EPT Index (%)	0.4	0.4	0.3	0.3	0.4	0.2	0.2	0.3	0.2	0.3	0.2	0.2	0.2	0.2	0.3	0.3
Percent *Baetis sp.*	1.0	1.0	1.0	1.0	1.0	1.0	0.8	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Number of non-*Baetis* Ephemeroptera	5	3	0	0	0	10	7	52	0	0	0	1	5	0	10	3
Percent Chironomidae	10.6	36.3	31.4	9.4	5.8	14.4	65.0	42.7	11.6	15.5	40.1	59.6	19.0	4.7	4.1	33.2
Number of Plecoptera Taxa	3	3	4	3	4	2	3	3	3	3	2	3	3	0	3	3
Percent Abundance of Oligochaetes & Hirudinea	0.9	4.0	1.7	0.6	0.0	0.0	1.0	0.0	1.9	0.0	1.9	3.6	1.3	0.7	0.5	1.8
Percent Dominant Taxon	31.9	26.9	14.4	36.1	36.8	38.9	17.1	14.7	33.5	27.9	24.9	16.7	19.2	36.3	32.9	15.2
Number of Common Taxa	11	13	22	13	15	22	21	15	14	N/A	N/A	9	9	11	25	19
Community Loss Index	0.6	0.8	0.2	0.8	0.4	0.5	0.3	0.3	0.5	N/A	N/A	0.4	0.6	0.6	0.5	0.7
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	3.77	4.83	4.56	3.58	3.72	3.71	5.38	4.56	3.88	3.86	4.36	4.78	4.52	5.04	3.20	3.85
Percent Tolerant Taxa	21.2	21.9	25.5	17.9	17.9	24.2	30.8	19.4	29.0	25.7	23.3	29.7	30.0	29.4	18.9	17.5
Percent Intolerant Taxa	42.4	53.1	43.1	46.4	50.0	48.5	48.7	44.4	32.3	40.0	50.0	51.4	37.5	35.3	54.1	45.0
Number of Intolerant Taxa	14	17	22	13	14	16	19	16	10	14	15	19	15	12	20	18
TROPHIC HABIT METRICS																
Number of Predator Taxa	8	8	15	5	7	6	13	12	8	9	7	11	12	9	7	12
Percent Collector-Gatherers	41.6	51.7	40.8	21.7	20.2	33.0	34.6	42.1	42.9	38.2	27.6	41.6	34.3	38.1	36.7	30.3
Number of Shredder Taxa	9	7	9	6	5	6	6	6	7	8	6	4	7	4	8	8
LIFE HISTORY METRICS																
Number of Univoltine Taxa	16	13	26	11	11	17	20	22	14	18	15	20	18	19	16	20
Number of Merovoltine Taxa	1	3	1	3	2	0	2	1	1	2	2	1	0	2	3	2
Number of Semivoltine Taxa	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent Semivoltine taxa	3.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table C-20: Select benthic macroinvertebrate population metrics for Site NG-2-BIO, South Dakota, 1995 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. NC = not calculated.

Site NG-2-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Total Density	1,317	8,007	3,726	1,429	4,245	6,100	1,736	9,759	2,940	4,334	1,860
Number of Taxa	30	41	35	35	46	24	30	28	47	39	27
Shannon-Weaver Diversity (H’)	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC
Number of EPT Taxa	10	15	10	11	15	6	9	4	10	10	6

Table C-21: Benthic macroinvertebrate population metrics for Site NG-2-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years. N/A = Not applicable as this was a background control site.

Site NG-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	1,800	1,773	2,340	1,410	2,913	1,383	1,163	843	2,365	8,014	1,435	4,033	1,115	1,535	355	1,621
Number of Taxa	31	38	33	35	25	37	32	25	31	29	27	29	37	35	25	37
Number of EPT Taxa	8	13	8	9	9	7	10	6	8	7	5	8	9	8	8	8
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H')	3.48	4.28	4.27	4.10	2.89	4.41	3.80	3.26	2.97	2.78	2.47	2.51	3.33	2.98	2.37	3.04
Percent Sensitive EPT Taxa	0.2	0.3	0.2	0.1	0.3	0.1	0.3	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
EPT Index (%)	0.3	0.3	0.2	0.3	0.4	0.2	0.3	0.2	0.3	0.2	0.2	0.3	0.2	0.2	0.3	0.2
Percent *Baetis sp.*	0.0	0.0	0.1	0.1	1.0	0.4	0.0	0.5	1.0	1.0	0.0	0.2	0.9	1.0	0.8	0.3
Number of non-*Baetis* Ephemeroptera	390	85	73	95	0	26	40	3	15	0	0	31	20	5	1	12
Percent Chironomidae	55.3	59.5	67.2	39.0	57.5	56.9	55.7	72.7	12.3	46.8	16.4	39.4	4.1	9.4	1.1	23.9
Number of Plecoptera Taxa	1	4	3	3	5	2	4	2	2	2	2	3	3	3	4	4
Percent Abundance of Oligochaetes & Hirudinea	0.6	0.8	0.6	1.8	0.0	0.7	0.1	0.0	0.8	0.0	0.3	0.2	2.3	5.9	3.9	4.7
Percent Dominant Taxon	30.1	17.1	15.5	14.9	49.8	15.4	17.9	31.8	40.2	39.3	64.8	52.1	29.6	46.3	45.6	44.7
Number of Common Taxa	N/A	N/A	0	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0	19	28	12	19
Community Loss Index	N/A	N/A	0.0	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	0.7	0.6	0.6	1.3	0.7
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	4.82	5.36	5.11	4.08	4.85	4.84	4.99	5.68	4.38	4.91	4.22	4.93	4.11	4.25	3.75	4.54
Percent Tolerant Taxa	32.3	34.2	27.3	28.6	16.0	21.6	28.1	28.0	32.3	24.1	29.6	31.0	27.0	28.6	24.0	27.0
Percent Intolerant Taxa	38.7	44.7	45.5	34.3	56.0	40.5	43.8	44.0	41.9	58.6	48.1	44.8	48.6	40.0	52.0	43.2
Number of Intolerant Taxa	12	17	15	12	14	15	14	11	13	17	13	13	18	14	13	16
TROPHIC HABIT METRICS																
Number of Predator Taxa	7	10	5	6	10	8	9	8	6	6	5	9	9	8	8	10
Percent Collector-Gatherers	67.1	42.4	50.4	29.4	10.1	40.9	41.7	13.8	56.9	19.5	12.5	15.6	23.5	33.2	7.6	17.5
Number of Shredder Taxa	6	8	5	7	4	6	7	5	3	6	6	5	5	7	4	6
LIFE HISTORY METRICS																
Number of Univoltine Taxa	16	21	14	13	9	17	14	12	10	12	13	13	15	13	9	17
Number of Merovoltine Taxa	3	2	2	1	1	1	3	0	1	2	3	1	1	3	1	3
Number of Semivoltine Taxa	1	0	0	0	1	0	1	1	1	0	0	2	0	0	0	1
Percent Semivoltine taxa	3.2	0.0	0.0	0.0	4.0	0.0	3.1	4.0	3.2	0.0	0.0	6.9	0.0	0.0	0.0	2.7

Table C-22: Select benthic macroinvertebrate population metrics for Site SG-1-BIO, South Dakota, 1995 - 2005. Quantitative sampling methods (i.e., Surber or Hess) were performed in these years. Data is shown for only years in which the site was sampled. NC = not calculated.

Site SG-1-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Total Density	5,977	4,509	2,737	467	4,791	8,957	5,116	13,225	5,464	4,213	9,921
Number of Taxa	31	24	34	21	29	34	37	42	45	44	36
Shannon-Weaver Diversity (H')	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC	NC
Number of EPT Taxa	10	13	10	9	13	12	12	12	14	13	11

Table C-23: Benthic macroinvertebrate population metrics for Site SG-1-BIO, South Dakota, 2006 - 2021. Semiquantitative sampling methods (Kick) were performed in these years.

Site SG-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Total Density	2,910	4,070	3,311	4,470	5,619	2,215	4,021	2,912	7,521	8,842	11,800	9,420	6,906	4,400	6,365	6,530
Number of Taxa	28	33	40	30	31	34	34	25	29	30	32	38	35	45	42	38
Number of EPT Taxa	11	10	12	9	15	11	12	11	10	11	13	10	12	13	12	12
COMPOSITION METRICS																
Shannon-Weaver Diversity Index (H')	3.83	3.91	3.99	3.62	3.94	4.48	3.82	3.69	2.83	2.98	3.42	3.99	3.40	3.77	3.41	3.20
Percent Sensitive EPT Taxa	0.3	0.2	0.3	0.3	0.4	0.2	0.3	0.3	0.3	0.3	0.3	0.2	0.3	0.2	0.2	0.3
EPT Index (%)	0.4	0.3	0.3	0.3	0.5	0.3	0.4	0.4	0.3	0.4	0.4	0.3	0.3	0.3	0.3	0.3
Percent *Baetis sp.*	0.9	1.0	1.0	1.0	0.9	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Number of non-*Baetis* Ephemeroptera	20	0	1	0	60	10	20	0	0	20	20	0	60	25	30	35
Percent Chironomidae	24.7	34.6	23.6	13.6	40.8	33.2	32.8	34.0	8.0	6.8	13.6	48.8	24.1	17.6	12.8	6.8
Number of Plecoptera Taxa	3	3	3	3	6	3	4	5	3	3	3	4	4	3	4	3
Percent Abundance of Oligochaetes & Hirudinea	0.0	0.5	0.7	1.1	0.0	1.1	0.2	0.0	0.5	0.2	0.5	0.4	0.4	0.8	0.5	0.5
Percent Dominant Taxon	17.2	22.4	22.0	23.3	13.6	13.1	18.2	27.1	42.8	39.5	25.0	18.0	27.7	24.9	30.3	28.3
Number of Common Taxa	12	18	21	19	20	22	17	16	17	15	22	23	11	25	19	23
Community Loss Index	0.6	0.7	0.6	0.7	0.6	0.4	0.5	0.8	0.7	0.6	0.6	0.7	0.6	0.6	0.6	0.8
TOLERANCE METRICS																
Hilsenhoff Biotic Index (HBI)	3.66	4.27	3.69	3.24	3.78	3.94	4.11	4.14	3.72	3.64	3.88	4.71	4.24	3.90	3.41	3.13
Percent Tolerant Taxa	17.9	24.2	15.0	26.7	12.9	20.6	14.7	20.0	13.8	10.0	3.1	15.8	20.0	26.7	23.8	23.7
Percent Intolerant Taxa	42.9	39.4	50.0	46.7	54.8	44.1	50.0	44.0	48.3	56.7	56.3	47.4	40.0	35.6	47.6	44.7
Number of Intolerant Taxa	12	13	20	14	17	15	17	11	14	17	18	18	14	16	20	17
TROPHIC HABIT METRICS																
Number of Predator Taxa	4	8	9	7	9	8	8	7	7	5	7	11	11	12	11	9
Percent Collector-Gatherers	48.7	57.2	56.8	42.9	63.0	51.2	69.1	72.5	57.7	69.4	66.7	63.8	54.0	54.1	47.0	55.2
Number of Shredder Taxa	5	7	6	6	6	6	6	6	6	6	5	7	5	7	5	7
LIFE HISTORY METRICS																
Number of Univoltine Taxa	15	21	23	12	14	20	20	12	16	17	22	21	17	19	19	16
Number of Merovoltine Taxa	1	0	1	1	1	0	1	1	0	1	0	1	0	3	1	1
Number of Semivoltine Taxa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Percent Semivoltine taxa	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

