Benthic Macroinvertebrate Monitoring in Whychus Creek (Sisters, OR), 2023

Whychus Canyon Phase 2a Restoration, August 2023

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Date: March 1, 2024

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Summary

The benthic macroinvertebrate community in Whychus Creek was sampled for the 16th year on 10-11 August 2023 in 13 reaches: Alder Springs (WC0150), Road 6360 (WC0600), Rimrock Ranch (WC1025), Whychus Canyon (WC1100, WC1150, WC1175, WC1200, WC1250), Camp Polk (WC1950), Willow Springs (WC2000, WC2050), City Park (WC2425), and Whychus Floodplain (WC2600). Sampling was done at a mix of unrestored and restored sites using proportional multihabitat (PM) and single-habitat riffle-targeted (RT) techniques. The ORDEQ PREDATOR predictive model and Index of Biotic Integrity (IBI) was applied to RT samples. Taxonomic and ecological traits assessed for all sample communities included community temperature and fine sediment optima; tolerance/sensitivity to fine sediment, pollution, and disturbance; functional feeding group; habit (locomotion); generation time (voltinism); flow (rheophily) and temperature association; and maximum body length.

A total of 143 taxa in 49 families was taken across all samples, which is similar to other recent years (133-156 total in 2018-2022). Order-level diversity was highest among Diptera (true flies), with 60 unique taxa in 10 families. EPT were also well-represented, with 20 mayfly taxa in five families, 19 caddisfly taxa in eight families, and 15 stonefly taxa in six families. Organismal abundance across the dataset was dominated by Naididae (tolerant, sediment-tolerant sludge worms associated with slower flows), which were found in every sample at abundances ranging from 7-220. Three taxa not found in any prior year were collected, all of which were found at WC1025 to WC1200: the aquatic mite *Wandesia* and the non-biting midges *Platysmittia bilyji* and *Nilotanypus fimbriatus*. Total richness at individual sites ranged from 21-62 taxa (mean 49.7, SD 10.1). RT samples had significantly more medium-bodied and cool/cold-associated organisms than PM samples; PM samples had significantly more total and mayfly taxa, Chironomidae (non-biting midges), sprawler organisms, and higher community sediment optima, reflecting the greater habitat and flow heterogeneity captured in this sampling method.

Macroinvertebrate community composition continues to be strongly influenced by reach location, with communities in most upstream reaches more similar to each other than to communities in downstream reaches. Few calculated community metrics were significantly different pre-and post-restoration at restored sites, but macroinvertebrate community composition differed significantly pre- and post-restoration at four of the six restored sites sampled in 2023. Significant unidirectional trends over time were seen for of 31 of 35 community metrics, with all but a single site (WC2050) having significant trends in 2-16 metrics. The greatest number of significant trends were seen at two sites where restoration resulted in a more dynamic primary channel reach (WC1100-2, WC2600). The direction of the significant trends for most metrics suggested improving habitat conditions, i., e., faster flows, cooler temperatures, more macrophytes, and greater habitat stability. However, other trends suggested declining habitat conditions, such as increases in community sediment optima or abundance of tolerant organisms. Except for a single site (WC2600), all significant trends in the negative community metrics were seen at sites in downstream reaches (WC1025-1 to WC1150), and increased habitat heterogeneity post-restoration in these reaches may account for changes in some negative metrics. While restoration activities have altered macroinvertebrate community composition and associated ecological traits, changes at unrestored reaches over time also suggest some degree of continuing uplift.

Background

Whychus Creek is a designated priority watershed for conservation and restoration in the upper Deschutes Basin. Projects implemented since 1999 have restored perennial flow to the creek and increased in-stream flow volume and channel complexity. Aquatic macroinvertebrates are monitored annually to assess community-level changes and their relationship to altered habitat conditions and, more recently, to creation of new heterogenous side channels as habitat for juvenile salmonids. The goals of macroinvertebrate monitoring in Whychus Creek include: 1. assessing ongoing changes at the watershed level through continued monitoring at selected long-term index sites; and 2. analyzing communities at the project level prior to and following restoration activities to increase fine-scale resolution at targeted sites.

Methods

Sampling sites

Benthic macroinvertebrate sampling was done 10-11 August 2023 in 13 reaches of Whychus Creek (Table 1): Alder Springs (WC0150), Road 6360 (WC0600), Rimrock Ranch (WC1025), Whychus Canyon (WC1100, WC1150, WC1175, WC1200, WC1250), Camp Polk (WC1950), Willow Springs (WC2000, WC2050) and Whychus Floodplain (WC2425, WC2600). This was the first year in which WC1175, WC1200, and WC1250 were sampled, and the first sampling event after a gap of five years at WC0150 and WC2425. Multiple samples were taken at WC1025 and WC1100 to assess different side channel reaches created following restoration done in 2021 and 2016, respectively. RT samples were taken in six reaches (WC0150, WC0600, WC1025-1, WC1950, WC2000).

Table 1. Whychus Creek sampling reaches in 2023

Site ID	Description	Years sampled	Site type
WC0150	d/s Alder Springs	2009, 2011-2017, 2023	Unrestored
WC0600	u/s Road 6360	2005, 2009, 2011-2023	Unrestored; long-term index
WC1025-1	Rimrock Ranch u/s	2011-2012, 2014-2017, 2019-2020, 2022-2023	Restored (2021)
WC1100-2[a]	Lower Whychus Canyon	2014-2015, 2017-2023	Restored (2016)
WC1150[b]	Upper Whychus Canyon	2014-2023	Unrestored
WC1175	Whychus Canyon reach 3	2023	Unrestored
WC1200	Whychus Canyon reach 2	2023	Unrestored
WC1250	Whychus Canyon reach 2	2023	Unrestored
WC1950	d/s Camp Polk bridge; WQ site coordinates	2009-2017, 2019, 2022-2023	Restored (2012)
WC2000	Willow Springs	2020-2021, 2023	Restored (2022)
WC2050	Willow Springs	2020-2021, 2023	Restored (2022)
WC2425	Sisters City Park	2005, 2009, 2011-2017, 2023	Restored (2022)
WC2600	Whychus floodplain; 4606 Road footbridge	2005, 2009, 2011-2023	Restored (2015); long-term index

[a] designated WC1025 in 2014. [b] designated WC1075 in 2014

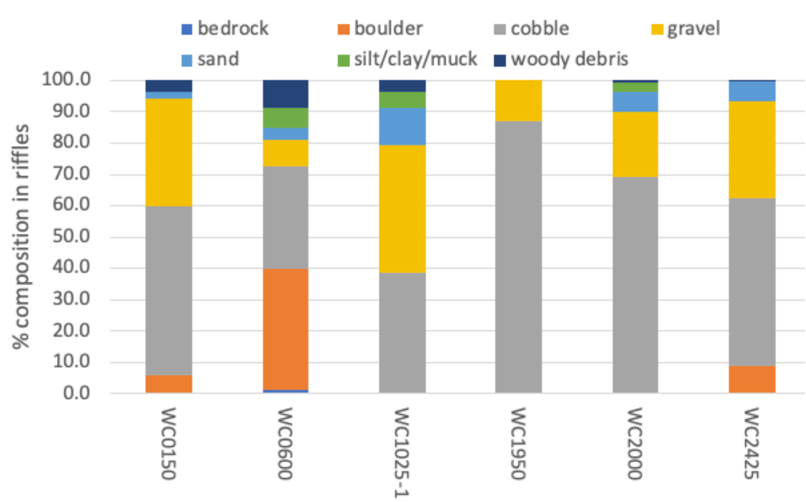
Macroinvertebrate sampling techniques

Sampling was done by CASM Environmental, UDWC staff, and volunteers from natural resource agencies and the surrounding community. After CASM Environmental staff demonstrated sampling techniques, teams received sampling kits and maps and dispersed into the field. Teams returned samples, data sheets, and equipment to CASM Environmental, who inspected each sample to ensure it was properly labeled and preserved.

Riffle-targeted protocol (RT)

Benthic macroinvertebrates were collected from riffle habitats according to Oregon Department of Environmental Quality (ORDEQ) protocols for Oregon's wadeable streams (ORDEQ, 2009). Reach lengths were calculated as 40 times the average wetted width of the active channel (minimum 500 ft. [150 m], maximum 1000 ft. [300 m]). The upstream and downstream limit of each reach and turning points along the channels were flagged by UDWC prior to sampling. A reach sample consisted of eight individual net sets, each collected in a 1 ft^2 area of riffle habitat using a D-frame kick net with 500 µm mesh and a 1 ft. (0.3 m) opening. In reaches with eight or more riffles, a single net set was taken in each of eight randomly selected riffles; in reaches with fewer riffles, two net sets were taken in each of four randomly selected riffles. Substrate composition was assessed at each sampling point (Figure 1).

Figure 1. Substrate types in riffle-targeted (RT) sample reaches.



Large rocks in the sampling area were rubbed and rinsed into the net to collect clinging organisms and set aside, and the remaining substrate was disturbed to a depth of 2-4 in. (6-10 cm) for 1-2 minutes. All net sets were pooled in a bucket, large debris was rinsed and removed, and sample material poured through a sieve lined with a 500 µm Nitex membrane. This concentrated sample was transferred to a 1 L Nalgene sample jar half-filled with 80% ethanol as a preservative. Jars were filled no more than 2/3 full; sample material was divided among multiple jars if needed. CASM Environmental replaced the 80% ethanol in all jars with fresh within 72 hours to ensure preservation.

Multihabitat protocol (PM)

To better assess the macroinvertebrate community in the heterogeneous habitats created in many reaches during restoration, multiple reaches were sampled using a proportional multihabitat protocol (Barbour et al., 2006; USEPA, 2009; Ode et al., 2016). Reach lengths were calculated and flagged as described above; at sites where both RT and PM samples were taken, two teams sampled the reach simultaneously, moving upstream as a unit. Before sampling, teams walked the reach to determine relative proportions of different in-stream habitats:

- bedrock/boulder (continuous rock; large mineral substrate >basketball size)
- cobble (tennis ball- to basketball-size)
- gravel (marble- to tennis ball-size)
- sand/silt (fine sediment)
- filamentous algae (long, flowing strands)
- aquatic vegetation (herbaceous plants rooted or floating in the channel)
- wood (tangles of small wood < 30 cm diameter and large woody debris ≥ 30 cm diameter in wetted channel)
- rootwads (root tangles extruding into flowing channel due to undercut banks

Each sample was a composite of 10 net sets; the number taken in each habitat was determined by its proportional representation in the reach (Figure 2). The flow where each net set was taken was recorded (rapid, riffle, run, glide, pool; Figure 3), but no flow types were targeted. Where there was sufficient current to carry suspended material into the net, cobble and gravel substrates were sampled as described for riffles. On bedrock and boulders, the D-net was held perpendicular to the substrate with the mouth facing upstream and the rock surface was rubbed clean in a 1 ft^2 (0.3 m) area in front of the net. In transects with little or no flow, the substrate was continuously disturbed to a depth of several inches while the D-net was swept repeatedly through the suspended material to capture dislodged invertebrates. In vegetation, the net was jabbed and swept through the vegetation repeatedly during the one-minute sampling time. Root wads, small wood tangles, and large woody debris were sampled similarly; invertebrates were picked off during a visual examination, then the net was held adjacent to and beneath the wood kicking it vigorously to dislodge invertebrates. Net sets were composited and processed as described for riffle samples.

Figure 2. Substrate types in proportional multihabitat (PM) sample reaches.

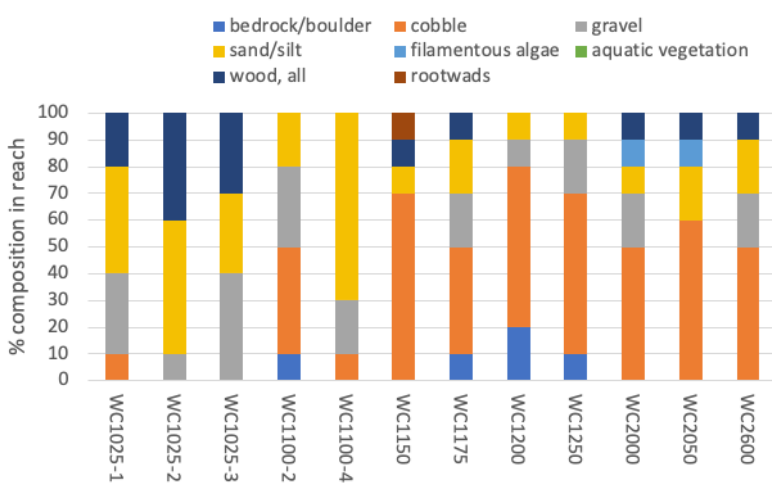
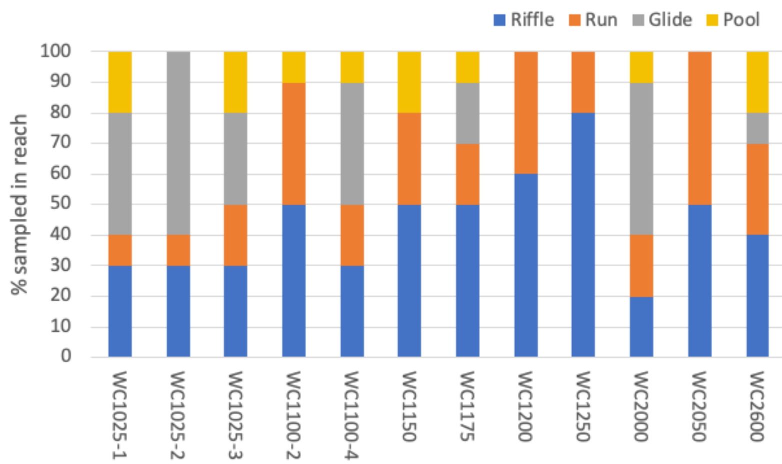


Figure 3. Flow types in composited PM samples.



Sample identification

Samples were identified by Cole Ecological, Inc. (www.coleecological.com). Each was first sub-sampled to a target count of 500 individuals by splitting the sample into equal aliquots; individual aliquots were selected randomly, and all organisms in each were picked out. An aliquot in which the target number was reached was picked to completion, which explains differences in organismal abundance between samples. Organisms were identified to the lowest practical taxonomic level using the standard taxonomic effort recommended by the Pacific Northwest Aquatic Monitoring Partnership (level 2; https://tinyurl.com/y6ynt4yo). Any changes that have occurred over time in the taxonomic nomenclature are noted with the historic name first and the current name in parentheses.

Biological/ecological traits of taxa

Assessing functional traits of macroinvertebrate taxa helps infer habitat conditions that shape the community, diagnose stressors or environmental filters, and relate restoration-related changes (Poff et al., 2006; Tullos et al., 2009; Culp et al., 2011; Van den Brink et al., 2011; White et al., 2017). Ecological and life history traits of the macroinvertebrate community were assigned to taxa where available; values for each trait are not known for every taxon. Trait data were drawn from sources specific to Oregon and/or the west (Vieira et al., 2006; Meyer & McCafferty, 2007; Huff et al., 2008; Richards & Rogers, 2011; Relyea et al., 2012; IDDEQ, 2015; SAFIT, 2016), and general and family-specific references (Pinder, 1986; Wiggins, 1996; Larson et al., 2000; Thorp & Rogers, 2016; Stewart & Stark, 2002; Anderson et al., 2013; Merritt et al., 2019; Twardochleb et al., 2020). Where multiple modalities existed for a trait, the primary one was used. Community measures calculated included:

- community optima for temperature and percent fine suspended sediment (weighted averages): Temperature and sediment are environmental filters on macroinvertebrate communities, and different taxa may have different tolerance ranges for cold vs. warm waters or for sedimentation. Increasing sedimentation can decrease richness and abundance of taxa that feed as scrapers or filterers, have a large maximum body size, soft exposed body, external exposed gills, associations with larger mineral substrates, and a crawling or sprawling habit. Taxa with operculate gills, smaller and more sclerotized bodies or cases/tubes, and a swimming, climbing, or clinging habit may become more abundant as sediment increases (Beche & Statzner, 2009; Sutherland et al., 2012; Buendia et al., 2013; Bona et al., 2015; Murphy et al., 2017; Doretto et al., 2018; Akamagwuna et al., 2019).

- trophic guild (functional feeding group), i.e., relative abundances of predator (PR), scraper (SC), shredder (SH), and collector (C; filterers and gatherers) organisms: Filterers are negatively impacted by sedimentation if their feeding structures become clogged (Rabení et al., 2005); predator abundance can increase as increasing habitat diversity and/or stability creates more abundant and diverse prey (Arce et al., 2014); scrapers can be more abundant on algae- and biofilm-coated mineral substrates; and shredders indicate more plant material and leaf litter input.

- habit (locomotion) i.e., relative abundance of swimmer (SW), clinger (CLG), burrower (BUR), climber (CLB), and sprawler (SPR) organisms: Swimmers can escape habitat disturbance more rapidly; climbers may indicate the presence of more aquatic macrophytes; burrowers are selected for in sedimented habitat, while sprawlers and crawlers can be smothered or lose habitat as interstitial spaces are filled (Mathers et al., 2017; Murphy et al., 2017).

- voltinism (# generations per year) i.e., relative abundance of multivoltine (>1 generation/year), univoltine (1 generation/year), and semivoltine <1 generation/yr) organisms. Multivoltinism is associated with more tolerant organisms and/or greater resilience in disturbed habitats, while semivoltine taxa require more stable conditions.

- rheophily (flow preference), i.e, relative abundance of organisms associated with erosional (fast/lotic), depositional (slow/lentic), and mixed flows (i.e., found in both lotic and lentic habitats);

- temperature associations, i.e., relative abundance of organisms with cool/cold or warm water temperature preferences (taxa with mixed or broad temperature range associations were omitted); and

- maximum length, i.e., relative abundance of organisms with small (< 9 mm), medium (9-16 mm), and large (>16 mm) body length: Small size is associated with faster development, greater tolerance, and rapid recolonization, which is an advantage in disturbed sites, while larger-bodied insects are slower to develop and can be more abundant in sites with greater habitat stability (Townsend & Hildrew, 1994; de Castro et al., 2018).

Data analysis

Analyses were done using PAST 4.0 (Hammer et al., 2001) and PRIMER-e v7 (Clarke et al., 2014) statistical software. CLUSTER dendrograms, SIMPER tests, and one-way ANOSIM were run on a Bray-Curtis similarity matrix of square-root transformed taxa abundances. Community evenness, a measure of ecosystem stability (Death, 1996; Wittebolle et al., 2009), was calculated on untransformed taxa abundances. Principal Component Analysis (PCA) was done using a variance-covariance matrix. At restored sites with more than two years of post-restoration data, statistically significant changes in community composition were assessed using one-way ANOVA on pre- and post-restoration communities with results reported at alpha = 0.05. Differences between mean values of a trait were examined using unpaired t tests with a cutoff value of $p \leq 0.05$ for statistical significance. Mann-Kendall trends analysis was done to assess site-level monotonic trends in calculated community metrics for sites sampled ≥4 years, with a cutoff value of $p \leq 0.05$ for statistical significance. Means are presented with standard deviation (SD).

Biological condition of RT samples was assessed using the ORDEQ multimetric invertebrate-based index of biotic integrity (IBI) and the probability-based PREDATOR model (Hubler, 2008). In the IBI, raw values of 10 metrics are assigned scaled values and then summed to give a score corresponding to a level of biological impairment (Table 2). These models were developed for riffle communities and cannot be applied to PM samples, but values of individual metrics in PM samples were calculated for comparisons. PREDATOR calculates the ratio of taxa observed at a site to taxa expected if the site is not impaired (O/E), based on comparison to established reference stream communities selected by the model. O/E scores correspond to condition categories of poor (most disturbed; ≤0.78); fair (moderately disturbed; 0.79-0.92); good (least disturbed; 0.93-1.23); or enriched (>1.23). Whychus Creek is an outlier for the PREDATOR model because it has lower annual precipitation than any reference streams the model selects.

Table 2. ORDEQ macroinvertebrate-based IBI metrics and scoring. [a] relative abundance of the most abundant taxon; [b] modified Hilsenhoff Biotic Index (Hilsenhoff, 1987); reflects tolerance to organic pollution and ranges from 1 (low tolerance) to 10 (high tolerance).

Metric	5	3	1
Taxa richness	>35	19-35	<19
Mayfly richness	>8	4-8	<4
Stonefly richness	>5	3-5	<3
Caddisfly richness	>8	4-8	<4
# sensitive taxa	>4	2-4	<2
# sediment-intolerant taxa	≥2	1	0
% dominance	<20	20-40	>40
% tolerant	<15	15-45	>45
% sediment-tolerant	<10	10-25	>25
MHBI	<4	4-5	>5
Summed Score and Condition			
<20 severely impaired; **20-29** moderately impaired; **30-39** slightly impaired; **>39** minimally/not impaired			

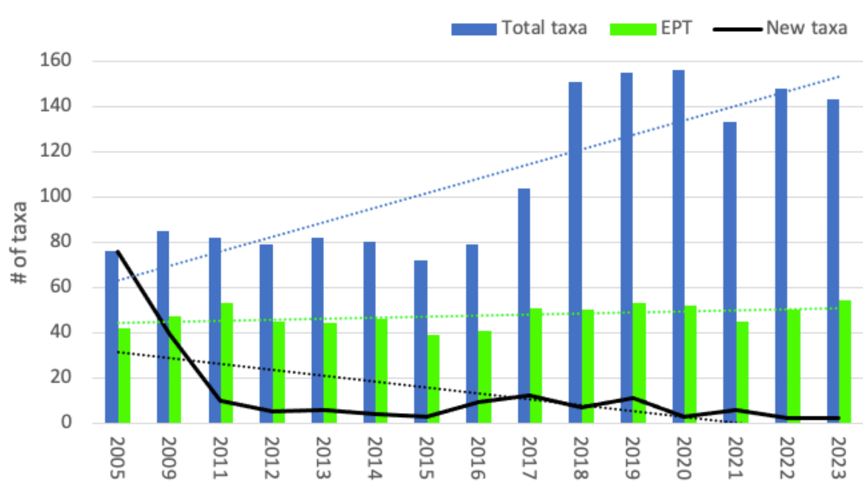
Results

Macroinvertebrate community in 2023 samples

Although sampling has been done in Whychus Creek in 15 years, a few new taxa are still found every year (Figure 4). The increase in total taxa from 2017 onwards was driven by restoration in different reaches; creation of new dynamic, heterogenous side channel habitats; and the introduction of PM sampling, which often captures a greater number of taxa per sample compared to RT sampling done in the same reach. The species accumulation curve is near saturation, but three taxa were new to the Whychus dataset in 2023, at abundances ranging from 3-10 individuals: *Wandesia*, an aquatic mite (WC1025-2 side channel PM sample); *Platysmittia bilyji*, a non-biting midge (WC1025-1 RT), and *Nilotanypus fimbriatus*, a non-biting midge (PM samples taken at WC1025-2, WC1025-3, WC1100-2, WC1200).

A total of 143 taxa in 49 families (34 insect, 15 non-insect) was collected across all sites in 2023, which is similar to recent years (Figure 4). Three taxa were taken in every sample: Naididae (widespread group of tolerant, sediment-tolerant sludge worms associated with slower flows), *Baetis tricaudatus* (small minnow mayfly associated with faster flows and clearer water), and *Rhithrogena* (sediment-sensitive flatheaded mayfly found on stones in faster flows). Order-level diversity was greatest among Diptera, with 60 unique taxa in 10 families, including 46 unique non-biting midge (Chironomidae) taxa. Other well-represented groups included Ephemeroptera (mayflies), with 20 unique taxa in five families; and Trichoptera (caddisflies), with 19 unique taxa in eight families. Organismal abundance was dominated by Naididae (tolerant, sediment-tolerant sludge worms associated with slower flows), which were found in every sample at abundances ranging from 7-220. Other highly abundant taxa included *Optioservus*, a tolerant riffle beetle associated with clear water and stable habitat in a variety of flows and temperatures, found in every sample except WC2425 and WC2600 at abundances ranging from 1-96; and *Baetis tricaudatus*, which was present in every sample at abundances ranging from 2-171.

Figure 4. Taxa accumulation in Whychus Creek samples. Total taxa = # of unique taxa taken among all samples in each year; EPT = # of Ephemeroptera (mayfly), Plecoptera (stonefly), and Trichoptera (caddisfly) taxa; new taxa = # of taxa taken for the first time at any Whychus site in each year. Linear trendlines are shown.



The target sub-sampling number of 500 individuals was attained in all but two samples (WC0600 RT, 206 organisms total; WC2425 RT, 170 organisms total), with 15-100% of the sample picked. In-channel restoration work at WC2425 occurred in 2022, so reduced organismal abundance in 2023 could be a result of restoration-related disturbance, as has been seen at other restored sites. Sample richness ranged from 21 to 62 taxa (mean 49.7, SD 10.1); this metric in the ORDEQ IBI receives the highest scaled score at >35 taxa, and only one site was below this threshold (WC2425, 21 taxa). Simpson's Diversity Index was high overall (0.8017 – 0.9311), indicating diverse and balanced communities.

PM samples are taken in all types of substrates, so composited samples contain more net sets from slower flows and/or softer substrates (i.e., sand/silt, vegetation, algae). However, all PM samples taken in 2023 included net sets taken in at least two riffles (Figure 3). Further, despite including still water habitat and softer substrates, PM samples consistently capture sensitive organisms, and this more heterogenous habitat array often results in greater sample richness. PM samples had significantly more total taxa and Ephemeroptera (mayfly) taxa, as well as greater relative abundances of non-biting midges (Chironomidae) and sprawlers and higher community sediment optima (Table 3). RT samples had significantly more organisms associated with cooler water temperatures and more medium-bodied organisms. No other calculated metrics differed significantly between sample types.

Table 3. Comparison of community metrics between riffle and multihabitat samples in 2023. Means are shown with standard deviation in parentheses. Metrics expressed as percentages reflect organismal relative abundance. Metrics that differed significantly between the two sampling types ($p \leq 0.05$) are in bold. RT = riffle-targeted, PM = proportional multihabitat.

	RT	PM			RT	PM
# total taxa	38.3 (9.5)	54.9 (4.9)		% small	66.2 (11.8)	75.5 (5.8)
Simpson's Diversity Index (1-D)	0.8775 (0.0423)	0.9096 (0.0263)		% multivoltine	23.0 (11.8)	34.1 (10.7)
# Ephemeroptera taxa	8.2 (1.6)	10.2 (1.6)		% univoltine	55.7 (12.5)	41.6 (7.3)
# Plecoptera taxa	4.0 (1.4)	5.2 (1.8)		% semivoltine	7.8 (7.6)	10.0(4.8)
# Trichoptera taxa	5.7 (1.5)	5.3 (1.6)		**% cool/cold-associated**	46.0 (16.2)	31.1 (9.6)
# sensitive taxa	3.3 (1.0)	4.6 (1.9)		% warm-associated	4.8 (4.4)	6.3 (3.6)
# sediment-sensitive taxa	1.8 (1.0)	1.4 (0.8)		% depositional-associated	12.7 (13.3)	14.8 (6.4)
% tolerant organisms	25.7 (20.5)	31.3 (14.7)		% erosional-associated	57.5 (18.5)	42.5 (13.3)
% sediment-tolerant organisms	13.2 (12.9)	13.0 (6.4)		% mixed flows	29.0 (12.5)	42.0 (12.9)
% dominance top taxon	23.5 (8.9)	17.7 (3.2)		% burrowers	12.9 (13.2)	14.8 (6.5)
Community temperature optima (^{o}C)	17.5 (0.9)	17.9 (0.6)		% climbers	1.7 (1.4)	5.6 (6.3)
Community fine sediment optima (% FSS)	6.6 (1.0)	8.1 (1.2)		% crawlers	0.6 (0.5)	0.9 (0.7)
% predators	13.1 (9.9)	8.6 (3.3)		% clingers	60.2 (12.2)	47.9 (3.8)
% scrapers	27.1 (8.8)	20.8 (5.5)		**% sprawlers (p = 0.0088)**	5.9 (4.3)	13.8 (5.7)
% shredders	1.7 (1.5)	4.9 (4.3)		% swimmers	16.8 (9.3)	14.3 (7.7)
% collectors	53.0 (6.4)	52.4 (10.9)		% non-insect	14.0 (13.1)	15.2 (6.5)
% large	2.3 (1.3)	3.9 (3.5)		**% Chironomidae**	7.4 (10.7)	20.2 (11.8)
% medium	27.6 (18.1)	14.3 (5.8)				

Macroinvertebrate community composition continues to be strongly influenced by reach location (Figure 5). Community composition was more similar among upstream reaches (WC1950 to WC2600) compared to the remaining downstream sites regardless of whether a PM or RT sample was taken, except for WC2425, which was least similar to all other sites. The communities at the two sites furthest downstream (WC0150 and WC0600) also differed more compared to all other samples. The paired PM samples taken simultaneously at WC2050 were most similar to each other (Bray Curtis Similarity Index = 0.79), confirming consistency and robustness of the sampling technique.

Differences between means of calculated trait values among all downstream (WC0600-WC1250) and upstream (WC1950-WC2600) sites were significant for 18 of 37 community metrics (Table 4). This is a greater difference than was seen in 2022, when only 7 of 37 community metrics differed significantly between downstream vs. upstream samples. In 2023, samples from downstream reaches had significantly more scrapers, large, semivoltine, warm-associated, and sprawler organisms, higher community sediment and temperature optima, and more organisms associated with a range of flow types. Upstream reaches had greater mean numbers of Trichoptera and sensitive taxa; higher relative diversity of EPT; more organisms associated with cool/cold and erosional flows; and more predator, collector, univoltine, and swimmer organisms. Fewer downstream-to-upstream trends across the entire sampling period were noted among sites sampled for multiple years (Appendix C); relative abundance of cold-associated organisms increased longitudinally from downstream to upstream, while relative abundance of tolerant organisms and community temperature optima were lower in upstream reaches.

Figure 5. CLUSTER dendrogram of the macroinvertebrate community among all 2023 samples. Blue = riffle-targeted, aqua = multihabitat; DUP = duplicate sample taken for QA. The number at the end indicates sampling year.

