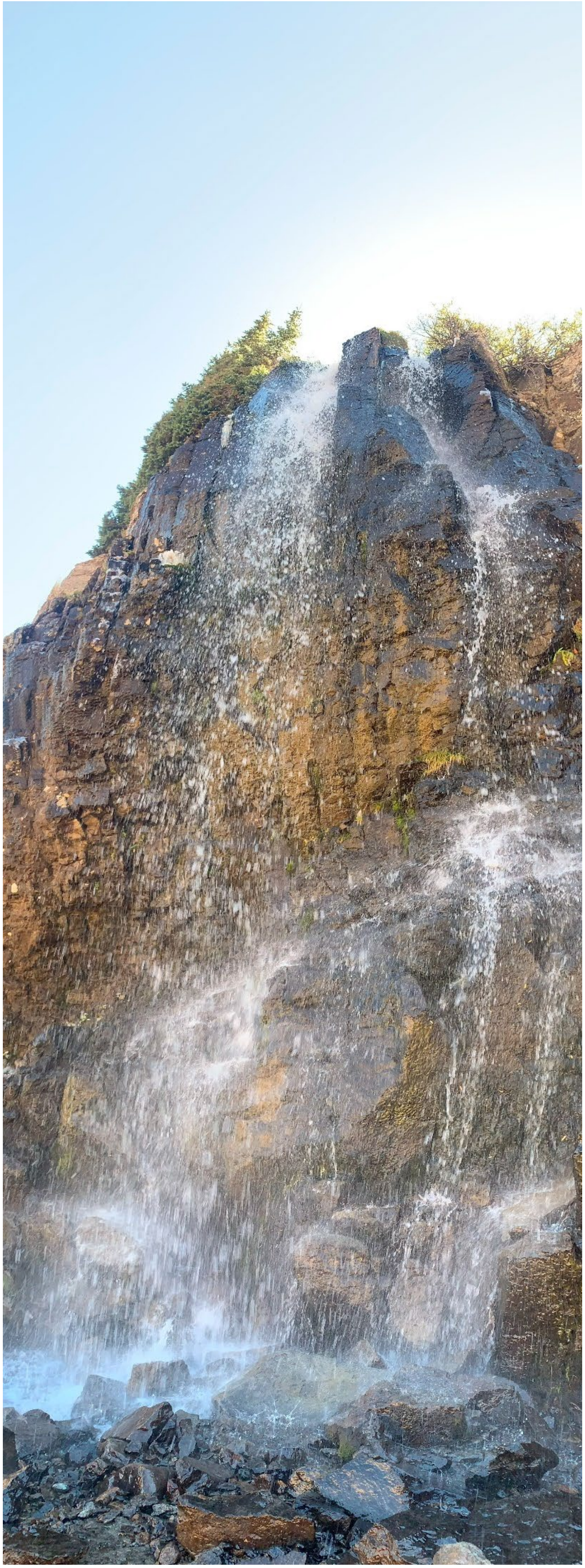

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SAN JUAN MOUNTAINS COLORADO

Water Quality and Benthic Macroinvertebrate Response Following Remediation of the Bullion King Mine Waste Rock Site

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Introduction

The Animas River Stakeholder Group (ARSG) characterized 330 abandoned mine sites in the 1990s in an effort to identify sources of metal loading in the Animas River Watershed. The Bullion King Mine site located at 12,300 feet in the Porphyry Gulch basin was identified by ARSG as a primary contributor of iron, cadmium, aluminum, and zinc to Mineral Creek, a tributary to the Animas River and was one of the top 33 waste piles identified for reclamation in the Upper Animas Use Attainability Analysis. In 2015 through 2016, the Colorado Division of Reclamation and Mining Safety (DRMS) remediated the Bullion King Mine. The remediation objectives included reducing the potential for snowmelt, storm runoff, and mine drainage to mobilize metals in the waste rock and enter Porphyry Gulch. Approximately 50,000 square of mine waste was capped with a polypropylene impervious liner and 10,000 square feet of over-steepened slopes were amended with Portland cement. The entire area was capped with on-site cover material and revegetated, and the mine drainage was re-routed away from the capped repository (Butler 2018).

The Bullion King waste rock project was started prior to the National Priority Listing of the Bonita Peak Superfund site, and was completed after the listing. Numerous remediation projects are ongoing or proposed in the Bonita Peak Mining District (BPMD), but it is unclear whether this type of remediation can have a demonstrable downstream benefit to aquatic life.

Objectives

Our primary objective is to determine if remediation of the Bullion King Mine site resulted in measurable improvements in downstream water quality and benthic macroinvertebrate community composition. Specific questions include:

1. Following remediation, did water quality improve downstream?

2. Prior to remediation, metal concentrations increased in Porphyry Gulch surface water as it flowed past the Bullion King Mine. Does this increase still occur and if so, does it occur at the same rate after remediation was complete?

3. Were there any shifts in benthic macroinvertebrate community composition at sites downstream of the Bullion King Mine following remediation?

Methods

Water quality data

We compiled water quality data for Porphyry Gulch sampling sites (**Figure 1**, **Table C- 1**) from various sources (**Table 1**) and grouped them into pre-remediation and post-remediation time periods. We considered all data collected prior to and during 2016 as pre-remediation and all data collected after 2016 as post-remediation. We synthesized pre-remediation data from online databases and previous reports. Post-remediation data were primarily collected by Mountain Studies Institute (MSI) and DRMS and supplemented with data collected by the US Environmental Protection Agency (EPA). Sample site names varied across sampling entities and were aligned prior to analysis based on site descriptions and coordinates (**Table C- 1**).

In 2022 and 2023, we collected grab water quality samples and manually measured discharge during high-flow and low-flow conditions at four locations:

- M06E: Porphyry Gulch above Bullion King Mine
- M06C: Bullion King Mine at adit
- M06B: Porphyry Gulch immediately below Bullion King Mine
- M06: Porphyry Gulch below Bullion King Mine, further downstream just upstream of Hwy 550

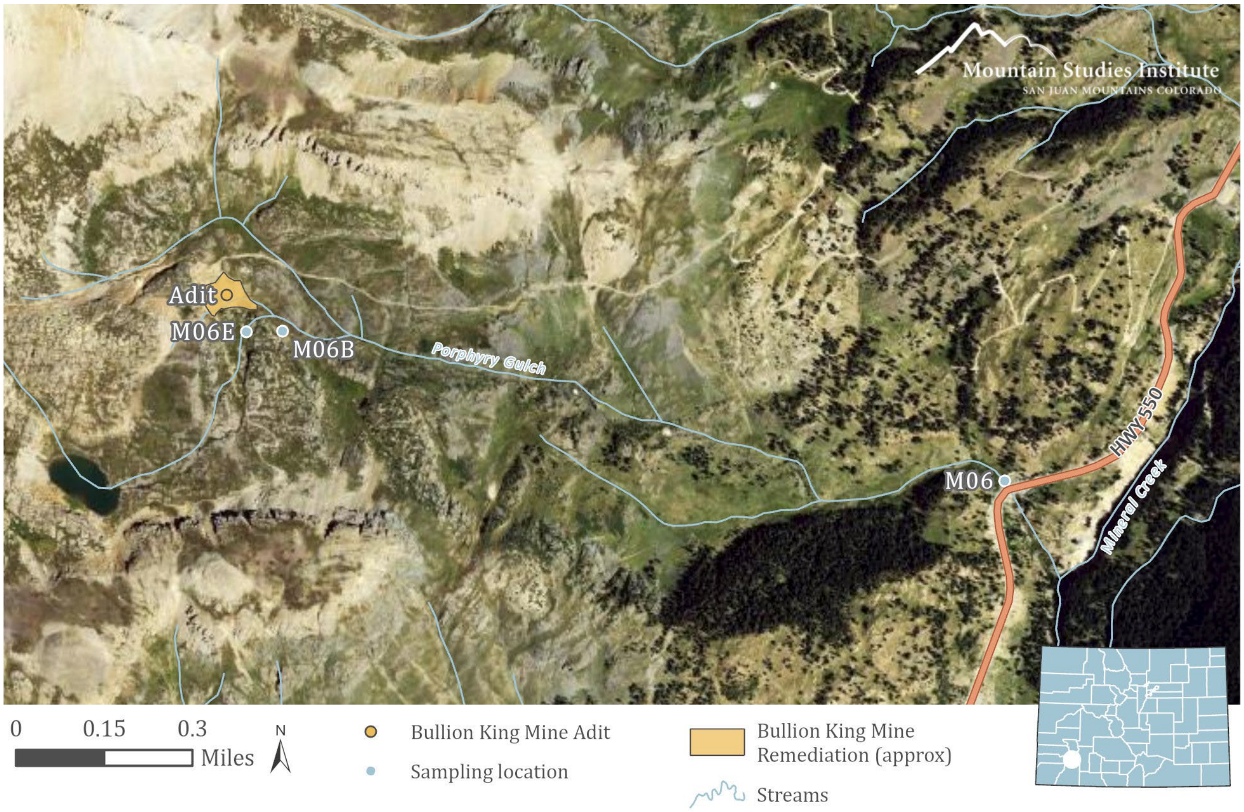

Figure 1. General Porphyry Gulch surface water sampling locations, see Table C- 1 for more information.

Table 1. Porphyry Gulch water quality data sources before and after remediation activities in 2015.

Data source	*Sampling agencies*	*Dates sampled*	*Sites sampled*
WQX database	Animas River Stakeholder Group (ARSG)	07/21/2015, 10/09/2015	M06
	Colorado Department of Health (CDPHE)	09/06/1991, 06/23/1992	M06
BPMD website	Bureau of Land Management (BLM)	9/21/2000*^	M06B, M06C, M06E
	USGS	8/24/1999, 8/25/1999	M06, M06B, M06C, M06E
ARSG Peter Butler, Ph.D	Animas River Stakeholder Group (ARSG)	11/3/2016	M06
	Bureau of Land Management (BLM)	9/21/2000*	M06E, M06B
	Chris Peltz	7/18/2013*, 8/17/2013	M06B, M06C, M06E
Scribe	EPA	06/19/2019	M06
Mountain Studies Institute (MSI)	Mountain Studies Institute (MSI)	6/15/2022, 7/21/2022, 9/27/2022, 07/22/2023, 09/22/2023	M06, M06B, M06E, M06 Seep

**Storm event samples; note that on 9/21/2000 samples were collected twice, before and during a storm event*
^Overlap with other dataset but contains additional sampling times

Pre-remediation data were limited. Samples collected prior to remediation that included Porphyry Gulch sites upstream and downstream of Bullion King mine were only collected on four dates. Of those pre-remediation data, two samples were collected during storm runoff conditions and thus are not directly comparable to the post-remediation data, which were not collected during storm runoff conditions. As such, we omitted storm samples from analysis but included these pre-remediation storm samples in charts in Appendix A and Appendix B for context. **Table 2** summarizes the number of sample events at each site pre- and post-remediation. Due to limited data, we combined all samples, regardless of seasonality, for assessment. We calculated mean concentrations at individual sites pre- and post-restoration as well as the difference in concentration between M06E (upstream of mine site) and M06B (downstream of mine site). Data analysis excluded data from M06C (the mine adit site) as our primary objectives were to identify downstream improvements and upstream to downstream changes in water quality.

Table 2. Number of sampling events pre and post remediation at Bullion King Mine.

Time period		*M06*	*M06B*	*M06C*	*M06E*
Pre-remediation	Runoff/summer flow	3	-	-	-
	Fall baseflow	5	4	4	4
	Storm event	-	2	2	2
Post-remediation	Runoff/summer flow	4	3	2	3
	Fall baseflow	2	3	1	2

Benthic macroinvertebrate sampling

Field sampling

We followed BMI sampling protocols developed by the Environmental Protection Agency (Barbour *et al.,* 1999) and Colorado Department of Public Health and Environment (CDPHE, 2016). Anderson (2007) assessed a variety of BMI sampling methods and determined that the most appropriate method for use in the Animas River watershed was a targeted riffle method that utilized a modified rectangular dip net coupled with a dolphin bucket. The net opening measured 46 cm by 25 cm or 0.115 m^2 (178 in^2). We collected each sample by placing the net securely on the bottom of the river with the net opening facing upstream. A biologist stood downstream of the net and disturbed the substrate on the river bottom that was immediately upstream of the net. We lifted and scrubbed rocks and gravel by hand for approximately 30 seconds so that benthic macroinvertebrates would be dislodged and drift downstream into the net opening. For each sample, we disturbed an area of approximately 0.115 m^2 of substrate, which was estimated in the field by using the size of the net opening as a guide (net opening is 46 cm by 25 cm; area of 0.115 m2). Within riffle habitat, we obtained twenty samples within an approximately 75 meter-long section of the Porphyry Gulch. We then made a composite of the twenty samples in a single sample container. In total, 2.3 m^2 of riffle habitat comprised the sample at each site (0.115m^2 x 20 samples).