This page intentionally left blank

Appendix D 2021 Periphyton Data

This page intentionally left blank

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/27/2021
Site: LABRADOR GULCH, LB-4-BIO

TOTAL CELLS/cm^2	183,479
NUMBER OF TAXA	22
SHANNON-WEAVER DIVERSITY (H')	3.92
TROPHIC STATE INDEX	77.3

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	3,276	1.8
Achnanthes linearis	4,915	2.7
Achnanthes minutissima	37,679	20.5
Amphora perpusilla	22,935	12.5
Cocconeis placentula	11,468	6.3
Cymbella affinis	1,638	0.9
Cymbella minuta	13,106	7.1
Fragilaria construens venter	1,638	0.9
Gomphonema angustatum	6,553	3.6
Navicula cryptocephala	9,829	5.4
Navicula cryptocephala veneta	1,638	0.9
Navicula minima	14,744	8.0
Navicula minuscula	8,191	4.5
Navicula radiosa	1,638	0.9
Navicula sp.	3,276	1.8
Navicula tripunctata	6,553	3.6
Navicula viridula	8,191	4.5
Nitzschia dissipata	3,276	1.8
Nitzschia frustulum	4,915	2.7
Nitzschia linearis	3,276	1.8
Rhoicosphenia curvata	11,468	6.3
Synedra rumpens	3,276	1.8

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/25/2021
Site: RENO CREEK, RC-1-BIO

TOTAL CELLS/cm^2	313,358
NUMBER OF TAXA	14
SHANNON-WEAVER DIVERSITY (H')	2.74
TROPHIC STATE INDEX	81.0

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	12,738	4.1
Achnanthes minutissima	53,500	17.1
Amphora perpusilla	114,643	36.6
Cocconeis placentula	66,238	21.1
Cymbella sinuata	7,643	2.4
Navicula cryptocephala	7,643	2.4
Navicula cryptocephala veneta	2,548	0.8
Navicula minima	5,095	1.6
Navicula viridula	7,643	2.4
Nitzschia communis	2,548	0.8
Nitzschia frustulum	5,095	1.6
Nitzschia linearis	5,095	1.6
Nitzschia palea	2,548	0.8
Rhoicosphenia curvata	20,381	6.5

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-1-BIO

TOTAL CELLS/cm^2	535,963
NUMBER OF TAXA	25
SHANNON-WEAVER DIVERSITY (H')	3.33
TROPHIC STATE INDEX	91.3

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	4,220	0.8
Amphora perpusilla	8,440	1.6
Caloneis ventricosa minuta	4,220	0.8
Cymbella affinis	16,881	3.1
Cymbella minuta	173,028	32.3
Fragilaria construens	4,220	0.8
Gomphonema angustatum	16,881	3.1
Gomphonema sp.	4,220	0.8
Navicula cryptocephala	16,881	3.1
Navicula cryptocephala veneta	4,220	0.8
Navicula graciloides	4,220	0.8
Navicula minima	12,661	2.4
Navicula sp.	8,440	1.6
Navicula tripunctata	12,661	2.4
Navicula viridula	4,220	0.8
Nitzschia frustulum	25,321	4.7
Nitzschia palea	4,220	0.8
Rhoicosphenia curvata	16,881	3.1
Surirella ovata	4,220	0.8
Synedra rumpens	8,440	1.6
Synedra tenera	4,220	0.8
CHLOROPHYTA		
Chlamydomonas sp.	4,220	0.8
Sphaerocystis schroeteri	4,220	0.8
CYANOPHYTA		
Anabaena circinalis	130,826	24.4
Oscillatoria sp.	37,982	7.1

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/26/2021
Site: ANNIE CREEK, AC-2-BIO

TOTAL CELLS/cm^2	452,432
NUMBER OF TAXA	21
SHANNON-WEAVER DIVERSITY (H')	3.28
TROPHIC STATE INDEX	84.3

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	22,461	5.0
Achnanthes linearis	12,835	2.8
Achnanthes minutissima	147,602	32.6
Amphora perpusilla	28,879	6.4
Caloneis ventricosa minuta	3,209	0.7
Cocconeis placentula	35,296	7.8
Cymbella minuta	9,626	2.1
Cymbella sinuata	6,417	1.4
Gomphonema angustatum	6,417	1.4
Gomphonema subclavatum	3,209	0.7
Navicula cryptocephala	16,044	3.5
Navicula graciloides	3,209	0.7
Navicula tripunctata	16,044	3.5
Navicula viridula	22,461	5.0
Nitzschia capitellata	3,209	0.7
Nitzschia linearis	6,417	1.4
Rhoicosphenia curvata	93,053	20.6
Synedra rumpens	3,209	0.7
Synedra ulna	6,417	1.4
CHLOROPHYTA		
Sphaerocystis schroeteri	3,209	0.7
CYANOPHYTA		
Oscillatoria sp.	3,209	0.7

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/26/2021
Site: ROSS VALLEY , RV-2-BIO

TOTAL CELLS/cm^2	136,844
NUMBER OF TAXA	18
SHANNON-WEAVER DIVERSITY (H')	3.53
TROPHIC STATE INDEX	75.0

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	9,123	6.7
Achnanthes linearis	3,649	2.7
Achnanthes minutissima	16,421	12.0
Amphora perpusilla	38,315	28.0
Caloneis ventricosa minuta	3,649	2.7
Cocconeis placentula	10,947	8.0
Navicula cascadensis	1,825	1.3
Navicula cryptocephala	10,947	8.0
Navicula cryptocephala veneta	9,123	6.7
Navicula gregaria	1,825	1.3
Navicula minima	5,474	4.0
Navicula rhynchocephala	1,825	1.3
Navicula tripunctata	5,474	4.0
Navicula viridula	3,649	2.7
Nitzschia linearis	1,825	1.3
Nitzschia palea	1,825	1.3
Rhoicosphenia curvata	9,123	6.7
CYANOPHYTA		
Oscillatoria sp.	1,825	1.3

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/26/2021
Site: LOST CAMP CREEK, LC-1-BIO

TOTAL CELLS/cm^2	178,236
NUMBER OF TAXA	18
SHANNON-WEAVER DIVERSITY (H')	3.00
TROPHIC STATE INDEX	72.9

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	6,812	3.8
Achnanthes linearis	10,217	5.7
Achnanthes minutissima	46,545	26.1
Amphora perpusilla	17,029	9.6
Cocconeis placentula	9,082	5.1
Cymbella minuta	1,135	0.6
Cymbella sinuata	3,406	1.9
Gomphonema angustatum	2,271	1.3
Gomphonema tenellum	1,135	0.6
Navicula cryptocephala	2,271	1.3
Navicula minima	1,135	0.6
Navicula minuscula	3,406	1.9
Nitzschia communis	3,406	1.9
Nitzschia frustulum	3,406	1.9
Nitzschia innominata	6,812	3.8
Nitzschia linearis	1,135	0.6
Nitzschia paleacea	1,135	0.6
Rhoicosphenia curvata	57,898	32.5

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/24/2021
Site: DEADWOOD CREEK, DC-2-BIO

TOTAL CELLS/cm^2	1,122,898
NUMBER OF TAXA	20
SHANNON-WEAVER DIVERSITY (H')	2.73
TROPHIC STATE INDEX	90.0

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	19,032	1.7
Achnanthes linearis	28,548	2.5
Achnanthes minutissima	609,031	54.2
Amphora perpusilla	9,516	0.8
Caloneis ventricosa	28,548	2.5
Cymbella minuta	76,129	6.8
Diatoma hiemale mesodon	9,516	0.8
Fragilaria vaucheria	57,097	5.1
Frustulia rhomboides	9,516	0.8
Gomphonema angustatum	19,032	1.7
Meridion circulare	28,548	2.5
Navicula cryptocephala	9,516	0.8
Navicula sp.	19,032	1.7
Neidium sp.	19,032	1.7
Nitzschia frustulum	19,032	1.7
Nitzschia linearis	9,516	0.8
Pinnularia sp.	9,516	0.8
Synedra rumpens	104,677	9.3
Synedra ulna	28,548	2.5
CHLOROPHYTA		
Cladophora sp.	9,516	0.8

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/23/2021
Site: EAST FORK FALSE BOTTOM CREEK, EFB-1-BIO

TOTAL CELLS/cm^2	<415
NUMBER OF TAXA	--
SHANNON-WEAVER DIVERSITY (H')	--
TROPHIC STATE INDEX	--

Organisms	Cells/cm^2	Rel % Conc.
NO ALGAE	<415	--

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/23/2021
Site: WEST FORK FALSE BOTTOM CREEK, WFB-1-BIO

TOTAL CELLS/cm^2	17,920
NUMBER OF TAXA	12
SHANNON-WEAVER DIVERSITY (H')	2.28
TROPHIC STATE INDEX	67.6

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes minutissima	597	3.3
Epithemia sorex	896	5.0
Eunotia pectinalis	10,453	58.3
Gomphonema angustatum	597	3.3
Gomphonema subclavatum	299	1.7
Meridion circulare	597	3.3
Nitzschia frustulum	299	1.7
Pinnularia sp.	299	1.7
Synedra rumpens	1,493	8.3
CHLOROPHYTA		
Ulothrix sp.	1,792	10.0
CRYPTOPHYTA		
Cryptomonas erosa	299	1.7
Rhodomonas minuta	299	1.7