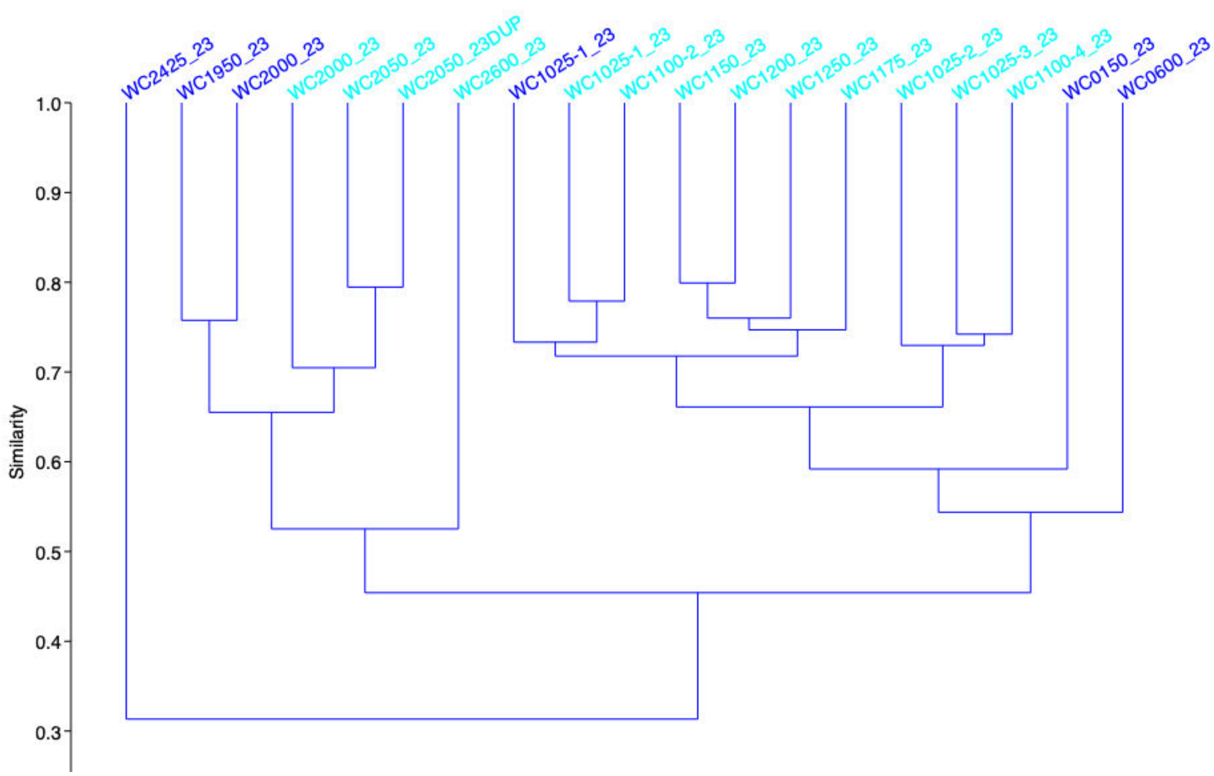


Table 4. Mean values of community metrics at downstream vs. upstream sites in 2023 samples. Bold type indicates a significant difference (p ≤0.05) between means. Metrics given as percentages reflect relative organismal abundance.

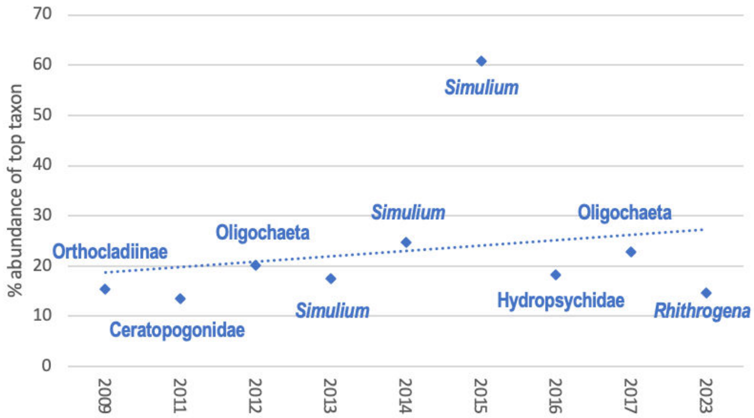
Community metric	Mean, downstream sites	Mean, upstream sites		Community metric	Mean, downstream sites	Mean, upstream sites
Taxa richness	51.30	46.90		**% large**	**4.40**	**1.40**
Ephemeroptera taxa	9.50	9.60		% medium	16.60	21.10
Plecoptera taxa	4.80	4.90		% small	73.50	72.20
Trichoptera taxa	**4.80**	**7.00**		% multivoltine	29.80	34.30
# sensitive taxa	3.90	4.70		**%ab univoltine**	**41.80**	**53.60**
# sediment-sensitive taxa	**1.20**	**2.30**		**%ab semivoltine**	**12.20**	**3.20**
% dominance top taxon	19.30	21.70		**% cool/cold-associated**	**28.90**	**49.00**
% tolerant organisms	**38.40**	**10.80**		**% warm-associated**	**7.50**	**2.80**
% sediment-tolerant organisms	15.70	8.10		% depositional-associated	15.80	9.90
Biotic Index	4.00	3.60		**% erosional-associated**	**41.60**	**60.80**
EPT richness	18.90	21.60		**% mixed flows**	**41.80**	**28.70**
% diversity EPT	**37.20**	**48.30**		% burrowers	15.90	9.80
community temp. optima	**18.20**	**16.80**		% climbers	5.80	1.30
community sediment optima	**8.30**	**6.30**		% crawlers	0.74	1.20
% predators	**7.30**	**15.20**		% clingers	50.20	53.50
% scrapers	**25.80**	**16.10**		**% sprawlers**	**13.40**	**6.80**
% shredders	4.30	3.20		**% swimmers**	**11.90**	**24.20**
% collectors	**49.50**	**59.70**				

Macroinvertebrate community characteristics at individual sampling sites

WC0150 (Alder Springs)

Riffle-targeted samples were taken in the primary channel at WC0150 in 2009-2017, and again in 2023. No active restoration was done at this site; however, it is subject to effects of restored perennial flow and surrounding land use and climate impacts, including locally declining groundwater levels, as well as potential impacts from restoration projects further upstream. The 2023 sample was taken primarily in cobble/gravel substrate (Figure 3). The macroinvertebrate community differed more from all other samples and was most similar to the communities at WC1100 through WC1250 (Figure 5). The community was dominated by *Rhithrogena*, a sediment-sensitive flatheaded mayfly found on stones in faster flows, at 14% relative abundance (Figure 6). In earlier sampling years the community at this site was dominated by more tolerant organisms, such as Orthocladiinae non-biting midges and oligochaetes (segmented worms), but except for 2015, the dominant taxon has occurred at fairly low relative abundances. Four taxa taken in 2023 were not found in this reach in any prior sampling year, all at abundances of 1-2 individuals: *Caudatella* (cool-adapted spiny crawler mayfly intolerant of fine sediment and low oxygen), *Claasenia* (common stonefly associated with larger, more open rivers), *Hesperoperla* (common stonefly associated with cool, rocky riffles and runs), *Wormaldia* (sediment-sensitive finger-net caddisfly associated with faster, cooler flows), and *Rhyacophila angelita* Gr. (free-living caddisfly that prefers clear water and is more common in lower elevation and warmer streams).

Figure 6. Relative abundance of the numerically dominant taxon at WC0150 in all sampling years. Riffle-targeted samples were taken every year. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.



Despite a lack of active restoration, trends analysis found significant decreases across time in relative abundance of predators, climbers, and crawlers (Appendix B). The target subsampling number of 500 organisms was attained every year (Figure 7). Taxa richness at this site has always been fairly high (>30 taxa); there were more total taxa in 2023 than in all prior years and more EPT than any year except 2011 (Figure 8). IBI scores varied across time but reflected slight to no disturbance every year except 2015, while PREDATOR scores reflected fair to good condition in five of nine sampling years, including 2023 (Figure 9). This site consistently has more sensitive than sediment-sensitive taxa (Figure 10), and although the relative abundance of tolerant organisms increased in recent years, abundance of tolerant organisms is generally low (Figure 11).

In 2023 this site had the greatest abundance of scraper, semivoltine, and clinger organisms of any 2023 sample, and the lowest abundances of burrowers, crawlers, and organisms associated with slower flows. Community temperature and sediment optima fluctuate but the community temperature optima was higher in 2023 than any prior year (18.1°C; Figure 12). However, relative abundance of warm-associated organisms is consistently much lower than that of cold-associated organisms (Figure 13). The majority of the 2023 community consisted of small, univoltine organisms that feed as collectors and move as clingers in cool, faster flows. The above traits suggest that habitat in the reach is stable, fast-flowing, and low in fine sediment.

Figure 7. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC0150. Riffle-targeted samples were taken every year. Target sub-sampling number is 500.

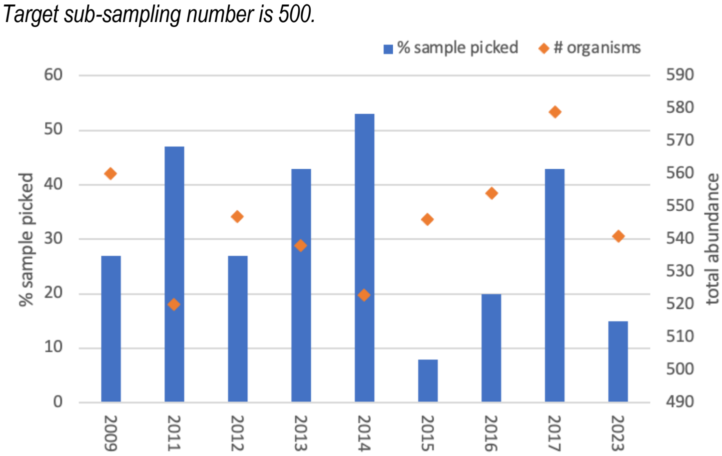


Figure 8. Sample richness and number of EPT taxa at WC0150. Riffle-targeted samples were taken every year. Linear trendlines are shown. In the ORDEQ IBI,>35 total taxa is assigned the highest scaled score.

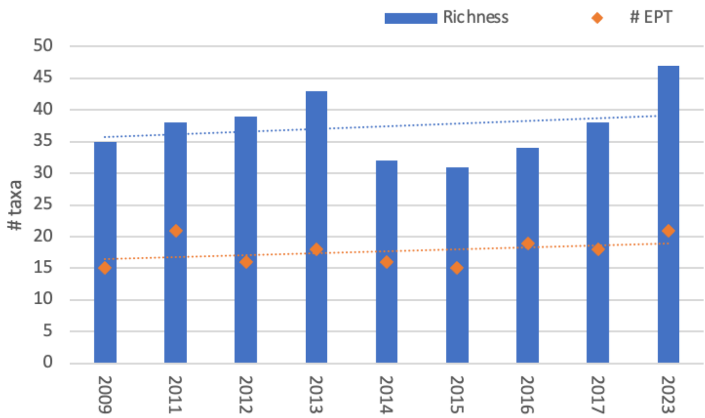


Figure 9. PREDATOR O/E and ORDEQ IBI scores at WC0150. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

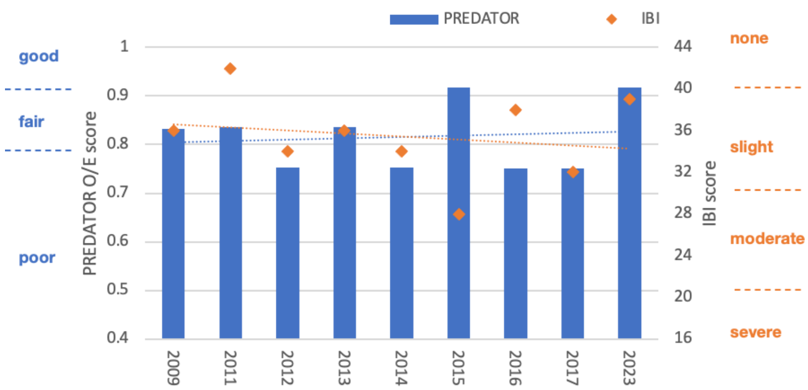


Figure 10. Sensitive and sediment-sensitive taxa at WC0150. Riffle-targeted samples were taken every year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.

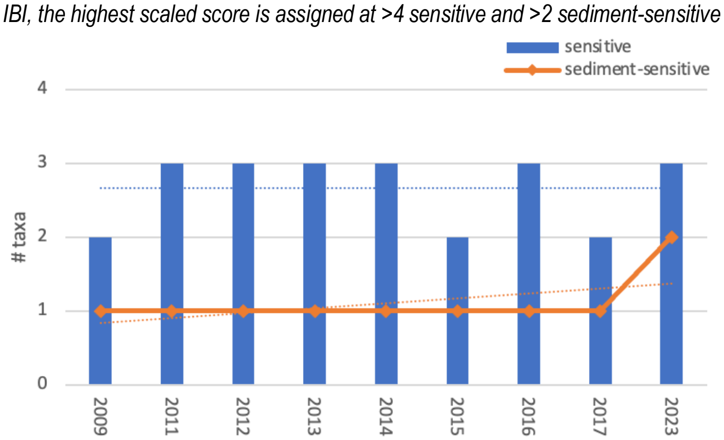


Figure 11. Relative abundance of tolerant and sediment-tolerant organisms at WC0150. Riffle-targeted samples were taken every year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

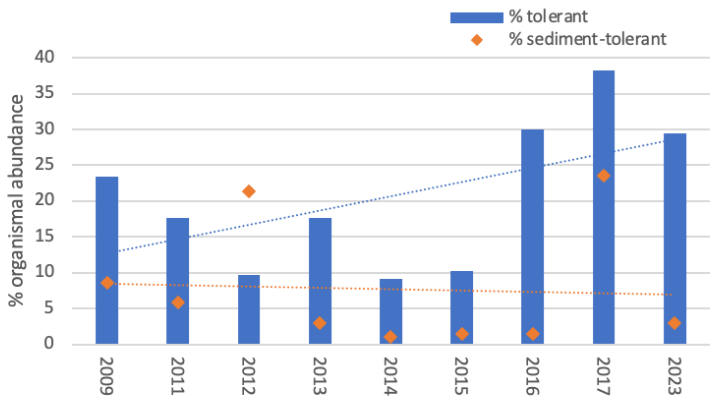


Figure 12. Temperature and fine sediment optima of the community (weighted means) at WC0150. Riffle-targeted samples were taken every year. Linear trendlines are shown.

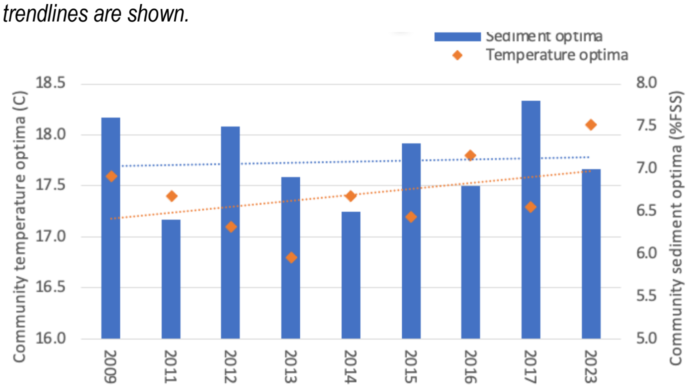
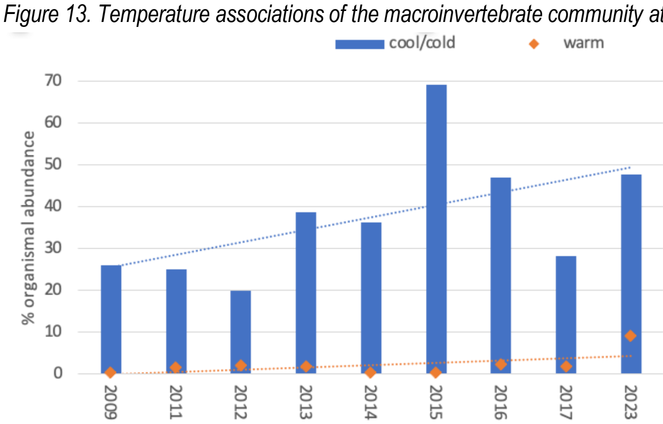


Figure 13. Temperature associations of the macroinvertebrate community at WC0150. RT samples were taken every year. Linear trendlines are shown.



Although several taxa were new to this site in 2023, there is no clear pattern to macroinvertebrate community similarity across different sampling years, and community composition was moderately similar (>55%) among all years (Figure 14). Differences in community composition (Figure 15) were driven primarily by *Simulium* (black fly associated with flowing water and recent habitat disturbance; dominant taxon in 2013-2015), Annelida (tolerant segmented worms; dominant taxon in 2012 and 2017), *Optioservus* (tolerant riffle beetle found in clear water and a variety of flows and temperatures; more abundant in recent years), and *Acentrella* (small minnow mayfly found in warmer streams; present in 2011 and 2012). Differences in traits calculated as relative abundances primarily distinguished between early and more recent sampling years, with the primary drivers being greater relative abundance of erosional, univoltine, and scraper organisms in recent years, and lower relative abundance of collectors in more recent years (Figure 16).

Figure 14. Cluster dendrogram of the WC0150 macroinvertebrate community in all sampling years. RT samples were taken in each year. The number at the end of each label indicates sampling year.

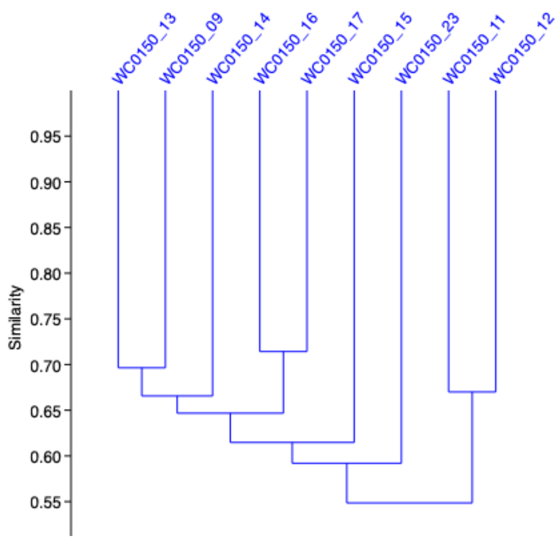


Figure 15. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0150 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. RT samples were taken in each year. Axis 1 explains 36% of total sample variation; axis 2 explains an additional 20% of variation.

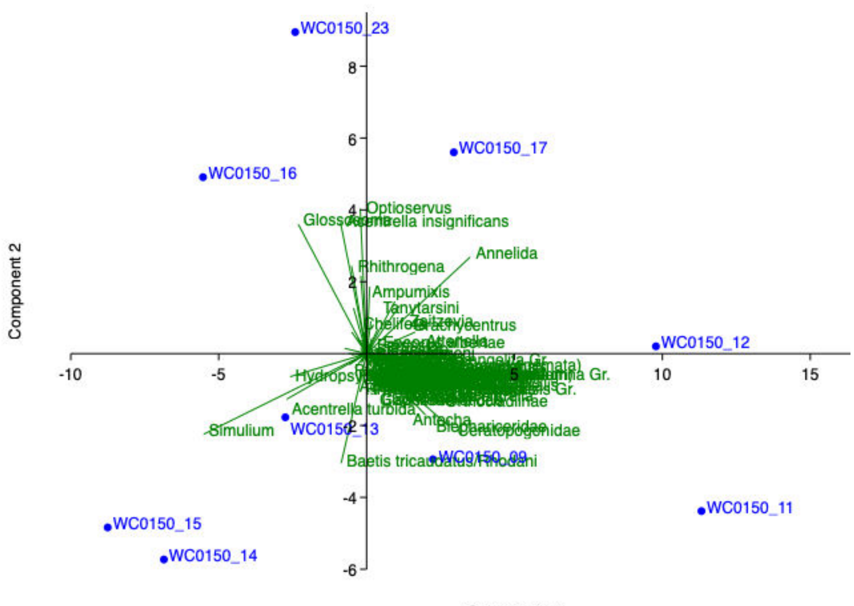
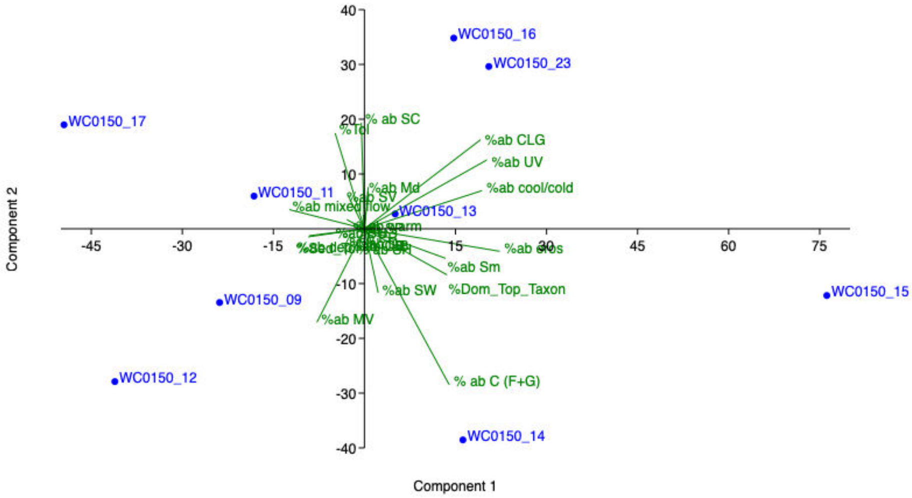


Figure 16. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC0150 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. RT samples were taken in all years. Axis 1 explains 56% of total sample variation; axis 2 explains an additional 24% of variation.



<u>WC0600 (Road 6360)</u>

RT samples were taken in the primary channel at WC0600 in every sampling year from 2005-2022; in 2018 and 2019, a PM sample was taken in the same reach. WC0600 is a long-term index site where no active restoration was done; however, it is subject to effects of restored perennial flow and surrounding land use and climate impacts. The 2023 sample was taken primarily in boulder/cobble substrate (Figure 3). The macroinvertebrate community was something of an outlier but was most similar to the WC0150 community (Figure 5). The community was dominated at 22.8% relative abundance by *Rhithrogena*, a sediment-sensitive flatheaded mayfly found on stones in faster flows (Figure 17). *Rhithrogena* dominated the 2019 sample at lower abundance, but this site has been dominated at moderate abundances by a variety of taxa (Figure 17), mostly associated with faster flows. All taxa taken in 2023 were found in at least one other year at this site.

Despite a lack of active restoration, trends analysis found significant increases across all sampling years in IBI score, total richness, number of sensitive taxa, and relative abundances of medium-bodied, univoltine, semivoltine, and climber organisms; and significant decreases in relative abundance of predators and crawlers (Appendix B). The majority of these trends suggest improvement in habitat quality and stability. The target subsampling number of 500 organisms was attained in all but three years (Figure 18), but the 2023 sample contained only 206 organisms. The reasons for this lower abundance are unclear, but this could impact calculated community metrics. Taxa richness increased over time; total and EPT richness in 2023 were both similar to the most recent sampling years, despite the smaller number of organisms (Figure 19), although EPT richness at this site was the lowest of any 2023 sample. IBI scores reflected slight to no disturbance in 13 of 15 sampling years, including 2023, while PREDATOR scores reflected fair to good condition in 10 sampling years, including 2023 (Figure 20). The number of sensitive taxa increased in recent years and was higher in 2023 than in any prior year (Figure 21), although the number of sediment-sensitive taxa was the lowest since 2017. However, there are consistently low relative abundances of sediment-tolerant organisms, and abundance of tolerant organisms decreased over time (Figure 22). Community temperature optima was lower in 2023 than in the prior two sampling years, and community sediment optima was similar to recent years (Figure 23). There are consistently fewer warm-associated than cool/cold-associated organisms at this site (Figure 24), but abundance of the latter decreased in recent years.

Figure 17. Relative abundance of the numerically dominant taxon at WC0060. Riffle-targeted samples were taken every year; multihabitat samples were taken in the same reach in 2018 and 2019. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

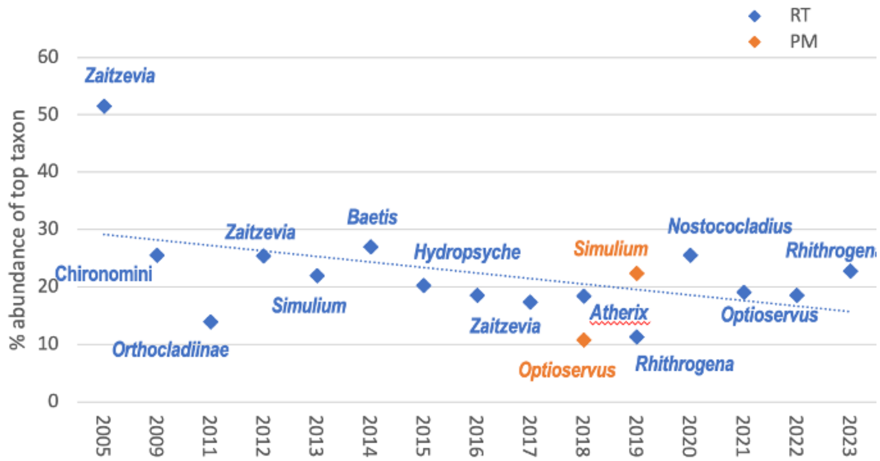


Figure 18. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC0600. Riffle-targeted samples were taken every year; multihabitat samples were taken in the same reach in 2018-2019. Target sub-sampling number is 500.

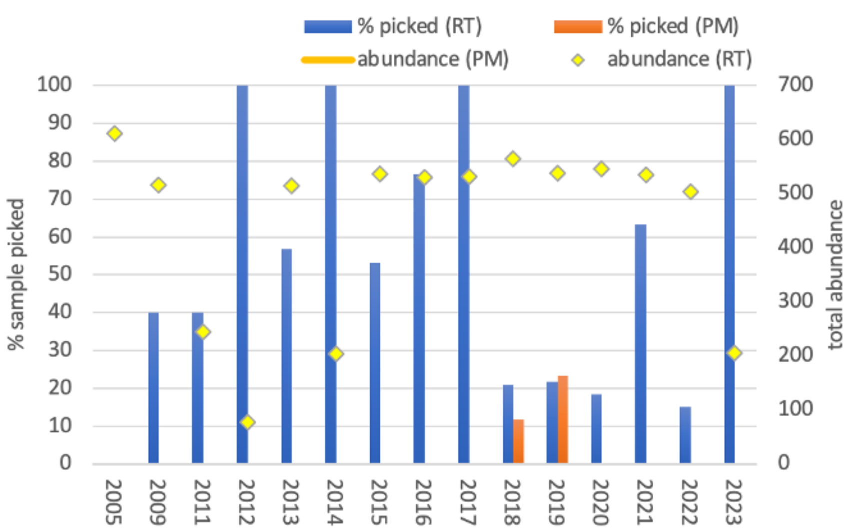


Figure 19. Sample richness and number of EPT taxa at WC0600. Riffle-targeted samples were taken every year; a multihabitat sample was taken in the same reach in 2018-2019. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

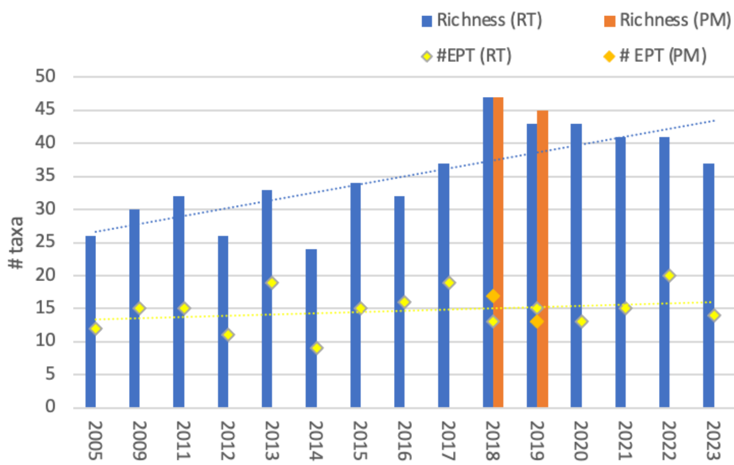


Figure 20. PREDATOR O/E and ORDEQ IBI scores at WC0600. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

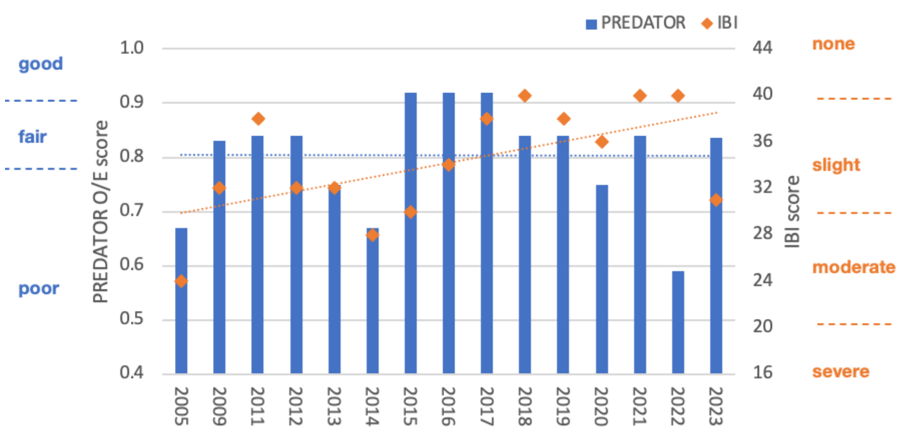


Figure 21. Sensitive and sediment-sensitive taxa at WC0600. Riffle-targeted samples were taken every year; multihabitat samples were taken in the same reach in 2018-2019; values were identical for years in which both sample types were taken. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and $\geq$2 sediment-sensitive taxa.

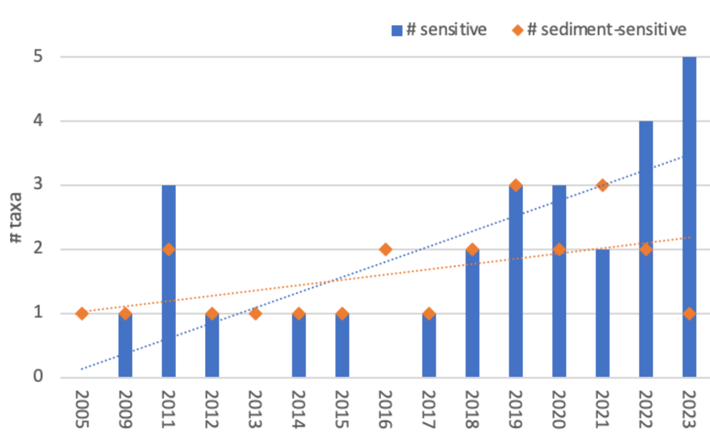


Figure 22. Relative abundance of tolerant and sediment-tolerant organisms at WC0600. Riffle-targeted samples were taken every year; multihabitat samples were also taken 2018-2019. Values were averaged for the years in which both sample types were taken. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

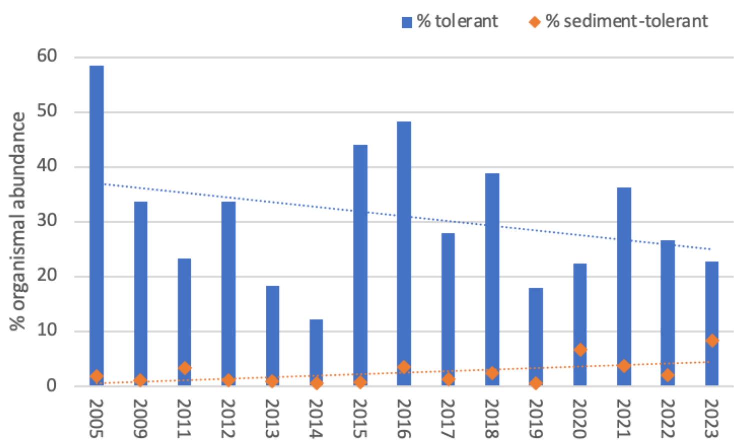


Figure 23. Temperature and fine sediment optima of the community (weighted means) at WC0600. Riffle-targeted samples were taken every year; multihabitat samples were also taken in 2018-2019. Values for years in which both sample types were taken were averaged. Linear trendlines are shown.