Laboratory Methods – BMI Community Samples

Samples were identified by Scott Roberts (Mountain Studies Institute) and Dr. Michael Bogan (University of Arizona). We sub-sampled each field sample using a rotating drum splitter until a minimum of 300 organisms was obtained. Using a 10x microscope, we identified organisms to the lowest practical taxonomic level based on Merritt, Cummins, and Berg (2019). Dr. Bogan identified all Chironomidae and Acari taxa and served as a second taxonomist for our quality assurance program by independently verifying at least 10% of all taxa. To eliminate potential bias from differing lab subsample sizes, we employed an algorithm to randomly subsample all samples to a fixed count of 300 individuals. All metrics discussed in this report are based on the 300 count subsampled data. We utilized the Ecological Data Application System

(EDAS) developed by Colorado Department of Public Health and the Environment (CDPHE) to calculate all metrics.

Several metrics have been developed to assess the composition and health of BMI communities. These relatively independent metrics provide multiple lines of evidence of the overall habitat condition and water quality of an aquatic system. We focus our analysis on metrics that Roberts (2017a) found to most strongly correlate with metal exposure in the BPMD and those that Roberts (2020) found to have the lowest inter-annual variability. These include the Multi-Metric Index (MMI); richness of metal-sensitive families (MSF); and the Modified Hilsenhoff Biotic Index (MHBI).

We applied non-metric multi-dimensional scaling ordination (NMS) within PC-ORD software (McCune and Mefford, 1999) to assess differences in benthic community structure among sites and years. Our NMS analysis was based on Bray-Curtis distance measures of species abundance. To reduce the influence of rare taxa on the ensuing ordination, we limited NMS analysis to species that occurred in at least five percent of samples (Peck, 2016).

Results

Water quality

We compared metal concentrations from surface water samples collected during the pre-remediation period to concentrations from samples collected during the post-remediation period. Many analytes had post-remediation concentrations that were within the range of data observed prior to remediation (e.g., dissolved zinc at M06). There were some instances where metal concentrations at a site were distinctly lower during the post-restoration period compared to the pre-remediation period. These instances often occurred both upstream (M06E) and downstream (M06B) of the remediation site, indicating that this phenomenon cannot be explained solely by remediation efforts (e.g., total arsenic, total and dissolved cadmium, and total and dissolved lead). However, several analytes had reduced concentrations from pre- to post-remediation that occurred downstream of the remediation site (M06B) without a corresponding reduction upstream of the remediation site (M06E). These included total aluminum, total and dissolved copper, total and dissolved manganese, and total zinc (Appendix A and B). We did not find evidence that metal concentrations were lower at the furthest downstream site, M06, following remediation.

In an attempt to minimize the influence of year-to-year variability in pre- and post-remediation data, we focused analysis on whether metal concentrations changed from upstream (M06E) to downstream (M06B) of the remediation site at the same rate during the pre- and post-remediation periods. We evaluated the magnitude of change from upstream to downstream as well as the percent change. We defined the percent change from upstream to downstream as the downstream concentration minus the upstream concentration, divided by the upstream value. Although this analysis was limited to very few pre-remediation data—1 to 3 samples for each analyte—we found that the rate of increase in concentrations of many metals diminished from upstream to downstream following restoration. For example, in the pre-remediation period, total aluminum concentrations increased 820% from M06E to M06B, but only increased 41% from M06E to M06B during the post-remediation period. This pattern of reduced rate of increase downstream of the Bullion King Mine post-remediation occurred for total aluminum, total arsenic, dissolved barium, total cadmium, dissolved calcium, total copper, total and dissolved iron, total lead, total and dissolved manganese, total nickel, dissolved potassium, dissolved sodium, sulfate, and total and dissolved zinc.

In some cases, mean concentrations upstream of the remediation site were greater than mean concentrations downstream of the remediation site. Total and dissolved magnesium and dissolved nickel had mean concentrations that were slightly higher upstream than downstream. The pattern of dissolved nickel concentrations from M06E to M06B appears to have shifted from pre- to post-remediation; pre-remediation concentrations at M06E were slightly higher than M06B (mean % change of -9.09%), while post-remediation, concentrations downstream were slightly higher (mean % change of 10.36%). For total and dissolved magnesium, concentrations upstream remained higher post-remediation but the percent change between sites declined (**Table 3**).

Table 3. Mean percent change and mean difference in concentrations from upstream (M06E) to downstream (M06B) of the Bullion King Mine before and after remediation activities. Positive percent change values reflect a mean % increase in concentration from upstream to downstream while negative percent change values reflect a reduction in concentrations from upstream to downstream. Green highlight indicates analytes where the mean change (magnitude or %) was reduced following remediation (i.e., the rate of change from upstream to downstream was lower following remediation). This analysis omits pre-remediation data collected during storm runoff conditions.

Analyte	***Pre-remediation***					***Post-remediation***				
	Mean % change from M06E to M06B	*Mean difference (M06B-M06E)*	*Mean concentration (mg/L)* M06E	M06B	*n*	*Mean % change from M06E to M06B*	*Mean difference (M06B-M06E)*	*Mean concentration (mg/L)* M06E	M06B	*n*
Aluminum,Diss	32.21	0.0090	0.0286	0.0377	3	34.63	0.0154	0.0724	0.0878	5
Aluminum,Tot	820.41	0.1260	0.2330	0.3590	2	38.25	0.0480	0.1396	0.1876	5
Arsenic,Diss	0.00	0.0000	0.0006	0.0006	2	10.00	0.00002	0.0004	0.0004	5
Arsenic,Tot	171.67	0.0013	0.0006	0.0019	2	0.00	0.0000	0.0006	0.0006	5
Barium,Diss	9.09	0.0010	0.0110	0.0120	1	2.07	0.0003	0.0142	0.0145	5
Beryllium,Diss	0.00	0.0000	0.0010	0.0010	1	4.67	0.000002	0.0003	0.0003	5
Cadmium,Diss	473.33	0.0007	0.0008	0.0014	3	1,018.67	0.0004	0.0002	0.0006	5
Cadmium,Tot	815.00	0.0012	0.0002	0.0014	2	328.00	0.0003	0.0003	0.0005	5
Calcium,Diss	8.77	0.6915	12.7945	13.4860	2	4.93	0.4460	10.9600	11.4060	5
Calcium,Tot	2.87	0.5000	17.4000	17.9000	1	4.67	0.3500	11.0900	11.4400	5
Chromium,Diss	0.00	0.0000	0.0004	0.0004	1	0.00	0.0000	0.0007	0.0007	5
Chromium,Tot	0.00	0.0000	0.0004	0.0004	1	0.00	0.0000	0.0014	0.0014	5
Copper,Diss	203.33	0.0017	0.0021	0.0038	3	405.00	0.0021	0.0005	0.0026	5
Copper,Tot	888.89	0.0064	0.0011	0.0075	2	403.98	0.0027	0.0007	0.0034	5
Iron,Diss	122.50	0.0049	0.0147	0.0196	3	24.53	0.0048	0.0332	0.0380	5
Iron,Tot	4,141.08	0.6235	0.0183	0.6418	2	44.37	0.0218	0.0480	0.0698	5
Lead,Diss	55.00	0.0001	0.0106	0.0107	3	184.44	0.0001	0.0002	0.0003	5
Lead,Tot	765.00	0.0062	0.0009	0.0071	2	87.00	0.0002	0.0004	0.0006	5
Magnesium,Diss	-3.50	-0.0550	0.7705	0.7155	2	-1.63	-0.0122	0.6248	0.6126	5
Magnesium,Tot	-9.45	-0.1040	1.1000	0.9960	1	-0.20	-0.0088	0.6330	0.6242	5
Manganese,Diss	7,321.76	0.1050	0.0241	0.1291	3	797.83	0.0358	0.0136	0.0494	5
Manganese,Tot	5,311.85	0.1490	0.0390	0.1880	2	201.04	0.0374	0.0237	0.0611	5
Molybdenum,Diss	0.00	0.0000	0.0100	0.0100	1	0.00	0.0000	0.0003	0.0003	5
Nickel,Diss	-9.09	-0.0001	0.0106	0.0105	2	10.36	0.00008	0.0008	0.0008	5
Nickel,Tot	75.00	0.0003	0.0004	0.0007	1	45.51	0.0002	0.0008	0.0010	5
Potassium,Diss	36.65	0.0900	0.2150	0.3050	2	3.48	0.0084	0.5632	0.5716	5
Potassium,Tot	0.00	0.0000	0.2000	0.2000	1	1.80	0.0048	0.6602	0.6650	5
Silver,Diss	0.00	0.0000	0.0008	0.0008	1	0.00	0.0000	0.0001	0.0001	5
Silver,Tot	0.00	0.0000	0.0008	0.0008	1	0.00	0.0000	0.0003	0.0003	5
Sodium,Diss	19.47	0.0740	0.3800	0.4540	1	-2.27	-0.0228	0.7946	0.7718	5
Sulfate	19.27	3.7333	26.9333	30.6667	3	11.27	2.4000	24.9400	27.3400	5
Vanadium,Diss	0.00	0.0000	0.0040	0.0040	1	-7.14	-0.0001	0.0027	0.0026	5
Zinc,Diss	4,562.28	0.2100	0.0137	0.2237	3	1,436.68	0.0916	0.0068	0.0984	5
Zinc,Tot	7,340.53	0.3054	0.0114	0.3168	2	1,059.88	0.0806	0.0083	0.0888	5
	n = 1-3					*n = 5*				

It appears that remediation activities reduced metal concentrations in surface water immediately downstream of the mine site (M06B), however, the patterns described above are based on severely limited data. Only one to three sampling events (depending on the analyte) captured both upstream and downstream conditions pre-remediation while post-remediation means are based on five sampling events. The limited pre-remediation data limits the degree of conclusivity of our interpretation.

Benthic Macroinvertebrates

Benthic Metrics

We collected benthic samples during pre-remediation conditions in 2015 and 2016 at M06 and M06B and during post-remediation conditions in 2022 and 2023 at M06, M06B, and M06E.

The Colorado Multi-Metric Index (MMI) was developed by CDPHE to assess the extent to which biological communities may have been altered by environmental stressors and to evaluate whether a water body is in attainment or impairment of designated aquatic life use (CDPHE 2020). All samples collected from Porphyry Gulch pre- and post-remediation are considered in attainment of aquatic life use (**Figure 2**).