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/25/2021
Site: FANTAIL CREEK, FC-1-BIO

TOTAL CELLS/cm^2	5,388
NUMBER OF TAXA	16
SHANNON-WEAVER DIVERSITY (H')	3.11
TROPHIC STATE INDEX	52.6

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	703	13.0
Achnanthes minutissima	2,227	41.3
Amphora ovalis	117	2.2
Amphora perpusilla	117	2.2
Caloneis ventricosa	117	2.2
Cocconeis placentula	234	4.3
Cymbella affinis	117	2.2
Diatoma tenue	117	2.2
Epithemia sorex	234	4.3
Gomphonema angustatum	352	6.5
Gomphonema sp.	117	2.2
Navicula cryptocephala	234	4.3
Navicula minima	117	2.2
Navicula viridula	117	2.2
Rhoicosphenia curvata	234	4.3
Synedra ulna contracta	234	4.3

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/24/2021
Site: NEVADA GULCH, NG-2-BIO

TOTAL CELLS/cm^2	1,653,691
NUMBER OF TAXA	17
SHANNON-WEAVER DIVERSITY (H')	2.64
TROPHIC STATE INDEX	91.1

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes linearis	28,760	1.7
Achnanthes minutissima	834,035	50.4
Amphora perpusilla	14,380	0.9
Caloneis ventricosa minuta	14,380	0.9
Cocconeis placentula	14,380	0.9
Cymbella sinuata	14,380	0.9
Gomphonema angustatum	86,279	5.2
Gomphonema subclavatum	86,279	5.2
Navicula cryptocephala	71,900	4.3
Navicula viridula	28,760	1.7
Nitzschia dissipata	14,380	0.9
Nitzschia frustulum	258,838	15.7
Nitzschia linearis	57,520	3.5
Nitzschia palea	71,900	4.3
Nitzschia paleacea	14,380	0.9
Rhoicosphenia curvata	28,760	1.7
CRYPTOPHYTA		
Rhodomonas minuta	14,380	0.9

DATA: PERIPHYTON ANALYSES
Client: WHARF
Sampled: 8/24/2021
Site: STEWART GULCH, SG-1-BIO

TOTAL CELLS/cm^2	219,091
NUMBER OF TAXA	16
SHANNON-WEAVER DIVERSITY (H')	2.15
TROPHIC STATE INDEX	73.5

Organisms	Cells/cm^2	Rel % Conc.
BACILLARIOPHYTA		
Order Pennales		
Achnanthes lanceolata	51,305	23.4
Achnanthes linearis	16,640	7.6
Achnanthes minutissima	119,251	54.4
Amphora perpusilla	2,773	1.3
Caloneis ventricosa	1,387	0.6
Diatoma hiemale mesodon	2,773	1.3
Fragilaria leptostauron	4,160	1.9
Fragilaria vaucheria	1,387	0.6
Gomphonema angustatum	1,387	0.6
Navicula gregaria	1,387	0.6
Navicula minima	4,160	1.9
Navicula minuscula	6,933	3.2
Navicula viridula	1,387	0.6
Nitzschia communis	1,387	0.6
Nitzschia frustulum	1,387	0.6
Nitzschia linearis	1,387	0.6

Appendix E Historical Periphyton Data

This page intentionally left blank

Table E-1: Periphyton population metrics for Site LB-4-BIO, South Dakota, 2005. Data is shown for the only year in which the site was sampled prior to 2006. NC = not calculated.

Site LB-4-BIO	2005
Density (cells/cm^2)	NC
Number of Taxa	54

Table E-2: Periphyton population metrics for Site LB-4-BIO, South Dakota, 2006 - 2021.

Site LB-4-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	96,692	268,422	47,162	579,611	1,030,889	85,328	107,845	38,956	44,352	43,610	180,874	261,688	229,142	200,277	33,888	183,479
Relative Density (%)	73.7	100.0	100.0	37.8	97.3	53.1	96.4	96.6	98.0	100.0	100.0	100.0	99.2	100.0	100.0	100.0
Number of Taxa (species)	20	11	14	18	18	20	21	19	22	18	23	26	23	19	15	22
Number of Diatom Taxa	17	11	14	17	16	18	20	18	21	18	23	26	22	19	15	22
Number of Periphyton Divisions	7	1	1	2	5	4	3	3	3	1	1	1	2	1	1	1
Number of Periphyton Genera	18	10	13	15	16	15	17	13	16	6	19	22	21	15	9	10
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	2.88	2.42	2.90	3.18	2.58	2.79	3.20	3.05	3.43	3.23	3.82	3.87	3.51	3.47	2.60	3.92
Autotrophic Index	195	1,061	829	407	481	531	671	749	309	292	1,354	536	653	785	1,228	519
TOLERANCE METRICS																
Percent Tolerant Diatoms	2.4	4.6	10.2	1.3	5.5	1.7	2.8	1.8	5.2	9.7	19.0	12.2	7.4	11.4	3.7	13.0
Lange-Bertalot Pollution Index	2.85	2.41	2.63	2.75	2.72	2.82	2.75	2.75	2.58	2.53	2.42	2.56	2.64	2.58	2.83	2.48
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	52.9	54.5	57.1	58.8	50.0	50.0	70.0	38.9	57.1	61.1	60.9	53.8	50.0	42.1	46.7	50.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	64.7	63.6	64.3	58.8	50.0	44.4	70.0	44.4	57.1	55.6	56.5	53.8	54.5	57.9	46.7	50.0
Percent Nitrogen Heterotrophs	11.8	18.2	14.3	11.8	6.3	11.1	10.0	5.6	19.0	22.2	17.4	19.2	13.6	15.8	20.0	9.0
Percent High Oxygen Diatoms	41.2	27.3	42.9	52.9	37.5	44.4	50.0	22.2	33.3	33.3	34.8	26.9	22.7	26.3	20.0	41.0
Percent Motile Diatoms	58.8	45.5	42.9	41.2	43.8	50.0	55.0	44.4	61.9	72.2	52.2	53.8	54.5	57.9	66.7	55.0
Percent Saprobic Diatoms	11.8	18.2	21.4	11.8	12.5	16.7	10.0	5.6	19.0	16.7	17.4	15.4	4.5	15.8	13.3	9.0

Table E-3: Periphyton population metrics for Site RC-1-BIO, South Dakota, 2017 - 2021. Data is shown for only years in which the site was sampled.

Site RC-1-BIO	2017	2018	2019	2020	2021
RICHNESS METRICS					
Density (cells/cm^2)	95,449	71,399	47,771	115,261	313,358
Relative Density (%)	100.0	100.0	100.0	99.0	100.0
Number of Taxa (species)	13	14	14	24	14
Number of Diatom Taxa	13	14	14	23	14
Number of Periphyton Divisions	1	1	1	2	1
Number of Periphyton Genera	11	12	11	13	7
COMPOSITION METRICS					
Shannon-Weaver Diversity Index for Diatoms (H')	2.30	2.24	2.52	3.14	2.74
Autotrophic Index	3,328	621	6,488	3,443	410
TOLERANCE METRICS					
Percent Tolerant Diatoms	2.8	5.0	9.1	6.2	3.0
Lange-Bertalot Pollution Index	2.76	2.62	2.54	2.64	2.83
TROPHIC HABIT METRICS					
Percent Eutrophic Diatoms	38.5	50.0	28.6	47.8	64.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	38.5	64.3	42.9	52.2	50.0
Percent Nitrogen Heterotrophs	15.4	14.3	7.1	17.4	29.0
Percent High Oxygen Diatoms	23.1	28.6	35.7	39.1	29.0
Percent Motile Diatoms	38.5	50.0	50.0	56.5	64.0
Percent Saprobic Diatoms	23.1	7.1	7.1	8.7	29.0

Table E-4: Periphyton population metrics for Site AC-1-BIO, South Dakota, 1993 - 2005.

Site AC-1-BIO	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density(cells/cm²)	2,100	205,900	611,500	67,300	75,000	862,800	33,900	18,271,800	108,205,900	240,100	457,700	1,894,000	288,500
Number of Taxa	6	13	11	4	2	13	3	21	22	12	13	11	11

Table E-5: Periphyton population metrics for Site AC-1-BIO, South Dakota, 2006 - 2021. N/A = no reference site established.

Site AC-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm²)	168,635	1,932,352	116,238	44,016	42,707	143,406	108,653	249,253	17,896,821	397,730	952,328	60,300	448,390	156,885	173,223	535,963
Relative Density (%)	87.5	100.0	100.0	100.0	29.4	19.9	100.0	99.0	100.0	100.0	100.0	65.7	99.4	97.8	94.0	67.0
Number of Taxa (species)	29	15	21	13	10	18	26	13	25	16	16	17	13	13	22	25
Number of Diatom Taxa	26	15	21	13	7	16	26	12	25	16	16	13	12	10	19	21
Number of Periphyton Divisions	6	1	1	1	3	4	1	3	1	1	1	7	3	4	3	3
Number of Periphyton Genera	20	13	13	10	9	13	21	11	20	7	13	16	12	11	12	15
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	3.59	3.04	3.04	2.54	2.64	3.00	3.88	1.98	3.58	2.26	3.33	3.02	2.29	1.84	3.28	3.08
Bahls Similarity Index (%)	29.2	34.0	15.9	35.9	13.6	10.7	42.4	34.0	11.0	N/A	N/A	24.9	22.5	31.9	38.4	14.0
Autotrophic Index	505	541	634	374	564	282	979	264	239	434	611	1,181	335	373	236	268
TOLERANCE METRICS																
Percent Tolerant Diatoms	6.1	24.8	3.5	5.9	26.7	7.3	15.4	12.6	15.5	3.2	3.8	0.0	17.2	18.4	12.8	5.0
Lange-Bertalot Pollution Index	2.65	2.28	2.47	2.66	2.40	2.67	2.42	2.61	2.18	2.71	2.66	2.82	2.56	2.57	2.57	2.20
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	61.5	60.0	61.9	61.5	42.9	50.0	38.5	50.0	44.0	50.0	56.3	46.2	41.7	40.0	52.6	48.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	3.8	0.0	8.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	46.2	60.0	47.6	46.2	71.4	50.0	50.0	66.7	36.0	56.3	43.8	46.2	50.0	50.0	47.4	38.0
Percent Nitrogen Heterotrophs	19.2	26.7	19.0	23.1	14.3	6.3	11.5	25.0	16.0	12.5	18.8	0.0	8.3	20.0	15.8	14.0
Percent High Oxygen Diatoms	30.8	26.7	33.3	30.8	42.9	37.5	34.6	50.0	32.0	50.0	25.0	38.5	33.3	30.0	36.8	33.0
Percent Motile Diatoms	61.5	66.7	76.2	61.5	71.4	56.3	38.5	41.7	48.0	50.0	68.8	30.8	41.7	60.0	52.6	57.0
Percent Saprobic Diatoms	19.2	13.3	19.0	23.1	14.3	12.5	7.7	16.7	12.0	12.5	18.8	0.0	8.3	10.0	10.5	14.0

Table E-6: Periphyton population metrics for Site AC-2-BIO, South Dakota, 1993 - 2005.