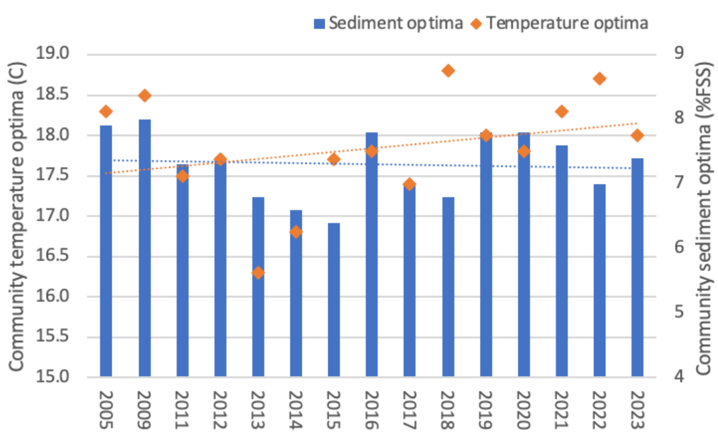
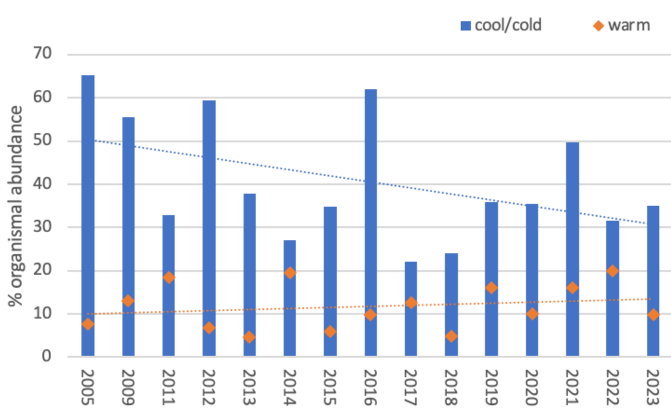


Figure 24. Temperature associations of the macroinvertebrate community at WC0600. Riffle-targeted samples were taken every year; multihabitat samples were also taken in 2018-2019. Values for years in which both sample types were taken were averaged. Linear trendlines are shown.



Macroinvertebrate community similarity was greater among recent samples (2018-2022), although the 2023 sample differed more (Figure 25). Differences in community composition between early and later sampling years (Figure 26) included greater abundance of *Rhithrogena* (sediment-sensitive flatheaded mayfly found on stones in faster flows) and *Acentrella insignificans* (small minnow mayfly found in warmer streams) in later sampling years (including 2023), as well as *Zaitzevia* (riffle beetle associated with fast flows and stable habitat; most abundant in 2005 and 2009) and Chironomini (tolerant, sediment-tolerant tribe of non-biting midge; abundant only in 2018 and 2019). Differences in traits calculated as relative abundances (Figure 27) included more multivoltine and medium-bodied organisms in more recent sampling years (2018-2023), and more small-bodied organisms in earlier years (2005-2014).

Figure 25. Cluster dendrogram of the WC0600 macroinvertebrate community. Blue = RT, aqua = PM. The number at the end of each label indicates sampling year.

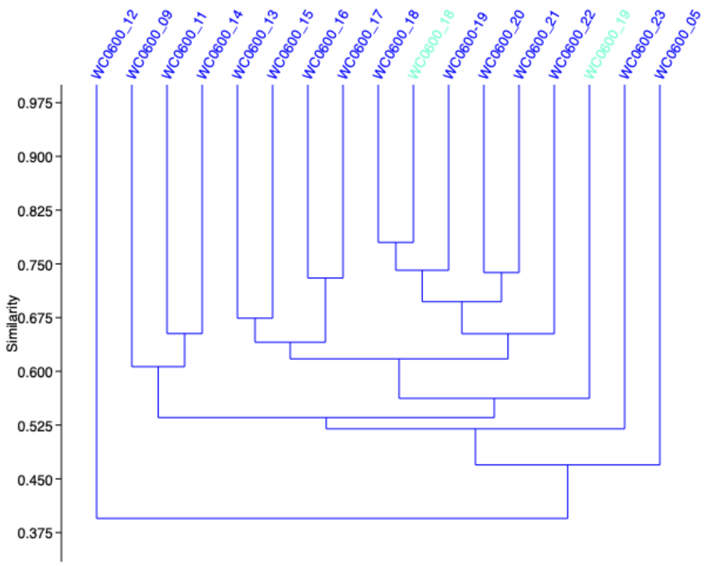


Figure 26. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0600 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 23% of total sample variation; axis 2 explains an additional 15% of variation.

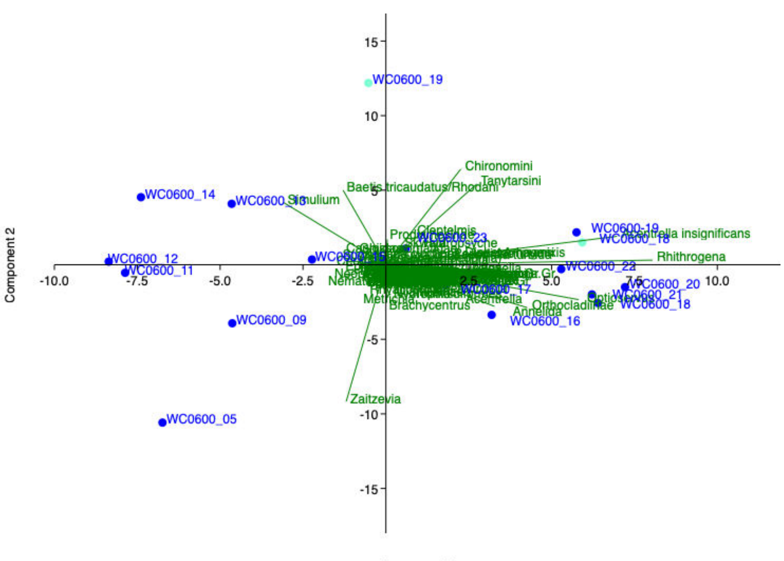
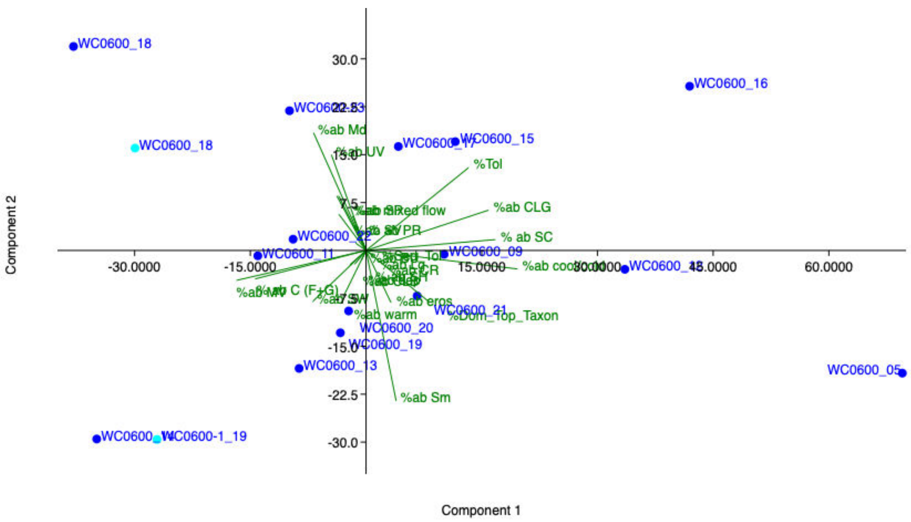


Figure 27. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0600 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 39% of total sample variation; axis 2 explains an additional 17% of variation.



<u>WC1025 (Rimrock Ranch)</u>

RT samples were taken in the primary channel of Whychus Creek (WC1025-1) around RM10.25 in 2011-2017, and 2022-2023; PM samples were taken in the same reach in 2019-2020 and 2022-2023 to provide additional baseline and post-restoration data for restoration actions implemented in 2021 as part of the Rimrock Phase II restoration project. PM samples were taken in two newly-formed side channels in 2022 and 2023 (WC1025-2, WC1025-3). Substrate in the primary channel RT and PM sample reach was primarily cobble/gravel (Figures 1 and 2); side channel samples were taken mainly in glides (Figure 3) with sand/silt (WC1025-2) or gravel (WC1025-3) substrate, though both had a greater proportion of wood substrate than any other 2023 sampling reach.

The macroinvertebrate community in the primary channel PM and RT samples was most similar to the community at WC1100, while the side channel sample communities were more similar to WC1175 and WC1250. However, this entire cluster of samples (WC1025-1 through WC1250) had similar community composition (Figure 5). The dominant taxon in all WC1025 samples was either Naididae (tolerant, sediment-tolerant sludge worm in softer sediments and slower flows; WC1025-1 RT, WC1025-3) or *Optioservus* (tolerant riffle beetle associated with clear water and stable habitat in a variety of flows and temperatures; WC1025-1 PM, WC1025-2), at low to moderate relative abundances (Figure 28). Organismal abundance in samples was generally high, with the target subsampling number of 500 attained every year in all side channel samples and in the primary channel except for 2017 (which may have been related to disturbance from restoration at WC1100 upstream in 2016) (Figure 29).

Five taxa were taken at this site for the first time, each at low abundance (1-2 individuals): Ostracoda (widespread, ubiquitous seed clam), *Onocosmoecus* (Northern caddisfly associated with slow-flowing sections of cool rivers), Physidae (common, tolerant bladder snail usually found on soft silty substrate) at WC1025-2; *Dixa* (meniscus midge found in slower sections of cool flowing water), and *Neoleptophlebia* (prong-gilled mayfly found in sediment and detritus in flowing water) at WC1025-1. The primary channel RT sample had the highest relative abundance of the top taxon, burrowers, organisms associated with slower flows, and of multivoltine, tolerant, and sediment-tolerant organisms and the lowest abundance of clingers among all 2023 samples, which suggests higher sediment levels and increased disturbance. In contrast, the primary channel PM sample had the greatest abundance of large-bodied organisms in 2023, which implies more stable habitat conditions, as larger organisms often have a longer developmental time. The WC1025-2 side channel community had the greatest abundance of shredders, sprawlers, the highest community sediment optima, fewest swimmers, and was the only 2023 sample to lack sediment-sensitive taxa, suggesting both higher sediment levels and a more intact riparian zone. The WC1025-3 community had the fewest caddisfly and sensitive taxa of all 2023 samples.

A trends analysis of primary channel samples (Appendix B) revealed significant unidirectional increases across time in the numbers of total, Ephemeroptera, and EPT taxa (Figure 30) and in relative abundance of tolerant and sediment-tolerant organisms, scrapers, univoltine organisms, organisms associated with both erosional and depositional flows, and climbers; and a significant decrease in the relative abundance of swimmers. These results are mixed, as some significant trends suggest improving habitat conditions (i.e., taxa richness metrics) while others suggest a decline (i.e., tolerant and sediment-tolerant organisms).

IBI scores for the primary channel communities reflected moderate to slight disturbance in all years; PREDATOR scores varied more, reflecting poor, fair, or good conditions in different years (Figure 31). Numbers of sensitive and sediment-sensitive taxa increased since 2017, with the new side channels supporting sensitive taxa as well, especially WC1025-2 (Figure 32). However, relative abundance of tolerant and sediment-tolerant organisms increased significantly (Figure 33), so these mixed results may be influenced by increased sample richness in this span. Community sediment and temperature optima increased over time, though not significantly, and are similar in primary and side channel samples (Figure 34). Relative abundance of organisms associated with cooler waters decreased over time though not significantly, and cool-associated organisms greatly outnumber warm-associated in all years and channel types (Figure 35).

Figure 28. Relative abundance of the numerically dominant taxon at WC1025. Blue = RT; green = PM; RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

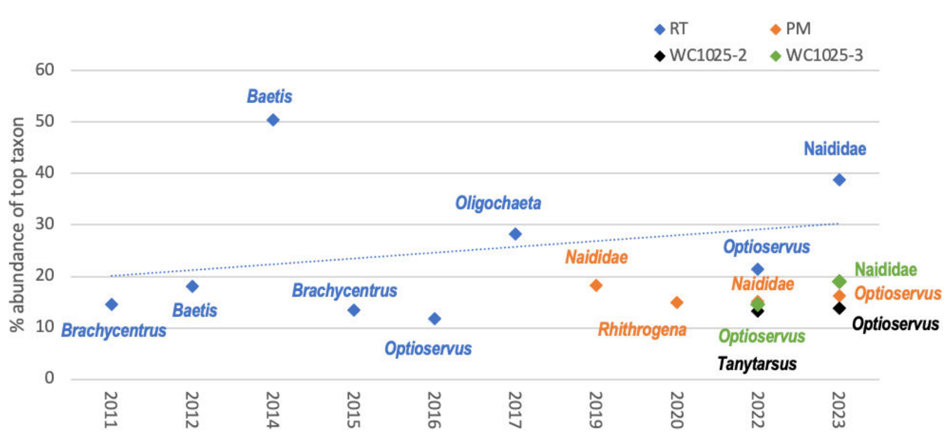


Figure 29. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1025 in all sampling years. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Target sub-sampling number is 500.

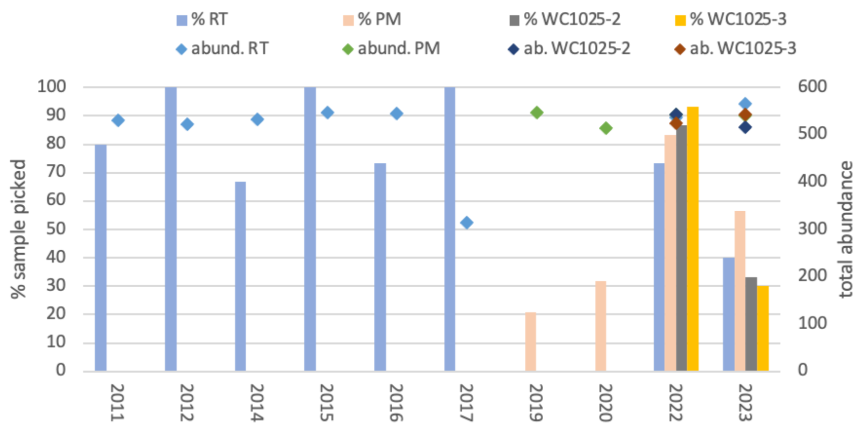


Figure 30. Sample richness and number of EPT taxa at WC1025. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

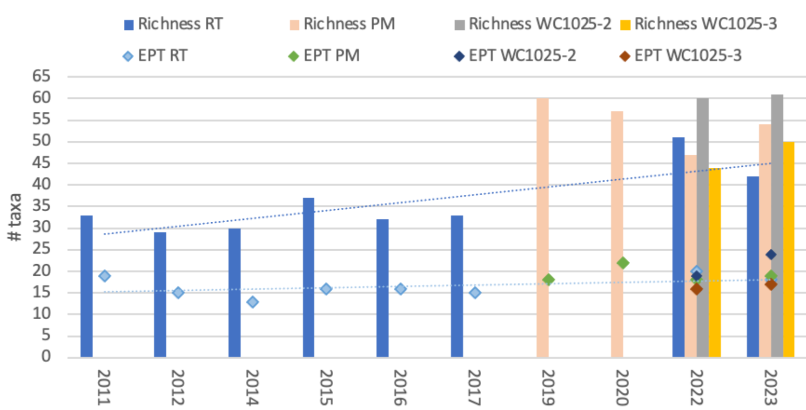


Figure 31. PREDATOR O/E and ORDEQ IBI scores at WC1025 in years RT samples were taken. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

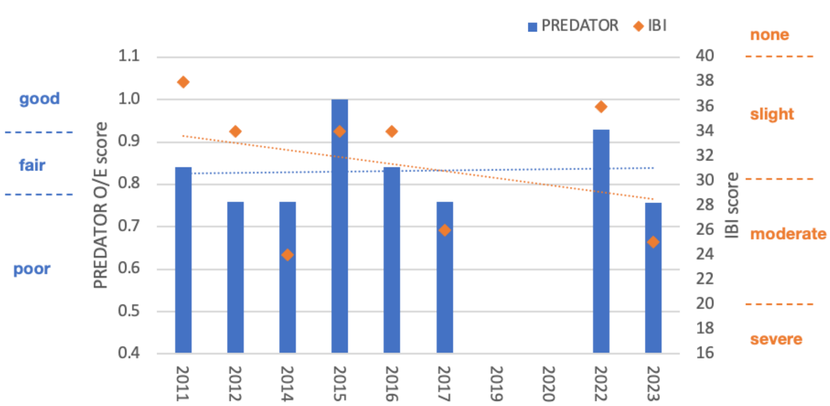


Figure 32. Sensitive and sediment-sensitive taxa at WC1025. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.

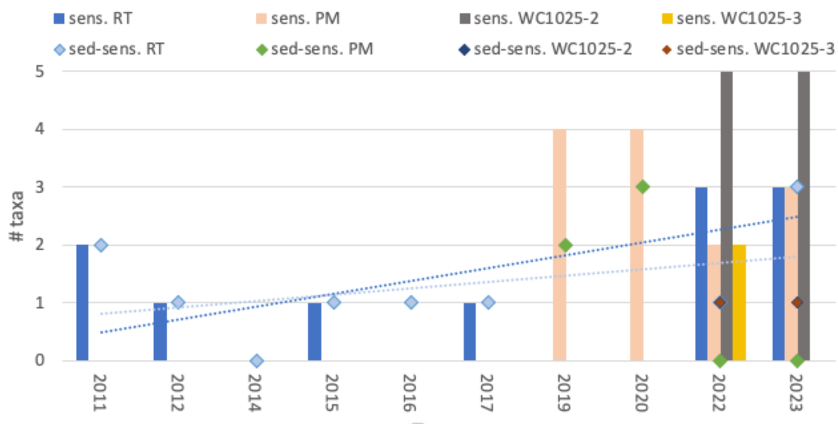


Figure 33. Relative abundance of tolerant and sediment-tolerant organisms at WC1025 in all sampling years. RT samples were taken 2011-2012 and 2014-2017; PM samples were taken 2019-2020; RT and PM samples were taken in 2022-2023. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

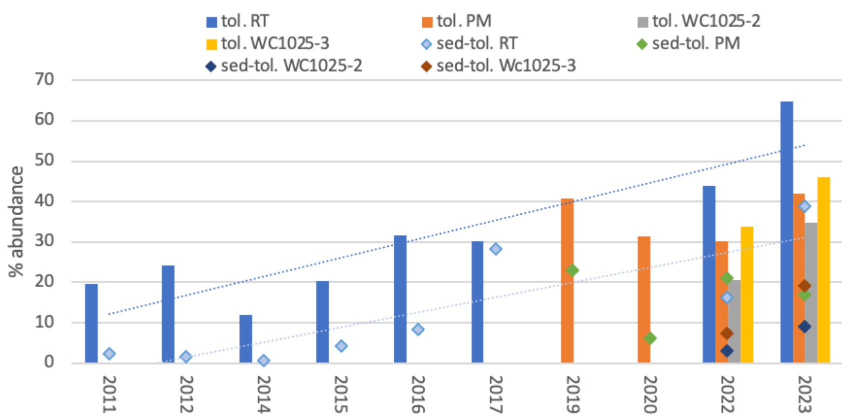


Figure 34. Temperature and fine sediment optima of the community (weighted means) at WC1025. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Linear trendlines are shown.

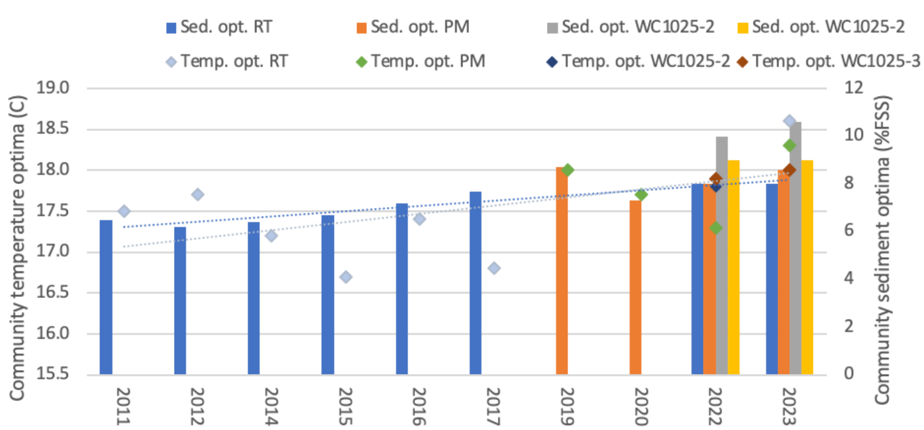
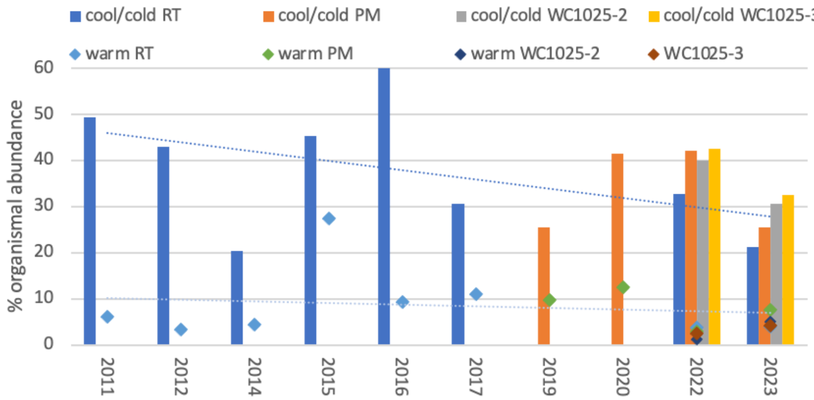


Figure 35. Temperature associations of the macroinvertebrate community at WC1025. RT and PM show primary channel data; WC labels indicate side channels formed post-restoration. Linear trendlines are shown.



Macroinvertebrate community composition was significantly different pre- and post-restoration. The community was more similar in all samples taken in 2022 and 2023, and more similar among samples taken in recent (2019-2020) compared to earlier years (Figure 35). Taxa differences between samples reflected differences between years and sampling techniques, with more Annelida (tolerant segmented worms) in PM samples, more *Baetis tricaudatus* (small minnow mayfly associated with faster flows and clearer water) in earlier years, and more of the nonbiting midge tribes Tanytarsini (builds tubes in soft sediments) and Chironomini (tolerant, warm-associated) in recent years (Figure 36). Differences in traits calculated were driven mainly by greater abundances of clingers and organisms associated with faster and cooler flows in RT samples, and more univoltine organisms in recent sampling years (Figure 37).

Figure 35. Cluster dendrogram of the WC1025 macroinvertebrate community. Blue = RT, aqua = PM. WC1025-1 is the primary channel; WC1025-2 and WC1025-3 are side channels formed post-restoration. The number at the end of each label indicates sampling year.

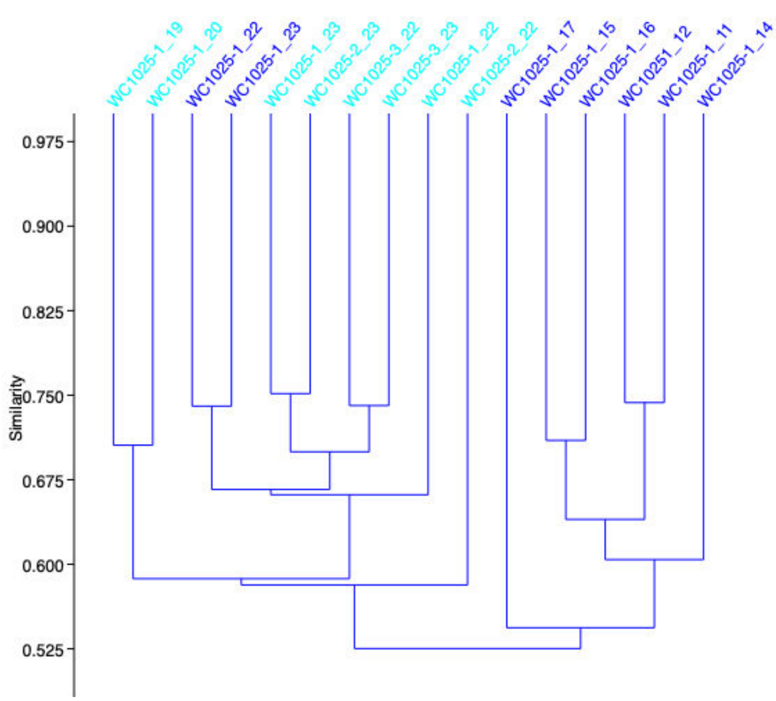


Figure 36. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among WC1025 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. WC1025-1 is the primary channel; WC1025-2 and WC1025-3 are side channels formed post-restoration. Axis 1 explains 30% of total sample variation; axis 2 explains an additional 15% of variation.

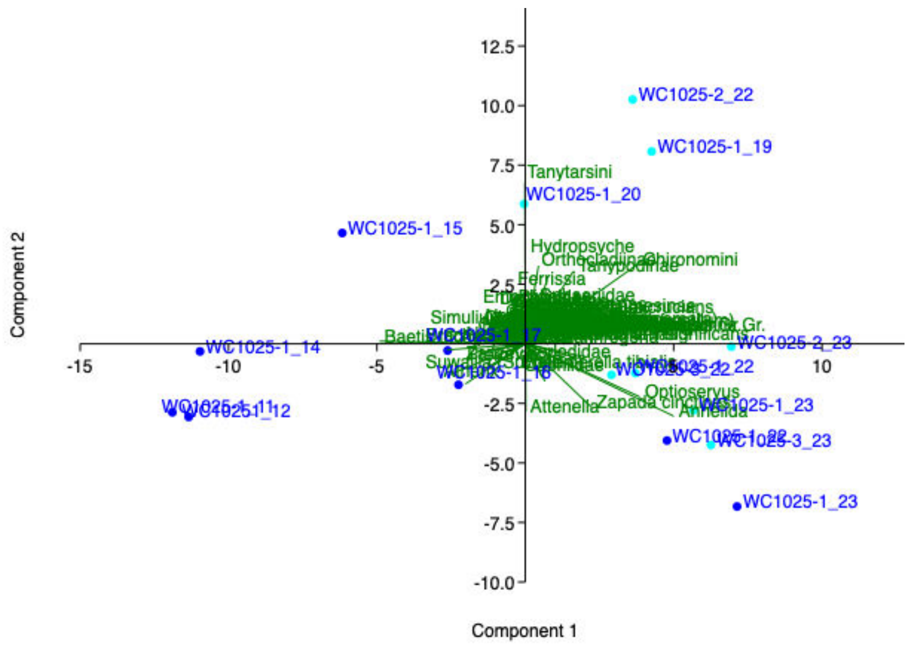
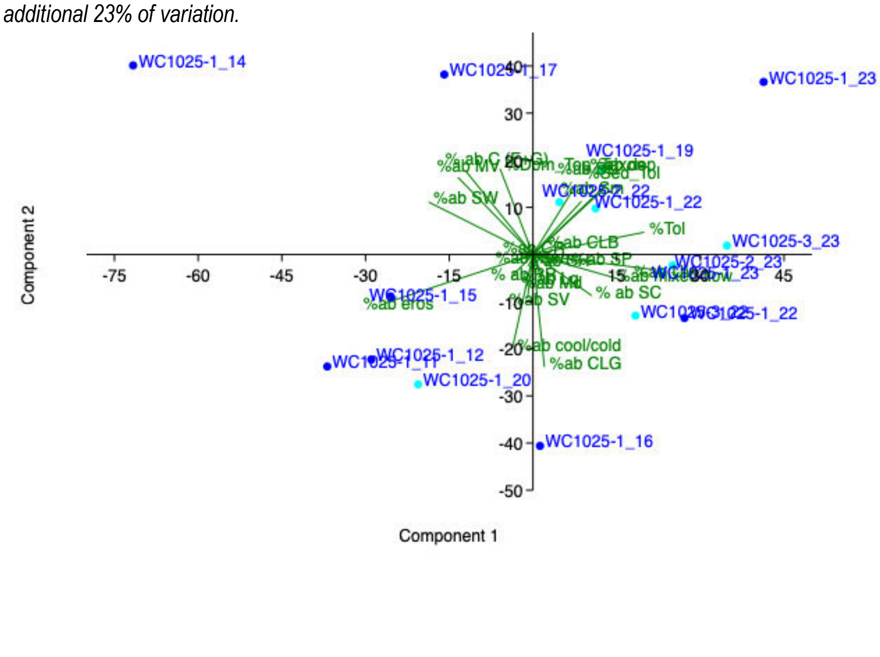


Figure 37. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1025 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. WC1025-1 is the primary channel; WC1025-2 and WC1025-3 are side channels formed post-restoration. Axis 1 explains 37% of total sample variation; axis 2 explains an additional 23% of variation.



WC1100 (Whychus Canyon reach 4)

Riffle-targeted samples were taken in the primary channel of Whychus Creek (WC1100-2) at RM 11.0 in 2014-2015 to collect baseline data prior to a 2016 Whychus Canyon restoration project that created a braided, dynamic system with new side channel habitats. After restoration, RT and PM samples were taken in the same longitudinal reach of the new primary channel (2017-2022), although extensive channel braiding post-restoration resulted in the original primary channel also having secondary channel characteristics. PM samples were taken in different side channels in 2017-2023, and the only side channel sampled in 2023 was WC1100-4. The primary channel PM sample in 2023 was taken primarily in riffles and runs with cobble substrate; and the side channel sample was taken primarily in glides with sand/silt substrate (Figures 2 and 3).

Primary and side channel communities at WC1100 in 2023 were most similar to primary and side channel communities (respectively) at WC1025 (Figure 5). The communities in both channels consisted mainly of small organisms that move as clingers, feed as collectors, prefer cooler water, and tolerate a variety of flow types. Only one taxon was taken in this reach for the first time in 2023: *Leucotrichia*, a tolerant pursecase-making caddisfly found on the upper surfaces of rocks in flowing portions of large open streams (1 individual, primary channel). The dominant taxon in the primary and side channel samples was present at similar and fairly low relative abundance (Figure 38) with Naidinae (tolerant, sediment-tolerant sludge worms associated with slower flows) dominating the PC community and *Polypedilum* (widespread non-biting midge found in a range of flows and temperatures) dominating the SC community. The PC sample had the greatest abundance of warm-associated organisms among all 2023 samples, while the SC sample had the highest MHBI value, the most Chironomini (tolerant non-biting midges), and the greatest abundance of organisms that are small and associated with a variety of flow types, as well as the fewest predators, collectors, and organisms associated with faster flows.