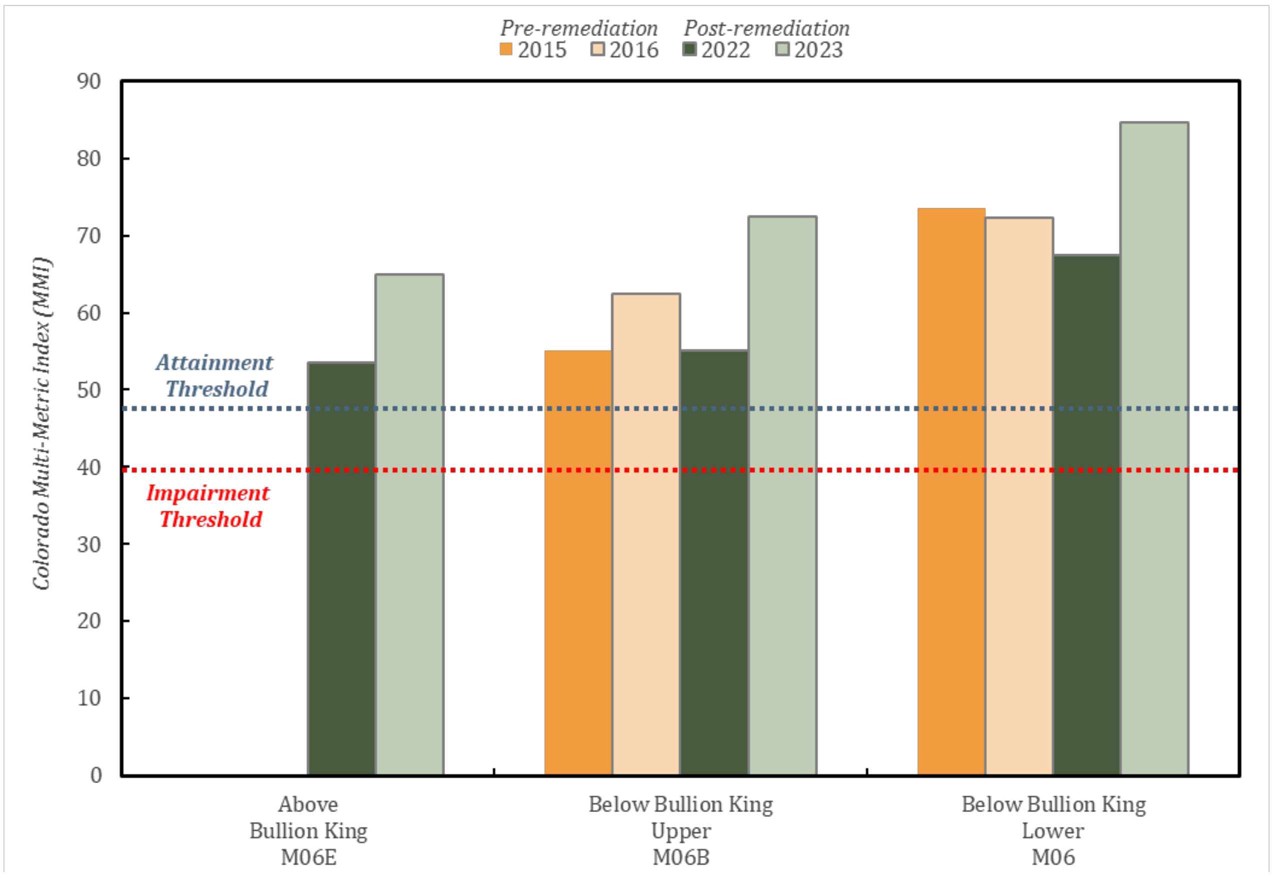

Figure 2. Colorado Multi-Metric Index (MMI) for samples collected at Porphyry Gulch.

With numerous metrics available to characterize benthic communities, it is often useful to look at multiple lines of evidence when assessing trends over time. Several benthic metrics that reflect community composition across all taxa do not readily convey a substantial change in benthic community composition downstream of Bullion King Mine following mine remediation (e.g., MMI, total taxa richness, MHBI). However, benthic metrics that reflect taxa known to be most sensitive to elevated metal contamination (Courtney and Clements, 2002) do suggest a shift in the benthic community downstream of the Bullion

King Mine following remediation. The number of metal sensitive taxa at M06B increased following remediation with the metal sensitive mayfly, *Rhithrogena*, occurring in samples collected in 2022 and 2023 and absent in samples collected in 2015 and 2016 (**Figure 3**). The relative abundance of metal sensitive taxa was higher in post-remediation samples at M06B and further downstream at M06 (**Figure 4**).

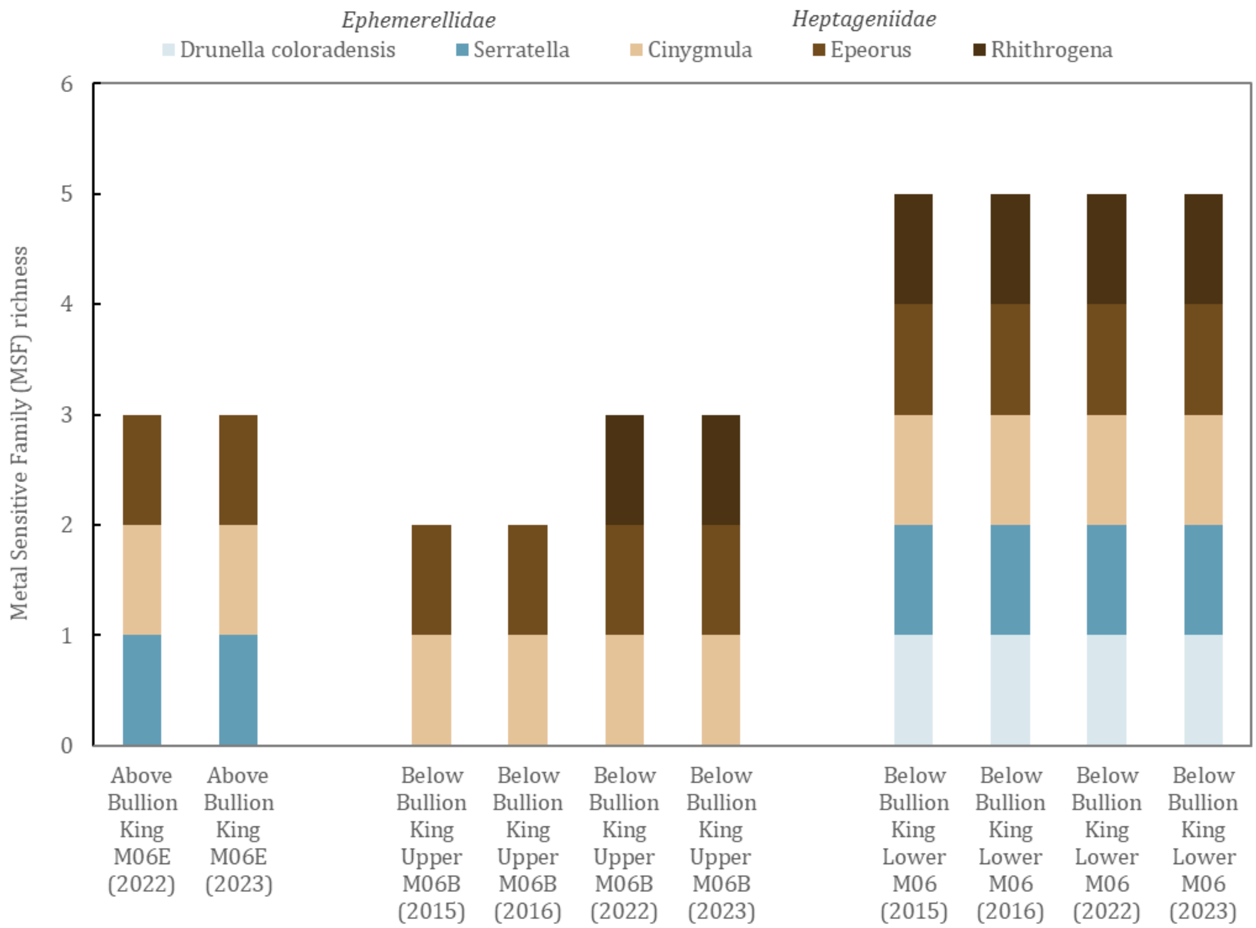

Figure 3*.* Richness of metal sensitive families for samples collected in Porphyry Gulch.

Ephemeroptera (mayflies), Plecoptera (stoneflies), and Trichoptera (caddisflies) (EPT) are known to be generally sensitive to degraded water quality (Maret et al. 2003). EPT relative abundance increased incrementally between monitoring years at M06B. Roberts (2017) noted that prior to remediation, M06B had fewer insects per square meter than other sites in the Animas River watershed. Following remediation, benthic density increased from about 150 insects per square meter in 2015-16 to 1,068 insects per square meter in 2022 and 367 insects per square meter in 2023.

Non-metric Multidimensional Scaling Ordination (NMS)

To further examine how benthic community structure may have shifted pre- and post-remediation, we used non-metric multidimensional scaling ordination (NMS), a statistical technique that plots each sample along axes in ordination space that represent gradients in community composition. Samples plotted closer to one another in ordination space have more similar community composition than samples plotted far from one another. We conducted ordination of a benthic dataset that included all Porphyry Gulch samples (M06E, M06B, and M06) as well as samples collected from nearby representative “stressed” and “reference” sites.

To represent the benthic community composition of relatively undisturbed reference sites, we included samples collected from Picayune Gulch, a reference site tributary to the Animas River, and from Boulder

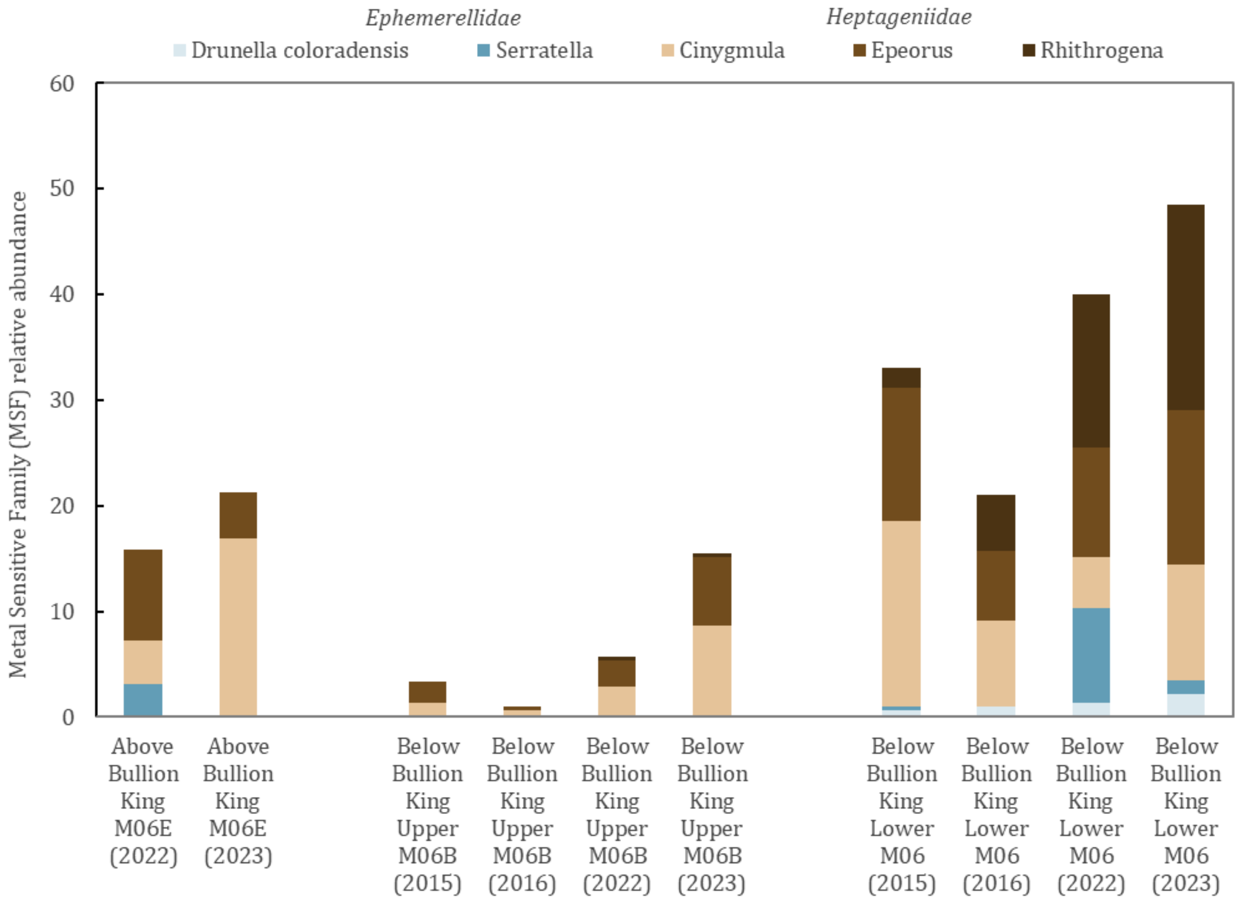

Figure 4. Relative abundance of metal sensitive families for samples collected in Porphyry Gulch

Creek and Waterfall Creek, two waterbodies that due to their good water quality were recently designated Outstanding Waters reaches and serve as drinking water sources for the towns of Silverton and Ophir. To represent benthic community composition from surface waters with elevated metal concentrations, we included a sample collected from the drainage pathway below the Bullion King Mine adit prior to remediation as well as a sample collected from Mineral Creek above South Mineral Creek. This combined dataset consisted of fifteen samples. We found that a two-dimensional solution provided the optimal ordination. The majority of the variability (61%) in benthic communities among samples was explained by NMS axis one (61%), while another roughly quarter of the variability was explained by axis two (23%). The gradient represented by axis one from left to right largely reflects an increase in the abundance of metal sensitive taxa (*Rhithrogena*, *Epeorus*, *Cinygmula*, and *Drunella coloradensis*) (Courtney and Clements, 2002) and a decrease in metal tolerant taxa (*Paraphaenocladius* and *Eukiefferiella claripennis*) (Ruse *et al.,* 2000).