Site AC-2-BIO	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density(cells/cm^2)	1,900	133,800	168,500	57,800	153,600	142,300	70,200	7,804,700	7,910,400	7,500	200,500	166,700	422,800
Number of Taxa	10	14	8	3	3	13	11	19	30	12	21	11	12

Table E-7: Periphyton population metrics for from Site AC-2-BIO, South Dakota, 2006 - 2021.

Site AC-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	325,533	837,091	17,061	69,203	150,849	16,947	112,637	914,563	874,668	23,229	187,319	215,024	89,724	95,896	22,535	452,432
Relative Density (%)	100.0	100.0	98.8	90.5	95.0	92.4	93.7	100.0	100.0	96.3	100.0	98.4	100.0	100.0	100.0	99.0
Number of Taxa (species)	16	19	20	15	15	12	18	16	18	19	14	17	16	18	16	21
Number of Diatom Taxa	16	19	19	14	14	11	16	16	18	18	14	16	16	18	16	19
Number of Periphyton Divisions	1	1	3	3	3	3	4	1	1	2	1	3	1	1	1	3
Number of Periphyton Genera	13	16	20	14	12	10	17	13	16	14	12	15	13	17	8	12
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	2.70	3.47	3.34	2.65	2.95	2.23	2.86	3.12	3.66	3.59	2.40	2.98	3.67	3.77	2.82	3.20
Bahls Similarity Index (%)	46.0	31.2	25.2	39.7	32.5	35.4	27.6	74.5	44.7	36.0	40.6	39.2	47.4	50.7	53.3	61.0
Autotrophic Index	770	2,288	1,153	430	827	209	253	619	219	551	330	4,329	176	1,933	301	279
TOLERANCE METRICS																
Percent Tolerant Diatoms	0.8	28.9	25.0	1.1	5.3	1.0	3.3	12.3	10.9	3.8	1.1	2.5	22.2	8.7	7.7	0.0
Lange-Bertalot Pollution Index	2.84	2.20	2.10	2.76	2.74	2.93	2.72	2.57	2.39	2.58	2.89	2.79	2.26	2.43	2.68	2.81
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	50.0	47.4	47.4	64.3	50.0	54.5	56.3	43.8	55.6	50.0	42.9	56.3	56.3	50.0	50.0	42.0
Percent Acidiphilic Diatoms	0.0	0.0	5.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.6	0.0	0.0
Percent Alkaliphilic Diatoms	62.5	57.9	52.6	50.0	57.1	45.5	50.0	50.0	55.6	50.0	50.0	50.0	56.3	50.0	43.8	32.0
Percent Nitrogen Heterotrophs	6.3	15.8	15.8	7.1	7.1	9.1	25.0	18.8	22.2	11.1	0.0	18.8	18.8	16.7	18.8	0.0
Percent High Oxygen Diatoms	31.3	31.6	26.3	42.9	35.7	54.5	18.8	18.8	38.9	38.9	35.7	31.3	31.3	33.3	25.0	42.0
Percent Motile Diatoms	50.0	47.4	52.6	57.1	50.0	54.5	31.3	50.0	50.0	38.9	35.7	50.0	62.5	55.6	56.3	42.0
Percent Saprobic Diatoms	6.3	10.5	15.8	7.1	14.3	18.2	6.3	18.8	22.2	5.6	7.1	18.8	18.8	11.1	18.8	5.0

Table E-8: Periphyton population metrics for Site AC-3-BIO, South Dakota, 2000 - 2005.

Site AC-3-BIO	2000	2001	2002	2003	2004	2005
Density (cells/cm^2)	5,411,300	1,524,700	165,200	15,400	311,800	8,300
Number of Taxa	17	33	24	11	19	15

Table E-9: Periphyton population metrics for Site AC-3-BIO, South Dakota, 2006 - 2021. Data is shown for only years in which the site was sampled.

Site AC-3-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019
RICHNESS METRICS														
Density (cells/cm^2)	173,810	1,276,416	1,183,845	517,242	217,265	57,222	2,624,784	95,914	3,025,171	81,904	680,902	343,931	393,760	138,661
Relative Density (%)	99.1	100.0	100.0	100.0	96.2	95.1	100.0	100.0	100.0	97.0	100.0	100.0	99.1	99.1
Number of Taxa (species)	11	15	21	18	19	20	14	18	18	23	17	14	17	26
Number of Diatom Taxa	10	15	21	18	18	19	14	18	18	22	17	14	16	25
Number of Periphyton Divisions	3	1	1	1	3	3	1	1	1	2	1	1	3	3
Number of Periphyton Genera	11	13	19	15	16	17	12	16	16	11	13	10	16	19
COMPOSITION METRICS														
Shannon-Weaver Diversity Index for Diatoms (H’)	1.81	3.28	3.83	3.39	3.40	3.16	2.57	2.43	2.77	3.91	2.64	3.05	3.68	3.56
Bahls Similarity Index (%)	26.2	29.2	47.9	45.3	43.9	40.4	36.7	55.5	29.9	54.7	40.7	46.9	46.1	50.1
Autotrophic Index	419	532	181	376	276	210	146	168	212	397	242	3,501	257	300
TOLERANCE METRICS														
Percent Tolerant Diatoms	0.0	14.3	27.8	1.8	5.0	0.9	3.4	2.4	0.0	16.3	2.1	8.7	31.8	7.6
Lange-Bertalot Pollution Index	2.91	2.41	2.04	2.44	2.57	2.75	2.75	2.81	2.39	2.28	2.83	2.66	2.13	2.54
TROPHIC HABIT METRICS														
Percent Eutrophic Diatoms	40.0	60.0	61.9	61.1	50.0	57.9	64.3	38.9	72.2	50.0	47.1	64.3	50.0	52.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	40.0	66.7	61.9	61.1	55.6	57.9	57.1	44.4	66.7	54.5	58.8	57.1	50.0	48.0
Percent Nitrogen Heterotrophs	0.0	20.0	19.0	11.1	16.7	10.5	21.4	16.7	5.6	13.6	17.6	21.4	18.8	16.0
Percent High Oxygen Diatoms	10.0	26.7	33.3	38.9	38.9	47.4	28.6	27.8	61.1	36.4	29.4	28.6	18.8	28.0
Percent Motile Diatoms	60.0	66.7	52.4	66.7	61.1	57.9	42.9	38.9	44.4	59.1	64.7	57.1	68.8	60.0
Percent Saprobic Diatoms	10.0	20.0	14.3	5.6	16.7	5.3	21.4	11.1	5.6	13.6	11.8	21.4	18.8	16.0

Table E-10: Periphyton population metrics for Site RV-2-BIO, South Dakota, 2006 – 2021. N/A = no reference site established.

Site RV-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	161,738	21,860	129,924	162,131	577,133	64,890	409,829	86,781	2,557,674	340,575	579,489	509,783	971,987	491,793	52,101	136,844
Relative Density (%)	98.7	91.2	91.1	69.1	56.7	62.4	100.0	97.8	100.0	98.8	95.0	98.0	100.0	97.4	95.0	99.0
Number of Taxa (species)	19	14	18	18	24	22	14	21	19	21	15	17	15	18	19	18
Number of Diatom Taxa	18	13	17	17	21	20	14	20	19	20	14	15	15	17	17	17
Number of Periphyton Divisions	3	3	3	2	5	4	1	3	1	2	3	4	1	3	3	2
Number of Periphyton Genera	15	12	12	14	19	19	10	15	12	9	14	14	11	15	12	8
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H’)	3.71	3.23	3.24	3.29	3.73	3.39	2.33	3.75	3.33	3.84	3.37	2.62	2.19	3.20	3.58	3.48
Bahls Similarity Index (%)	33.9	26.5	40.6	51.3	26.0	22.4	36.2	48.0	18.7	N/A	N/A	32.3	21.8	37.5	40.2	68.0
Autotrophic Index	658	3,602	964	657	942	890	787	518	374	6,428	824	680	268	329	1,284	416
TOLERANCE METRICS																
Percent Tolerant Diatoms	1.3	3.2	8.7	1.2	1.5	1.0	3.5	6.7	7.5	5.1	0.0	0.7	6.8	10.8	1.4	5.0
Lange-Bertalot Pollution Index	2.40	2.45	2.57	2.59	2.63	2.66	2.82	2.54	2.65	2.43	2.72	2.87	2.78	2.56	2.68	2.70
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	61.1	38.5	52.9	64.7	47.6	60.0	50.0	50.0	42.1	55.0	50.0	66.7	53.3	47.1	35.3	53.0
Percent Acidiphilic Diatoms	0.0	23.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	61.1	30.8	52.9	58.8	52.4	55.0	57.1	55.0	42.1	45.0	42.9	60.0	53.3	47.1	35.3	47.0
Percent Nitrogen Heterotrophs	11.1	7.7	23.5	11.8	9.5	10.0	14.3	15.0	15.8	15.0	7.1	6.7	20.0	17.6	0.0	12.0
Percent High Oxygen Diatoms	27.8	38.5	23.5	35.3	33.3	25.0	21.4	30.0	31.6	40.0	28.6	53.3	26.7	23.5	29.4	24.0
Percent Motile Diatoms	61.1	38.5	58.8	52.9	57.1	70.0	64.3	65.0	57.9	65.0	64.3	60.0	66.7	58.8	64.7	71.0
Percent Saprobic Diatoms	11.1	7.7	23.5	11.8	9.5	10.0	14.3	15.0	21.1	15.0	7.1	13.3	13.3	11.8	5.9	18.0

Table E-11: Periphyton population metrics for Site LC-1-BIO, South Dakota, 2010 - 2021. Data is shown for only years in which the site was sampled. N/A = no reference site established.