Organismal abundance in samples from this reach is generally high, with the target sub-sampling number of 500 attained in WC1100-2 and WC1100-4 in all years except the 2019 and 2020 PM samples (Figure 39). A trends analysis of primary channel samples revealed significant unidirectional increases across time in IBI score; numbers of total, mayfly, EPT, sensitive, and sediment-sensitive taxa; community sediment optima; relative abundance of tolerant and sediment-tolerant organisms; relative abundance of scrapers, univoltine, semivoltine, and cool/cold-associated organisms; organisms that move as climbers and clingers; and a significant decrease across time in relative abundance of multivoltine organisms. Except for a single trait (community sediment optima), these trends suggest improvement in habitat quality and stability. A trends analysis of the WC1100-4 side channel revealed significant unidirectional increases across time in the number of sensitive taxa and relative abundance of tolerant and sediment-tolerant organisms; and significant decreases in relative abundance of shredders, collectors, and swimmers.

Consistent with the trends analysis, total and EPT taxa richness in the primary channel increased overall since 2018, and the side channel sample values for these traits have been similar to the primary channel (Figure 40). No RT samples were taken in this reach in 2023 so there are no new PREDATOR or IBI scores, but IBI scores in past years reflected moderate to slight disturbance in all years while PREDATOR scores varied more, reflecting poor, fair, or good conditions. Few sediment-sensitive taxa were taken in PC or SC samples, but sensitive taxa richness increased over time, with more taken in the primary channel PM sample and SC sample in 2023 than in prior years (Figure 41).

Abundance of tolerant organisms generally exceeds that of sediment-tolerant organisms in each sampling year and has increased in recent years (Figure 42). Community sediment optima are generally higher in SC vs. PC samples, and both sediment and community temperature optima increased in recent years (Figure 43). Relative abundance of cool/cold-associated organisms decreased in the most recent sampling years, but since 2018 there are generally more cold-associated than warm-associated organisms each year in both PC and SC samples (Figure 44).

Figure 38. Relative abundance of the numerically dominant taxon at WC1100-2 (primary channel) and WC1100-4 (side channel). RT and PM = primary channel samples; only PM samples were taken at WC1100-4. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

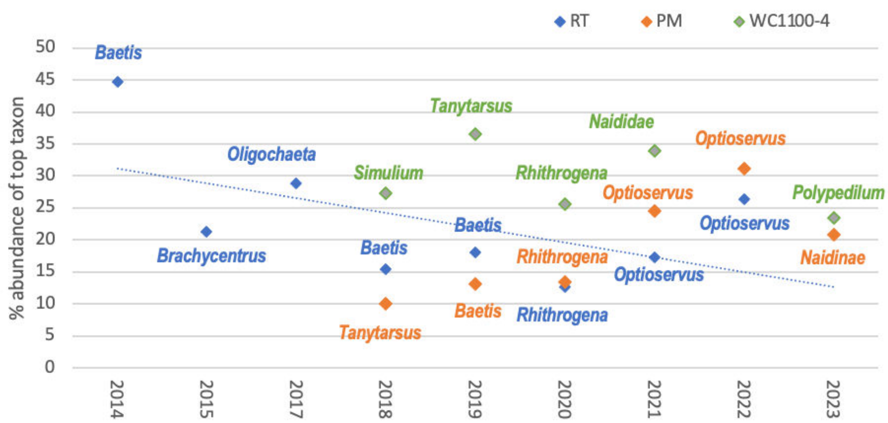


Figure 39. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1100. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Target sub-sampling number is 500.

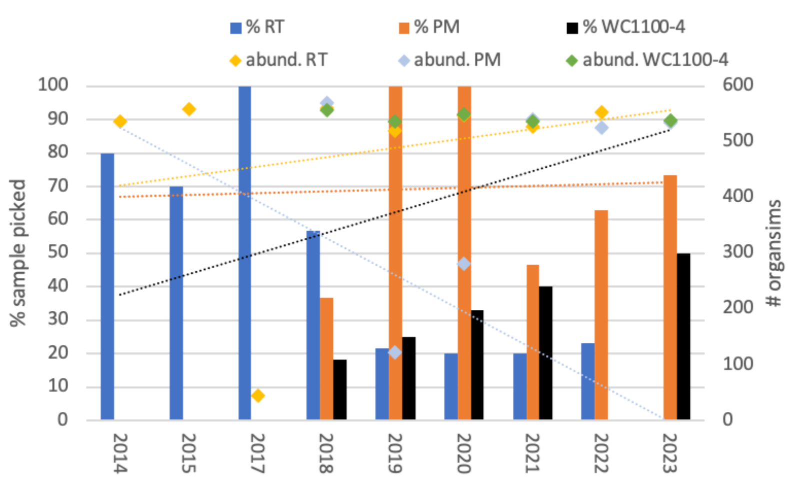


Figure 40. Sample richness and number of EPT taxa at WC1100. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

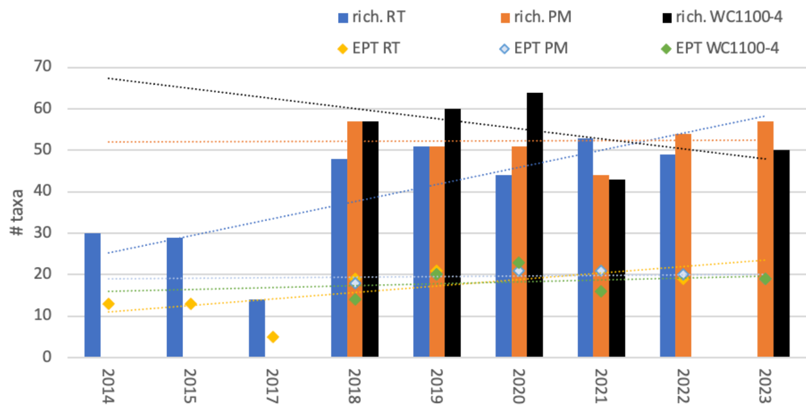


Figure 41. Numbers of sensitive and sediment-sensitive taxa at WC1100. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

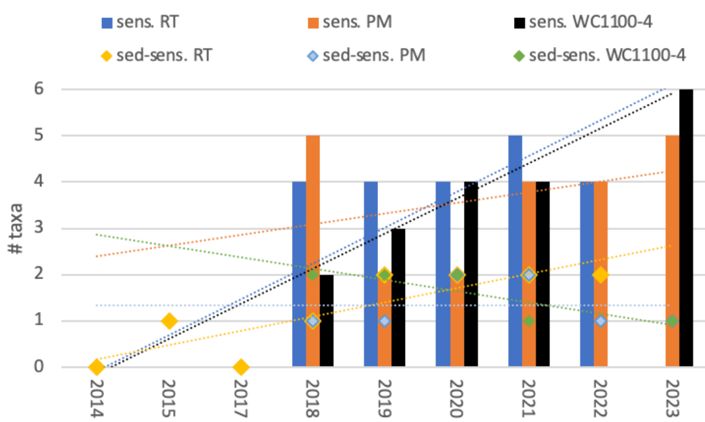


Figure 42. Relative abundance of tolerant and sediment-tolerant organisms at WC1100 in all sampling years. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

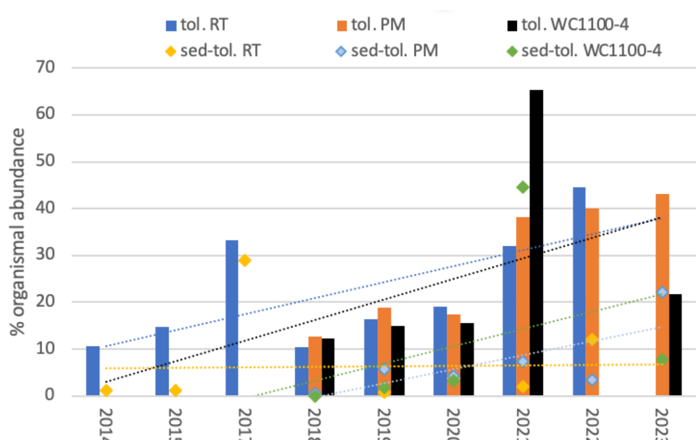


Figure 43. Temperature and fine sediment optima of the community (weighted means) at WC1100. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Linear trendlines are shown.

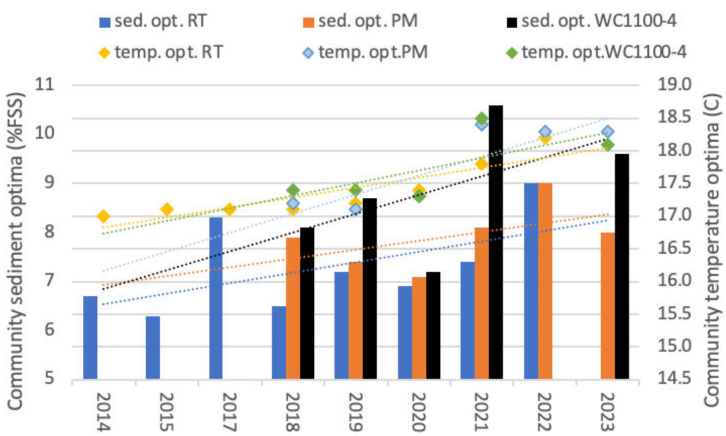
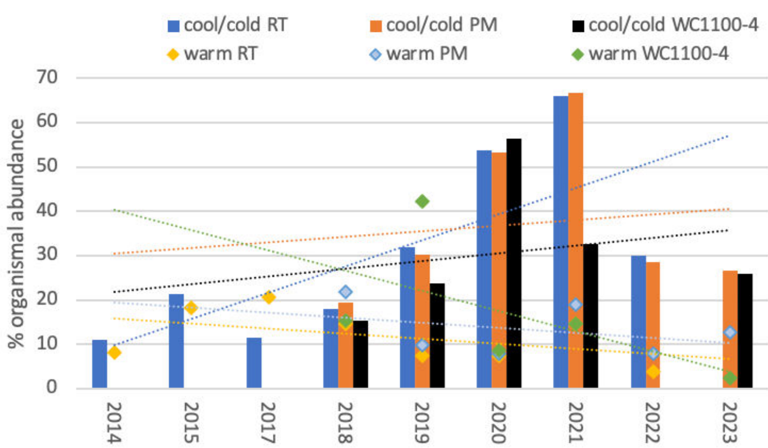


Figure 44. Temperature associations of the macroinvertebrate community at WC1100 in all sampling years. RT and PM = primary channel samples; only PM samples were taken at WC1100-4. Linear trendlines are shown.



Macroinvertebrate community composition was significantly different pre- and post-restoration, and community differences were influenced more by sampling year than by sampling method or channel type. Overall similarity was greater among the 2018-2020 communities and the 2021-2023 communities (Figure 45), suggesting larger community changes following restoration. The 2017 community differed more from all other years, likely due to restoration-related disturbance. Community differences (Figure 46) were largely driven by *Baetis tricaudatus* (small minnow mayfly associated with faster flows and clearer water; more abundant pre-restoration), Annelida (tolerant segmented worms ; more abundant in post-restoration PM samples), Chironomini (tolerant non-biting midge tribe; more abundant in SC samples) and *Brachycentrus* (humpless casemaking caddisfly with long developmental time found in flowing areas of larger streams on woody material and vascular hydrophytes; more abundant in PC samples). Differences in traits calculated as relative abundances distinguished between pre- and post-restoration communities, with more clingers and organisms associated with cooler, faster flows post-restoration, and more tolerant organisms in PM samples from recent years (Figure 47).

Figure 45. Cluster dendrogram of the WC1100 macroinvertebrate community in all sampling years. Blue = RT, aqua = PM; WC1100-2 is the primary channel. The number at the end of each label indicates sampling year.

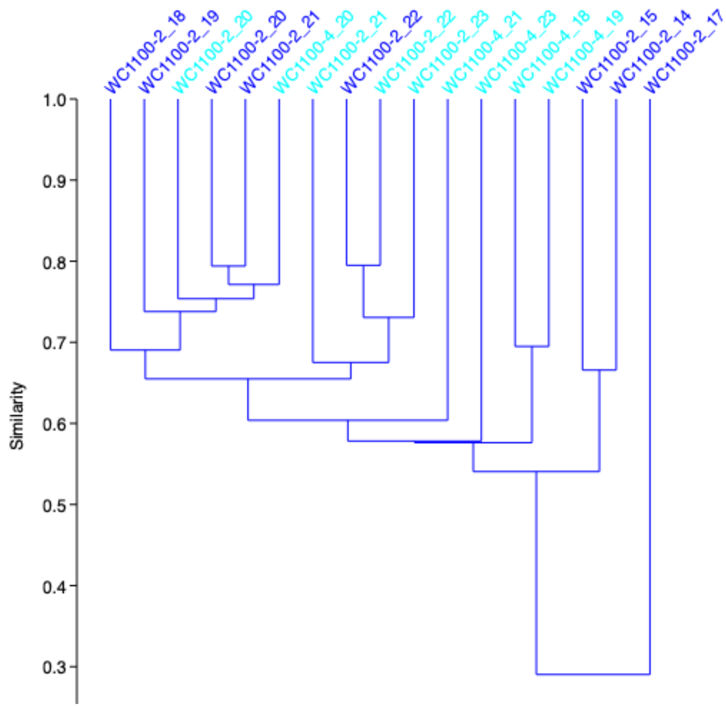


Figure 46. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among WC1100 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. WC1100-2 is the primary channel; WC1100-4 is a primary/side channel that developed post-restoration. Axis 1 explains 30% of total sample variation; axis 2 explains an additional 16% of variation.

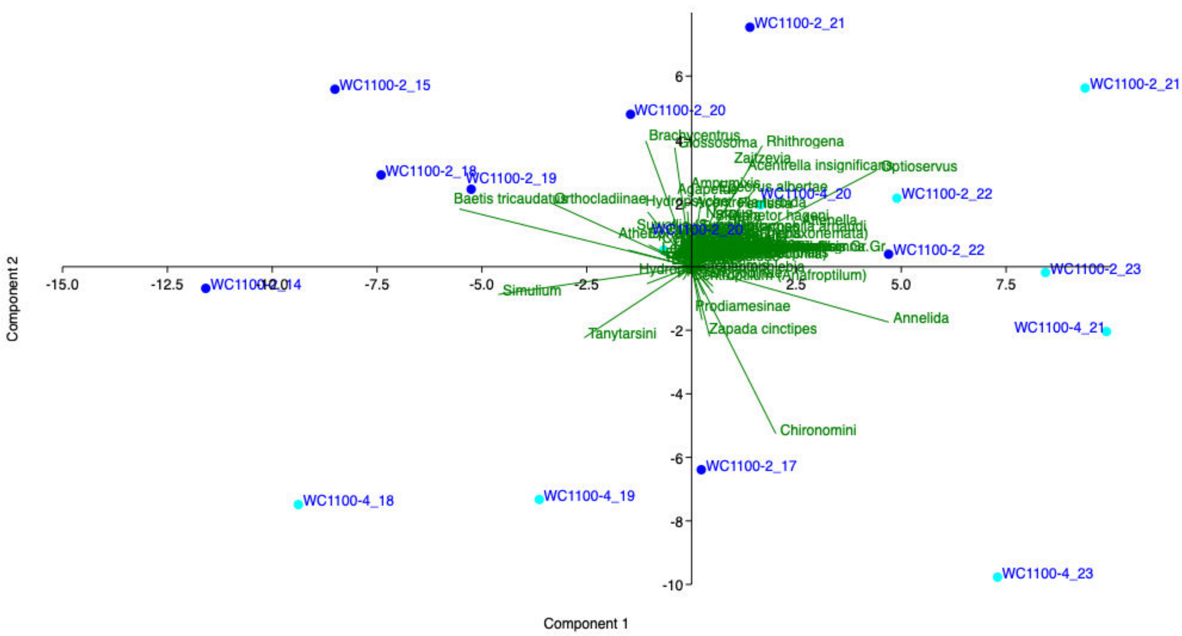
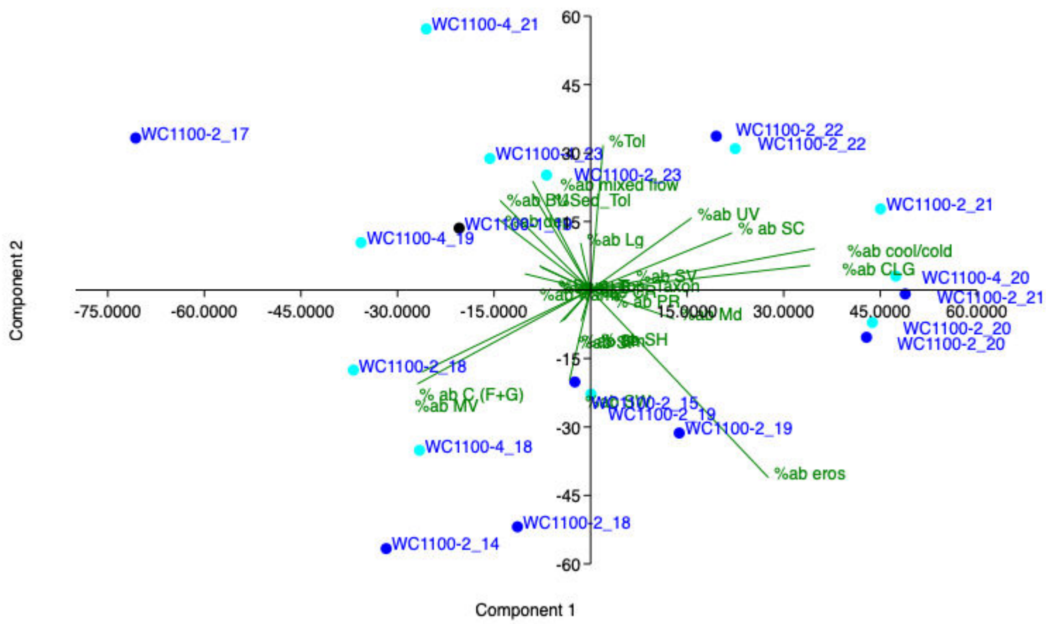


Figure 47. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1100 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. WC1100-2 is the primary channel; WC1100-4 is a primary/side channel that developed post-restoration. Axis 1 explains 34% of total sample variation; axis 2 explains an additional 27% of variation.



Comparison to adjacent unrestored reach

WC1150 was designated as an upstream control for the restored WC1100 reach. While taxa and trait differences in the WC1150 community in 2017 suggest that activities at WC1100 in 2016 may have affected the control reach, it is the most spatially appropriate reach for comparison. The only metric that differed significantly between the two reaches pre- and post-restoration was the number of EPT taxa, which was significantly greater in the control reach prior to but not following restoration (Table 4). However, mean values of all positive metrics (i.e., increasing score corresponds to improving condition) for model scores, richness, and temperature association were higher post-restoration at the restored reach, and were the same or greater post-restoration in the control reach. In contrast, the mean number of warm-associated organisms was similar and low in both reaches pre- and post-restoration.

Table 5. Differences in selected community metrics between restored (WC1100-2) and upstream control (WC1150) reaches pre- and post-restoration. Mean values shown with standard deviation in parentheses. Bold type indicates a significant difference (p <0.05).

	Pre-restoration			Post-restoration		
	WC1100-2	**WC1150**	**Sig.?**	**WC1100-2**	**WC1150**	**Sig.?**
PREDATOR	0.845 (0.120)	0.785 (0.078)	N	0.917 (0.219)	0.813 (0.046)	N
IBI	28 (5.7)	32 (2.8)	N	37 (10.5)	32 (10.6)	N
# total taxa	29.5 (0.71)	31.5 (2.12)	N	47.8 (11.4)	44.7 (12.7)	N
# EPT taxa	**13 (0)**	**15.5 (0.7)**	**Y**	18.8 (4.5)	17.4 (2.8)	N
Cool/cold-associated	16.2 (7.2)	26.5 (14.0)	N	36.3 (18.8)	40.7 (20.9)	N
Warm-associated	13.2 (7.2)	10.0 (3.7)	N	12.3 (5.8)	10.1 (7.1)	N

WC1150 (Whychus Canyon reach 3)

Sampling began at WC1150 in 2014 to provide an upstream reference for WC1100, as no active restoration was done here. RT samples were taken in 2014-2018 and 2022; PM samples were taken in 2018-2021 and 2023. The 2023 sample was taken primarily in riffles with cobble substrate (Figures 2 and 3), and the community was most similar to those at sites immediately upstream (WC1175-WC1250; Figure 5). The 2023 community consisted mainly of small, univoltine organisms that move as clingers, feed as collectors, prefer cooler water, and tolerate a variety of flow types. The community was fairly balanced; it was dominated at a low relative abundance by tolerant Naidinae sludge worms (Figure 48), but this site was dominated in most years by taxa associated with cool flowing water including *Glossosoma* (saddlecase caddisfly), *Ampumixis* (riffle beetle), *Rhithrogena* (flatheaded mayfly), and *Nostococladius* (nonbiting midge that mines in *Nostoc* algae). Three taxa were taken in this reach for the first time in 2023, at abundances ranging from 1-3 individuals: Prodiamesinae (non-biting midge subfamily associated with cooler faster flows and tolerant of high levels of entrained fine sediments), *Paraleptophlebia* (prong-gilled mayfly associated with sediments and detritus in fast cool flows), and *Protoptila* (saddlecase-maker caddisfly associated with larger warmer rivers in both clear and sedimented waters).

Organismal abundance in this reach is generally high, with the target sub-sampling number of 500 attained in all but two years (2017, 2022; Figure 49). A trends analysis (Appendix B) revealed significant unidirectional increases across time in mayfly taxa richness, community temperature optima, relative abundance of tolerant, large, univoltine, semivoltine, and climber organisms; and significant unidirectional decreases in relative abundance of predators, multivoltine, and crawler organisms. These results are mixed, as the direction of the trends in voltinism, body length, and taxonomic metrics suggest improved and more stable habitat conditions, while the trends in feeding guild and tolerance metrics suggest increased stressors.

Numbers of total and EPT taxa increased since sampling began, especially after 2017 (Figure 50); from 2018-2023 there were more total taxa (mean = 48, SD 10.2) and EPT taxa (mean = 18.2, SD 2.2) compared to earlier years (2014-217; mean total taxa = 30.5, SD 4.04; mean EPT = 15.3, SD 1.7), though the difference was significant only for total taxa. No RT samples were taken in 2023 so there are no new model scores, but IBI scores reflected moderate to no disturbance in most years, while PREDATOR scores reflected fair to good conditions in four of the six years that RT samples were taken. There were significantly more sensitive and sediment-sensitive taxa in 2018-2023 compared to prior years (Figure 51). Relative abundance of tolerant organisms was greater in later sampling years while mean abundance of sediment-tolerant organisms is consistently lower, but the differences were not significant, although values for both metrics increased in recent sampling years (Figure 52). Community temperature and sediment optima increased overall since sampling began (Figure 53), though the trend was significant only for community temperature optima, which was also significantly greater in 2018-2023 compared to the earlier years. Abundance of cool/cold-associated organisms generally exceeded that of warm-associated in each year, especially since 2018, though neither differed significantly between the two time ranges.

Figure 48. Relative abundance of the numerically dominant taxon at WC1150. RT = riffle-targeted; PM = multihabitat. In the ORDEQ IBI, <20% abundance of the top taxon receives the highest scaled score.

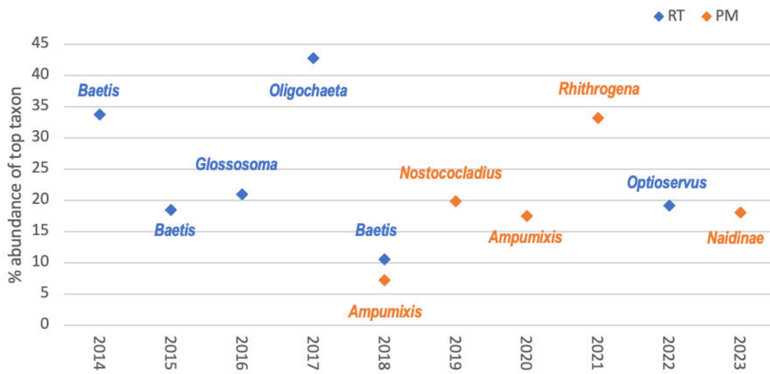


Figure 49. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1150. RT = riffle-targeted; PM = multihabitat. Target sub-sampling number is 500.

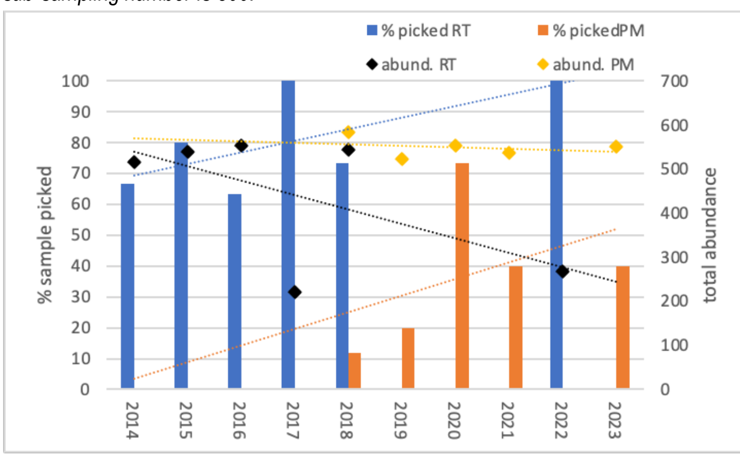


Figure 50. Sample richness and number of EPT taxa at WC1150. RT = riffle-targeted, PM = multihabitat. For the year that both a PM and RT sample were taken (2018), values were averaged. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

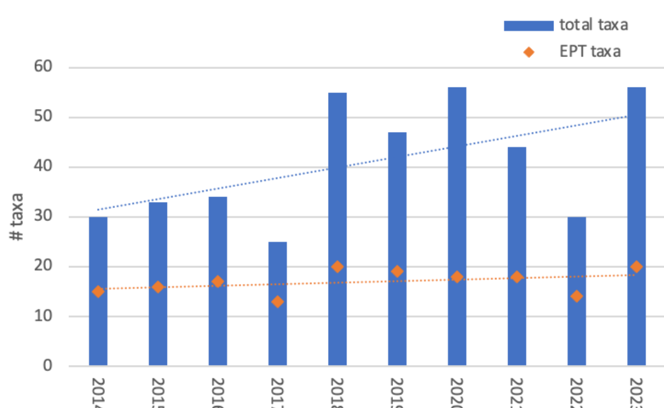


Figure 51. Numbers of sensitive and sediment-sensitive taxa at WC1150. RT = riffle-targeted, PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

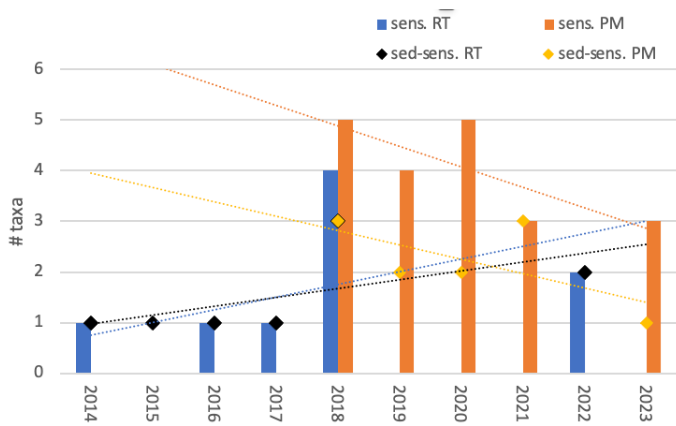


Figure 52. Relative abundance of tolerant and sediment-tolerant organisms at WC1150. RT = riffle-targeted, PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled scores are assigned at <15% tolerant and <10% sediment-tolerant.

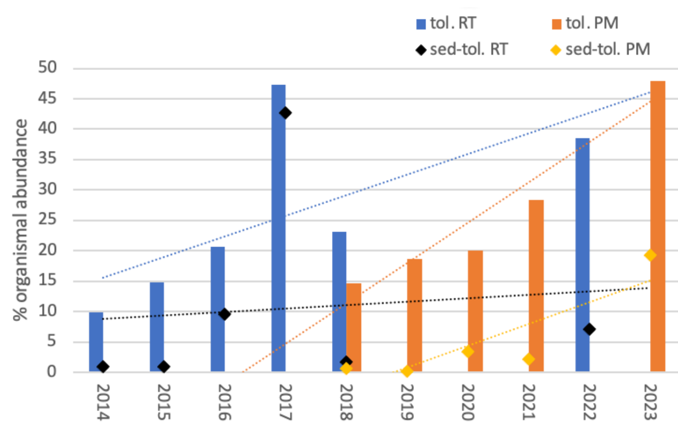


Figure 53. Temperature and fine sediment optima of the community (weighted means) at WC1150. RT = riffle-targeted, PM = multihabitat. Linear trendlines are shown.

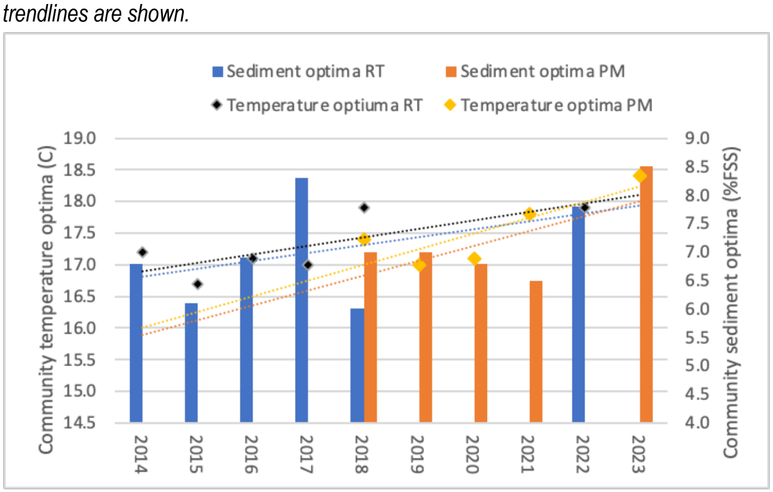
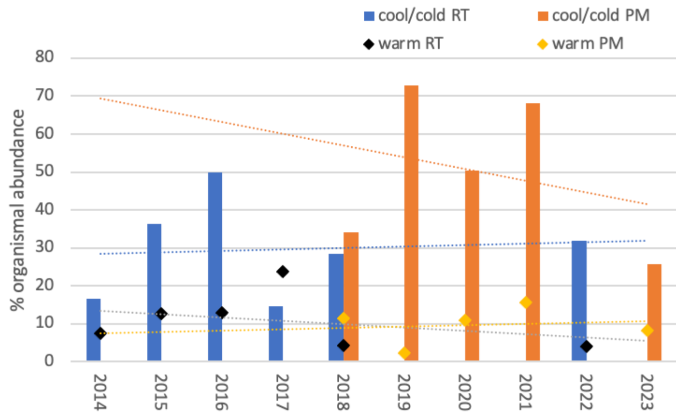


Figure 54. Temperature associations of the macroinvertebrate community at WC1150. RT = riffle-targeted, PM = multihabitat. Linear trendlines are shown.



