## **CASM ENVIRONMENTAL, LLC**

Benthic Macroinvertebrate Monitoring in Whychus Creek (Sisters, OR), 2022



Sampling crew in Whychus Creek; CASM Environmental

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## Summary

The benthic macroinvertebrate community in Whychus Creek was sampled for the 14th year on 11-12 August 2022 in 12 reaches: Road 6360 (WC0600), Rimrock Ranch (WC0850, WC0900, WC1025), Whychus Canyon (WC1100, WC1150), Camp Polk (WC1825, WC1850, C1900, WC1925, WC1950), and Whychus Floodplain (WC2600). Samples were a mix of unrestored and restored sites and were taken 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 length.

A total of 148 unique taxa in 50 families was taken across all samples, which is similar to recent years (133-156 total in 2018-2021). Order-level diversity was highest among Diptera (true flies), with 63 unique taxa in 10 families, and the EPT were also well-represented, with 21 mayfly taxa in five families, 17 caddisfly taxa in eight families, and 11 stonefly taxa in five families. Only two taxa not found in any prior year were collected, each as a single individual: the nonbiting midge *Parochlus* (WC2600) and the true fly family Muscidae (WC1025 primary and side channels). Sample richness ranged from 30-67 taxa (mean = 48.9, SD 8.7). PM samples had significantly more total taxa, more chironomid midges, and more organisms that are multivoltine, sprawlers, and tolerate a variety of flow types, while RT samples had significantly more organisms associated with faster flows. There was no significant difference between RT and PM samples for other calculated community metrics. Macroinvertebrate community composition continues to be strongly influenced by reach location, with 60% overall community similarity among most of the downstream sites (WC0600 through WC1150), regardless of sampling method. Abundance across the dataset was dominated by *Optioservus*, a tolerant riffle beetle that inhabits sediments and detritus in flowing and still habitats in streams, found in every sample except WC2600 at abundances ranging from 7-164 individuals.

Restoration in Whychus Creek restored perennial flow and created more dynamic, heterogeneous habitat. This was accompanied by changes in the macroinvertebrate community, even at sites with no active restoration, in ways that suggest overall uplift. PREDATOR and IBI scores are generally less informative than taxonomic and ecological metrics. Five of the nine RT samples taken in 2022 had O/E scores of 'most disturbed' (poor), but seven scored as slightly or not disturbed based on IBI score. No significant unidirectional trends were detected in O/E scores over time, and a significant trend in IBI score was seen at only WC0600 (increasing; unrestored site), WC1100-2 (increasing; restored site), and WC1825 (decreasing; restored site). Many sites had significant unidirectional trends in positive taxonomic and ecological traits, such as more total taxa (six sites), mayfly taxa (five sites), sediment-sensitive taxa (six sites), and more semivoltine and large-bodied organisms (indicative of stable habitat conditions; four sites each). Fewer sites had significant trends in metrics relating to temperature or flow associations, although WC1100-2 had significantly increasing community temperature optima and WC1025 and WC2600 had significant increases in

community fine sediment optima. Abundance of tolerant organisms also increased significantly across time at WC1025, WC1100-2, and WC1150.

Restored sites show a pattern of severe community disturbance immediately after restoration followed by recovery across one to three years. Side channels are colonized quickly and support a diversity of taxa with a range of tolerances and flow and temperature associations. Recent restoration at WC1025 (2021) seemed less disruptive, as many 2022 metrics were in the range of prior years and differed little from WC0900 downstream. More taxa new to the site (8) were taken here than in any other 2022 sample; most were in the side channels, and the number of total and EPT taxa in the side channels was comparable to the primary channel. The community at WC1100, which was restored in 2016, was severely disturbed in 2017 but subsequent post-restoration recovery included significantly more total and EPT taxa, cool/cold-associated organisms, and higher IBI scores. The Camp Polk reaches, restored in 2012, had the lowest abundances of warm-associated organisms among the entire sample set, which may be a reflection of groundwater sporing inputs in the upstream Camp Polk reach, although post-restoration recovery included increases in both positive and negative metrics. Several years after restoration, the WC1950 community showed significant increases across time in the number of total, mayfly, EPT, sensitive, and sediment-sensitive taxa, and in the number of shredders, large-bodied, and cool/cold-associated organisms.

## Background

Whychus Creek is a designated priority watershed for conservation and restoration in the upper Deschutes Basin. Projects implemented since 1999 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. 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 11-12 August 2022 in multiple regions of Whychus Creek: Road 6360 (WC0600), Rimrock Ranch (WC0850, WC0900, WC1025), Whychus Canyon (WC1100, WC1150), Camp Polk (WC1825, WC1850, WC1900, WC1925, WC1950), and Whychus Floodplain (WC2600). Multiple samples were taken at WC1025 and WC1925 to assess different side channel reaches following restoration done in 2021 and 2012, respectively. RT samples were taken in nine reaches (WC0600, WC0900, WC1025-1, WC1100-2, WC1150,

WC1825, WC1850, WC1900, WC1950), including a duplicate sample (WC1950DUP) for quality assurance. PM samples were taken in seven reaches (WC0850, WC1025 primary and side channels, WC1100-2, WC1925 primary and side channels, WC2600).