NMS axis two was less intuitively related to metals and is more likely related to differences in geography and elevation among sites.

Ordination revealed that although there is inter-annual variability in benthic community structure, there is a clear difference in the community composition between reference and non-reference locations; samples from reference sites were distributed within the lower right corner of the plot and stressed sites were distributed on the left (**Figure 5**). The Porphyry Gulch samples were distributed in the mid- and upper-right portion of the plot with a distinct separation between the M06 samples and the higher elevation M06E and M06B samples.

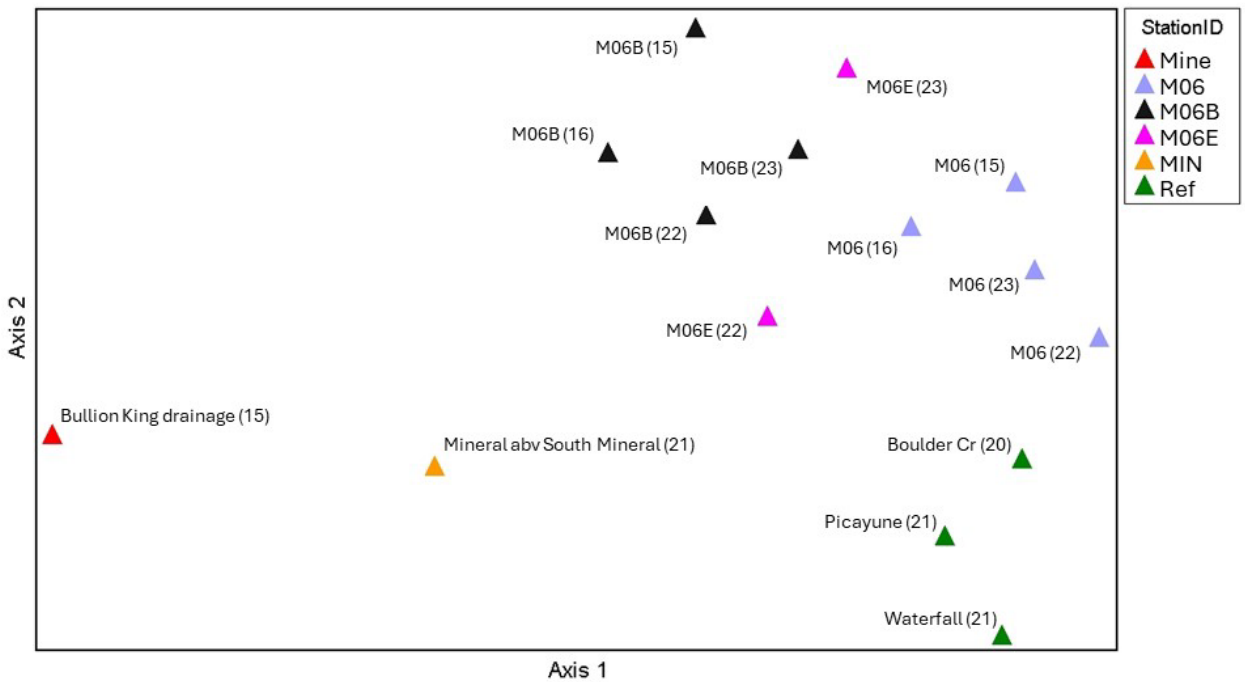

Figure 5. NMS Ordination for samples collected in the Mineral Creek basin. Note: Numbers in parenthesis indicate two-digit year sample was collected

NMS can be a useful tool for assessing how a specific benthic community sample compares to previous years and also to a corresponding reference site. Trends in benthic community structure over time can be evaluated by assessing whether the community at a particular site is trending toward or away from other sites. Following remediation, samples from M06B shifted closer in ordination space to samples from M06E and M06, indicating that community composition immediately below the remediation site became more similar to community composition upstream of the remediation site as well as the community composition further downstream of the remediation site. In ordination we can also assess whether benthic community composition of each site trended toward or away from a reference centroid (i.e., whether the quantifiable distance from the reference centroid is increasing or decreasing). We calculated distances in ordination space between M06B and M06 benthic samples and the centroid of samples collected from reference sites (**Table 4**). Comparing distances to reference centroids across years, we found that after remediation, M06B and M06 were closer in ordination space to the reference centroid than prior to remediation.

Table 4. Distance in ordination space from each sample to the reference site centroid.

Stream Name	Site Name	Reference Centroid	2015	2016	2022	2023
Porphyry Gulch	Below Bullion King Mine - Upper - M06B	Centroid of Boulder, Picayune, and Waterfall Creeks	1.87	1.72	1.37	1.38
	Below Bullion King Mine - Lower - M06		1.14	1.03	0.74	0.87

Note*: Greater distance equates to less similarity; Shorter distance equates to greater similarity. For example, M06B had benthic community composition that was more similar (shorter distance) to the reference centroid in 2022 and was less similar in 2015.*

Discussion

The limited availability of pre-remediation water quality data from Porphyry Gulch obscures our ability to detect the influence of remediation on downstream water quality conditions. Although some improvement in water quality appears to have occurred following remediation, it is difficult to conclusively attribute the improvement solely to remediation due to the limited amount of water quality data. Concentrations of several metals (total aluminum, total and dissolved copper, total and dissolved manganese, and total zinc) decreased distinctly from pre- to post-remediation without similar trends noted at the upstream control site. Additionally, the rate of change from upstream of the mine site to downstream of the mine site decreased following remediation for nearly half of all metals analyzed. ARSG characterized the Bullion King Mine as the highest contributor of iron, cadmium, and zinc and the second highest contributor of aluminum and manganese of the eight mine waste rock sites they assessed in the Mineral Creek basin. The site contributed almost half the total iron, cadmium, and zinc loads of the eight sites ARSG assessed. Following remediation there was a reduction in the rate of increase in each of these metals of concern as surface water flowed past the Bullion King Mine from M06E to M06B (Table 3). These trends in water quality improvement following remediation were not readily observed at the most downstream site, M06, suggesting that improvements may have been highly localized. Generally, concentration ranges at M06 pre-remediation appear to be within the range of or slightly lower than M06B (Appendix B).

A 2009 study examined a reach of Mineral Creek for post-remediation improvements noted circumneutral inflow from Porphyry Gulch (Runkel, 2009). The limited pre-remediation pH data for the Bullion King Mine and Porphyry Gulch suggest that pH values are roughly neutral. Many abandoned mine sites in the larger Animas River Basin drain much more acidic water, which increases the solubility of most metallic cations (e.g., copper, nickel, cobalt, and manganese). The circumneutral pH of Porphyry Gulch likely has implications for how potential post-remediation water quality changes are assessed. In circumneutral conditions, some metals can become mobile or precipitate (Tamoto *et al*., 2015). For example, in circumneutral conditions aluminum hydroxide precipitates replace dissolved aluminum as the main contributor to aluminum toxicity (Gensemer *et al*., 2018). Zinc similarly changes its speciation from a dissolved cation to a carbonate complex in circumneutral or net-alkaline waters (Nuttal and Younger, 2000). Interestingly, after remediation, the reduction of the rate of increase as surface water passes Bullion King Mine was 2 to 18 times greater for total concentrations of the five metals identified by ARSG compared to dissolved concentrations.

Due to data limitations, we were not able to differentiate seasonal flow conditions (e.g., high-flow vs. low-flow). Waste rock piles, such as the Bullion King site, may contribute to metal loading differently depending on the season. Runoff from and infiltration into waste rock piles tend to act as the primary contaminate transport mechanisms from these materials and occur most often during spring run-off or storm events. Runkel (2009) notes that waste rock pile removal may not drastically improve water quality at baseflow conditions for these reasons. With that in mind, results presented here may be missing a crucial seasonality component that, due to limited data, we could not assess. Conducting sampling during storm events and run-off conditions both pre- and post-remediation at future waste rock pile remediation sites may better capture the potential water quality improvement benefits. Pre-remediation storm samples at the Bullion King site in September 2000 and July 2013 captured 2-to 10-fold increases in metal concentrations during and after the storm event (Butler, 2018). Both rainwater and snowmelt water exhibit slightly acidic pH values (with pH values ranging between 5 and 6), which may partially explain the increased mobilization of metals from waste rock piles (Brooks *et al*., 2001; Demers *et al*., 2010; Jefferies *et al*., 1979;). Unfortunately, at the time of this report, no post-remediation storm samples have been collected. Capturing concentrations during and after storm conditions post-remediation at the Bullion King site may provide a clearer picture of remediation success. In the absence of pre- and post-remediation storm event water quality data, it is possible that summer and fall baseflow water quality may residually reflect antecedent storm events to some degree. When storm events mobilize metals from waste rock or redistribute stream bottom precipitates, these metals may accumulate, persist, and influence water quality at lower flows for perhaps an extended time period until higher flows occur that re-mobilize, dilute, or disperse these metals (Butler 2018). The concept of delayed post-restoration recovery due to residual sediment contamination has been demonstrated on the Arkansas River (Clements *et al.,* 2010).

Our clearest evidence of improvement in water quality came from assessing the rate of change in concentration as water flows from upstream of the remediation site to downstream of the remediation site. This approach allowed us to minimize the influence of year-to-year variability in pre- and post-remediation data and served as a more focused analytical metric than comparing data to a reference site from a neighboring watershed. In future assessments of mine remediation, we recommend collecting pre- and post-remediation water quality and benthic macroinvertebrate data from upstream and downstream of the remediation site. This approach is possible when the mine site is situated such that it drains to the middle of a perennial stream reach but is more problematic when mine sites are situated upslope of all perennial stream reaches and bracketing is not feasible.

We present evidence that there were measurable changes in Porphyry Gulch benthic community composition following remediation. Specifically, five years after the Bullion King Mine remediation was completed, we found an increase in benthic density, EPT relative abundance, and richness and relative abundance of metal sensitive taxa. We demonstrated through NMS ordination that benthic community composition at sites downstream of the Bullion King Mine became more similar to known regional reference sites following remediation. These post-remediation changes appear most pronounced at M06B, the site in closest downstream proximity to the mine site. The benthic community immediately downstream of the Bullion King Mine (M06B) continues to differ from the benthic communities upstream of the mine site (M06E) and further downstream of the mine site (M06). For example, post-remediation benthic density is still lower at M06B compared to M06E and M06. Additionally, metal sensitive Ephemerellidae mayfly taxa occur at M06E and M06 but were still not observed at M06B following remediation. It could be informative to continue benthic sample collection at M06B to see if further improvements in water quality and habitat conditions for aquatic life related to Bullion King remediation allow Ephemerellidae taxa to occupy M06B in the future.

With similar mine remediation projects planned for BPMD and other mineralized regions, this work has broad implications and relevant monitoring recommendations that may be applicable elsewhere. Specifically, we recommend collecting multiple lines of evidence before and after remediation that are reflective of longer-term water quality and habitat conditions at stations both immediately upstream and downstream of the remediation activity. Recommended lines of evidence include benthic macroinvertebrate community composition, streambed sediment concentrations, and autosampler devices capable of capturing continuous water quality data including storm events.

References

Anderson, C. 2007. Effects of mining on benthic macroinvertebrate communities and monitoring strategy. In S. Church, P. von Guerard, and S. Finger (Eds.), Integrated Investigations of Environmental Effects of Historical Mining in the Animas River Watershed, San Juan County, Colorado. USGS Professional Paper 1651.

Barbour, M., J. Gerritsen, B. Snyder, and J. Stribling. (1999). Rapid bioassessment protocols for use in streams and wadeable rivers: periphyton, benthic macroinvertebrates and fish, second edition. EPA 841-B-99-002. US Environmental Protection Agency; Office of Water; Washington, D.C.

Brooks, P., McKnight, D. and Bencala, K. (2001). Annual maxima in Zn concentrations during spring snowmelt in streams impacted by mine drainage. *Env Geol, 40*():1447–1454. doi: 10.1007/s002540100338

Butler, Peter. (2018). Bullion King Mine Waste Remediation Project WQC 60522. Animas River Stakeholders Group.https://static1.squarespace.com/static/53f664ede4b032c1fade347d/t/5ed94c97502173035c80d357/1591299234756/Bullion+King+Final+Report.pdf

Clements, W., N. Vieira, and S. Church. (2010). Quantifying restoration success and recovery in a metal-polluted stream: a 17-year assessment of physicochemical and biological responses. Journal of Applied Ecology, 47(4): 899-910.