Macroinvertebrate community composition was also more similar among samples from recent years (2018-2023) compared to prior years (Figure 55). Differences in community composition reflected the trait differences between earlier and later sampling periods and were driven mainly by *Baetis tricaudatus* (small minnow mayfly associated with faster flows and clearer water), *Simulium* (black fly associated with flowing water and recent habitat disturbance), and *Ampumixis* (sediment-intolerant riffle beetle associated with clear, cold flowing water), all of which were more abundant in earlier years (Figure 56). Differences in traits calculated as relative abundances distinguished between early and later sampling periods, with greater abundance of clingers, tolerant, and erosional flow-associated organisms in recent years and fewer multivoltine organisms. These differences suggest increased flow and greater habitat stability during the 2018-2023 span.

Figure 55. Cluster dendrogram of the WC1150 community in all sampling years. Blue = riffle-targeted, aqua = multihabitat. The number at the end of each label indicates sampling year.

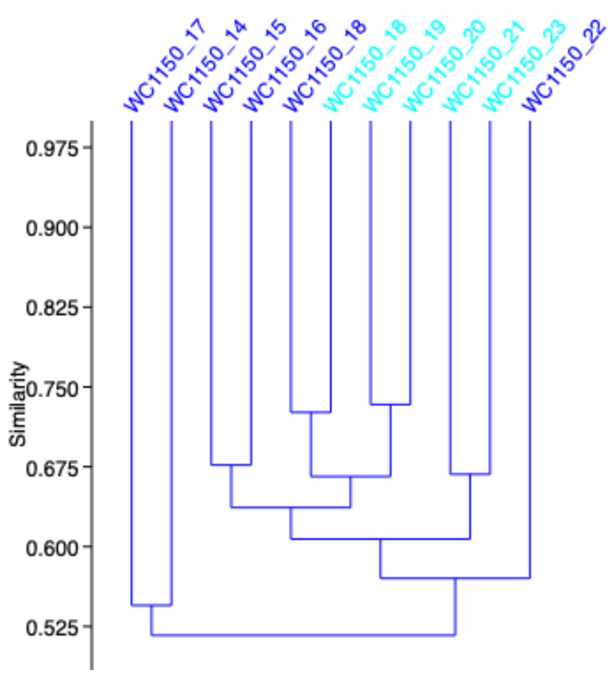


Figure 56. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1150 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 25% of total sample variation; axis 2 explains an additional 19% of variation.

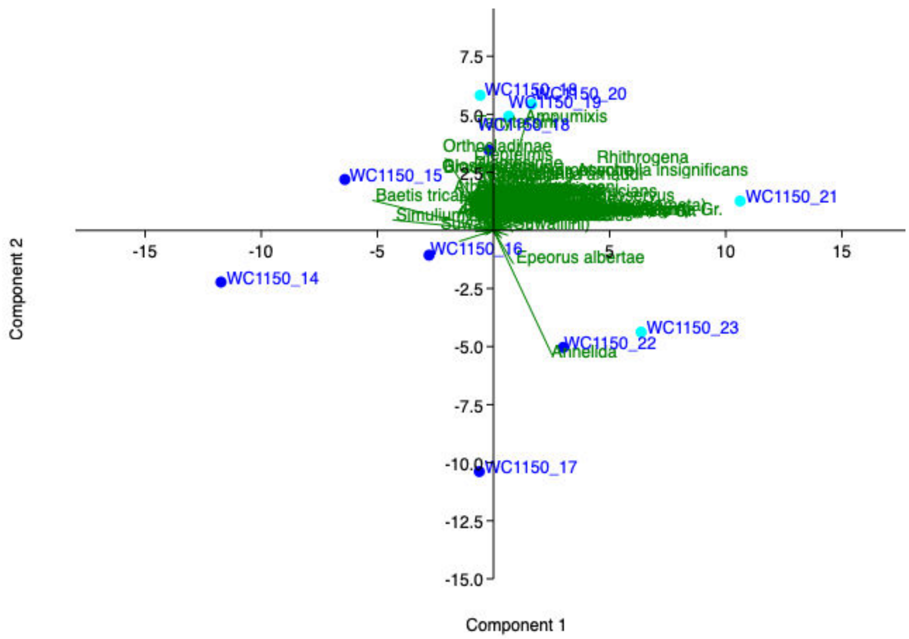
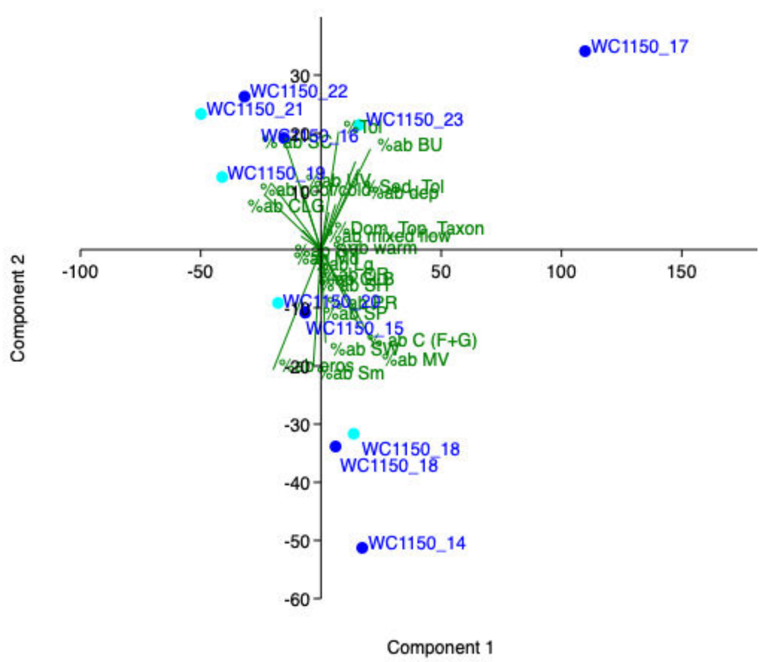


Figure 57. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1150 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 51% of total sample variation; axis 2 explains an additional 23% of variation.



WC1175, WC1200, WC1250 (Whychus Canyon reaches 3 and 2)

PM samples were taken at these sites for the first time in 2023 to obtain baseline data and to provide additional control samples for WC1100, as no restoration has been done in these reaches. All samples were taken primarily in riffles with cobble substrate (Figures 2 and 3). Macroinvertebrate community composition among all three was most similar to the unrestored WC1150 reach immediately downstream (Figure 5). All communities consisted mainly of small organisms that move as clingers and feed as collectors in faster, cooler flows. WC1175 had more multivoltine organisms, while WC1200 and WC1250 had a greater abundance of univoltine organisms. The communities were dominated at low relative abundance (Figure 58) by either *Optioservus* (tolerant riffle beetle associated with clear water and stable habitat in a variety of flows and temperatures; WC1175, WC1250) or Naidinae (tolerant sludge worm; WC1200). The WC1175 sample had more total taxa and the lowest relative abundance of the dominant taxon and of EPT taxa among all 2023 samples, while WC1250 had the highest community temperature optima of all 2023 samples.

Organismal abundance was high, with the target subsampling number of 500 attained in each sample. WC1175 had more total taxa than the other two reaches while WC1200 had more EPT taxa (Figure 59). There were more sensitive taxa compared to sediment-sensitive in each reach (Figure 60), and more tolerant than sediment-tolerant organisms (Figure 61). WC1250 had the highest community temperature optima and the lowest community sediment optima (Figure 62), but all three reaches had a greater abundance of cold-associated organisms compared to warm-associated (Figure 63). Macroinvertebrate community compositions was similar among all the sites, but greater between WC1200 and WC1250 compared to WC1175 (Figure 64).

Figure 58. Relative abundance of the numerically dominant taxon at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken. In the ORDEQ IBI, <20% abundance of the top taxon receives the highest scaled score.

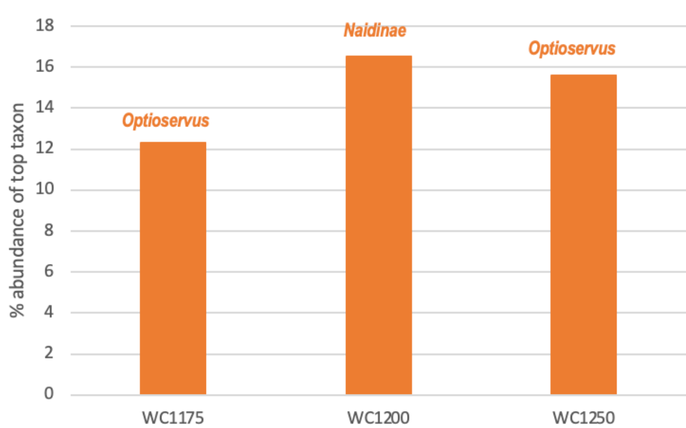


Figure 59. Sample richness and number of EPT taxa at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score

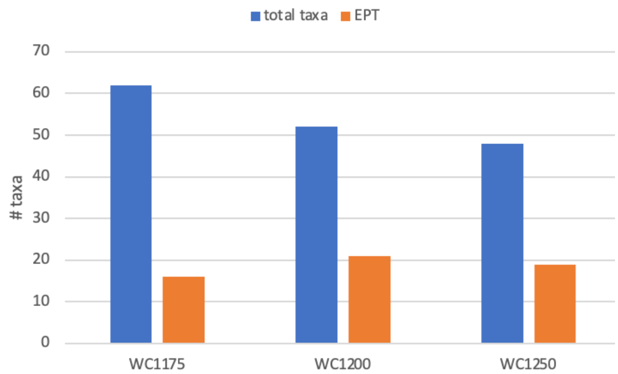


Figure 60. Numbers of sensitive and sediment-sensitive taxa at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

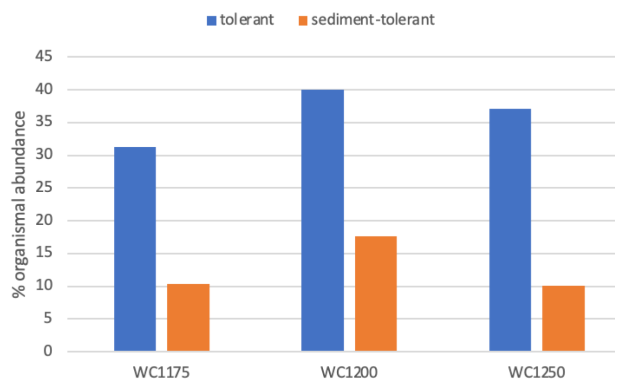


Figure 61. Relative abundance of tolerant and sediment-tolerant organisms at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken. In the ORDEQ IBI, the highest scaled scores are assigned at <15% tolerant and <10% sediment-tolerant.

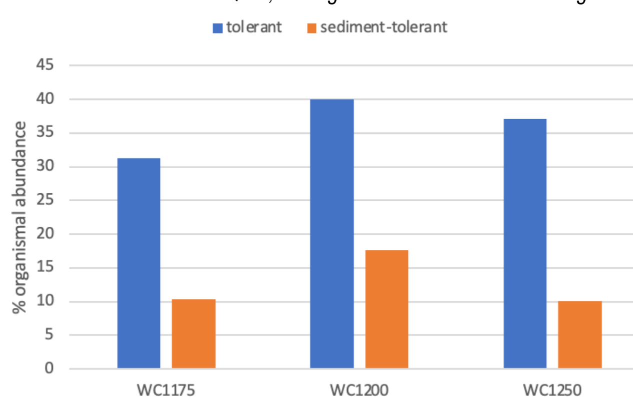


Figure 62. Temperature and fine sediment optima of the community (weighted means) at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken.

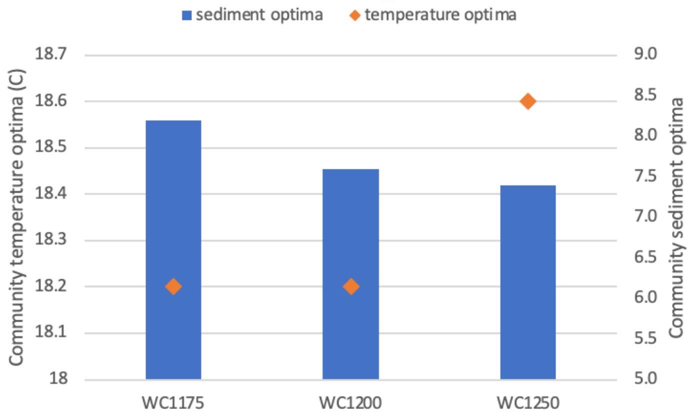


Figure 63. Temperature associations of the macroinvertebrate community at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken.

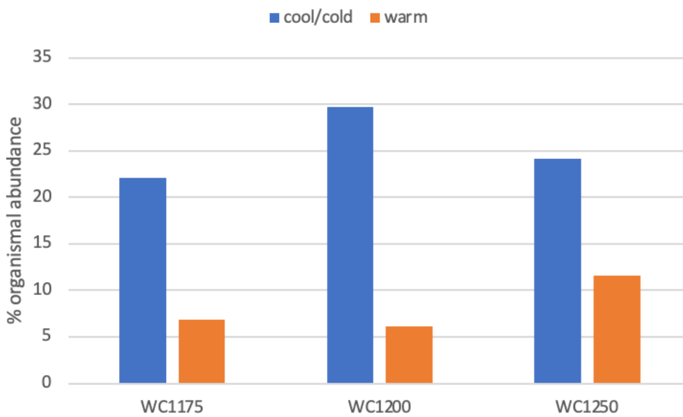
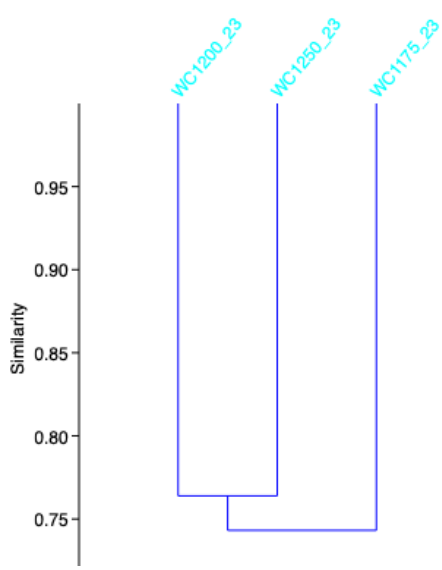


Figure 64. Cluster dendrogram of the community at WC1175, WC1200, and WC1250 in their first sampling year. PM samples were taken. The number at the end of each label indicates sampling year.



WC1950 (Camp Polk)

The reach at WC1950 is one of five restored in 2012 at Camp Polk, but the only one in this restoration project sampled in 2023. RT samples were taken in 2009-2017, 2019, and 2022-2023; PM samples were taken in 2020 and 2021. The 2023 sample was taken in primarily cobble substrate (Figure 1). The macroinvertebrate community was most similar to the adjacent WC2000 reach (Figure 5) and consisted mainly of small, univoltine organisms that move as clingers and feed as collectors in cooler, faster flows. Organismal abundance was dominated at low relative abundance by *Simulium* (black fly associated with faster flows; Figure 65). All taxa taken in this reach in 2023 were found in samples in at least one prior year.

Organismal abundance in this reach is generally high, with the target subsampling number of 500 attained in all but a single year (Figure 66). A trends analysis (Appendix B) revealed significant unidirectional increases across time in number of EPT taxa and sediment-intolerant taxa and relative abundance of climbers, and significant unidirectional decreases across time in relative abundance of multivoltine and burrower organisms. These trends suggest decreased sedimentation and increased habitat stability and quality sustaining a greater diversity of sensitive taxa.

Taxa richness increased since sampling began, especially after 2017 (Figure 67), with significantly more total taxa taken in 2019-2023 (mean 50.5, SD 8.3) than in 2009-2017 (mean 31.8, SD 1.8). The mean number of EPT taxa in recent years was also greater (20.2 vs. 17.6 in earlier years) but not significantly so. IBI scores reflected slight to no disturbance in most sampling years and changed little over time (Figure 68); O/E scores varied more, especially in early sampling years, but stabilized around the cutoff point between poor and fair condition in more recent years. A similar pattern of overall increased metric values in 2019-2023 compared to 2009-2017 was seen for the numbers of sensitive and sediment-sensitive taxa (Figure 69) and relative abundance of tolerant and sediment-tolerant organisms (Figure 70), though the differences were significant only for sensitive taxa (2.3 vs 4.3) and sediment-sensitive taxa (1.1 vs. 2.5). Community temperature and sediment optima (Figure 71) varied more across time, with no significant difference between early and recent sampling periods, although both were higher in recent years. However, cool/cold-associated organisms outnumbered warm-associated in four of the five most recent sampling years (Figure 72). There were also more cool/cold associated and fewer warm-associated in recent years, but the difference between the two periods was not significant for either metric.

Figure 65. Relative abundance of the numerically dominant taxon at WC1950. Blue = riffle-targeted, orange = multihabitat. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

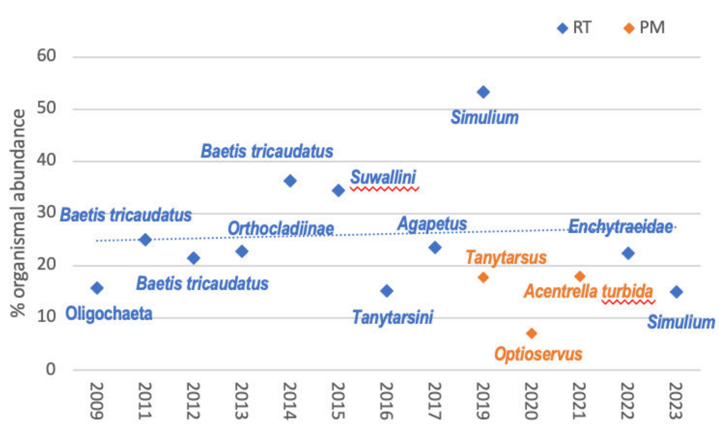


Figure 66. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1950. RT = riffle-targeted; PM = multihabitat. Target sub-sampling number is 500.

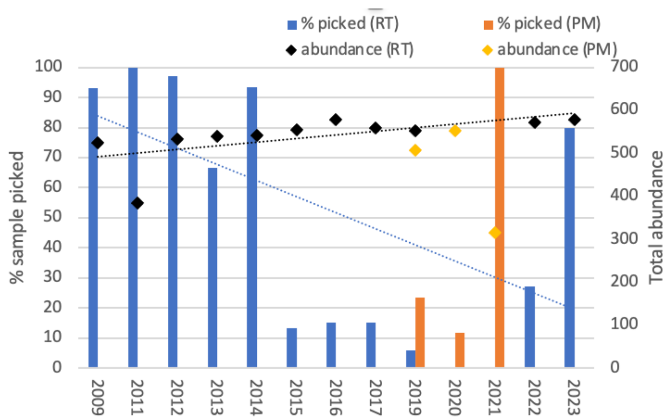


Figure 67. Sample richness and number of EPT taxa at WC1950. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

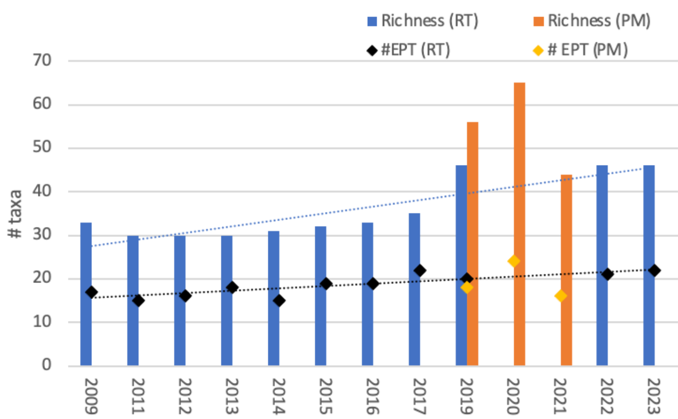


Figure 68. PREDATOR and IBI scores at WC1950 in years when riffle-targeted samples were taken. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

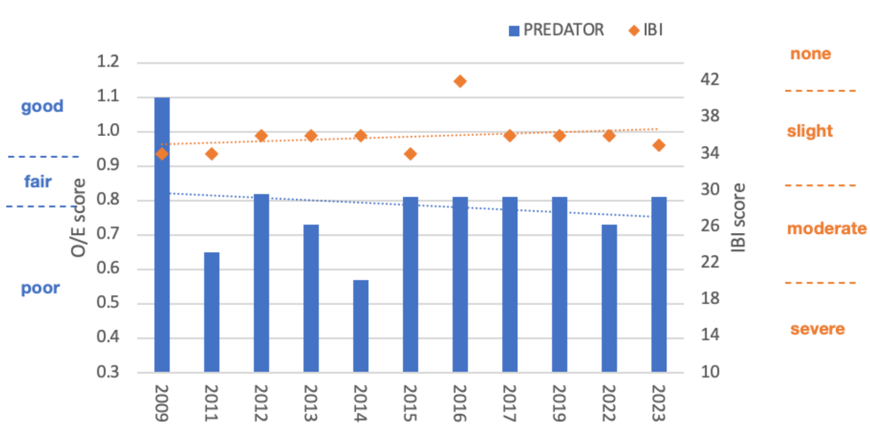


Figure 69. Numbers of sensitive and sediment-sensitive taxa at WC1950. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

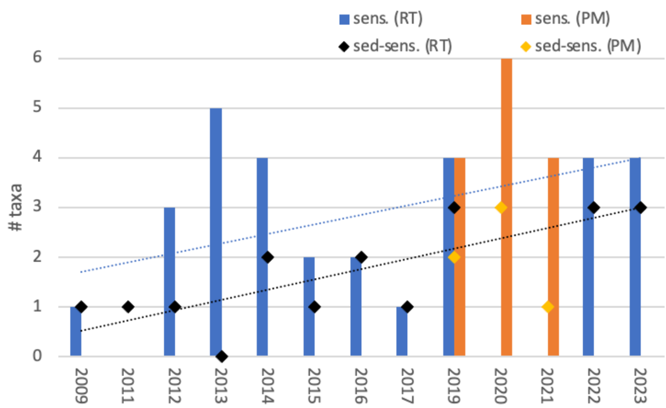


Figure 70. Relative abundance of tolerant and sediment-tolerant organisms at WC1950 RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

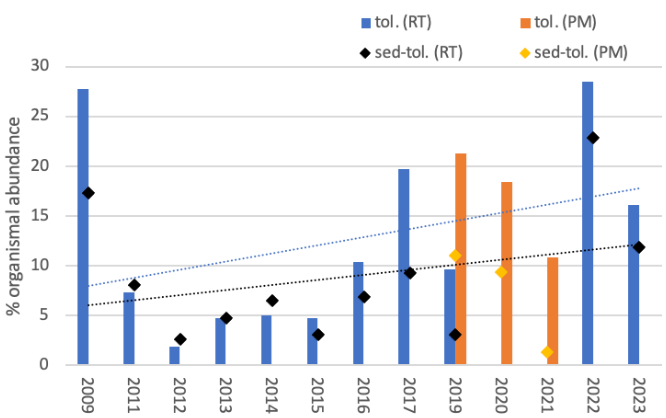


Figure 71. Temperature and fine sediment optima of the community (weighted means) at WC1950. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown.

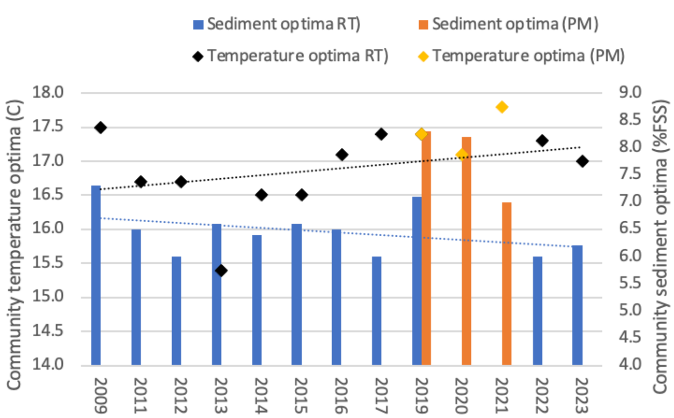
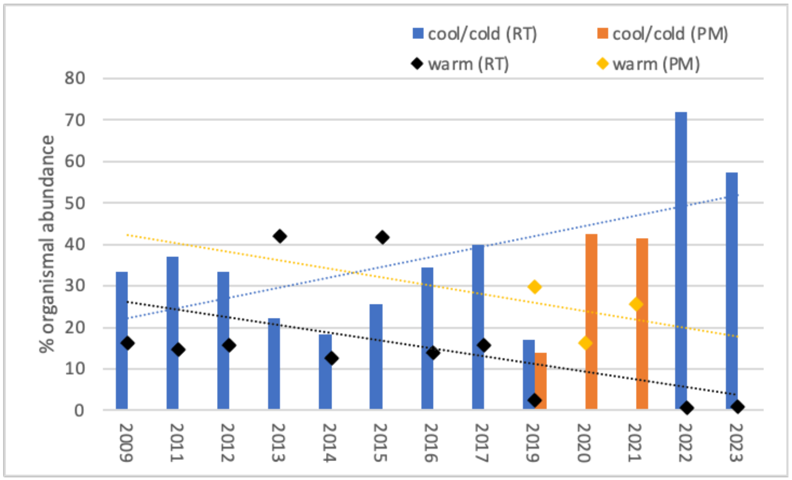


Figure 72. Temperature associations of the macroinvertebrate community at WC1950. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown.



Macroinvertebrate community composition was significantly different pre- vs. post-restoration. The community was more similar among PM samples taken in recent years (2019-2021), which clustered separately from all other years and samples (Figure 73). The 2022 and 2023 RT sample communities were most similar to each other but were more closely related to RT samples taken in the years following restoration than they were to PM samples in recent years. Differences in community composition (Figure 74) were driven mainly by Tanytarsini (non-biting midge tribe that builds tubes in soft sediments; most abundant 2015-2020), *Baetis tricaudatus* (small minnow mayfly associated with faster flows, clearer water, and recent disturbance; most abundant in the years immediately following restoration), *Simulium* (black fly associated with flowing water and recent disturbance; more abundant in post-restoration RT samples), and Orthocladiinae (widespread non-biting midge subfamily; most abundant immediately prior to and for a few years after restoration).

Differences in traits calculated as relative abundances (Figure 75) reflected an increase in univoltine and cool/cold-associated organisms in 2022 and 2023, as well as more organisms associated with faster flows (except in PM samples) and a more balanced community (lower top taxon abundance) in most years following 2017 (Figure 76) These differences suggest faster, cooler flows and greater habitat stability during the 2019-2023 span.

Figure 73. Cluster dendrogram of the WC1950 macroinvertebrate community. Blue = riffle-targeted, aqua = multihabitat. The number at the end of each label indicates sampling year.

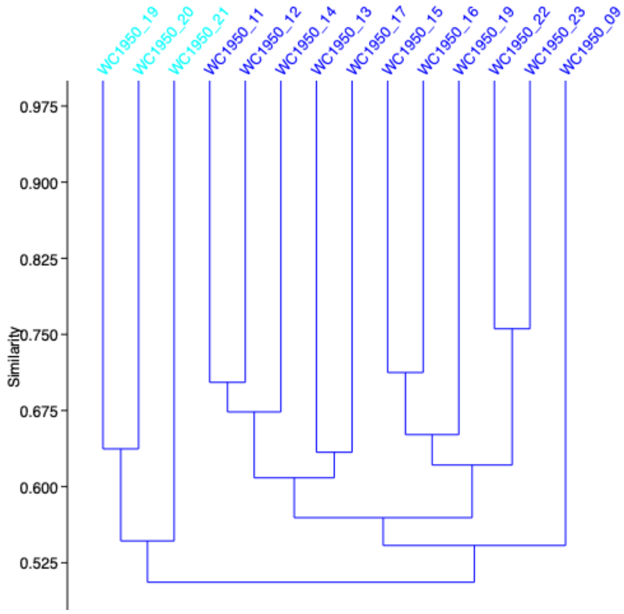


Figure 74. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1950 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 27% of total sample variation; axis 2 explains an additional 16% of variation.

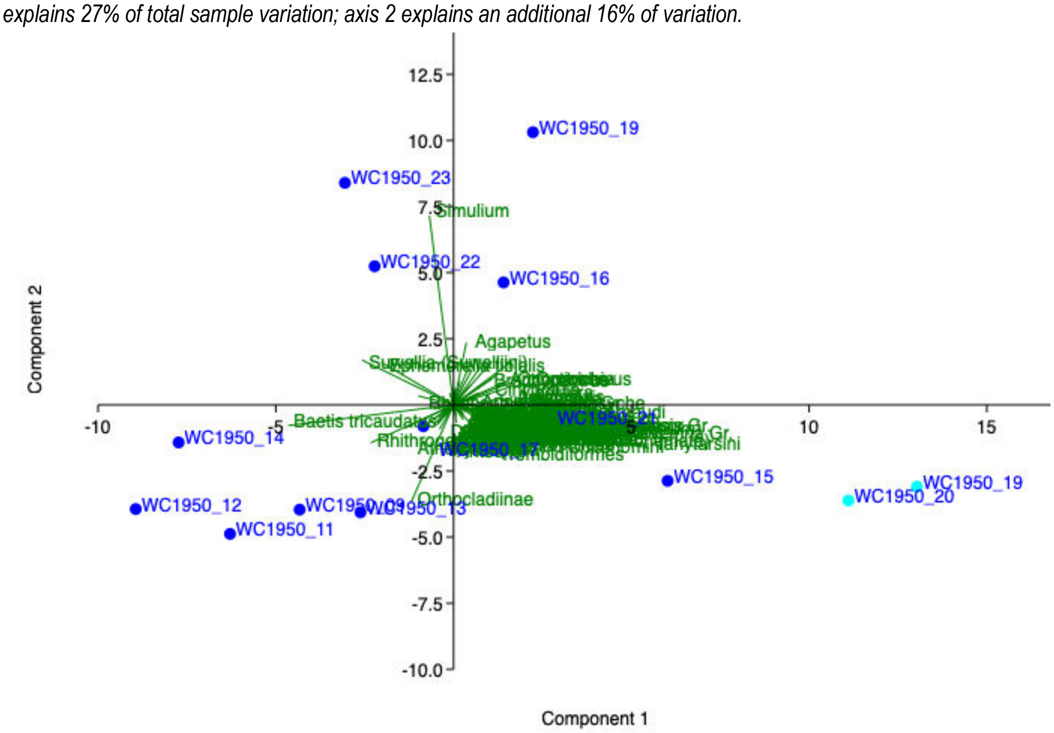
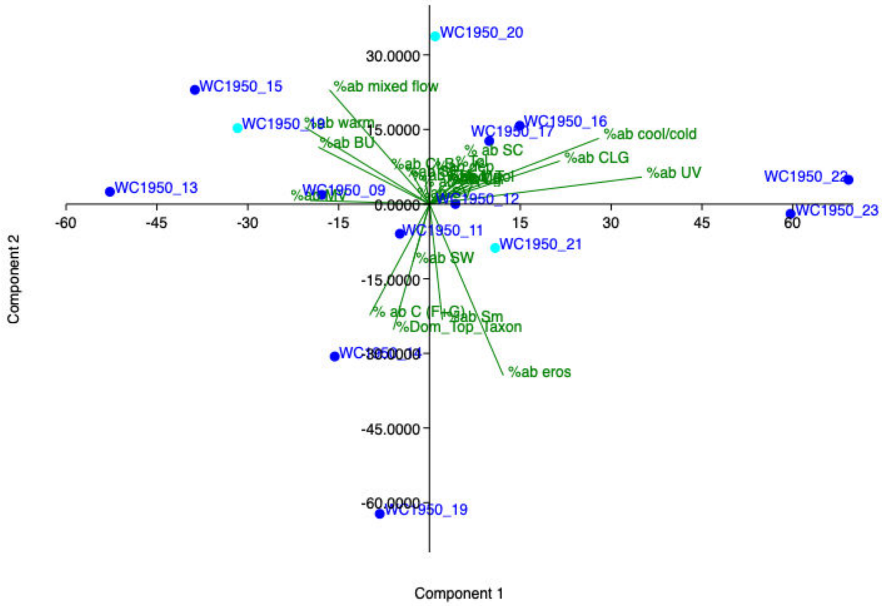


Figure 75. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC1950 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 40% of total sample variation; axis 2 explains an additional 20% of variation.