Site LC-1-BIO	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS												
Density (cells/cm^2)	77,822	19,460	151,202	576,992	13,158,786	41,065	284,581	92,030	172,369	67,790	36,665	178,236
Relative Density (%)	97.6	100.0	100.0	100.0	100.0	98.5	100.0	99.2	100.0	98.9	100.0	100.0
Number of Taxa (species)	18	10	17	18	22	16	18	18	15	18	16	18
Number of Diatom Taxa	17	10	17	18	22	15	18	17	15	17	16	18
Number of Periphyton Divisions	3	1	1	1	1	2	1	3	1	3	1	1
Number of Periphyton Genera	17	9	15	16	19	12	16	16	14	15	8	8
COMPOSITION METRICS												
Shannon-Weaver Diversity Index for Diatoms (H')	2.99	1.84	2.74	3.62	3.91	3.28	2.45	2.41	3.62	3.14	3.34	3.00
Bahls Similarity Index (%)	26.1	18.1	36.5	43.4	32.0	N/A	N/A	23.3	33.7	33.9	40.4	49.0
Autotrophic Index	228	406	660	579	144	-9,640	367	5,050	247	488	418	365
TOLERANCE METRICS												
Percent Tolerant Diatoms	1.2	0.9	0.9	4.7	12.3	13.8	5.1	8.3	26.7	10.3	25.5	4.0
Lange-Bertalot Pollution Index	2.81	2.95	2.88	2.41	2.19	2.46	2.78	2.75	2.18	2.62	2.28	2.78
TROPHIC HABIT METRICS												
Percent Eutrophic Diatoms	35.3	60.0	52.9	55.6	45.5	60.0	50.0	41.2	46.7	47.1	31.3	44.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	47.1	60.0	58.8	55.6	40.9	66.7	61.1	41.2	53.3	47.1	43.8	50.0
Percent Nitrogen Heterotrophs	11.8	10.0	11.8	16.7	13.6	20.0	16.7	17.6	20.0	11.8	12.5	22.0
Percent High Oxygen Diatoms	29.4	50.0	52.9	38.9	31.8	46.7	33.3	29.4	26.7	29.4	18.8	22.0
Percent Motile Diatoms	47.1	40.0	47.1	50.0	50.0	46.7	44.4	47.1	53.3	52.9	50.0	50.0
Percent Saprobic Diatoms	5.9	10.0	5.9	16.7	18.2	20.0	11.1	11.8	13.3	11.8	18.8	11.0

Table E-12: Periphyton population metrics for Site DC-2-BIO, South Dakota, 2000 - 2005.

Site DC-2-BIO	2000	2001	2002	2003	2004	2005
Density (cells/cm^2)	24,300,000	3,138,300	836,700	159,100	255,500	143,500
Number of Taxa	22	23	14	20	24	15

Table E-13: Periphyton population metrics for Site DC-2-BIO, South Dakota, 2011 - 2021. Data is shown for only years in which the site was sampled.

Site DC-2-BIO	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS											
Density (cells/cm^2)	161,425	1,825,935	954,051	8,851,146	1,014,763	1,469,829	3,523,436	350,055	115,714	589,501	1,122,898
Relative Density (%)	85.0	100.0	100.0	97.2	96.5	96.2	100.0	100.0	96.7	99.0	99.0
Number of Taxa (species)	19	17	17	16	18	18	12	20	25	21	20
Number of Diatom Taxa	17	17	17	15	17	17	12	20	24	20	19
Number of Periphyton Divisions	5	1	1	3	2	3	1	1	3	3	2
Number of Periphyton Genera	17	16	12	14	13	14	10	18	21	12	15
COMPOSITION METRICS											
Shannon-Weaver Diversity Index for Diatoms (H')	3.04	2.60	2.93	2.63	2.49	2.78	1.91	2.81	3.52	3.37	2.68
Bahls Similarity Index (%)	21.9	13.9	40.5	30.4	19.0	28.5	12.8	51.7	37.5	38.4	41.0
Autotrophic Index	549	278	802	350	1,285	1,347	16,357	1,535	1,788	411	1,006
TOLERANCE METRICS											
Percent Tolerant Diatoms	10.2	2.5	4.3	2.2	5.4	2.0	0.9	0.0	1.4	2.6	0.0
Lange-Bertalot Pollution Index	2.26	2.26	2.53	2.60	2.59	2.35	2.68	2.65	2.46	2.51	2.66
TROPHIC HABIT METRICS											
Percent Eutrophic Diatoms	47.1	23.5	52.9	46.7	47.1	47.1	41.7	30.0	25.0	35.0	21.0
Percent Acidiphilic Diatoms	5.9	11.8	0.0	0.0	5.9	5.9	8.3	5.0	12.5	0.0	11.0
Percent Alkaliphilic Diatoms	47.1	23.5	64.7	53.3	47.1	47.1	41.7	35.0	37.5	40.0	26.0
Percent Nitrogen Heterotrophs	17.6	11.8	11.8	6.7	11.8	17.6	16.7	5.0	4.2	10.0	5.0
Percent High Oxygen Diatoms	35.3	29.4	52.9	60.0	35.3	47.1	16.7	50.0	54.2	40.0	32.0
Percent Motile Diatoms	41.2	47.1	35.3	33.3	41.2	41.2	33.3	35.0	50.0	30.0	42.0
Percent Saprobic Diatoms	5.9	5.9	11.8	6.7	5.9	17.6	16.7	5.0	4.2	5.0	0.0

Table E-14 : Periphyton population metrics for Site EFB-1-BIO, South Dakota, 1995 - 2005.

Site EFB-1-BIO	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density (cells/cm^2)	87,600	49,400	56,000	70,400	66,700	8,786,200	2,088,600	56,100	8,600	435,500	186,500
Number of Taxa	8	2	2	5	13	18	32	20	11	20	5

Table E-15: Periphyton population metrics for Site EFB-1-BIO, South Dakota, 2006 - 2021. Data is shown for only years in which the site was sampled. No algae were observed in 2021.

Site EFB-1-BIO	2006	2009	2010	2018	2019	2020	2021
RICHNESS METRICS							
Density (cells/cm^2)	501,365	15,135	118,510	47,427	47,387	145,259	<415
Relative Density (%)	99.1	100.0	92.6	94.8	89.5	99.0	--
Number of Taxa (species)	15	18	19	17	13	20	--
Number of Diatom Taxa	14	18	18	16	11	19	--
Number of Periphyton Divisions	3	1	3	3	4	2	--
Number of Periphyton Genera	15	17	18	15	13	11	--
COMPOSITION METRICS							
Shannon-Weaver Diversity Index for Diatoms (H')	2.37	3.19	3.29	2.77	2.06	3.56	--
Bahls Similarity Index (%)	27.2	43.4	32.5	51.7	29.1	43.3	--
Autotrophic Index	6,225	1,764	656	918	2,120	1,039	--
TOLERANCE METRICS							
Percent Tolerant Diatoms	1.8	1.2	0.0	1.1	0.0	7.5	--
Lange-Bertalot Pollution Index	2.78	2.56	2.67	2.53	2.47	2.39	--
TROPHIC HABIT METRICS							
Percent Eutrophic Diatoms	42.9	44.4	44.4	43.8	18.2	47.4	--
Percent Acidiphilic Diatoms	0.0	0.0	5.6	6.3	18.2	0.0	--
Percent Alkaliphilic Diatoms	50.0	50.0	55.6	50.0	9.1	47.4	--
Percent Nitrogen Heterotrophs	7.1	11.1	0.0	6.3	0.0	15.8	--
Percent High Oxygen Diatoms	50.0	50.0	61.1	43.8	45.5	47.4	--
Percent Motile Diatoms	28.6	38.9	38.9	25.0	36.4	47.4	--
Percent Saprobic Diatoms	7.1	16.7	5.6	12.5	0.0	21.1	--

Table E-16: Periphyton population metrics for Site WFB-1-BIO, South Dakota, 2007 - 2021. Data is shown for only years in which the site was sampled.

Site WFB-1-BIO	2007	2008	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS													
Density (cells/cm^2)	360,080	44,542	36,272	98,482	113,584	1,486,017	130,377	309,968	120,204	231,781	102,572	38,391	17,920
Relative Density (%)	98.2	85.3	100.0	96.0	96.0	99.2	100.0	90.7	60.9	48.4	100.0	100.0	87.0
Number of Taxa (species)	25	16	12	8	11	7	8	14	11	11	13	11	12
Number of Diatom Taxa	23	15	12	7	9	6	8	13	9	10	13	11	9
Number of Periphyton Divisions	4	3	1	3	5	3	1	3	5	2	1	1	3
Number of Periphyton Genera	20	16	12	7	11	7	7	13	10	10	12	8	11
COMPOSITION METRICS													
Shannon-Weaver Diversity Index for Diatoms (H')	3.70	3.04	2.62	1.14	1.76	1.05	2.55	2.26	1.86	2.50	2.60	1.41	1.82
Bahls Similarity Index (%)	25.1	8.5	15.2	3.5	3.8	6.3	11.2	12.4	1.6	2.7	6.5	2.2	10.0
Autotrophic Index	2,180	2,442	1,477	455	974	507	942	1,039	18,940	451	2,176	826	1,055
TOLERANCE METRICS													
Percent Tolerant Diatoms	7.3	4.6	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	1.1	0.0
Lange-Bertalot Pollution Index	2.35	2.37	2.29	2.90	2.56	2.12	2.22	2.68	2.77	2.53	2.27	2.84	2.83
TROPHIC HABIT METRICS													
Percent Eutrophic Diatoms	65.2	33.3	41.7	14.3	22.2	16.7	25.0	30.8	22.2	10.0	30.8	18.2	22.0
Percent Acidiphilic Diatoms	0.0	20.0	16.7	28.6	33.3	16.7	12.5	23.1	22.2	20.0	23.1	27.3	22.0
Percent Alkaliphilic Diatoms	56.5	46.7	50.0	28.6	11.1	16.7	25.0	23.1	22.2	20.0	38.5	27.3	33.0
Percent Nitrogen Heterotrophs	17.4	20.0	8.3	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	11.0
Percent High Oxygen Diatoms	39.1	40.0	33.3	57.1	55.6	50.0	62.5	53.8	44.4	50.0	53.8	63.6	67.0
Percent Motile Diatoms	65.2	40.0	16.7	14.3	22.2	16.7	12.5	30.8	22.2	10.0	15.4	18.2	33.0
Percent Saprobic Diatoms	17.4	13.3	0.0	0.0	11.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0

Table E-17: Periphyton population metrics for Site CC-1A-BIO, South Dakota, 2006 - 2021. Data is shown for only years in which the site was sampled.