Courtney, L. and W. Clements. (2002). Assessing the influence of water and substratum quality on benthic macroinvertebrate communities in a metal-polluted stream: an experimental approach. Freshwater Biology, 47: 1766-1778.

Demers, J. D., Driscoll, C. T. , and Shanley, J. B. (2010). Mercury mobilization and episodic stream acidification during snowmelt: Role of hydrologic flow paths, source areas, and supply of dissolved organic carbon, *Water Resour. Res*., 46, W01511, doi:10.1029/2008WR007021.

Jeffries, D. S., Cox, C. M. and Dillon, P. J. (1979). Depression of pH in Lakes and Streams in Central Ontario During Snowmelt. *Journal of the Fisheries Research Board of Canada*, *36*(6): 640-646. doi: 10.1139/ f79-093

Gensemer, R.W., Gondek, J.C., Rodriquez, P.H., Arbildua, J.J., Stubblefield, W.A., Cardwell, A.S., Santore, R.C., Ryan, A.C., Adams, W.J. and Nordheim, E. (2018), Evaluating the effects of pH, hardness, and dissolved organic carbon on the toxicity of aluminum to freshwater aquatic organisms under circumneutral conditions. *Environ Toxicol. Chem., 37*: 49-60. doi: 10.1002/etc.3920

Maret, T, D. Cain, D. MacCoy, and T. Short. (2003) Response of benthic macroinvertebrate assemblages to metal exposure and bioaccumulation associated with hard-rock mining in northwestern streams, USA. *Journal of North American Benthological Society, 22*(4): 598-620.

Nuttel, C.A. and Younger, P.L. (2000). Zinc removal from hard, circum-neutral mine waters using a novel closed-bed limestone reactor. *Water Research, 34*(4): 1262-1268. doi: 10.1016/S0043-1354(99)00252-3.

Roberts, S. (2017). Porphyry Gulch 2017 Benthic Macroinvertebrate Assessment. Prepared by Mountain Studies Institute for Animas River Stakeholders Group.

Runkel, R. L., Bencala, K. E., Kimball, B. A., Walton-Day, K., and Verplanck, P. L. (2009). A comparison of pre- and post-remediation water quality, Mineral Creek, Colorado. *Hydrological Processes, 23*(23), 3319–3333. doi:10.1002/hyp.7427

Ruse, L. P., Herrmann, S. J., and Sublette, J. E. (2000). Chironomidae (Diptera) species distribution related to environmental characteristics of the metal-polluted Arkansas River, Colorado. *Western North American Naturalist*, *60*(1, 4).

Tamoto, S., Tabelin, C.B., Igarashi, T., Ito, M., and Hirojohsi, N. (2015). Short and long term release mechanisms of arsenic, selenium and boron from a tunnel-excavated sedimentary rock under in situ conditions. *J. Cont. Hydr.* 175–176, 60–71.

Appendix A – Analyte Concentrations

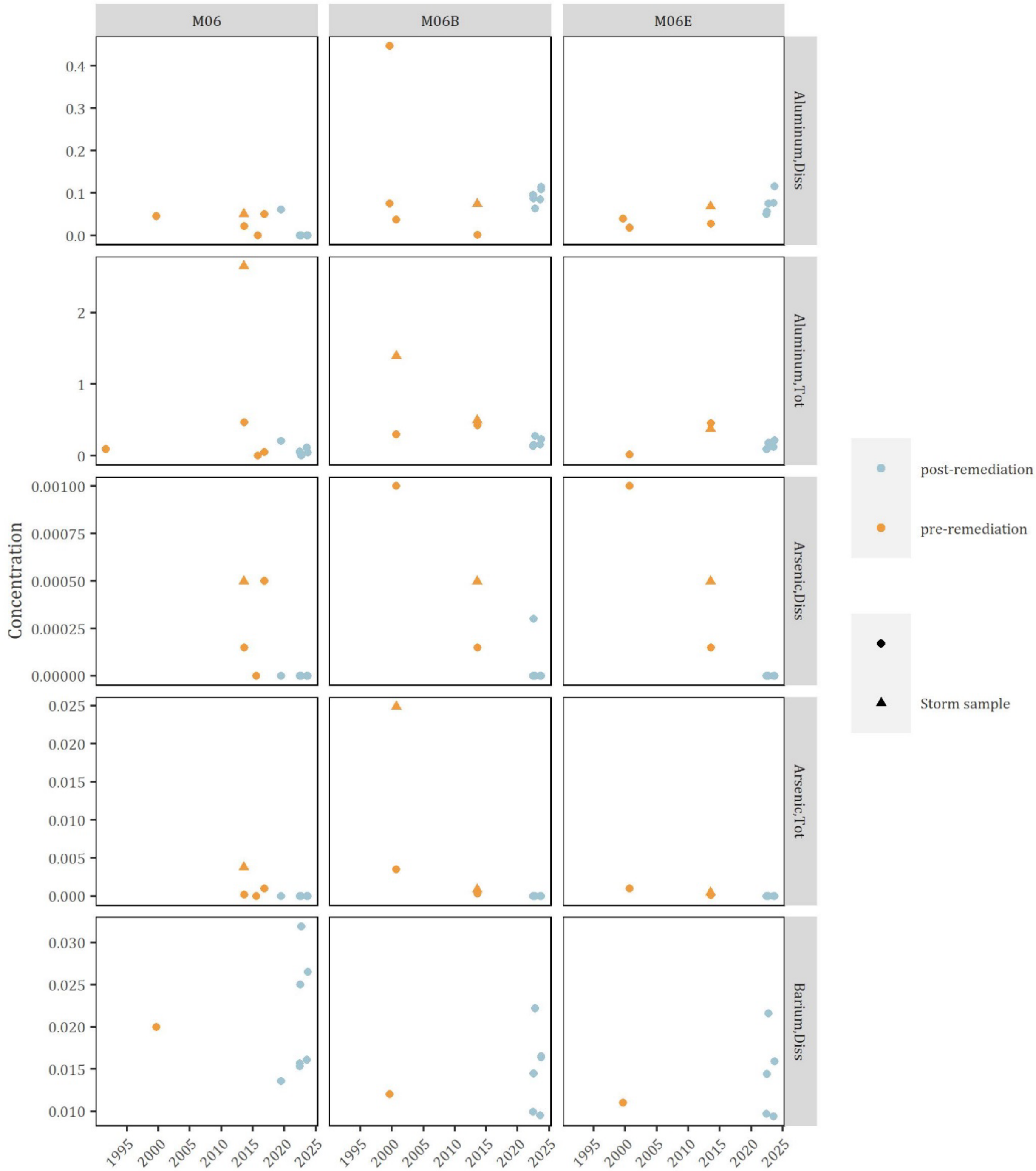

Figure A- 1. Aluminum, arsenic, and barium concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

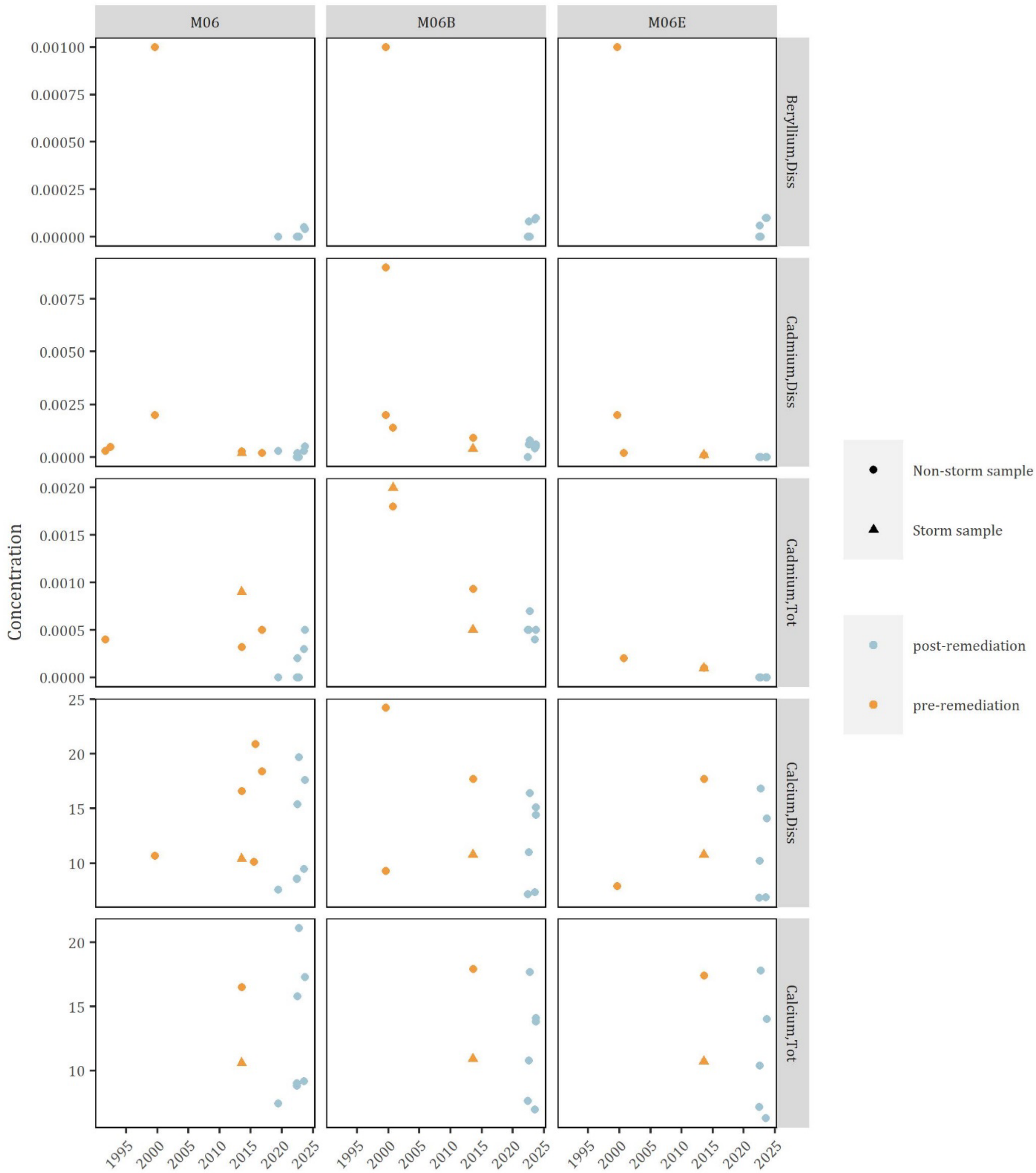

Figure A- 2. Beryllium, cadmium, and calcium concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