WC2000 (Willow Springs)

WC2000 is one of two reaches at Willow Springs that was restored in 2022. A PM and RT sample were taken in 2020-2021 and 2023. Both 2023 samples were taken in primarily cobble substrate (Figure 1 and 2), with most of the PM sample net sets taken in glides (Figure 3). Both sample communities were more closely related to those in nearby reaches, with the WC1950 through WC2600 sample communities clustering separately from the remaining 2023 samples (Figure 5). The 2023 PM and RT communities consisted mainly of small, univoltine organisms that move as clingers and feed as collectors in cooler, faster flows. Samples were dominated at low relative abundance by tolerant Naidinae sludge worms (PM sample) or the small minnow mayfly *Baetis tricaudatus* (RT sample; Figure 76).

The RT sample had the greatest relative abundance among all 2023 samples of organisms associated with faster flows, the fewest that tolerate a variety of flows, and was the only 2023 sample to lack nonbiting midges in the tolerant Chironomini tribe. Eleven taxa were taken in this reach for the first time in 2023, all at abundances ranging from 1-9: *Zaitzevia* (riffle beetle associated with faster flows and stable habitat), *Atherix* (tolerant water snipe fly common in montane streams), Ceratopogonidae (biting fly family found in a variety of flowing and still habitats), *Dicranota* (sediment-tolerant crane fly found in detritus, sediment, and leaf packs in cooler water and a range of flows), *Hesperoconopa* (sensitive crane fly found in bottom sediments of fast clear cold streams), *Hexatoma* (sediment-tolerant crane fly found in detritus and moss in cold water and a range of flows), Muscidae (true fly family with tolerant aquatic and semi-aquatic members found in a range of flows and temperatures), *Ameletus* (combmouth mayfly found in cool water and a range of flows), *Megarcys* (sensitive spring stonefly found in cold, well-oxygenated, flowing montane streams), *Kogotus/Rickera* (sediment-sensitive spring stonefly found in rocks and leaf packs in cold riffles), and *Rhyacophila betteni* (green rockworm caddisfly found in cold, flowing, stable habitats).

Organismal abundance in this reach is high, with the target subsampling number of 500 attained in all years and sample types (Figure 77). A trends analysis (Appendix B) revealed a significant monotonic increase across time in relative abundance of small-bodied organisms and a significant decrease in crawlers, suggesting potentially increased sedimentation and habitat instability immediately following restoration. Total and EPT taxa numbers were lower in the RT sample after restoration, while the values for these metrics in the 2023 sample were the same or greater than pre-restoration (Figure 78). The PM sample in each year had more total taxa than the RT sample, while EPT taxa richness was slightly greater in RT samples. PREDATOR and IBI scores were lower in the 2023 RT sample compared to the prior two years (Figure 79) and may have been impacted by restoration activities. The number of sensitive taxa was higher in the PM sample in each year; there were more sensitive taxa in the 2023 PM sample than in any prior year, but fewer in the RT sample. However, the number of sediment-sensitive taxa was similar among all samples and years (Figure 80). Relative abundance of tolerant organisms exceeded that of sediment-tolerant in each year, and while both increased over time in both PM and RT samples, the overall values are fairly low (Figure 81). Community sediment optima are consistently lower in RT samples compared to PM in each year but decreased overall since sampling began (Figure 82); community temperature optima are largely unchanged and similar in both sample types. There are consistently more cool/cold-associated organisms and fewer warm-associated organisms in in RT samples, but cool/cold-associated are more abundant than warm in every year and sample type (Figure 83).

Figure 76. Relative abundance of the numerically dominant taxon at WC2000. Restoration occurred in 2022. Blue = riffle-targeted, orange = multihabitat. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

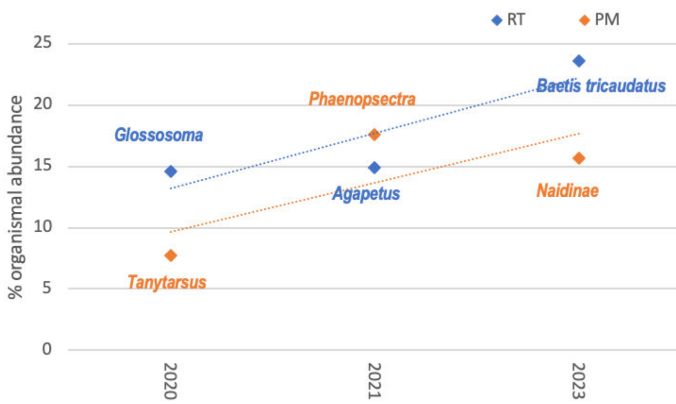


Figure 77. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC2000. RT = riffle-targeted; PM = multihabitat. Target sub-sampling number is 500.

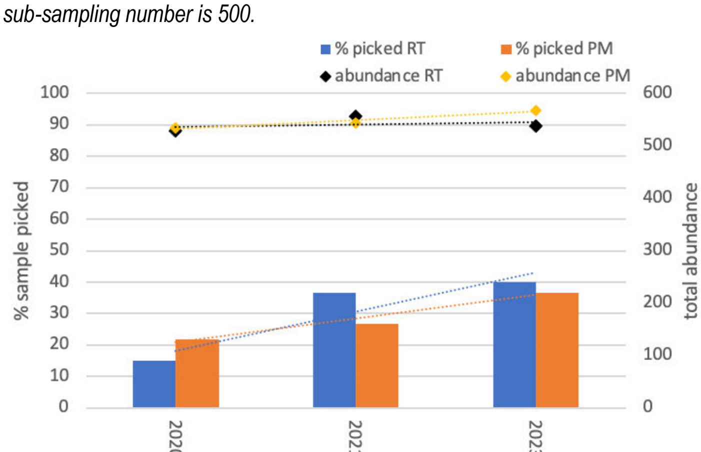


Figure 78. Sample richness and number of EPT taxa at WC2000. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

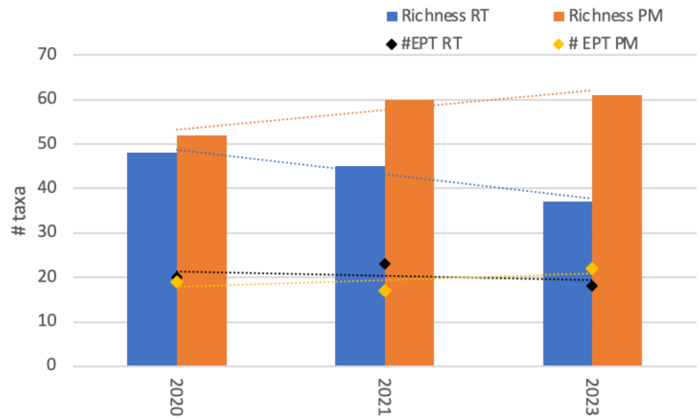


Figure 79. PREDATOR O/E and ORDEQ IBI scores at WC2000. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

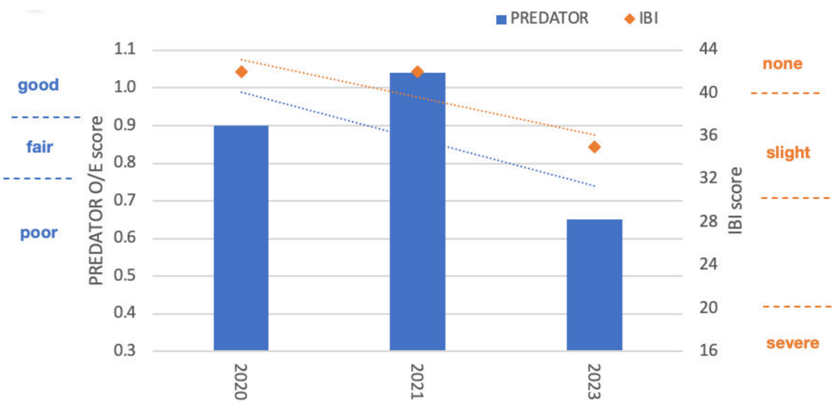


Figure 80. Numbers of sensitive and sediment-sensitive taxa at WC2000. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

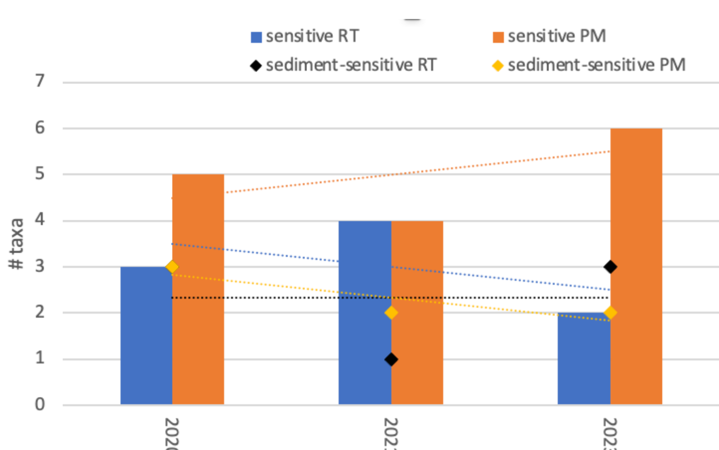


Figure 81. Relative abundance of tolerant and sediment-tolerant organisms at WC2000. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

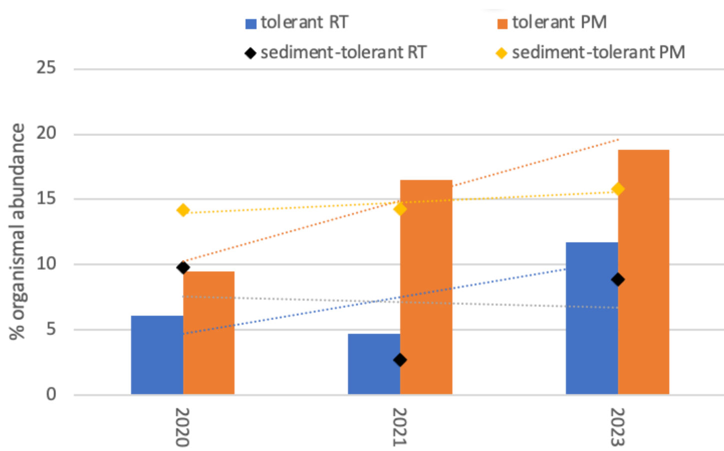


Figure 82. Temperature and fine sediment optima of the community (weighted means) at WC2000. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. Note that the 2023 RT and PM sample had the same community temperature optima.

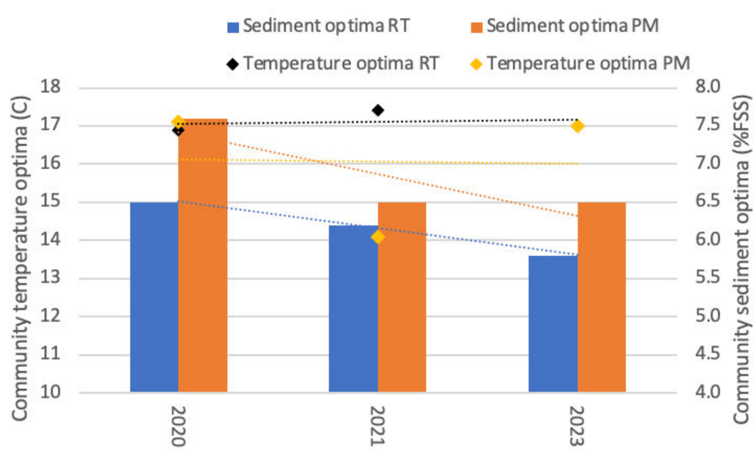
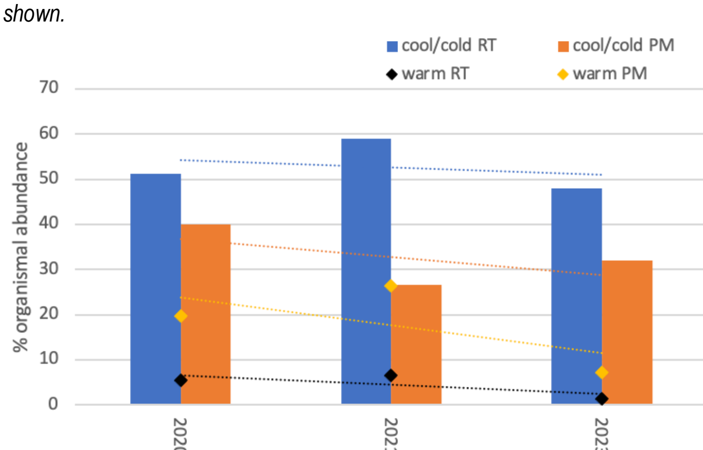


Figure 83. Temperature associations of the macroinvertebrate community at WC2000. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown.



Macroinvertebrate community composition was fairly similar among all years, but the PM and RT sample taken after restoration differed more from all prior years (Figure 84). Differences in community composition (Figure 85) reflected both differences in sampling type and in restoration impacts, with increased abundance of *Baetis tricaudatus* (small minnow mayfly associated with faster flows, clearer water, and recent disturbance) after restoration and more tolerant Chironomini and Annelida in PM samples. Differences in traits calculated as relative abundances made similar distinctions, with greater abundance of organisms associated with faster flows in all RT samples, and more univoltine and small organisms in both 2023 samples (Figure 86).

Figure 84. Cluster dendrogram of the WC2000 community. Blue = riffle-targeted, aqua = multihabitat. The number at the end of each label indicates sampling year.

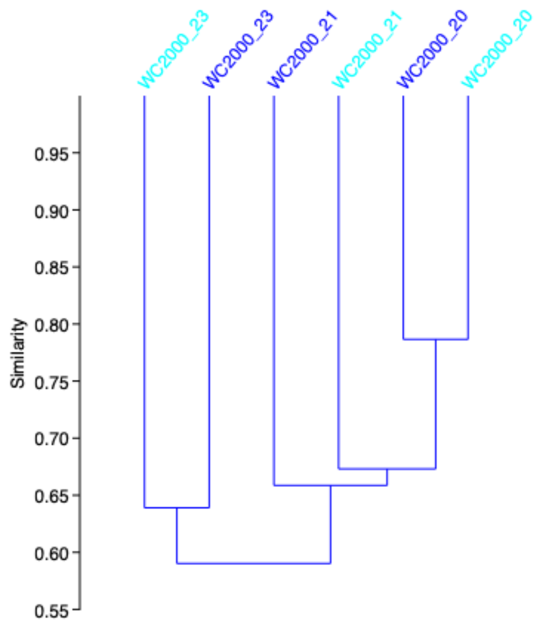


Figure 85. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC2000 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 43% of total sample variation; axis 2 explains an additional 30% of variation.

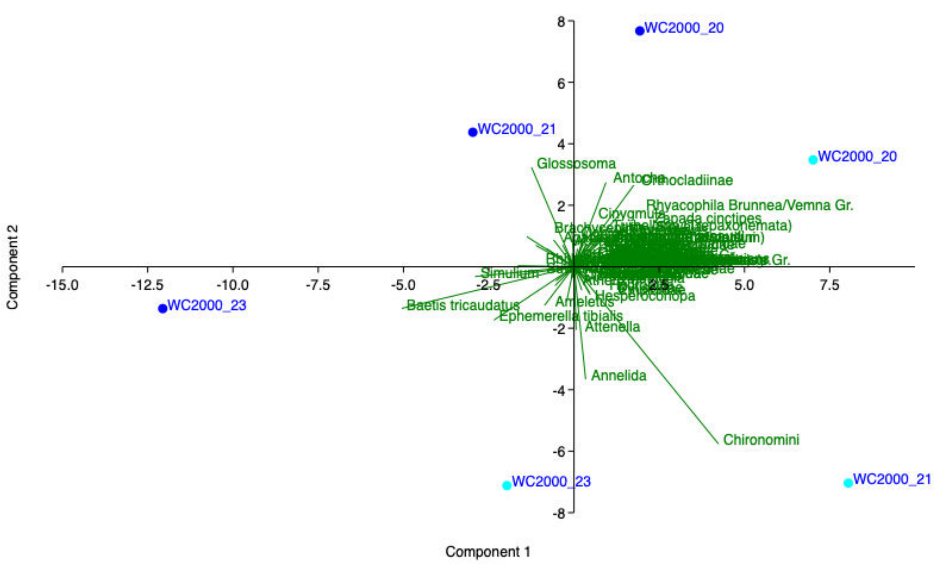
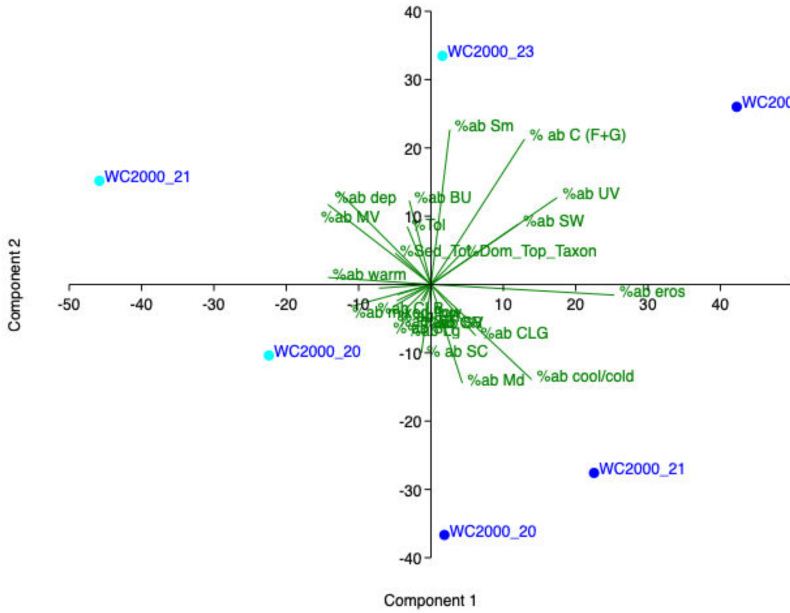


Figure 86. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC2000 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 44% of total sample variation; axis 2 explains an additional 38% of variation.



WC2050 (Willow Springs)

WC2050 is one of two reaches at Willow Springs that was restored in 2022. A PM and site duplicate sample were taken in 2020-2021 and in 2023. The 2023 sample was taken in primarily in cobble substrate in riffles and runs (Figure 2 and 3). The duplicate samples taken in this reach were most similar to each other (Figure 5), confirming the robustness and reliability of the sampling protocol. Community composition in this reach was most similar to that at the adjacent WC2000. The 2023 community consisted mainly of small multivoltine organisms that move as clingers and feed as collectors in cool fast flows. The community was dominated at low relative abundance by *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water, whose abundance can increase following recent disturbance (Figure 87).

The 2023 sample had fewer scrapers and more collectors than any other 2023 sample, as well as the most multivoltine and swimmer organisms, suggesting potentially more disturbed conditions and less exposed or biofilm-coated mineral material. Seven taxa were taken in this reach for the first time in 2023: Muscidae (true fly family with tolerant aquatic and semi-aquatic members in a range of flows and temperatures), *Drunella coloradensis* (sensitive, sediment-sensitive spiny crawler mayfly), *Cinygma* (sensitive flatheaded mayfly that prefers woody substrates), *Epeorus longimanus* (flatheaded mayfly found on large ricks in cool streams), *Paraleptophlebia* (prong-gilled mayfly associated with sediments and detritus in fast cool flows), *Kogotus/Rickera* (cold-adapted, sediment-sensitive stonefly), and *Rhyacophila angelita* (sediment-sensitive free-living caddisfly).

Organismal abundance in this reach is high, with the target subsampling number of 500 attained in all years (Figure 88). Trends analysis revealed no significant monotonic changes across time in any community metrics (Appendix B). Total and EPT taxa numbers were high overall and increased slightly after restoration (Figure 89). Numbers of sensitive and sediment-sensitive taxa were relatively high in this reach prior to and after restoration (Figure 90), but relative abundance of tolerant and sediment-tolerant organisms was lower in 2023 compared to prior years (Figure 91). There are no apparent trends in community sediment or temperature optima (Figure 92). Cool/cold-associated organisms are generally well represented in this reach, and abundance of warm-associated organisms was lower in 2023 than in any prior year (Figure 93).

The 2023 community was more similar to that in 2021 than in 2020 (Figure 94), and differences in community composition (Figure 95) were driven mainly by *Baetis tricaudatus*, which dominated the 2023 community, as well as *Ochrotrichia* (microcaddisfly found in a variety of flows and temperatures; absent in 2021), *Zapada cinctipes* (forest stonefly associated with cool waters whose presence indicates ongoing contributions from the riparian zone to the instream food base; absent in 2021), and *Micrasema* (cool-adapted humpless casemaker caddisfly; absent in 2023). Differences in traits calculated as relative abundances (Figure 96) were driven mainly by greater abundance of multivoltine and univoltine organisms and fewer warm-associated organisms after restoration.

Figure 87. Relative abundance of the numerically dominant taxon at WC2050. Multihabitat samples were taken each year. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

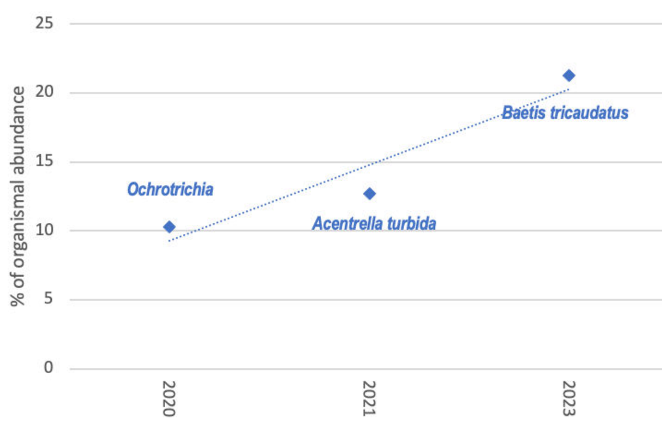


Figure 88. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC2050. Multihabitat samples were taken each year. Target sub-sampling number is 500.

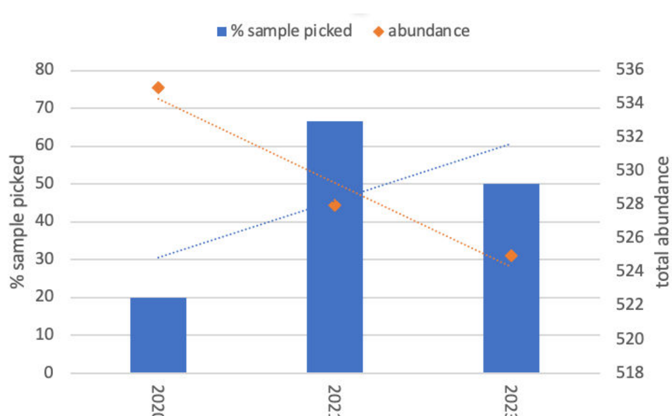


Figure 89. Sample richness and number of EPT taxa at WC2050. Multihabitat samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

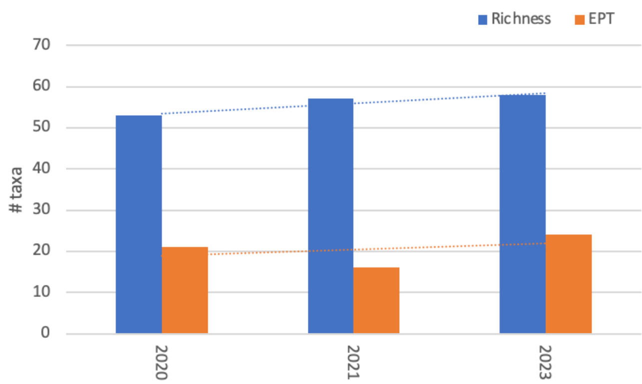


Figure 90. Numbers of sensitive and sediment-sensitive taxa at WC2050. Multihabitat samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

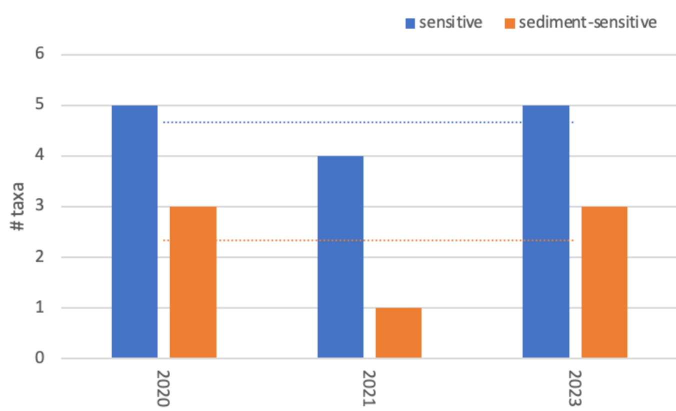


Figure 91. Relative abundance of tolerant and sediment-tolerant organisms at WC2050. Multihabitat samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

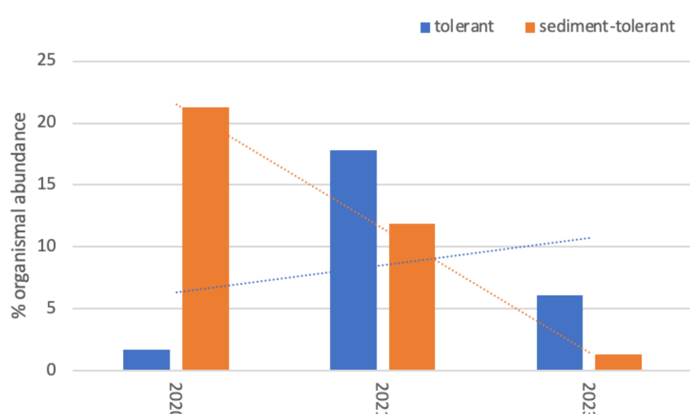


Figure 92. Temperature and fine sediment optima of the community (weighted means) at WC2050. Multihabitat samples were taken each year. Linear trendlines are shown.

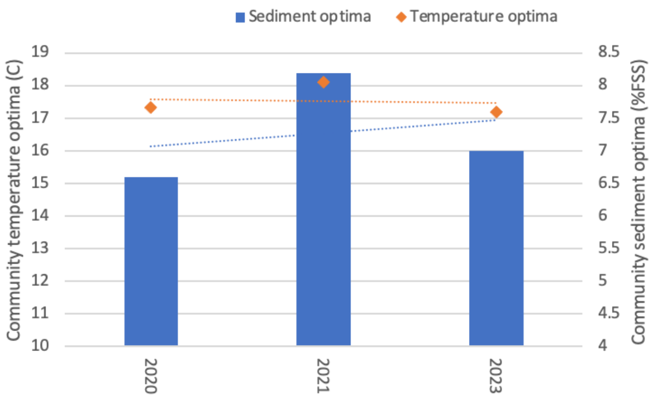


Figure 93. Temperature associations of the macroinvertebrate community at WC2050. Multihabitat samples were taken each year. Linear trendlines are shown.

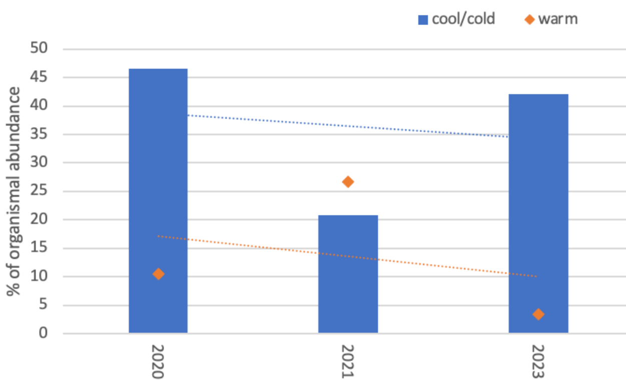


Figure 94. Cluster dendrogram of the WC2050 community. Multihabitat samples were taken each year. The number at the end of each label indicates sampling year.

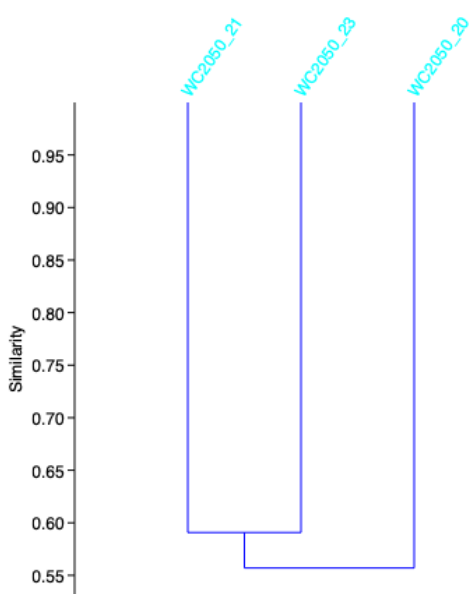


Figure 95. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC2050 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Multihabitat samples were taken each year. Axis 1 explains 60% of total sample variation; axis 2 explains an additional 40% of variation.