Site CC-1A-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2018
RICHNESS METRICS											
Density (cells/cm^2)	5,663,748	3,001,940	1,539,484	83,866	231,339	72,316	355,256	1,561,471	3,851,376	91,714	1,384,744
Relative Density (%)	100.0	100.0	100.0	100.0	83.1	100.0	100.0	100.0	100.0	89.6	99.2
Number of Taxa (species)	10	11	17	13	15	18	15	14	13	19	13
Number of Diatom Taxa	10	11	17	13	14	18	15	14	13	18	12
Number of Periphyton Divisions	1	1	1	1	3	1	1	1	1	2	2
Number of Periphyton Genera	10	10	13	13	15	14	12	12	13	10	12
COMPOSITION METRICS											
Shannon-Weaver Diversity Index for Diatoms (H')	2.03	2.90	2.78	3.22	2.82	3.04	2.28	3.10	2.32	3.58	2.99
Bahls Similarity Index (%)	41.5	42.5	24.8	27.3	32.3	46.8	33.6	25.6	8.2	N/A	28.4
Autotrophic Index	2,350	223	714	434	758	909	440	352	608	N/A	240
TOLERANCE METRICS											
Percent Tolerant Diatoms	29.3	39.4	39.4	15.1	19.4	23.6	4.9	45.9	24.3	22.1	19.4
Lange-Bertalot Pollution Index	1.89	1.87	1.83	2.13	2.05	2.10	2.54	1.70	1.91	2.10	2.19
TROPHIC HABIT METRICS											
Percent Eutrophic Diatoms	30.0	27.3	41.2	30.8	35.7	33.3	33.3	50.0	30.8	33.3	41.7
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	6.7	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	40.0	45.5	47.1	38.5	57.1	44.4	40.0	57.1	38.5	44.4	50.0
Percent Nitrogen Heterotrophs	10.0	18.2	11.8	23.1	21.4	16.7	20.0	35.7	23.1	16.7	33.3
Percent High Oxygen Diatoms	40.0	36.4	35.3	30.8	42.9	33.3	40.0	35.7	38.5	38.9	16.7
Percent Motile Diatoms	30.0	36.4	64.7	46.2	42.9	44.4	53.3	42.9	38.5	50.0	58.3
Percent Saprobic Diatoms	20.0	18.2	17.6	15.4	21.4	11.1	20.0	21.4	15.4	16.7	16.7

Table E-18: Periphyton population metrics for Site FC-1-BIO, South Dakota, 1993 - 2005. * = cells/sample.

Site FC-1-BIO	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density (cells/cm^2)	194,100*	616,000	102,300	64,300	106,500	95,800	67,500	4,755,200	17,113,700	74,700	142,600	600,400	118,900
Number of Taxa	10	7	4	2	8	10	11	20	20	18	15	17	7

Table E-19: Periphyton population metrics for Site FC-1-BIO, South Dakota, 2006 – 2021. Data is shown for only years in which the site was sampled. N/A = no reference site established.

Site FC-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	21,480	45,744	63,819	11,317	22,039	1,425	41,443	188,546	19,736	17,642	217,582	209,787	24,302	33,872	33,939	5,388
Relative Density (%)	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	98.0	92.9	100.0	99.0	100.0
Number of Taxa (species)	10	14	15	15	17	3	13	20	9	12	12	22	16	20	23	16
Number of Diatom Taxa	10	14	15	15	17	3	13	20	9	12	12	21	15	20	22	16
Number of Periphyton Divisions	1	1	1	1	1	1	1	1	1	1	1	3	3	1	2	1
Number of Periphyton Genera	9	8	11	13	12	3	12	15	7	5	11	18	14	12	12	11
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	2.99	1.99	2.40	2.88	3.30	1.58	3.57	2.35	3.10	2.24	1.66	2.68	2.93	3.43	3.46	3.11
Bahls Similarity Index (%)	38.0	48.9	65.4	43.5	53.9	52.6	36.6	43.3	27.6	N/A	N/A	21.2	24.1	38.1	33.1	38.0
Autotrophic Index	802	2,035	8,817	522	1,159	3,409	2,940	529	1,102	432	909	4,018	607	1,266	446	771
TOLERANCE METRICS																
Percent Tolerant Diatoms	0.0	1.8	1.9	0.0	2.1	0.0	0.0	0.7	0.0	10.0	0.6	2.7	1.9	13.2	9.8	2.0
Lange-Bertalot Pollution Index	2.63	2.40	2.42	2.35	2.19	2.33	2.59	2.72	2.18	2.54	2.93	2.79	2.69	2.26	2.49	2.65
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	70.0	50.0	53.3	40.0	58.8	66.7	38.5	40.0	55.6	50.0	58.3	57.1	46.7	50.0	59.1	56.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	7.7	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	60.0	50.0	60.0	33.3	47.1	66.7	46.2	45.0	55.6	33.3	50.0	42.9	46.7	35.0	45.5	50.0
Percent Nitrogen Heterotrophs	0.0	21.4	13.3	6.7	11.8	0.0	0.0	10.0	22.2	25.0	8.3	9.5	13.3	15.0	18.2	6.0
Percent High Oxygen Diatoms	50.0	21.4	40.0	26.7	58.8	33.3	38.5	30.0	55.6	25.0	41.7	42.9	46.7	25.0	27.3	38.0
Percent Motile Diatoms	20.0	57.1	53.3	53.3	52.9	66.7	38.5	45.0	66.7	75.0	66.7	57.1	33.3	60.0	72.7	44.0
Percent Saprobic Diatoms	0.0	14.3	6.7	6.7	5.9	0.0	0.0	10.0	11.1	16.7	16.7	14.3	6.7	20.0	9.1	6.0

Table E-20: Periphyton population metrics for Site NG-2-BIO, South Dakota, 1993 - 2005. * = cells/sample.

Site NG-2-BIO	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density(cells/cm^2)	1,336,800*	460,900	130,800	341,000	235,600	56,500	52,500	6,206,200	6,204,700	1,062,900	255,200	1,515,100	955,600
Number of Taxa	6	11	7	3	9	2	6	29	11	11	28	11	13

Table E-21: Periphyton population metrics for Site NG-2-BIO, 2006 - 2021. Data is shown for only years in which the site was sampled. N/A = Not applicable as this was a background control site.

Site NG-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	880,052	1,209,235	1,686,978	267,725	244,357	220,060	583,332	2,107,682	391,223	670,054	2,779,570	2,002,287	169,710	298,884	563,447	1,653,691
Relative Density (%)	100.0	100.0	100.0	84.1	100.0	100.0	99.1	100.0	100.0	100.0	99.2	100.0	100.0	100.0	99.0	99.0
Number of Taxa (species)	17	20	21	21	25	18	21	22	12	20	21	23	15	17	21	17
Number of Diatom Taxa	17	20	21	20	25	18	20	22	12	20	20	23	15	17	20	16
Number of Periphyton Divisions	1	1	1	3	1	1	2	1	1	1	2	1	1	1	2	2
Number of Periphyton Genera	15	18	16	15	20	15	18	17	9	10	16	17	11	11	9	10
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	3.08	3.54	3.01	3.73	4.21	3.34	3.62	3.88	2.75	3.20	3.44	3.97	2.51	3.13	3.53	2.60
Bahls Similarity Index (%)	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	N/A	17.3	19.2	41.2	34.7	30.0
Autotrophic Index	538	541	455	445	496	513	213	206	204	95	204	1,806	435	505	142	382
TOLERANCE METRICS																
Percent Tolerant Diatoms	2.9	3.7	1.9	4.7	11.7	0.7	8.1	9.6	0.0	7.5	6.4	5.8	3.9	7.8	5.9	4.0
Lange-Bertalot Pollution Index	2.48	2.43	2.59	2.42	2.23	2.30	2.45	2.25	2.24	2.01	2.34	2.40	2.75	2.52	2.61	2.59

Site NG-2-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	41.2	45.0	61.9	65.0	56.0	55.6	65.0	54.5	66.7	50.0	60.0	60.9	53.3	41.2	45.0	50.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	47.1	40.0	52.4	55.0	48.0	55.6	65.0	50.0	58.3	45.0	50.0	47.8	53.3	41.2	35.0	38.0
Percent Nitrogen Heterotrophs	11.8	15.0	23.8	15.0	16.0	16.7	30.0	22.7	8.3	20.0	20.0	17.4	20.0	17.6	20.0	19.0
Percent High Oxygen Diatoms	35.3	30.0	42.9	40.0	44.0	44.4	30.0	36.4	66.7	25.0	40.0	39.1	40.0	35.3	30.0	44.0
Percent Motile Diatoms	35.3	35.0	57.1	65.0	56.0	61.1	55.0	59.1	50.0	55.0	65.0	56.5	53.3	58.8	60.0	56.0
Percent Saprobic Diatoms	5.9	20.0	14.3	10.0	12.0	0.0	15.0	22.7	0.0	10.0	10.0	8.7	13.3	11.8	15.0	6.0

Table E-22 Periphyton population metrics for Site SG-1-BIO, South Dakota, 1993 - 2005. * = cells/sample.

Site SG-1-BIO	1993	1994	1995	1996	1997	1998	1999	2000	2001	2002	2003	2004	2005
Density(cells/cm^2)	2,939,500*	247,700	207,100	77,500	214,100	56,600	100,800	5,806,500	1,646,600	445,700	274,900	242,900	1,354,400
Number of Taxa	13	7	6	4	11	2	9	26	19	10	10	7	11

Table E-23: Periphyton population metrics for Site SG-1-BIO, South Dakota, 2006 - 2021.