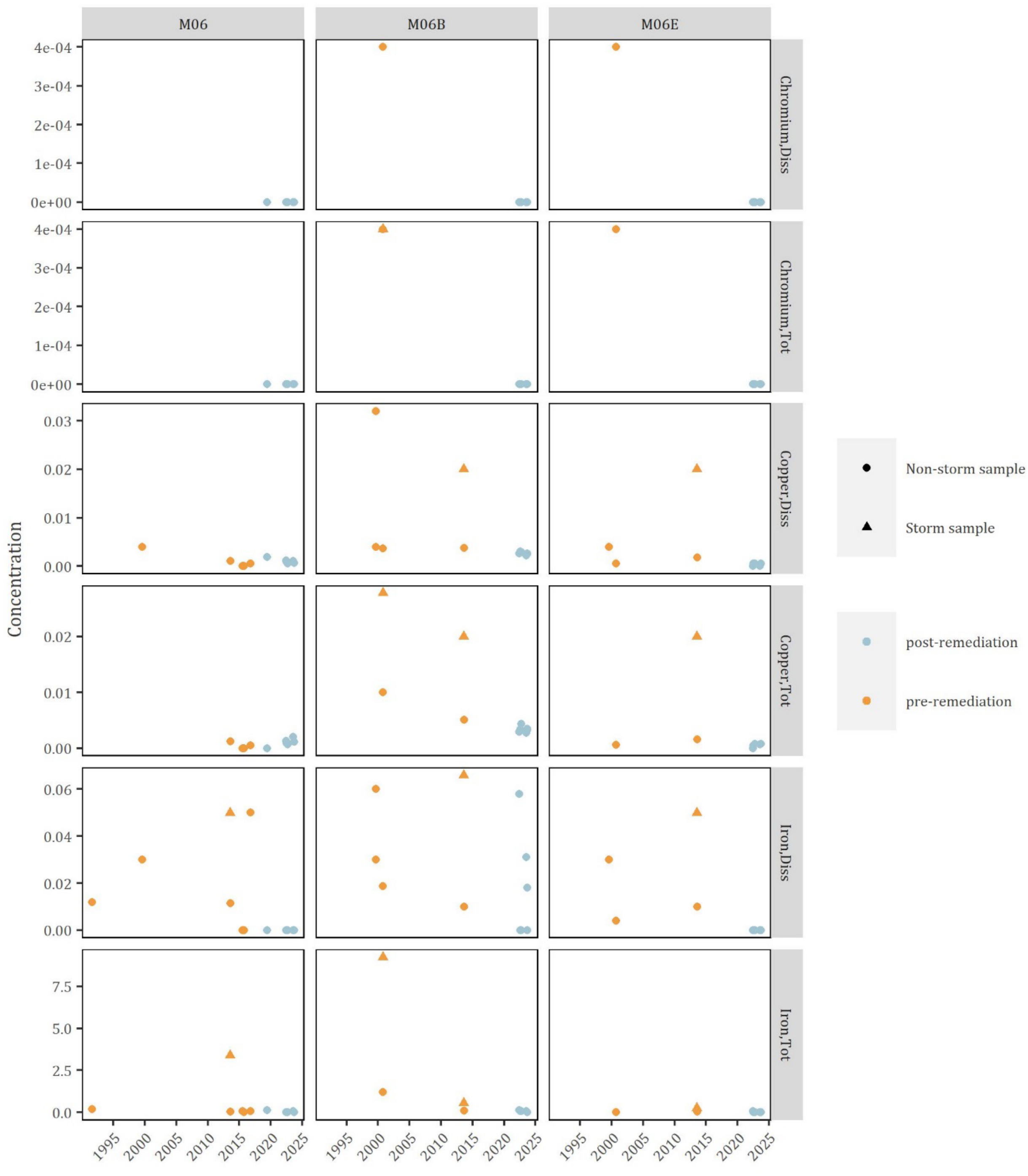

Figure A- 3. Chromium, copper, and iron concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

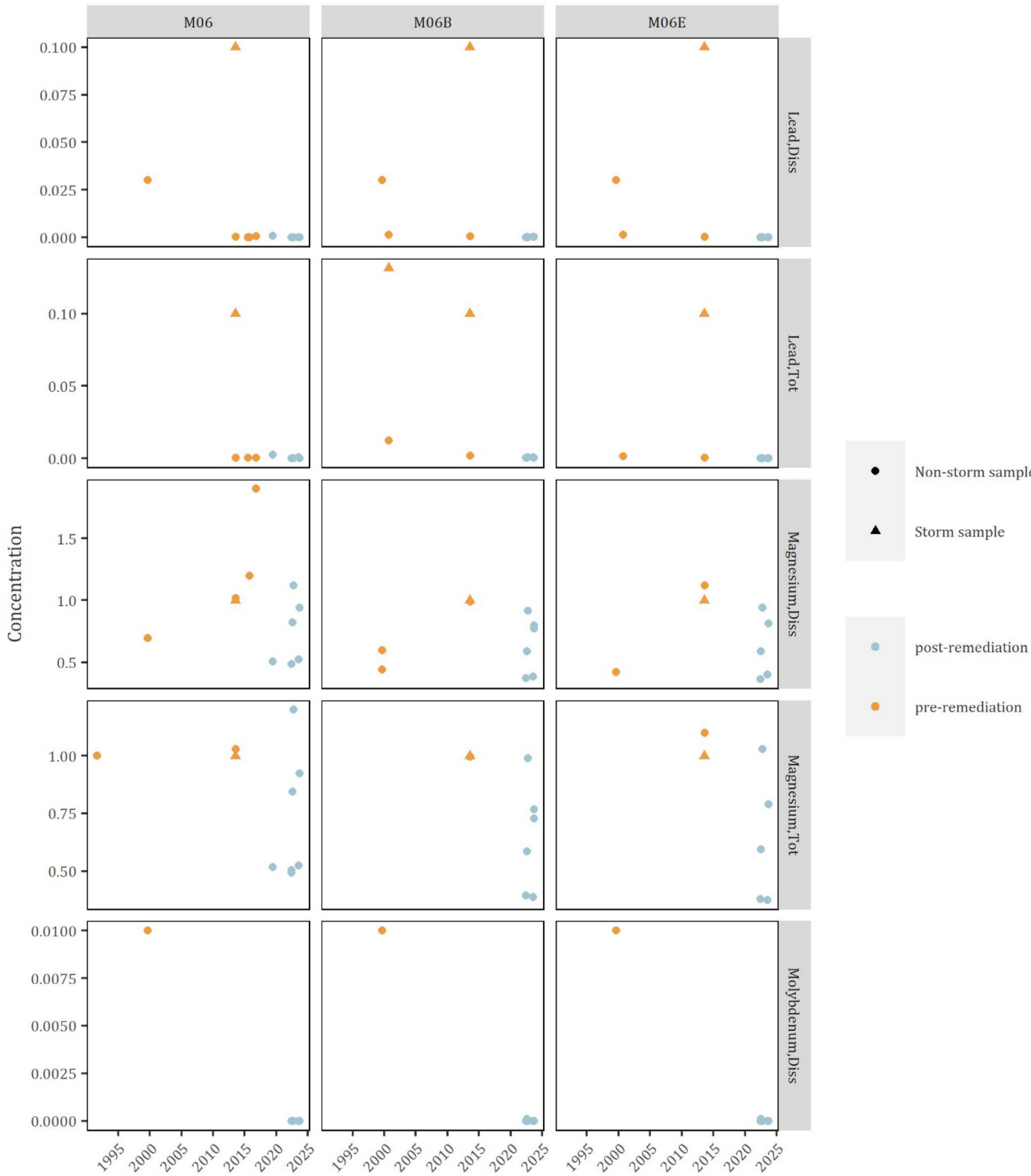

Figure A- 4. Lead, magnesium, and molybdenum concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

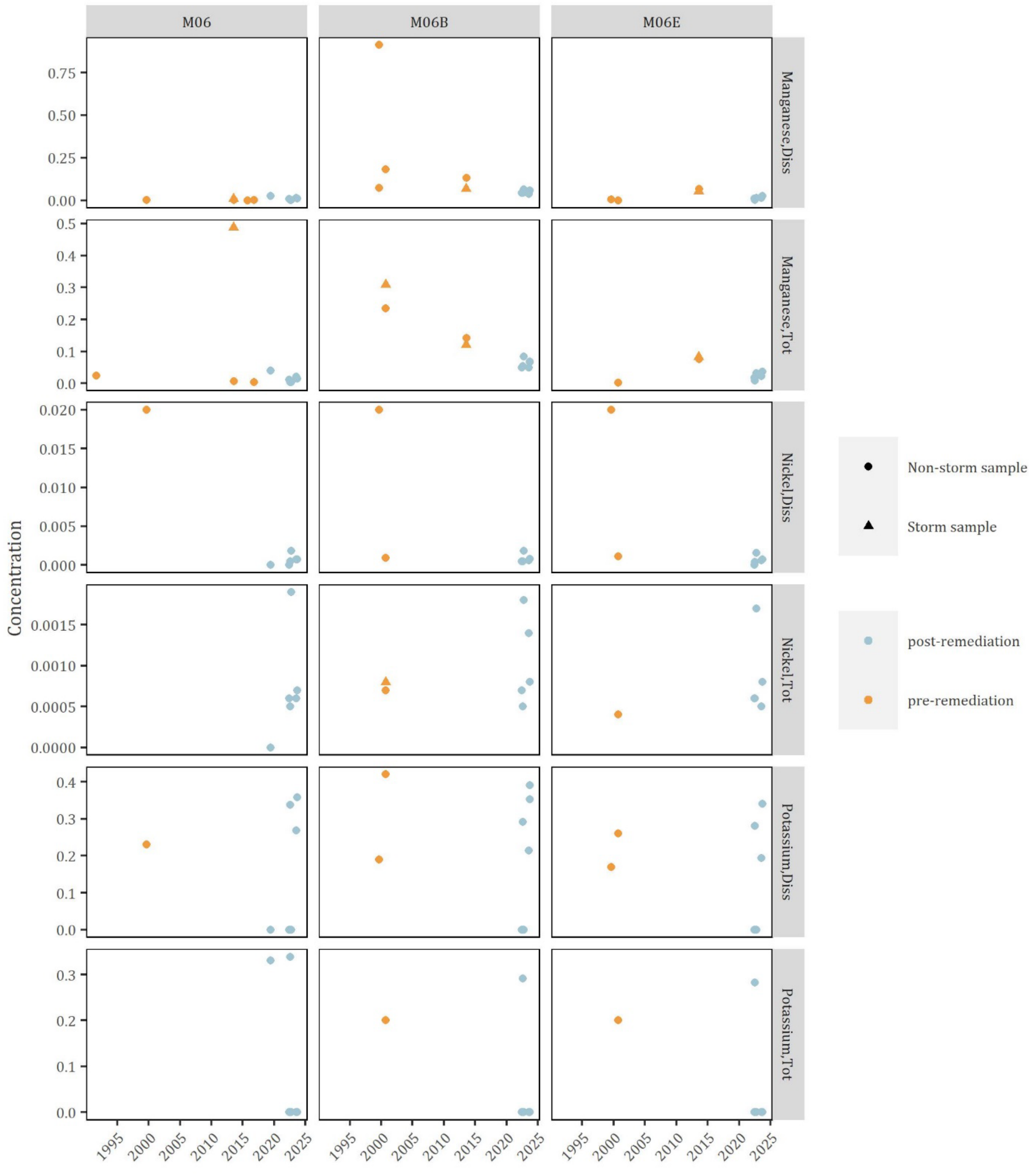

Figure A- 5. Manganese, nickel, and potassium concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

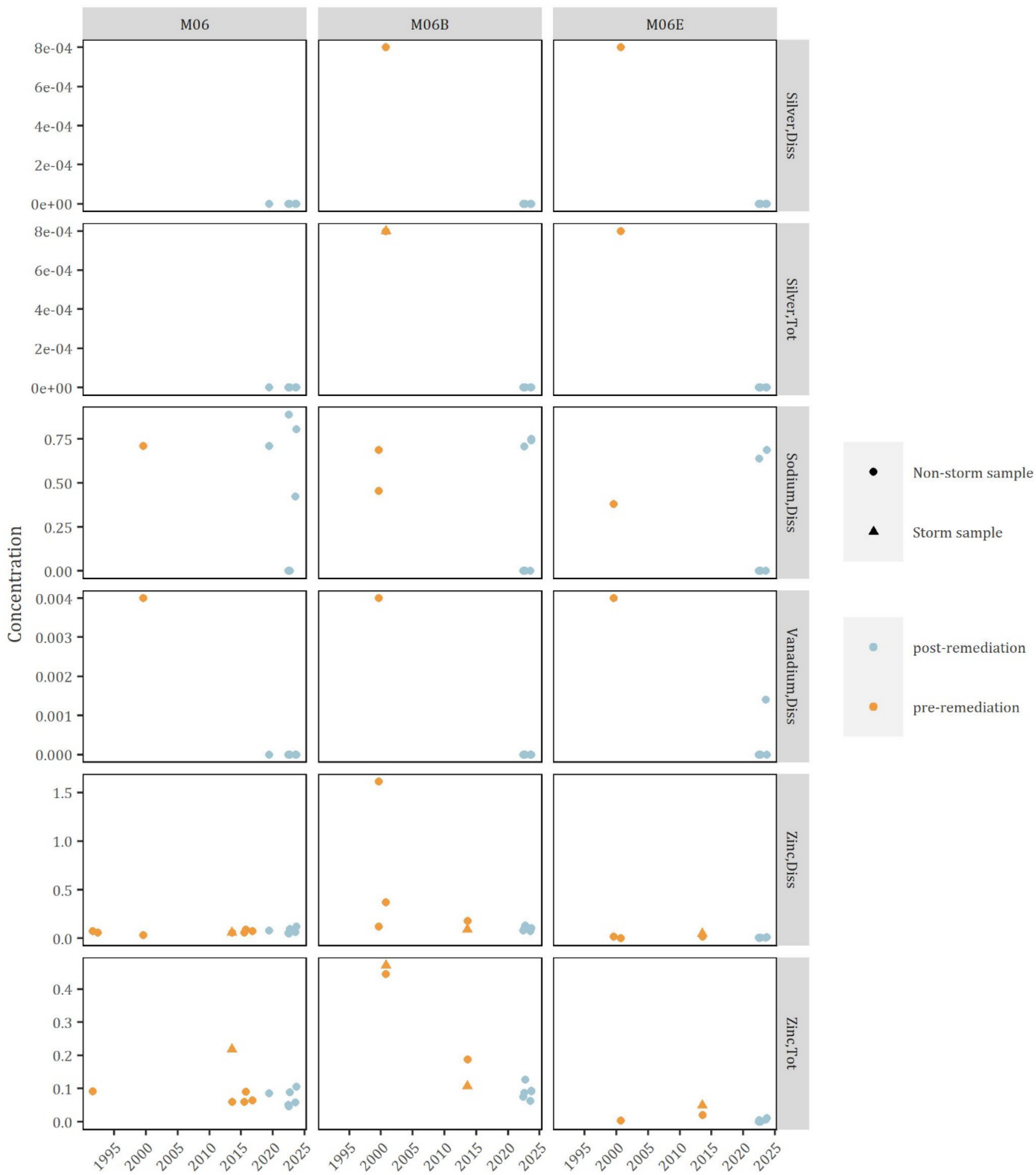

Figure A- 6. Silver, sodium, vanadium, and zinc concentrations at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent samples taken during or directly after a storm event.