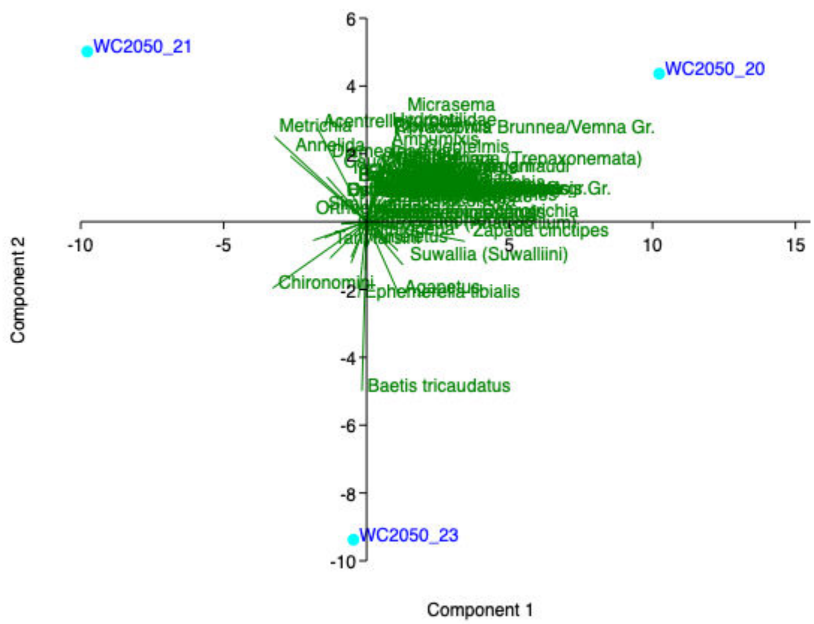
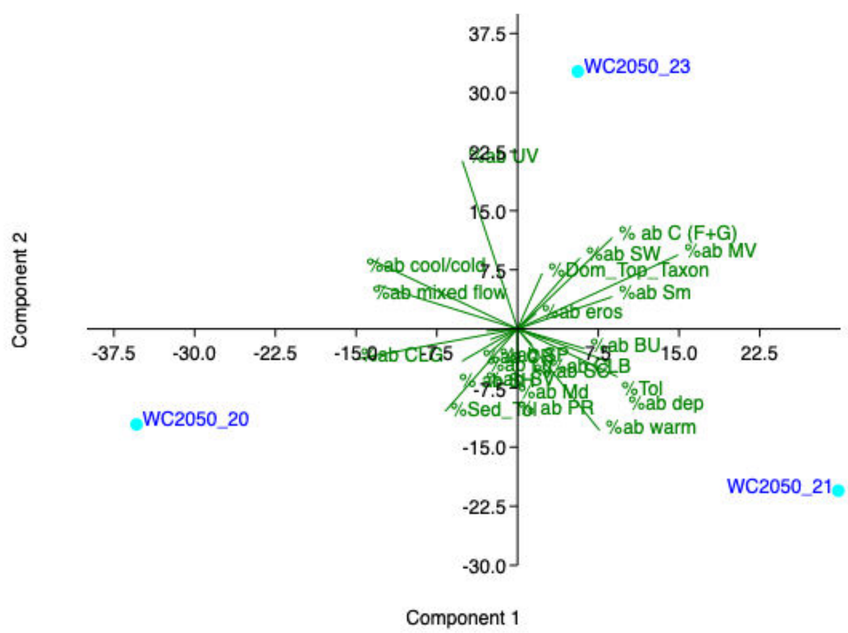


Figure 96. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC2050 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Multihabitat samples were taken each year. Axis 1 explains 57% of total sample variation; axis 2 explains an additional 43% of variation.



WC2425 (Sisters City Park)

WC2425 was sampled in each project year from 2005-2017 but was not sampled again until 2023, following an instream project done in 2022 to create fewer and more hardened access points to the creek, as this reach runs through a well-used campground with many humans and dogs traversing the banks and channel. RT samples were taken in all years, with the 2023 sample taken in primarily cobble substrate (Figure 1). The macroinvertebrate community was an outlier to all other 2023 samples (Figure 5) and consisted mainly of small multivoltine organisms that move as clingers and swimmers and feed as collectors in cool fast flows. The community was dominated at low relative abundance by *Suwaliini*, a green stonefly associated with cooler waters in a variety of flow types (Figure 97).

The 2023 sample had the fewest total and EPT taxa of all 2023 samples, but the proportion of total taxa comprised by EPT was the highest. The sample also had the lowest community temperature and sediment optima and MHBI of all 2023 samples; the fewest semivoltine, warm-associated, and sprawler organisms; and the greatest relative abundance of univoltine, predator, and cool/cold-associated organisms. While the low values of richness metrics suggest impaired habitat conditions, ranges of other metrics indicate cooler, faster flows with less fine suspended sediment and a sufficient prey base. A single taxon was taken for the first time here in 2023: *Agraylea* (one individual), a microcaddisfly associated with algae and vascular hydrophytes in a range of temperatures and flow types.

The low number of taxa in 2023 could be explained by low organismal abundance, as the entire sample had only 170 organisms. This may have been due to the impacts of recent restoration, although there have been other years in which fewer than 200 organisms total were taken (2011, 2015; Figure 98). Trends analysis revealed significant monotonic changes across time in seven community metrics (Appendix B), with significantly decreased relative abundance of the dominant taxon and significant increases in abundance of predators, clingers, swimmers, large-bodied organisms, and organisms associated with cooler and faster flows. These trends all suggest increased habitat stability and quality over time.

Total and EPT taxa richness fluctuated across time (Figure 99), and both were lower in 2023 compared to the most recent sampling years (2016-2017). PREDATOR and IBI scores also fluctuated, ranging from poor to fair condition (O/E) and moderate to no disturbance (IBI; Figure 100). Scores for both models were lower in 2023 compared to the most recent prior sampling years, although the IBI score was still in the range of slight disturbance. There are consistently more sensitive than sediment-sensitive taxa in each year, and values for both metrics in 2023 were largely unchanged from prior years (Figure 101). Relative abundance of tolerant and sediment-tolerant organisms fluctuated more, but the values of both metrics overall are low (Figure 102). Community sediment and temperature optima varied less (Figure 103) but community temperature optima was lower in 2023 compared to prior years, and the community sediment optima in 2023 was the lowest of any sampling year. Cool/cold-associated organisms are consistently abundant in this reach and there are few to no warm-adapted organisms in each year (Figure 104).

The 2023 community was most similar to the 2011 sample (Figure 105), and between-year differences in community composition were driven mainly by greater abundance of *Orthocladiinae* (widespread non-biting midge) in 2005 and 2009, fewer *Suwaliini* in 2005-2011, and more *Simulium* (black fly associated with faster flows and recent disturbance) in most years prior to 2023 (Figure 106). Differences in traits calculated as relative abundances (Figure 107) were driven mainly by greater abundance of multivoltine organisms in the earliest sampling years, and fewer organisms associated with faster flows and more associated with a range of flows in 2005 (early in restoration of perennial flow to the creek).

Figure 97. Relative abundance of the numerically dominant taxon at WC2425. Riffle-targeted samples were taken each year. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

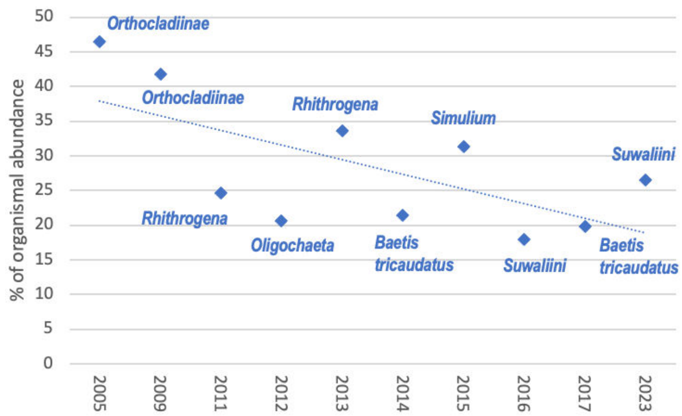


Figure 98. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC2425. Riffle-targeted samples were taken each year. Target sub-sampling number is 500.

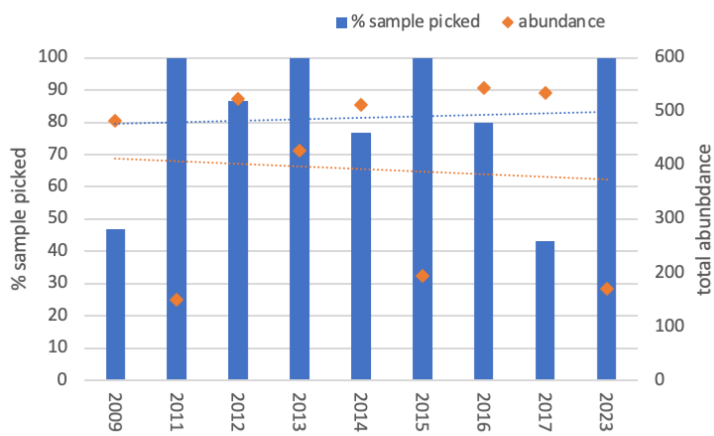


Figure 99. Sample richness and number of EPT taxa at WC2425. Riffle-targeted samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

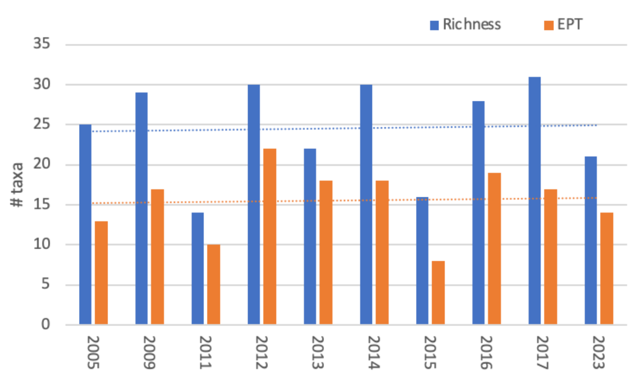


Figure 100. PREDATOR O/E and ORDEQ IBI scores at WC2425. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

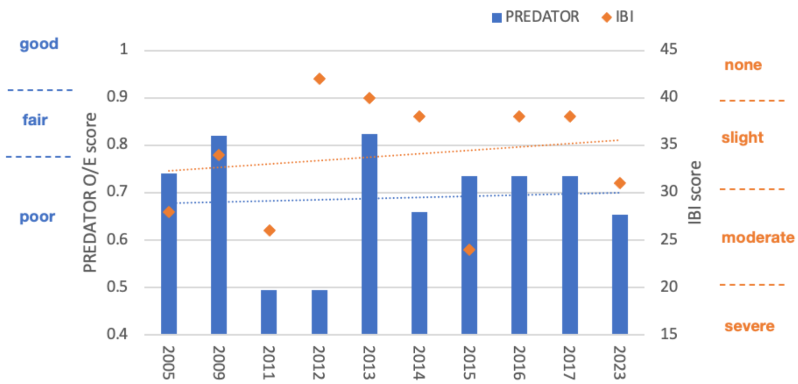


Figure 101. Numbers of sensitive and sediment-sensitive taxa at WC2425. Riffle-targeted samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.

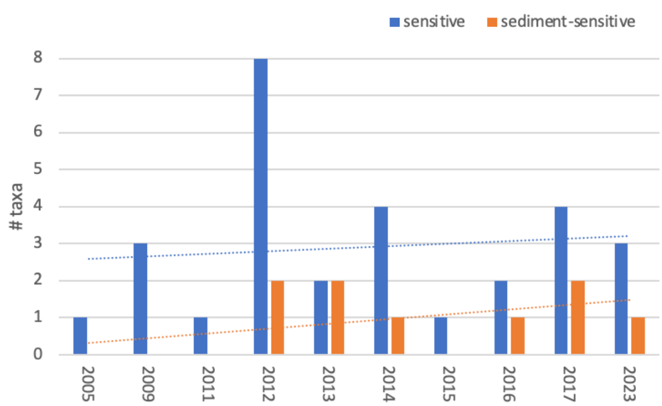


Figure 102. Relative abundance of tolerant and sediment-tolerant organisms at WC2425. Riffle-targeted samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

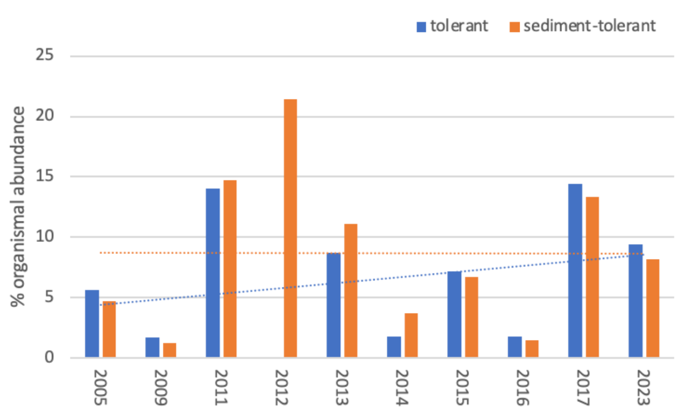


Figure 103. Temperature and fine sediment optima of the community (weighted means) at WC2425. Riffle-targeted samples were taken each year. Linear trendlines are shown.

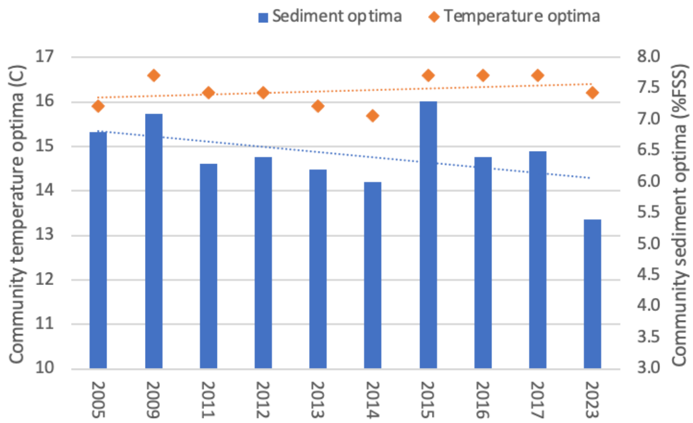


Figure 104. Temperature associations of the macroinvertebrate community at WC2425. Riffle-targeted samples were taken each year. Linear trendlines are shown.

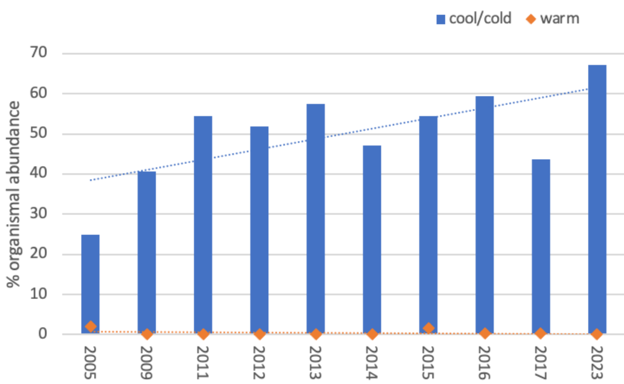


Figure 105. Cluster dendrogram of the WC2450 community in all sampling years. Riffle-targeted samples were taken each year. The number at the end of each label indicates sampling year.

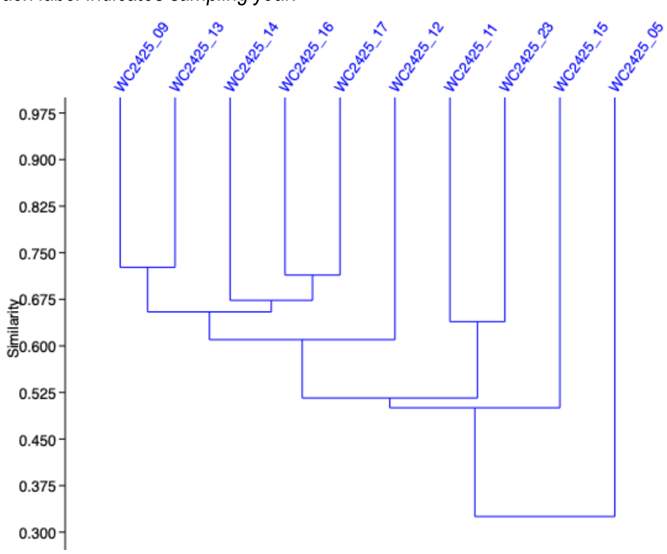


Figure 106. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC2425 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Riffle-targeted samples were taken each year. Axis 1 explains 41% of total sample variation; axis 2 explains an additional 23% of variation.

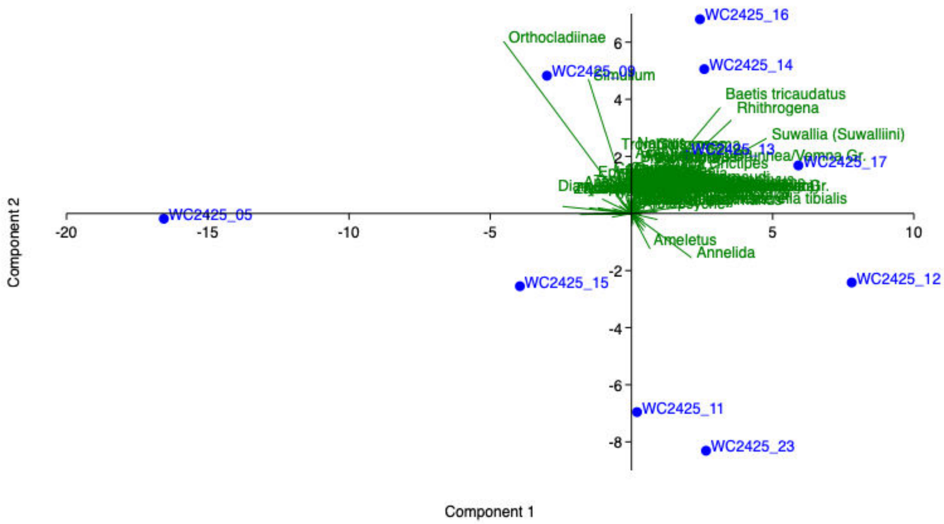
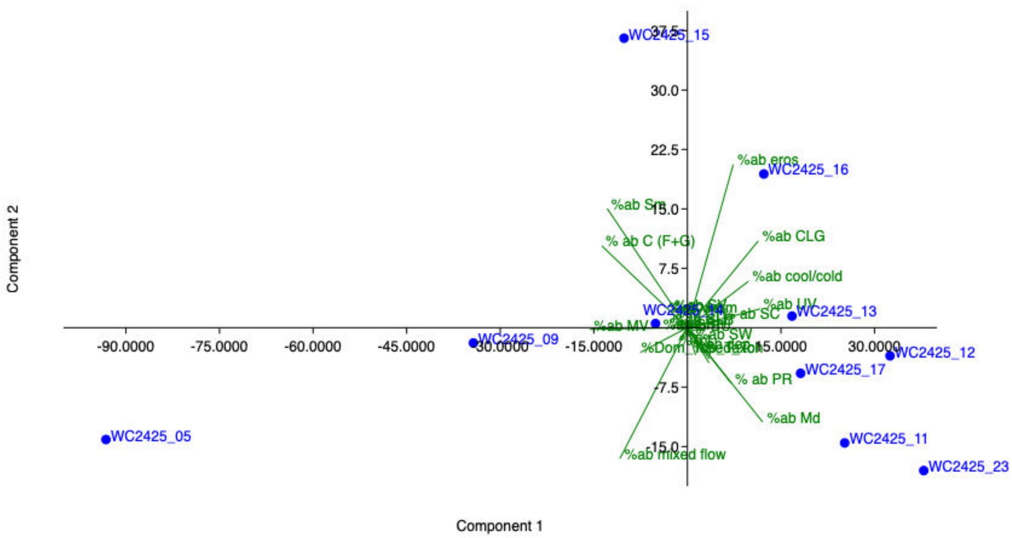


Figure 107. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC2425 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Riffle-targeted samples were taken each year. Axis 1 explains 67% of total sample variation; axis 2 explains an additional 12% of variation.



<u>WC2600 (Whychus floodplain)</u>

The WC2600 reach was sampled in 2005-2023, with RT samples taken through 2020 and PM samples taken 2018-2023. In 2015, the stream was directed into a network of new meandering channels, and additional PM samples were taken in new side channels in 2018. Sampling focused on the primary channel but as stream structure developed after restoration, that reach included elements of side channel habitat. The 2023 sample was taken in primarily cobble substrate in riffles and runs (Figure 2 and 3). The 2023 macroinvertebrate community was most similar to that at other upstream reaches (WC1950-WC2050; Figure 5) and consisted mainly of small univoltine organisms that feed as collectors and move as clingers in cooler, faster flows. The community was dominated at low relative abundance by *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water (Figure 108).

The WC2600 sample had more sensitive taxa than any other 2023 sample. Only a single taxon was taken for the first time in this reach in 2023: *Paraleptophlebia*, a prong-gilled mayfly associated with sediments and detritus in fast cool flows (one individual). Organismal abundance in samples from this reach fluctuated over time, but the target subsampling number of 500 was attained in RT and PM samples every year since 2018 (Figure 109). Trends analysis revealed significant monotonic changes across time in 16 metrics (Appendix B), including significant increases in numbers of total, mayfly, stonefly, and EPT taxa (Figure 110); sensitive and sediment-sensitive taxa (Figure 111); community sediment optima; and relative abundance of large-bodied, semivoltine, cool/cold-associated, predator, shredder, climber, and sprawler organisms. Significant decreases across time were seen in relative abundance of collectors and warm-associated organisms. Except for community sediment optima, these trends suggest cooler, less sedimented, and more stable habitat conditions over time, with a sufficient prey base and potentially more plant material.

Relative abundance of tolerant and sediment-tolerant organisms fluctuated over time but has consistently been low (<20%; Figure 112). Community temperature optima fluctuated greatly in early sampling years but stabilized more since 2018, while community sediment optima increased (Figure 113). Cool/cold-associated organisms were more abundant than warm-associated in every year since 2015 (Figure 114).

Macroinvertebrate community composition was significantly different pre- and post-restoration. The 2023 macroinvertebrate community was most similar to PM samples taken in 2018-2019 and 2022 (Figure 115), and between-year differences in community composition (Figure 116) were driven mainly by Orthocladiinae (widespread non-biting midge subfamily; less abundant in 2015-2016), Tanytarsini (non-biting midge tribe that builds tubes in soft sediments; abundant only since 2020), and *Simulium* (black fly associated with flowing water; more abundant since 2013). Differences in traits calculated as relative abundances (Figure 117) were driven mainly by greater abundance of burrowers and warm-associated organisms in early sampling years, and greater abundance of the dominant taxon and collectors in 2005 and 2016.

Figure 108. Relative abundance of the numerically dominant taxon at WC2600. Blue = riffle-targeted, orange = multihabitat. In the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

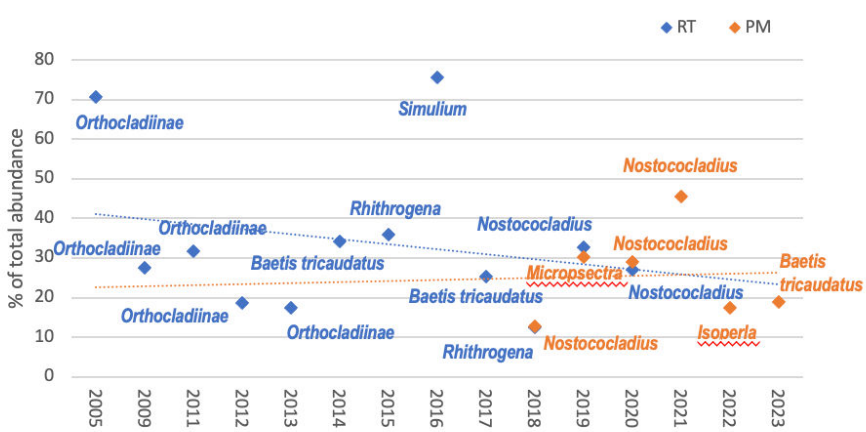


Figure 109. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC2600. RT = riffle-targeted; PM = multihabitat. Target sub-sampling number is 500.

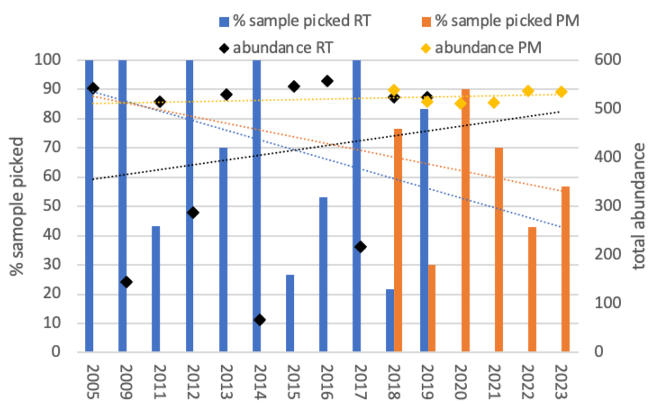


Figure 110. Sample richness and number of EPT taxa at WC2600. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

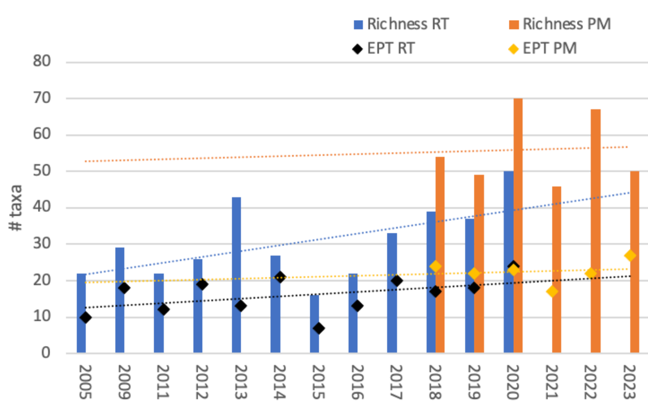


Figure 111. Numbers of sensitive and sediment-sensitive taxa at WC2600. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.

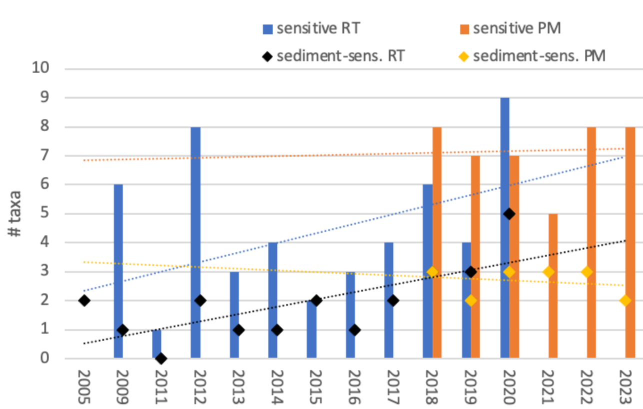


Figure 112. Relative abundance of tolerant and sediment-tolerant organisms at WC2600 RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. In the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

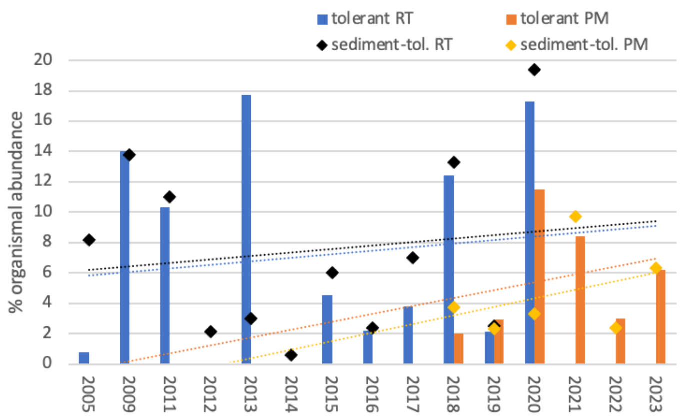


Figure 113. Temperature and fine sediment optima of the community (weighted means) at WC2600. RT = riffle-targeted; PM = multihabitat. Linear trendlines are shown. Note that the 2018 and 2019 RT /PM samples had the same community temperature optima.

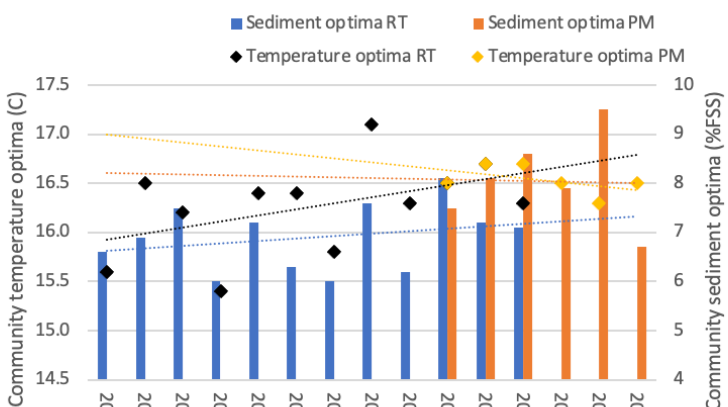


Figure 114. Temperature associations of the macroinvertebrate community at WC2600. RT = riffle-targeted; = multihabitat. Linear trendlines are shown.

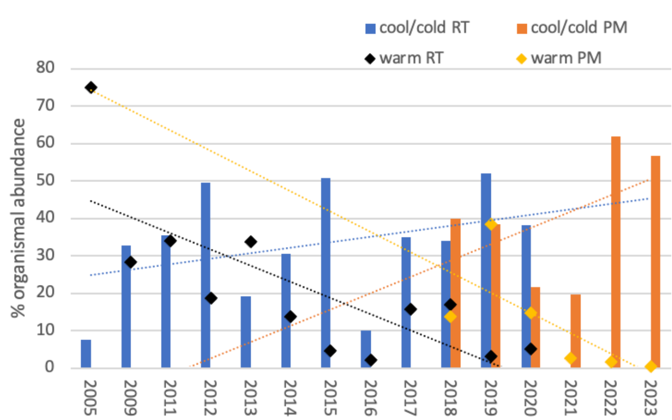


Figure 115. Cluster dendrogram of the WC2600 community in all sampling years. Blue = riffle-targeted, aqua = multihabitat. The number at the end of each label indicates sampling year.