Site SG-1-BIO	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015	2016	2017	2018	2019	2020	2021
RICHNESS METRICS																
Density (cells/cm^2)	1,794,085	191,330	138,576	597,915	1,679,861	71,932	470,367	1,754,926	3,473,607	188,461	554,651	280,592	847,012	83,582	192,701	219,091
Relative Density (%)	100.0	100.0	100.0	95.2	100.0	93.7	100.0	100.0	98.9	100.0	100.0	99.3	98.8	100.0	100.0	100.0
Number of Taxa (species)	11	16	12	14	17	31	14	18	15	18	15	10	18	14	10	16
Number of Diatom Taxa	11	16	12	13	17	30	14	18	14	18	15	9	17	14	10	16
Number of Periphyton Divisions	1	1	1	3	1	3	1	1	3	1	1	3	3	1	1	1
Number of Periphyton Genera	10	13	11	10	13	20	13	15	10	10	10	7	14	13	7	8
COMPOSITION METRICS																
Shannon-Weaver Diversity Index for Diatoms (H')	2.32	2.45	2.03	2.26	3.02	3.98	2.37	2.86	1.61	2.96	2.69	1.90	2.19	2.13	2.03	2.15
Bahls Similarity Index (%)	28.2	21.5	23.8	32.8	31.9	53.9	19.1	51.5	12.1	30.2	27.5	12.6	53.3	42.1	47.4	34.0
Autotrophic Index	854	613	1,221	159	182	340	150	359	186	1,377	467	10,102	224	436	385	334
TOLERANCE METRICS																
Percent Tolerant Diatoms	6.8	5.4	14.8	3.3	4.2	5.0	3.1	4.4	0.6	4.5	10.8	0.7	6.6	3.2	7.4	6.0
Lange-Bertalot Pollution Index	2.56	2.31	2.56	2.44	2.52	2.47	2.66	2.55	2.76	2.50	2.48	2.84	2.59	2.52	2.58	2.61
TROPHIC HABIT METRICS																
Percent Eutrophic Diatoms	27.3	43.8	41.7	46.2	41.2	46.7	42.9	27.8	50.0	44.4	53.3	33.3	41.2	35.7	20.0	56.0
Percent Acidiphilic Diatoms	0.0	0.0	0.0	0.0	0.0	0.0	0.0	5.6	0.0	0.0	0.0	11.1	0.0	0.0	0.0	0.0
Percent Alkaliphilic Diatoms	45.5	50.0	41.7	46.2	41.2	50.0	50.0	38.9	42.9	38.9	46.7	33.3	47.1	42.9	40.0	56.0
Percent Nitrogen Heterotrophs	9.1	12.5	16.7	15.4	11.8	16.7	14.3	11.1	7.1	16.7	26.7	11.1	11.8	21.4	10.0	19.0
Percent High Oxygen Diatoms	54.5	43.8	33.3	46.2	52.9	40.0	42.9	38.9	35.7	33.3	33.3	55.6	29.4	42.9	40.0	38.0
Percent Motile Diatoms	36.4	43.8	58.3	61.5	47.1	56.7	50.0	50.0	42.9	55.6	66.7	11.1	41.2	50.0	40.0	56.0
Percent Saprobic Diatoms	9.1	12.5	8.3	15.4	11.8	10.0	14.3	11.1	14.3	16.7	13.3	11.1	11.8	14.3	10.0	13.0

This page intentionally left blank

Appendix F 2021 Water Quality Data

This page intentionally left blank

MIDCONTINENT TESTING LABORATORIES, INC.
2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: Labador Gulch Bio
Sampled: 08/21/21 at 03:30 PM by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825211
Received: 08/23/21 at 04:00 PM by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	348	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	192	mg/L	1			SM 2340 B	GAM	08/30/21
pH	8.61	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	164	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	< 0.050	mg/L	1	0.008	0.050	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	0.019	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	46.4	mg/L	1	0.070	1.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	18.6	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.009	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	< 0.050	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.006	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **Labador Gulch**
Bio

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	360	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	200	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	160	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.74	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	8.71	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	13.5	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: RC Bio
Sampled: 08/20/21 at 01:00 PM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825202
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	308	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	124	mg/L	1			SM 2340 B	GAM	08/30/21
pH	8.12	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	152	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	2.47	mg/L	4	0.032	0.200	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	0.037	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	33.3	mg/L	1	0.070	1.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	9.98	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	< 0.005	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	0.285	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.006	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **RC Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	320	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	50.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	197	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.49	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	7.65	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	12.0	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: NPDES001 - BIO
Sampled: 08/23/21 at 10:45 AM
by Darnell Witte
Sample Matrix: Water

Lab ID#: 20210826109
Received: 08/24/21 at 12:40 PM
by Julie Muzzy
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	484	μmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/27/21
Hardness	238	mg/L	1			SM 2340 B	GAM	08/31/21
pH	8.50	S.U.	1			SM 4500-H+ B	JAM	08/27/21
Total Dissolved Solids	242	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/26/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/26/21
Non-Metallics								
BOD, 5-Day	< 3.0	mg/L	150ml			SM 5210	SAA	08/25/21
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/27/21
Nitrogen, Ammonia (NH3)	< 0.050	mg/L	1	0.009	0.050	SM 4500-NH3 D	JAM	08/26/21
Nitrogen, Nitrate (NO3)	2.91	mg/L	5	0.039	0.250	SM 4500-NO3 F	BLL	08/30/21
Phosphorus (P) Dissolved	0.015	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	51.9	mg/L	2	0.140	2.00	SM 3111 B	GRT	08/27/21
Magnesium (Mg)	26.3	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/27/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.011	mg/L	10	0.00061	0.005	EPA 200.8	TNA	08/30/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	TNA	08/30/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	TNA	08/30/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	TNA	08/30/21
Iron (Fe)	< 0.050	mg/L	10	0.00093	0.050	EPA 200.8	TNA	08/30/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	TNA	08/30/21
Nickel (Ni)	< 0.005	mg/L	10	0.00008	0.005	EPA 200.8	TNA	08/30/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	TNA	08/30/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	TNA	08/30/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **NPDES001 - BIO**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Metals - Total Recoverable								
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	TNA	08/30/21
Field Test								
Field Conductivity	480	µmhos/cm	1			Field Conductivity	BLL	08/26/21
Field Flow Rate	0.300	gal/min	1			Field Flow	BLL	08/26/21
Field Oxygen (O2)	8.79	mg/L	1			Field Oxygen	BLL	08/26/21
Field pH	8.60	S.U.	1			Field pH	BLL	08/26/21
Field Temperature	10.9	°C	1			Field Temp.	BLL	08/26/21

Report Approved By:

Report Approved On: 9/2/2021 9:06:52 AM

MIDCONTINENT TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: NPDES002 - BIO
Sampled: 08/23/21 at 10:50 AM by Darnell Witte
Sample Matrix: Water

Lab ID#: 20210826110
Received: 08/24/21 at 12:40 PM by Julie Muzzy
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	498	μmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/27/21
Hardness	241	mg/L	1			SM 2340 B	GAM	08/31/21
pH	8.41	S.U.	1			SM 4500-H+ B	JAM	08/27/21
Total Dissolved Solids	264	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/26/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/26/21
Non-Metallics								
BOD, 5-Day	< 3.0	mg/L	150ml			SM 5210	SAA	08/25/21
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/27/21
Nitrogen, Ammonia (NH3)	< 0.050	mg/L	1	0.009	0.050	SM 4500-NH3 D	JAM	08/26/21
Nitrogen, Nitrate (NO3)	0.313	mg/L	1	0.008	0.050	SM 4500-NO3 F	BLL	08/30/21
Phosphorus (P) Dissolved	0.012	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	48.7	mg/L	2	0.140	2.00	SM 3111 B	GRT	08/27/21
Magnesium (Mg)	29.1	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/27/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.006	mg/L	10	0.00061	0.005	EPA 200.8	TNA	08/30/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	TNA	08/30/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	TNA	08/30/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	TNA	08/30/21
Iron (Fe)	< 0.050	mg/L	10	0.00093	0.050	EPA 200.8	TNA	08/30/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	TNA	08/30/21
Nickel (Ni)	< 0.005	mg/L	10	0.00008	0.005	EPA 200.8	TNA	08/30/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	TNA	08/30/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	TNA	08/30/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **NPDES002 - BIO**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst/Date	
Metals - Total Recoverable								
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	TNA	08/30/21
Field Test								
Field Conductivity	480	µmhos/cm	1			Field Conductivity	BLL	08/26/21
Field Flow Rate	0.040	gal/min	1			Field Flow	BLL	08/26/21
Field Oxygen (O2)	9.73	mg/L	1			Field Oxygen	BLL	08/26/21
Field pH	8.55	S.U.	1			Field pH	BLL	08/26/21
Field Temperature	11.9	° C	1			Field Temp.	BLL	08/26/21

Report Approved By:

Report Approved On: 9/2/2021 9:06:52 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: Lost Camp - BIO
Sampled: 08/23/21 at 10:35 AM
by Darnell Witte
Sample Matrix: Water

Lab ID#: 20210826111
Received: 08/24/21 at 12:40 PM
by Julie Muzzy
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	312	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/27/21
Hardness	147	mg/L	1			SM 2340 B	GAM	08/31/21
pH	8.27	S.U.	1			SM 4500-H+ B	JAM	08/27/21
Total Dissolved Solids	152	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/26/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/26/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/27/21
Nitrogen, Nitrate (NO3)	0.591	mg/L	1	0.008	0.050	SM 4500-NO3 F	BLL	08/30/21
Phosphorus (P) Dissolved	0.042	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	41.9	mg/L	1	0.070	1.00	SM 3111 B	GRT	08/27/21
Magnesium (Mg)	10.3	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/27/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	< 0.005	mg/L	10	0.00061	0.005	EPA 200.8	TNA	08/30/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	TNA	08/30/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	TNA	08/30/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	TNA	08/30/21
Iron (Fe)	0.063	mg/L	10	0.00093	0.050	EPA 200.8	TNA	08/30/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	TNA	08/30/21
Nickel (Ni)	< 0.005	mg/L	10	0.00008	0.005	EPA 200.8	TNA	08/30/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	TNA	08/30/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	TNA	08/30/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	TNA	08/30/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **Lost Camp - BIO**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst/Date	
Field Test								
Field Conductivity	310	µmhos/cm	1			Field Conductivity	BLL	08/26/21
Field Flow Rate	45.0	gal/min	1			Field Flow	BLL	08/26/21
Field ORP	204	mV	1			Field ORP	BLL	08/26/21
Field Oxygen (O2)	8.61	mg/L	1			Field Oxygen	BLL	08/26/21
Field pH	8.18	S.U.	1			Field pH	BLL	08/26/21
Field Temperature	11.8	°C	1			Field Temp.	BLL	08/26/21