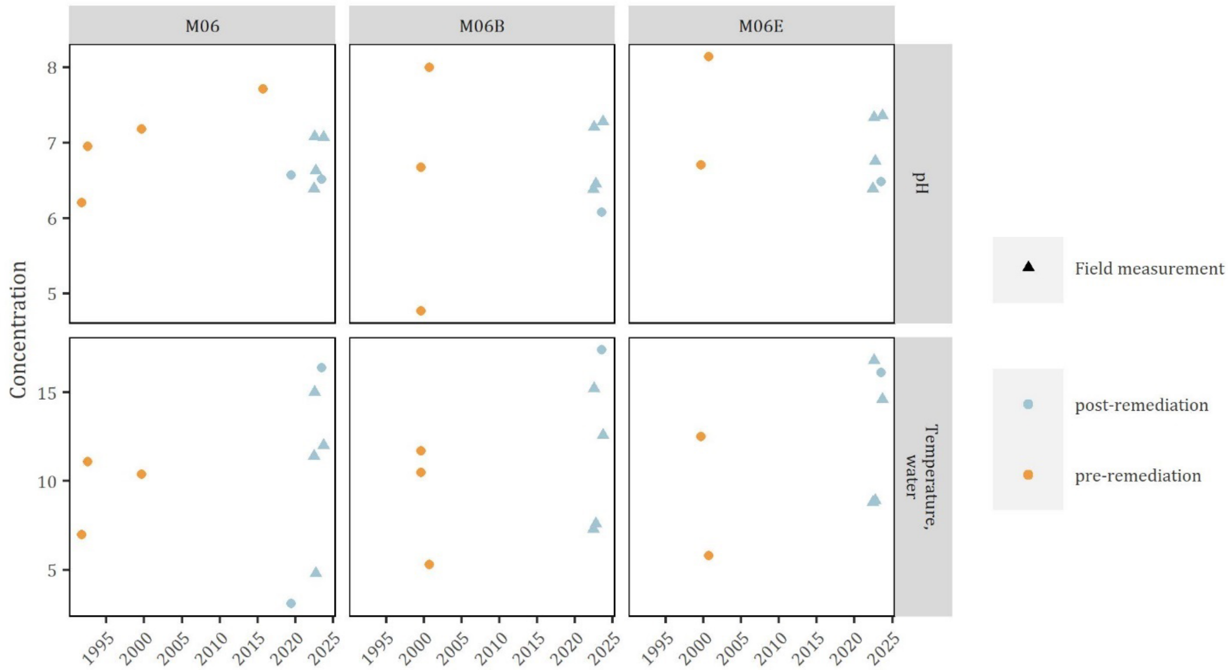

Figure A- 7. pH and temperature (*C) at three sampling sites pre and post remediation activities at Bullion King mine. Yellow represents pre-remediation samples and blue represents post-remediation samples. Triangles represent confirmed field measurements; circles represent samples that may or may not be field measurements.

Appendix B: Analyte Boxplots

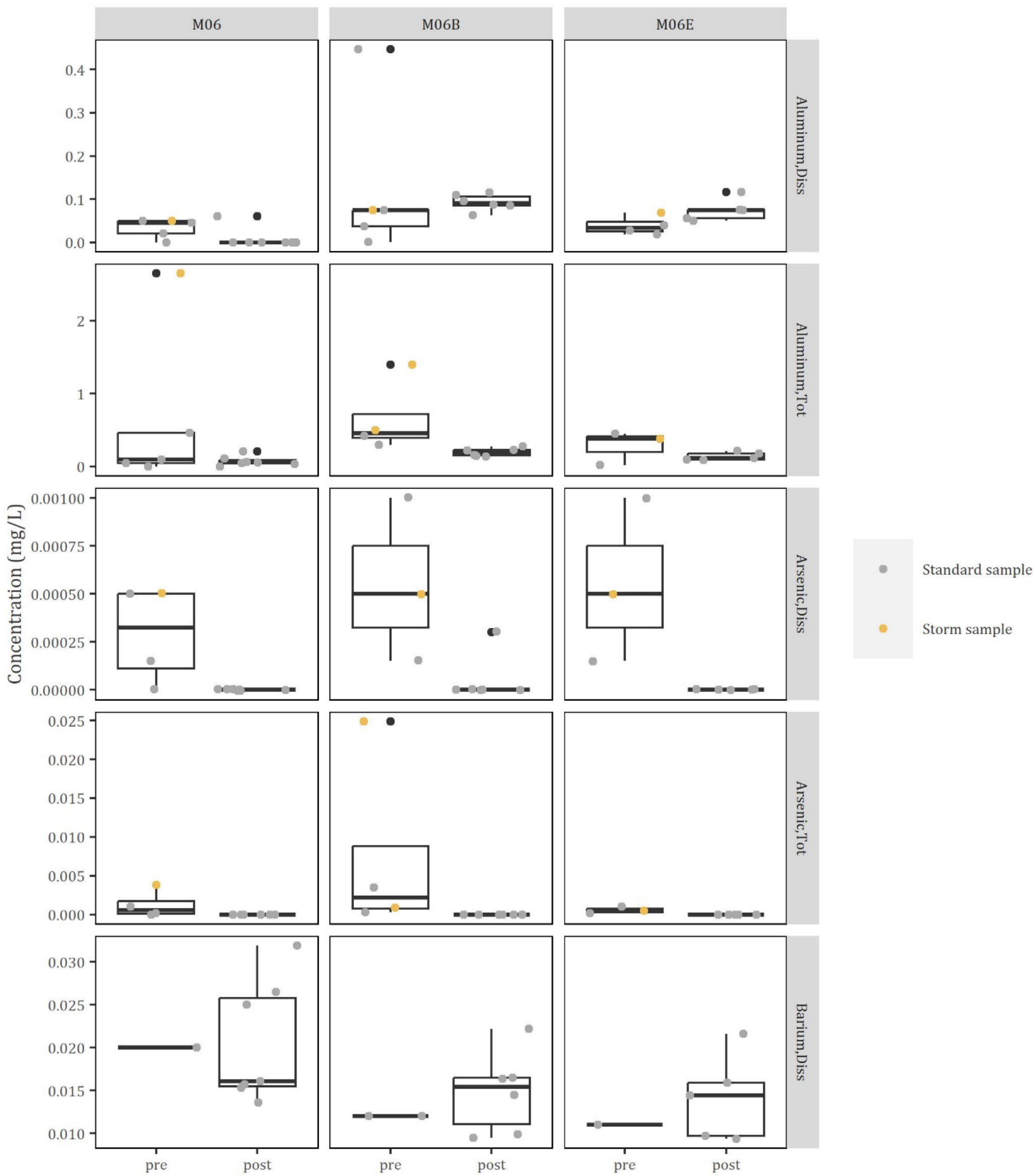

Figure B- 1. Boxplot of aluminum, arsenic, and barium concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Yellow points represent storm samples. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

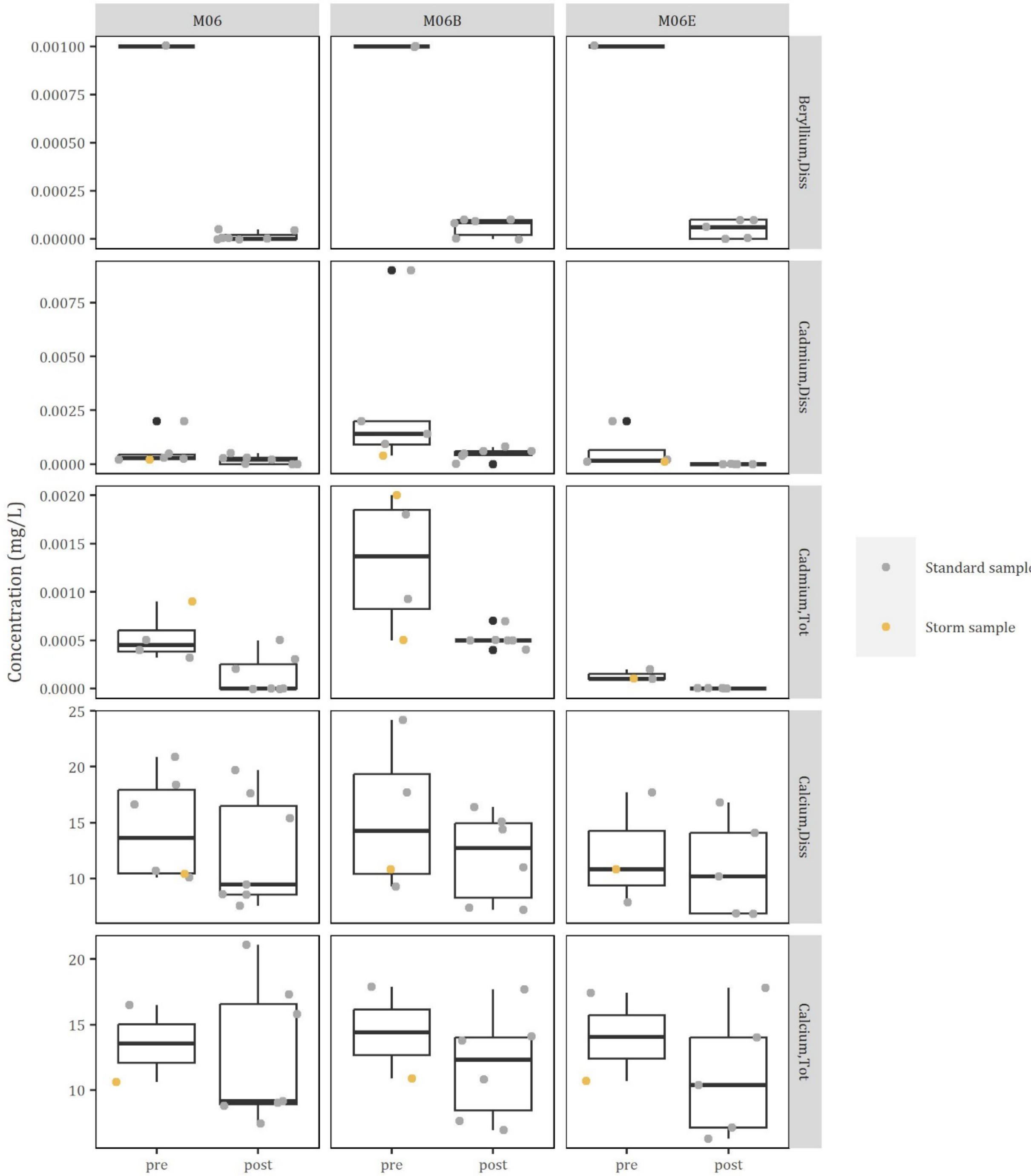

Figure B- 2. Boxplot of beryllium, cadmium, and calcium concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