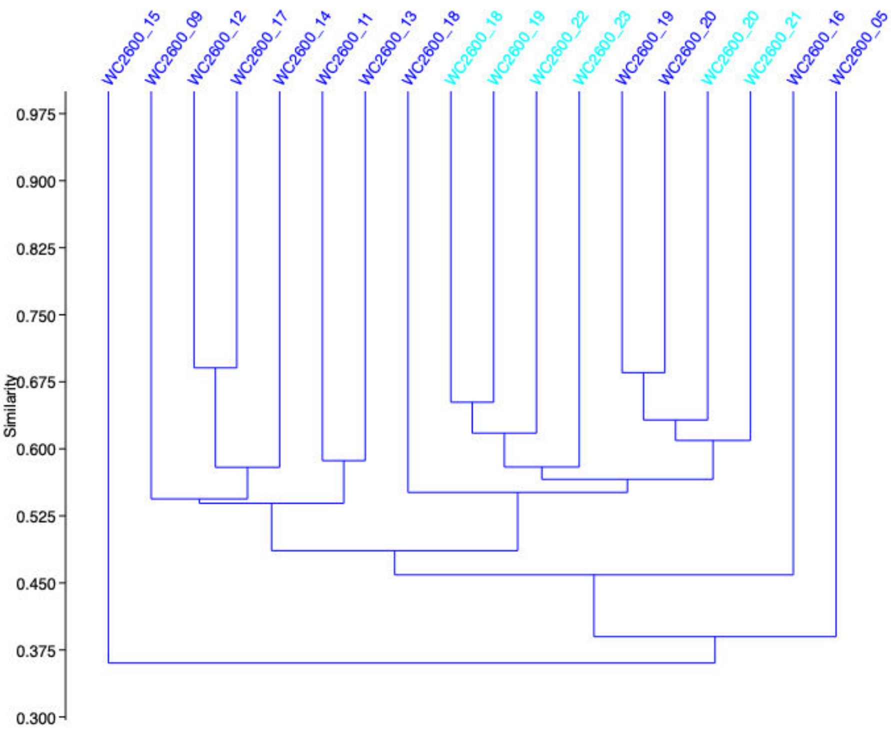


Figure 116. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC2600 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 24% of total sample variation; axis 2 explains an additional 18% of variation.

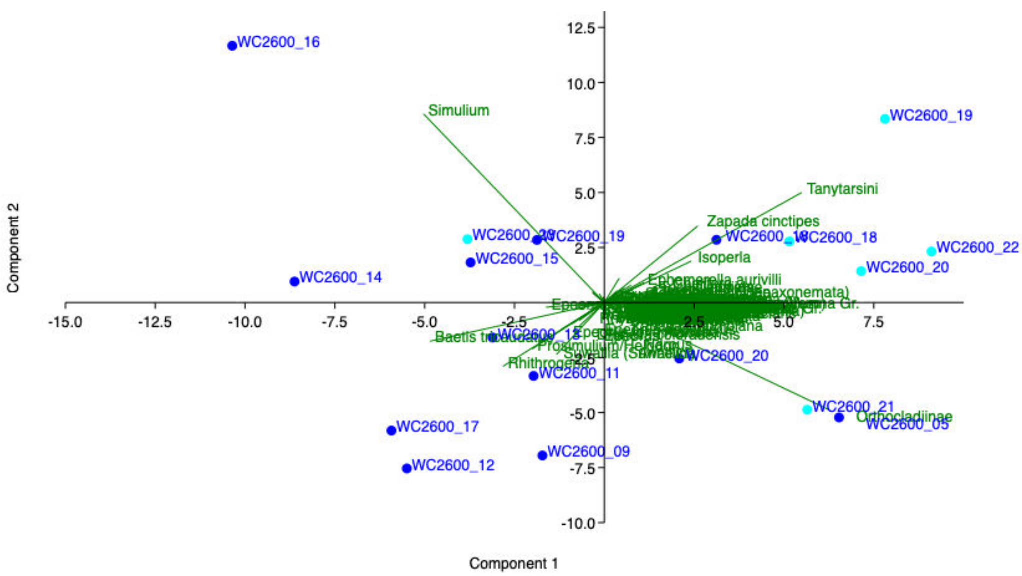
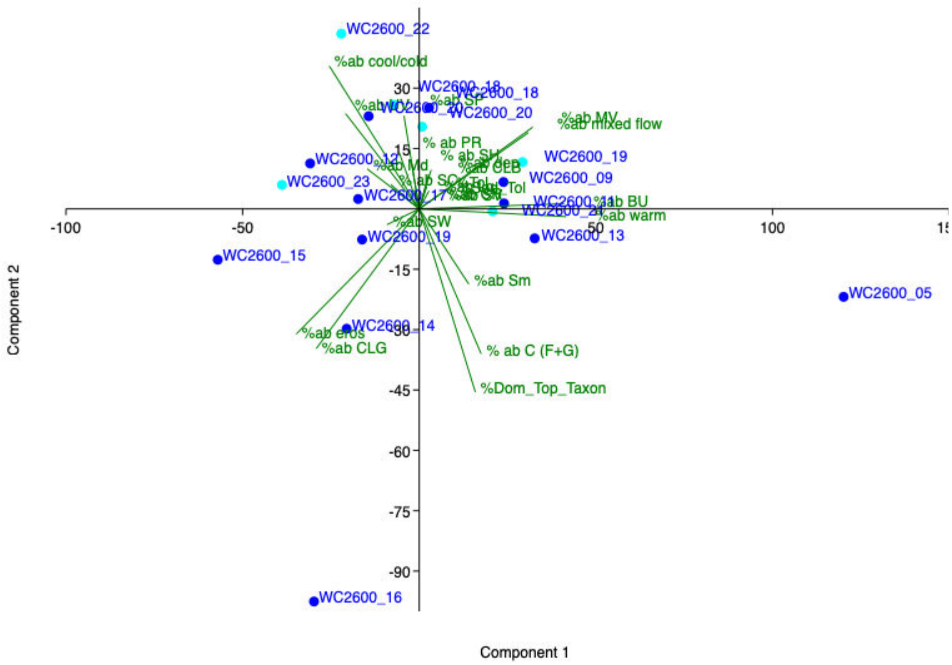


Figure 117. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC2600 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 37% of total sample variation; axis 2 explains an additional 22% of variation.



Discussion

Macroinvertebrate sampling and bioassessment in Whychus Creek has been ongoing since 2005. During that time, restoration activities were implemented at the basin level, such as restoring perennial flow; and at the reach level, including re-meandering mainstem channels, restoring floodplain connectivity, and creating new heterogeneous side channels and a more dynamic, mobile primary channel. The benthic macroinvertebrate community was expected to change in response to habitat changes due to restoration, but additional factors operating throughout this extended sampling period such as altered precipitation patterns, extreme heat, and widespread wildfires can also be expected to affect community composition. Given the significant changes in some community metrics at both upstream and downstream sites that were not actively restored, it is likely that both climate factors as well as potential general uplift from multiple restoration projects are affecting the communities in different reaches.

Samples were taken in Whychus Creek on 15 occasions spanning 17 years, and while the species accumulation curve is approaching saturation, new taxa are taken almost every year. In 2023, three taxa were new to the complete dataset, and all but two (WC0600, WC1950) of the reaches that were sampled in multiple years had at least one taxon new to the reach. New taxa are generally present at very low abundances and may represent taxa that are present consistently but in such low numbers they are rarely taken in samples, as well as newly colonizing taxa that may or may not persist. Macroinvertebrate community composition continues to be strongly influenced by reach location, with greater similarity among communities in upstream reaches (i.e., WC1950-WC2600) compared to those in downstream reaches (i.e., WC0600-WC1250), regardless of sampling technique or whether a primary or side channel reach was sampled.

Changes in macroinvertebrate community composition over time are reflected by changes in community metrics, even at sites that have not experienced active restoration. Significant unidirectional trends over time were seen in 31 of 35 calculated community metrics (see Appendix B), with all but a single site (WC2050) having significant trends in 2-16 metrics. The greatest number of significant trends (16 metrics) was seen at two sites (WC1100-2, WC2600) where restoration resulted in a more dynamic primary channel reach. The direction of the significant trends for most metrics, including taxonomy-based traits such as taxa richness (total, EPT, sensitive, sediment-sensitive taxa) and ecological traits such as functional feeding group, voltinism, temperature associations, and habit, suggested improving habitat conditions, i.,e., faster flows, cooler temperatures, more macrophytes, and greater habitat stability. However, other trends suggested declining habitat conditions; all significant trends in community temperature optima (significant at a single site) and community sediment optima (significant at three sites) showed an increase over time, as did significant trends in relative abundance of tolerant organisms (significant at four sites) and sediment-tolerant organisms (significant at three sites). Except for a single site (WC2600), all significant trends in the four negative community metrics above were seen at sites in downstream reaches (WC1025-1 to WC1150).

Unrestored reaches

Among the sites sampled in 2023, only WC0150, W0600, and WC1150 have not undergone active restoration. However, as these sites are among the furthest downstream on the creek, they may experience a general uplift due to habitat and macroinvertebrate community changes at multiple upstream restored reaches in addition to any impacts on community composition from climate-related factors. WC0150 was sampled in 2023 after a gap of five years. Three community metrics showed a significant decrease over time, with fewer predators, climbers, and crawlers. Predator abundance relies on a stable prey base, while climbers and crawlers can be impacted by the density of macrophytes. WC0600, which was sampled in every project year, showed significant trends in nine community metrics; this site also had decreasing predator and crawler abundance, but trends in other metrics suggested improving habitat conditions, and this was the only site with a significant trend in IBI score (increasing). Results at WC1150, where 10 community metrics showed a significant unidirectional trend, were also mixed, with some suggesting improving habitat conditions (i.e., mayfly richness, abundance of large-bodied organisms, semivoltine organisms, and climbers) and some suggesting declining conditions (i.e., fewer predators, more tolerant organisms). It is likely that environmental/climate factors are operating on the community at all the sites, but the greater number of significant trends at WC1150 may be due to greater proximity to multiple restoration projects.

Three sites were sampled for the first time in 2023: WC1175, WC1200, and WC1250. These sites are just upstream of the unrestored WC1150 reach, which was used as a comparison for WC1100-2 following restoration there. All three sample communities were similar to the WC1150 community (>70%), and were also more closely related to the communities at WC1025 and WC1100 than to any remaining samples further upstream or downstream. The community at WC1175 was perhaps the most notable of the three, as it had the most total taxa and the most balanced/even community among all 2023 samples (suggesting higher-quality habitat conditions), but also the lowest proportion of all taxa comprised by EPT and the fewest univoltine organisms (suggesting less stable and/or lower quality habitat conditions). The community at WC1250 had the highest community temperature optima among all 2023 samples, while all calculated trait values for WC1200 fell within the ranges seen among 2023 samples.

Restored reaches

Most of the restored reaches that were sampled in 2023 experienced restoration quite recently (2021 [WC1025] or 2022 [WC2000, WC2050, WC2425]), while restoration at some sites occurred 7-11 years ago (2012 [WC1950]; 2016 [WC1100]). There are few years of post-restoration data available for the three reaches restored most recently, so identifying trends is not yet possible. However, while some degree of community disturbance can be inferred from changes in macroinvertebrate community composition and community metric values post-restoration, multiple positive metrics improved at each site. The communities at WC2000 and WC2050 were less balanced in 2023 compared to pre-restoration, although the relative abundance of the top taxon in both was still close to the range that receives the highest score in the ORDEQ IBI (<20%), while the WC2425 community was more balanced in 2023 than in any prior year. PREDATOR and IBI scores were both lower at WC2000 and WC2425 following restoration (no RT samples have been taken at WC2050). Total and EPT taxa richness were similar pre- and post-restoration at WC2000 and WC2050, but values of both were lower at WC2425, although this site also had a more balanced community post-restoration.

The post-restoration macroinvertebrate community at WC2000 was fairly similar to pre-restoration years (~58% similar to all prior years), but the 2023 PM and RT samples were more similar to each other than to any prior year. Multiple community metrics also differed post-restoration, in ways that suggest increased disturbance. WC2000 had more sensitive taxa and more tolerant and sediment-tolerant organisms post-restoration, but fewer sediment-sensitive taxa. There were also fewer cool/cold-associated organisms and more warm-associated post-restoration. Relative abundance of shredders, climbers, and large organisms was much lower post-restoration, and there were few semivoltine organisms and more swimmers. These all suggest increased habitat disturbance and a reduction in aquatic macrophytes.

WC2050 had fewer years of pre-restoration sampling than the other two recently restored sites, and between-year similarity of the macroinvertebrate community in all three sampling years was relatively high. However, multiple community metrics differed post-restoration, in ways that suggest increased habitat quality. In the year following restoration, WC2050 had more sensitive and sediment-sensitive taxa, fewer tolerant and sediment-tolerant organisms, lower community temperature and sediment optima, and more cool/cold-associated and fewer warm-associated organisms. However, changes in other metrics post-restoration suggest some level of disturbance, with fewer predators, shredders, and semivoltine organisms, and more collectors and univoltine organisms.

The post-restoration community at WC2425 was among those in several prior years at this site without high similarity to other sampling years. Post-restoration results at this site were mixed; there were fewer sensitive and sediment-sensitive taxa but also fewer tolerant and sediment-tolerant organisms. Community temperature and sediment optima were slightly lower post-restoration, and there were more cool/cold-associated and very few warm-associated organisms. The 2023 community had more predators and fewer multivoltine organisms than any prior year, but also more burrowers (which can indicate increased sedimentation).

Restoration at WC1025 occurred in 2021 and thus there are currently few years of post-restoration data. Overall between-year community similarity is fairly high (>53%), but all post-restoration primary and side channel communities are more similar to each other than to pre-restoration communities. Changes in community composition are evident from post-restoration differences in taxonomic and ecological metrics, which reflect some level of post-restoration disturbance. In the primary channel, there were significantly more mayfly taxa but also significantly more tolerant organisms and a higher community sediment optima post-restoration, which suggests higher sediment levels. There was little change in the number of total and EPT taxa post-restoration in the primary channel and although values of some community metrics in the side channels relating to functional feeding group and habit indicate higher sediment levels and more riparian zone inputs, total and EPT taxa richness in the side channels has been similar to post-restoration values in the primary channel.

Restoration at WC1100 occurred in 2016. The 2017 macroinvertebrate community reflected restoration-related disturbance; a subsequent overall change in community composition was evident, with a significant difference between pre- and post-restoration communities. Several traits differed significantly pre-and post-restoration, with more sensitive taxa and large organisms and lower abundance of the dominant taxon post-restoration, as well as fewer collectors and swimmers, suggesting more stable and less sedimented habitat. This site also had significant unidirectional trends in more metrics (16) than any other site sampled in 2023 except for WC2600. Prior to restoration, there were significantly fewer EPT taxa at WC1100 compared to the upstream control WC1150, but this difference disappeared post-restoration. Although other trait values did not differ significantly between the project and reference reaches pre- and post-restoration, many were comparable to or greater than those at the reference site in the years after restoration occurred.

Restoration at WC1950 occurred in 2012, and macroinvertebrate community composition was significantly different pre- and post-restoration. The only community metric that differed significantly pre- and post-restoration at this site was the number of sensitive taxa, which was greater following restoration, but there were significant unidirectional increases across time in several metrics that suggest improved habitat conditions, with more EPT and sediment-intolerant taxa and fewer multivoltine and burrower organisms.

WC2600 is downstream of restoration done in the Whychus floodplain in 2014. In the years since restoration the primary channel has taken on secondary channel characteristics, and macroinvertebrate community composition is significantly different post-restoration. Several community metrics differed significantly post-restoration, and this site also had significant unidirectional trends in more metrics (16) than any other site sampled in 2023 except for WC1100-2.

While restoration activities have altered macroinvertebrate community composition and associated ecological traits, changes at unrestored reaches over time also suggest some degree of overall uplift. Among the 2023 sites that were sampled in multiple years, 82 of 385 community metrics (11 sites sampled x 35 metrics; Appendix B) showed a significant unidirectional trend over time, with 68% of the individual trends in a direction that indicated improved habitat conditions among both restored and unrestored sites. Metrics with the greatest number of negative trends (i.e., trend direction indicated declining habitat condition) included tolerant organisms and crawlers, suggesting that sedimentation may be an issue in the basin.

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Appendix A. Range, mean, and standard deviation (Std. Dev.) of all 2023 sample community metrics.

	Min.	Max.	Mean	Std. Dev.			Min.	Max.	Mean	Std. Dev.
Taxa richness	21	62	49.7	10.1		% medium	6.1	60.6	18.3	12.4
Ephemeroptera taxa	6	14	9.5	1.8		% small	35.9	84.8	73.0	11.7
Plecoptera taxa	2	9	4.8	1.7		% multivoltine	13.1	53.5	31.4	12.5
Trichoptera taxa	3	9	5.6	1.7		%ab univoltine	28.9	72.4	46.1	11.0
# sensitive taxa	1	8	4.2	1.7		%ab semivoltine	2.4	23.1	8.9	5.8
# sediment-sensitive taxa	0	3	1.6	0.8		% cool/cold-associated	21.2	67.1	36.3	13.4
% dominance top taxon	12.3	38.7	20.2	6.5		% warm-associated	0.0	12.7	5.8	3.7
% tolerant	6.1	64.8	28.2	16.8		% depositional-associated	2.8	39.0	13.6	8.9
% sediment-tolerant	1.3	38.9	12.5	8.8		% erosional-associated	19.9	77.5	48.6	16.7
MHBI	2.1	4.6	3.9	0.6		% mixed flows	13.6	69.1	37.0	13.8
EPT richness	14	27	19.9	3.5		% burrowers	2.8	39.0	13.6	8.9
% diversity EPT	25.8	66.7	41.3	8.8		% climbers	0.0	23.6	4.1	5.3
community temperature optima	16.2	18.6	17.7	0.7		% crawlers	0.0	2.7	0.9	0.8
community sediment optima	5.4	10.6	7.6	1.3		% clingers	39.8	75.8	51.4	9.5
% predators	3.9	31.2	10.2	6.2		% sprawlers	1.2	28.5	11.0	6.2
% scrapers	9.9	38.1	22.2	7.6		% swimmers	3.5	40.1	16.5	9.7
% shredders	0.4	12.4	3.9	3.8		% non-insect	3.6	40.1	14.3	8.9
% collectors	27.5	65.5	53.3	9.6		% Chironomidae	1.7	47.2	15.5	12.5
% large	0.6	13.3	3.3	2.9						

Appendix B. Significant unidirectional site-level trends across time in calculated metrics

I = increasing over time; D = decreasing over time; N = no significant trends. Green indicates change in a direction that suggests improving habitat conditions; yellow indicates change in a direction that suggests declining habitat conditions. However, note that trends in some metrics such as flow association or habit may reflect the greater habitat heterogeneity and/or slower flows in new side channels following restoration at some sites.

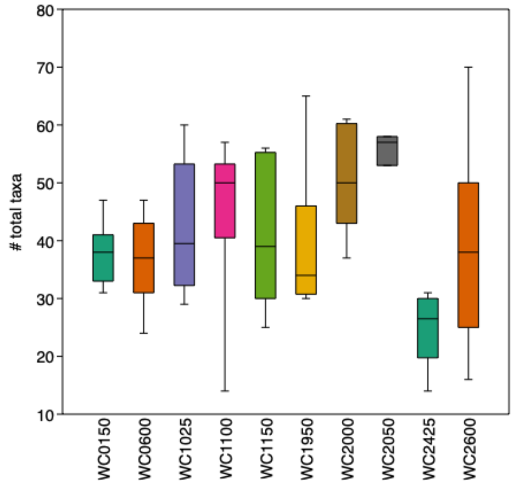
	WC1050	WC0600	WC1025-1	WC1100-2	WC1100-4	WC1150	WC1950	WC2000	WC2050	WC2425	WC2600
PREDATOR	N	N	N	N	—	N	N	N	—	N	N
IBI	N	I	N	I	—	N	N	N	—	N	N
# taxa	N	N	I	I	N	N	I	N	N	N	I
# Ephemeroptera	N	N	I	I	N	I	N	N	N	N	I
# Plecoptera	N	N	N	N	N	N	N	N	N	N	I
# Trichoptera	N	N	N	N	N	N	N	N	N	N	N
#EPT Taxa	N	N	N	I	N	N	I	N	N	N	I
# Sensitive	N	N	N	I	N	N	N	N	N	N	I
#Sed-sensitive	N	N	N	I	N	N	I	N	N	N	I
Temp. Optima	N	N	N	N	N	I	N	N	N	N	N
Sed. Optima	N	N	I	I	N	N	N	N	N	N	I
% Top Taxon	N	N	N	N	N	N	N	N	N	D	N
%Tolerant	N	N	I	I	I	I	N	N	N	N	N
%Sed-tolerant	N	N	I	I	N	N	N	N	N	N	N
% ab Predator	D	D	N	N	N	D	N	N	N	I	I
% ab Scraper	N	N	I	N	N	N	N	N	N	N	N
% ab Shredder	N	N	N	N	D	N	N	N	N	N	I
% ab Collector	N	N	N	D	D	N	N	N	N	N	D
%ab Large	N	N	N	N	N	I	N	N	N	I	I
%ab Medium	N	I	N	N	N	N	N	N	N	N	N
%ab Small	N	N	N	N	N	N	N	I	N	N	N
%ab Multivoltine	N	N	N	D	N	D	D	N	N	N	N
%ab Univoltine	N	I	I	I	N	I	N	N	N	N	N
%ab Semivoltine	N	I	N	I	N	I	N	N	N	N	I
%ab cool/cold	N	N	N	I	N	N	N	N	N	I	I
%ab warm	N	N	N	N	N	N	N	N	N	N	D
%ab depositional	N	N	I	N	N	N	N	N	N	N	N
%ab erosional	N	N	D	N	N	N	N	N	N	I	N
%ab mixed flow	N	N	N	N	N	N	N	N	N	N	N
%ab Burrower	N	N	N	N	N	N	D	N	N	N	N
%ab Climber	D	I	I	I	N	I	I	N	N	N	I
%ab Crawler	D	D	N	N	N	D	N	D	N	N	N

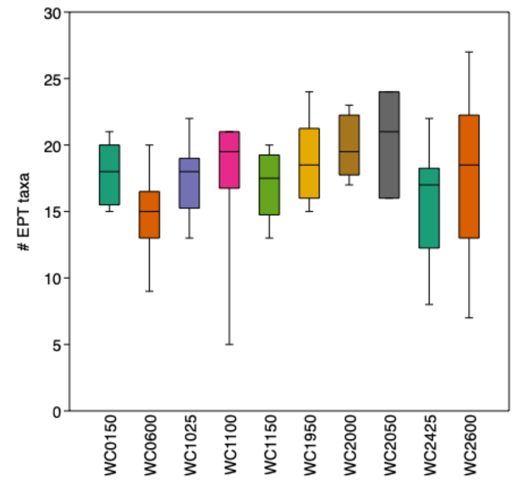
%ab Clinger	N	N	N	I	N	N	N	N	N	I	N
%ab Sprawler	N	N	N	N	N	N	N	N	N	N	I
%ab swimmer	N	N	D	N	D	N	N	N	N	I	N
total sig trends	**3**	**7**	**11**	**16**	**4**	**10**	**6**	**2**	**0**	**7**	**16**

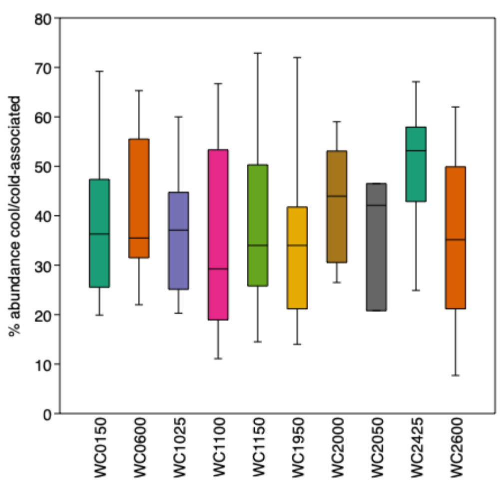
Appendix C. Significant differences in community metrics pre- vs. post-restoration

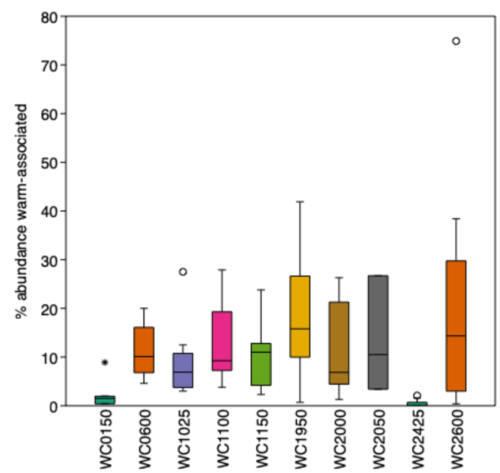
	WC1025-1	**WC1100-2**	**WC1950**	**WC2600**			**WC1025-1**	**WC1100-2**	**WC1950**	**WC2600**
# total taxa	--	--	--	--		% collectors	--	D	--	D
# Ephemeroptera	I	--	--	--		% large	--	I	--	--
# Plecoptera taxa	--	--	--	--		% medium	--	--	--	--
# Trichoptera taxa	--	--	--	--		% small	I	--	--	--
# sensitive taxa	--	I	I	--		% multivoltine	--	--	--	--
# sediment-sensitive taxa	--	--	--	I		%ab univoltine	I	--	--	--
% tolerant organisms	--	--	--	--		%ab semivoltine	--	--	--	--
% sediment-tolerant organisms	--	--	--	--		% cool/cold-associated	--	--	--	--
% top taxon	--	--	--	--		% warm-associated	--	--	--	D
Community temperature optima	--	--	--	I		% depositional-associated	--	--	--	--
Community sediment optima	I	--	--	--		% erosional-associated	D	--	--	I
% top taxon	--	D	--	--		% mixed flows	--	--	--	D
% tolerant organisms	I	--	--	--		% burrowers	--	--	--	D
% sediment-tolerant organisms	--	--	--	--		% climbers	--	--	--	--
% predators	--	--	--	--		% crawlers	--	--	--	--
% scrapers	--	--	--	--		% clingers	--	--	--	--
% shredders	--	--	--	--		% sprawlers	--	--	--	--
						% swimmers	--	D	--	--

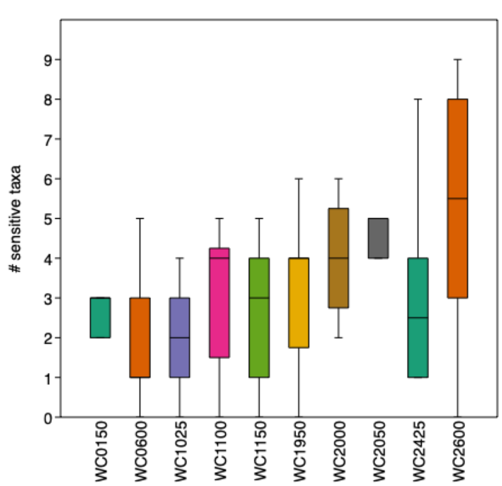
Appendix D. Boxplots of metric value ranges among all sites from downstream to upstream, 2005-2023

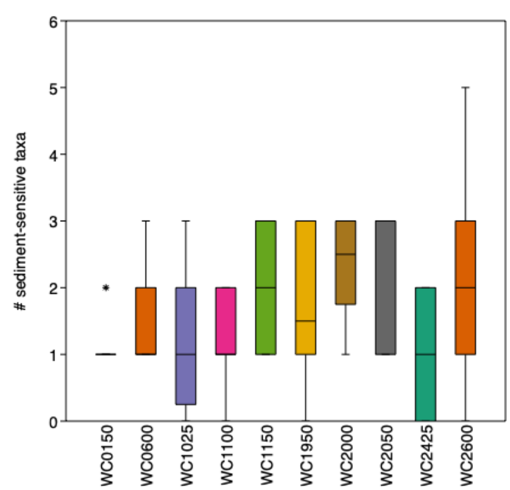


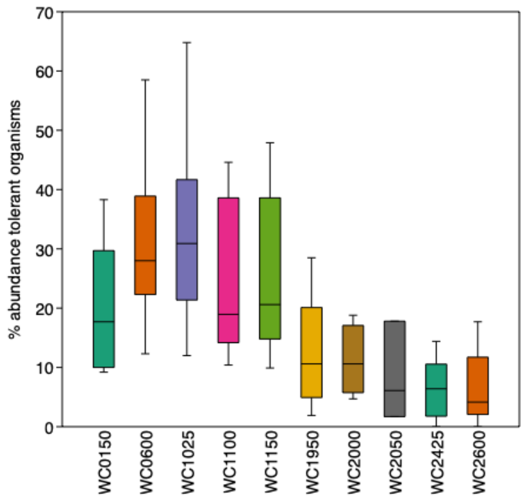


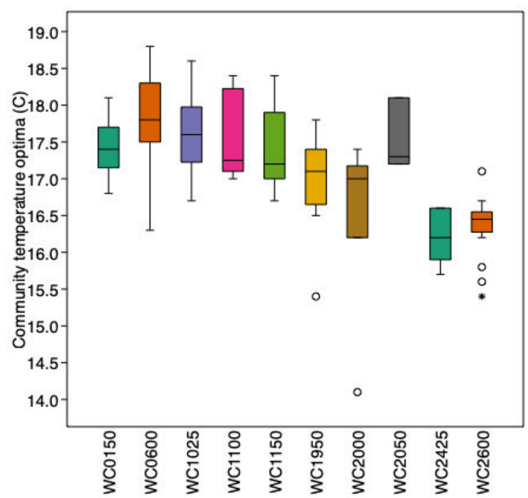


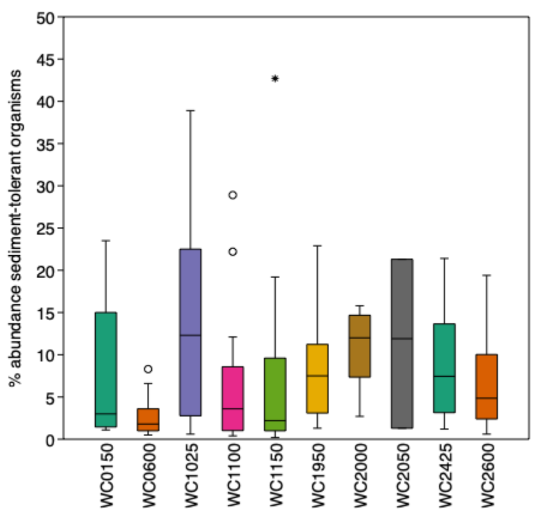


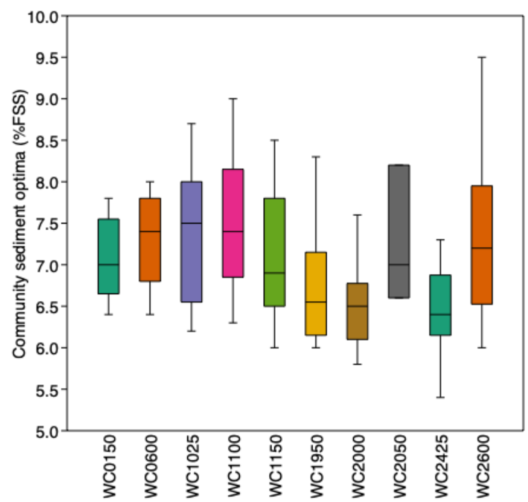












Cool/cold- and warm-associated organisms at WC0600 (long-term monitoring site, low in watershed, downstream of flow restoration and habitat restoration), WC1100-2 (within restoration), WC1825 (downstream of restoration), and WC2600 (long-term monitoring site, high in watershed, downstream of restoration).