Report Approved By:

Report Approved On: 9/2/2021 9:06:52 AM

MIDCONTINENT TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: NPDES005 - BIO
Sampled: 08/23/21 at 10:25 AM by Darnell Witte
Sample Matrix: Water

Lab ID#: 20210826107
Received: 08/24/21 at 12:40 PM by Julie Muzzy
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	455	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/27/21
Hardness	230	mg/L	1			SM 2340 B	GAM	08/31/21
pH	8.52	S.U.	1			SM 4500-H+ B	JAM	08/27/21
Total Dissolved Solids	231	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/26/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/26/21
Non-Metallics								
BOD, 5-Day	< 3.0	mg/L	150ml			SM 5210	SAA	08/25/21
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/27/21
Nitrogen, Ammonia (NH3)	< 0.050	mg/L	1	0.009	0.050	SM 4500-NH3 D	JAM	08/26/21
Nitrogen, Nitrate (NO3)	2.05	mg/L	2	0.016	0.100	SM 4500-NO3 F	BLL	08/30/21
Phosphorus (P) Dissolved	0.026	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	54.1	mg/L	2	0.140	2.00	SM 3111 B	GRT	08/27/21
Magnesium (Mg)	23.1	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/27/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.010	mg/L	10	0.00061	0.005	EPA 200.8	TNA	08/30/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	TNA	08/30/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	TNA	08/30/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	TNA	08/30/21
Iron (Fe)	0.061	mg/L	10	0.00093	0.050	EPA 200.8	TNA	08/30/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	TNA	08/30/21
Nickel (Ni)	< 0.005	mg/L	10	0.00008	0.005	EPA 200.8	TNA	08/30/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	TNA	08/30/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	TNA	08/30/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **NPDES005 - BIO**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst/Date	
Metals - Total Recoverable								
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	TNA	08/30/21
Field Test								
Field Conductivity	450	µmhos/cm	1			Field Conductivity	BLL	08/26/21
Field Flow Rate	175	gal/min	1			Field Flow	BLL	08/26/21
Field Oxygen (O2)	9.26	mg/L	1			Field Oxygen	BLL	08/26/21
Field pH	8.55	S.U.	1			Field pH	BLL	08/26/21
Field Temperature	11.8	°C	1			Field Temp.	BLL	08/26/21

Report Approved By:

Report Approved On: 9/2/2021 9:06:52 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: AC-3 Biomon
Sampled: 08/20/21 at 01:40 PM by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825203
Received: 08/23/21 at 04:00 PM by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	442	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	226	mg/L	1			SM 2340 B	GAM	08/30/21
pH	8.53	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	231	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	4.84	mg/L	5	0.039	0.250	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	0.049	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	56.0	mg/L	2	0.140	2.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	21.0	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.017	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	0.144	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	0.002	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.388	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	0.083	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **AC-3 Biomon**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	440	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	222	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	187	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	9.05	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	8.37	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	12.4	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

[signature]

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: DC-2-Bio
Sampled: 08/20/21 at 11:30 AM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825201
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	268	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	113	mg/L	1			SM 2340 B	GAM	08/30/21
pH	7.58	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	147	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	< 0.050	mg/L	1	0.008	0.050	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	< 0.010	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	32.7	mg/L	1	0.070	1.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	7.64	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	< 0.005	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	1.63	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.014	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **DC-2-Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	280	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	65.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	196	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.33	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	7.23	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	12.7	°C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: EFB - Bio
Sampled: 08/21/21 at 11:50 AM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825210
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	620	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	295	mg/L	1			SM 2340 B	GAM	08/30/21
pH	7.79	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	392	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	5.98	mg/L	4	0.032	0.200	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	< 0.010	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	94.6	mg/L	3	0.210	3.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	14.4	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	< 0.005	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	0.386	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.012	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	0.009	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **EFB - Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	630	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	95.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	154	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.33	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	7.50	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	13.2	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: WFB - Bio
Sampled: 08/21/21 at 11:40 AM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825209
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1002 - WHARF RESOURCES(USA),INC

MATT ZIETLOW
WHARF RESOURCES(USA),INC.
10928 WHARF ROAD
LEAD, SD 577549710

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	169	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	51.4	mg/L	1			SM 2340 B	GAM	08/30/21
pH	5.05	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	106	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	< 0.050	mg/L	1	0.008	0.050	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	< 0.010	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	12.4	mg/L	1	0.070	1.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	4.97	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	< 0.005	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	< 0.050	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.036	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	0.061	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **WHARF RESOURCES(USA),INC** Sample Site: **WFB - Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	170	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	15.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	199	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.30	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	5.82	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	13.8	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: NG-2-Bio
Sampled: 08/21/21 at 10:45 AM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825207
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1007 - GOLDEN REWARD MINING CO.

MATT ZIETLOW
GOLDEN REWARD MINING CO.
10928 WHARF ROAD
LEAD, SD 57754

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	1170	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	414	mg/L	1			SM 2340 B	GAM	08/30/21
pH	8.10	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	661	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	14.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	0.267	mg/L	1	0.008	0.050	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	< 0.010	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	117	mg/L	4	0.280	4.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	29.6	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.015	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	0.002	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	0.943	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	0.002	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.009	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **GOLDEN REWARD MINING CO.** Sample Site: **NG-2-Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	1090	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	20.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	196	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.88	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	8.13	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	12.2	° C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: FC-Bio
Sampled: 08/21/21 at 10:30 AM by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825206
Received: 08/23/21 at 04:00 PM by Eric Fuehrer
Account: W1007 - GOLDEN REWARD MINING CO.

MATT ZIETLOW
GOLDEN REWARD MINING CO.
10928 WHARF ROAD
LEAD, SD 57754

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	781	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	424	mg/L	1			SM 2340 B	GAM	08/30/21
pH	7.79	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	518	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	1.14	mg/L	2	0.016	0.100	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	< 0.010	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	107	mg/L	4	0.280	4.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	38.2	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.009	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	0.344	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.009	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	< 0.005	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **GOLDEN REWARD MINING CO.** Sample Site: **FC-Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	740	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	20.0	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	214	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	7.72	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	7.64	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	12.7	°C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM

MIDCONTINENT
TESTING LABORATORIES, INC.

2381 South Plaza Drive P.O. Box 3388 Rapid City, SD 57709
(605) 348-0111 -- www.thechemistrylab.com

Sample Site: SG-Bio
Sampled: 08/21/21 at 09:50 AM
by Brett Scheeder
Sample Matrix: Water

Lab ID#: 20210825204
Received: 08/23/21 at 04:00 PM
by Eric Fuehrer
Account: W1007 - GOLDEN REWARD MINING CO.

MATT ZIETLOW
GOLDEN REWARD MINING CO.
10928 WHARF ROAD
LEAD, SD 57754

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Physical Properties								
Electrical Conductivity	681	µmhos/cm	1	0.153	5.00	SM 2510B	JAM	08/25/21
Hardness	358	mg/L	1			SM 2340 B	GAM	08/30/21
pH	7.97	S.U.	1			SM 4500-H+ B	JAM	08/25/21
Total Dissolved Solids	430	mg/L	100ml	13.0	50.0	SM 2540 C	JNG	08/25/21
Total Suspended Solids	< 10.0	mg/L	100ml	2.37	10.0	SM 2540 D	JNG	08/25/21
Non-Metallics								
Cyanide, WAD	< 0.010	mg/L	1	0.00054	0.010	Kelada 01	TMN	08/25/21
Nitrogen, Nitrate (NO3)	4.52	mg/L	4	0.032	0.200	SM 4500-NO3 F	JEP	08/25/21
Phosphorus (P) Dissolved	0.016	mg/L	1			SM 4500-P E	SAA	08/27/21
Metals - Dissolved								
Calcium (Ca)	111	mg/L	4	0.280	4.00	SM 3111 B	GRT	08/25/21
Magnesium (Mg)	19.7	mg/L	1	0.036	0.500	SM 3111 B	GRT	08/25/21
Metals - Total								
Mercury (Hg)	< 0.0002	mg/L	1	0.00005	0.0002	EPA 245.1	GRT	08/26/21
Metals - Total Recoverable								
Arsenic (As)	0.015	mg/L	10	0.00061	0.005	EPA 200.8	EJF	08/25/21
Cadmium (Cd)	< 0.001	mg/L	10	0.00024	0.001	EPA 200.8	EJF	08/25/21
Chromium (Cr)	< 0.001	mg/L	10	0.00012	0.001	EPA 200.8 DRC	EJF	08/25/21
Copper (Cu)	< 0.005	mg/L	10	0.00016	0.005	EPA 200.8	EJF	08/25/21
Iron (Fe)	< 0.050	mg/L	10	0.00093	0.050	EPA 200.8	EJF	08/25/21
Lead (Pb)	< 0.001	mg/L	10	0.00019	0.001	EPA 200.8	EJF	08/25/21
Nickel (Ni)	0.006	mg/L	10	0.00008	0.005	EPA 200.8	EJF	08/25/21
Selenium (Se)	0.006	mg/L	10	0.00077	0.005	EPA 200.8	EJF	08/25/21
Silver (Ag)	< 0.001	mg/L	10	0.00008	0.001	EPA 200.8	EJF	08/25/21
Zinc (Zn)	< 0.050	mg/L	10	0.00044	0.050	EPA 200.8	EJF	08/25/21

Report of Analysis for: **GOLDEN REWARD MINING CO.** Sample Site: **SG-Bio**

Parameter	Result	Units	DF	MDL	PQL	Method	Analyst	Date
Field Test								
Field Conductivity	690	µmhos/cm	1			Field Conductivity	BLL	08/24/21
Field Flow Rate	532	gal/min	1			Field Flow	BLL	08/24/21
Field ORP	229	mV	1			Field ORP	BLL	08/24/21
Field Oxygen (O2)	8.83	mg/L	1			Field Oxygen	BLL	08/24/21
Field pH	7.39	S.U.	1			Field pH	BLL	08/24/21
Field Temperature	10.7	°C	1			Field Temp.	BLL	08/24/21

Report Approved By:

Report Approved On: 8/30/2021 10:13:40 AM