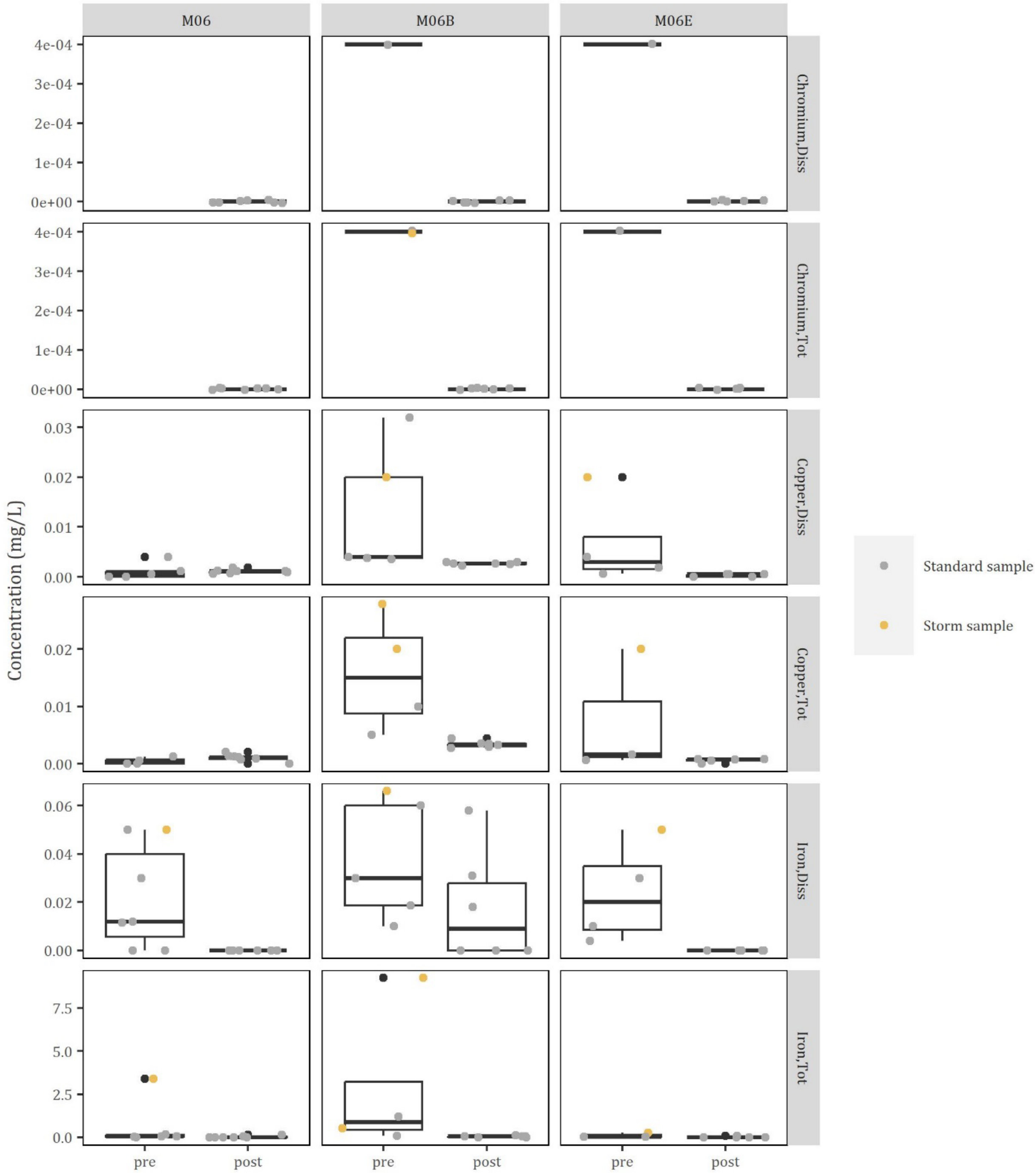

Figure B- 3. Boxplot of chromium, copper, and iron concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

Figure B- 4. Boxplot of lead, magnesium, and molybdenum concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

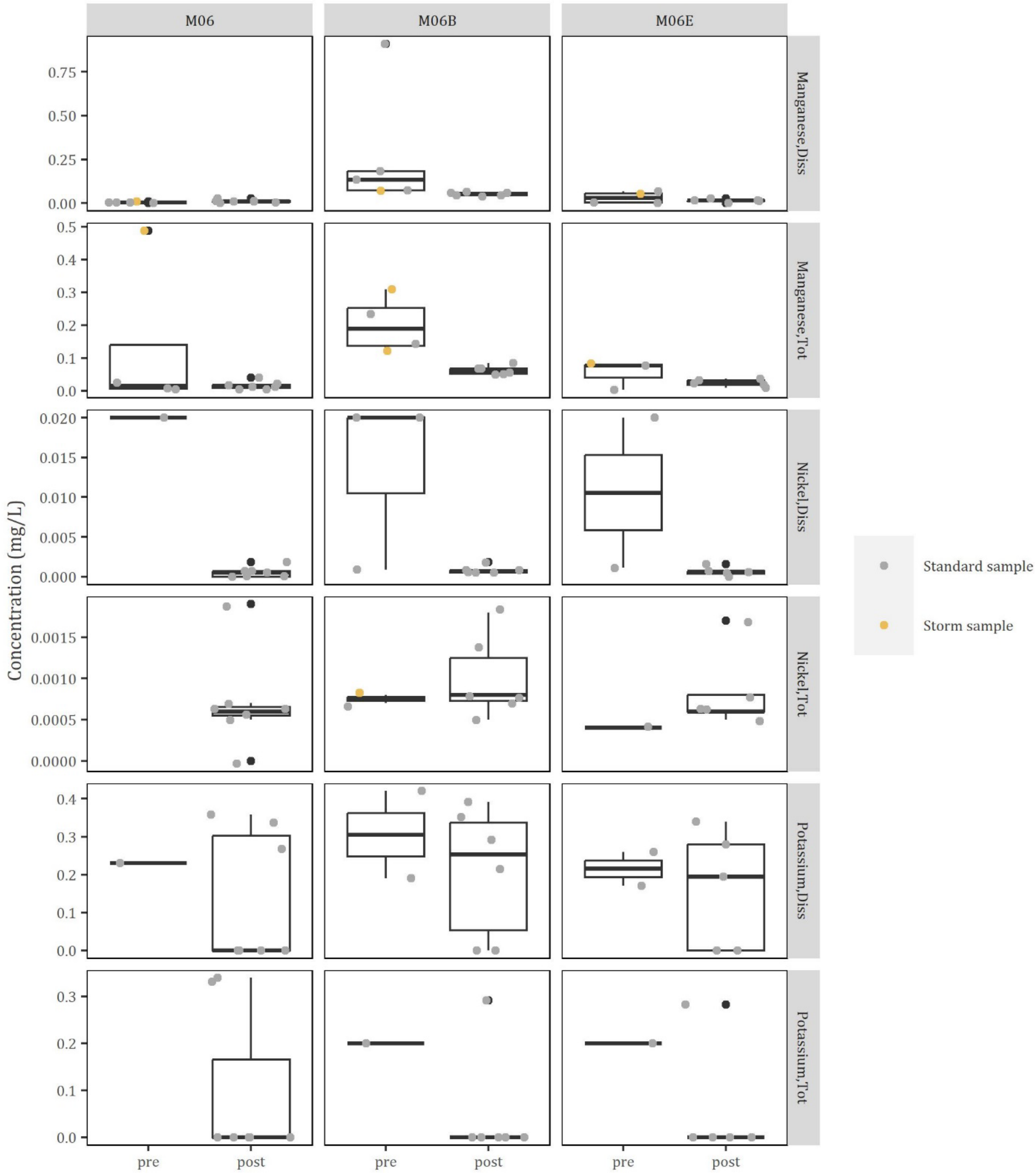

Figure B- 5. Boxplot of manganese, nickel, and potassium concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

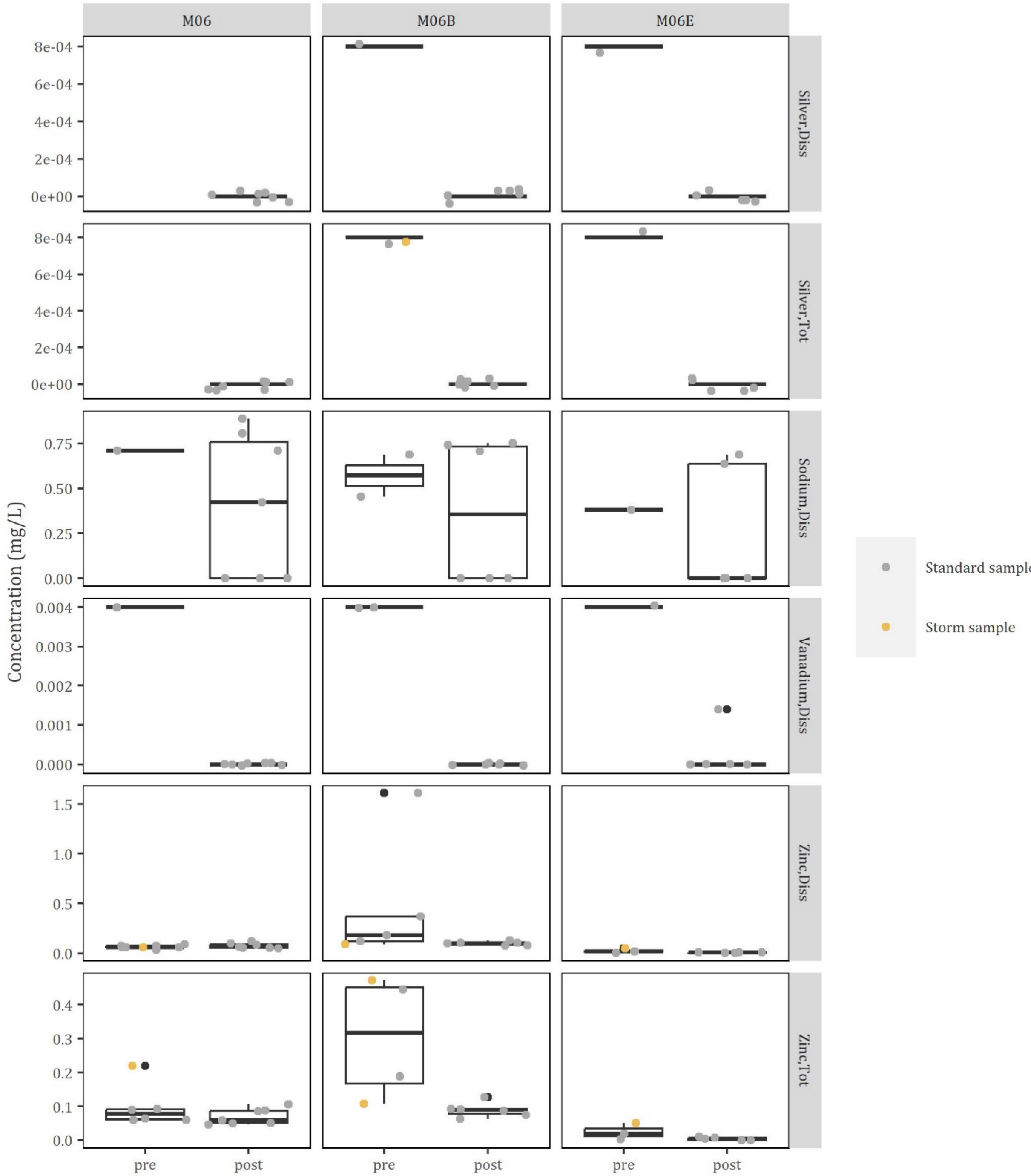

Figure B- 6. Boxplot of silver, sodium, vanadium, and zinc concentrations across three samplings sites (M06, M06E, and M06B) pre and post remediation activities at Bullion King mine. Black points represent boxplot outliers and correspond with adjacent points. Sites are presented in order from downstream (left) to upstream (right).

Appendix C – Supplemental Tables and Figures

Table C- 1. Porphyry Gulch sampling site descriptions used by various agencies and corresponding sample site code

Site name	*Lat*	*Long*	Site description
M06	37.88501	-107.72309	Porphyry Gulch, Porphyry at HWY 550, Porphyry abv HWY 550, Porphyry below 550, M06
M06B	37.88791	-107.74084	Porphyry below Bullion King, Bullion King Mine below Dump, M06B
M06C	37.88861	-107.74222	Bullion King Mine, Bullion King lower, Bullion King Adit
M06E	37.88790	-107.74174	Porphyry above Bullion King, M06E