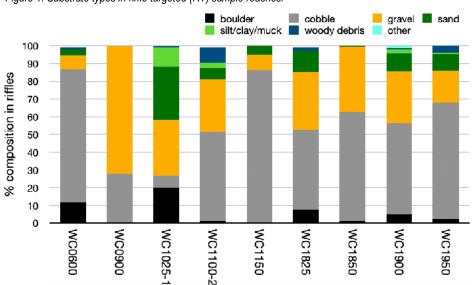
## Macroinvertebrate sampling techniques

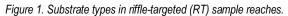
Sampling was done by CASM Environmental, UDWC staff, and volunteers from natural resource agencies and the surrounding community. After CASM Environmental staff demonstrated sampling techniques, teams received sampling kits and maps and dispersed into the field. Teams returned samples, data sheets, and 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 channel were flagged by UDWC prior to sampling. A reach sample consisted of eight individual net sets, each collected in a 1 ft<sup>2</sup> 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).

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 staff 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 of net sets 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<sup>2</sup> (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 using hands or feet while the D-net was swept repeatedly through the suspended material to capture disturbed/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 while the material was kicked vigorously to dislodge invertebrates. Net sets were composited and processed as described for riffle samples.

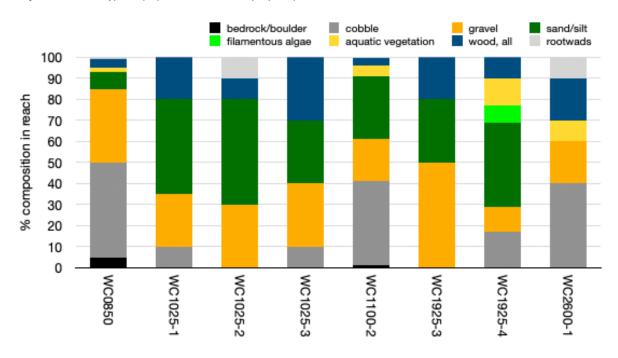


Figure 2. Substrate types in proportional multihabitat (PM) sample reaches.

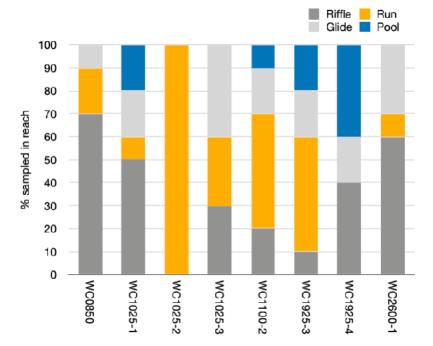


Figure 3. Flow types in composited PM samples.

#### 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 which were then selected randomly, and all individuals in each selected aliquot 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 & Covich, 2001; 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; burrowers are selected for in

sedimented habitat, while sprawlers and crawlers can be smothered and/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.</li>
- 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).

In 2022, the numbers of ORDEQ-designated temperature and sediment indicator taxa were not calculated. These represent only a small proportion of the total taxa in a sample and metrics of temperature association and sediment tolerance are already measured in a way that incorporates more of the taxa in a sample.

## 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, non-metric multidimensional scaling (nMDS) ordinations, 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. 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 analysis was done to assess site-level trends in calculated metrics, 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 1). 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;  $\leq$ 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 1. ORDEQ macroinvertebrate-based IBI metrics and scoring. <sup>a</sup> relative abundance of the most abundant taxon; <sup>b</sup> 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-sensitive taxa	≥2	1	0		
% dominance <sup>a</sup>	<20	20-40	>40		
% tolerant taxa	<15	15-45	>45		
% sediment-tolerant taxa	<10	10-25	>25		
MHBIÞ	<4	4-5	>5		
Summed score & condition					

<20 Severely impaired; 20-29 moderately impaired; 30-39 slightly impaired; >39 Minimally/not impaired

## **Results**

## Macroinvertebrate community in 2022 samples

This is the 14th year of macroinvertebrate sampling in Whychus Creek and some new taxa are found every year (Figure 4). The increase in total taxa taken annually since 2016 was driven by restoration at different sites, creation of new dynamic, heterogeneous side channels, and use of PM sampling, which often captures more taxa than RT samples in the same reach. The rate of accumulation is slowing as the species curve approaches saturation, and only two taxa were new to the Whychus dataset in 2022: *Parochlus*, a non-biting midge found in a variety of running waters, which is a member of a chironomid sub-family (Podonominae) that was also seen for first time in the Whychus dataset (one individual at WC2600); and Muscidae, a true fly family whose tolerant aquatic and semi-aquatic members are found in a range of flows and temperatures (one individual each in WC1025-1 RT, WC1025-1 PM and WC1025-2 PM).

A total of 148 unique taxa in 50 families (33 insect families, 17 non-insect) was collected across all sites, similar to other recent years (Figure 4). Two taxa were seen in every sample: *Ampumixis dispar*, a sediment-intolerant riffle beetle associated with clear, cold flowing water; and *Simulium*, a black fly associated with flowing water. Order-level diversity was highest among Diptera, with 63 unique taxa in 10 families, including 43 non-biting midge (Chironomidae) genera. Other well-represented groups included Ephemeroptera (mayfly; 21 unique taxa in five families), Trichoptera (caddisfly; 17 unique taxa in eight families), and Plecoptera (stonefly; 11 unique taxa in five families). Abundance was dominated by *Optioservus*, a tolerant riffle beetle that inhabits sediments and detritus in a variety of flows, found in every sample at abundances ranging from 7-164 except at WC2600. Other abundant taxa included *Simulium*, a black fly associated with flowing water found in every sample at abundances ranging from 1-198; and Naidinae, a widespread subfamily of tolerant, sediment-tolerant sludge worms associated with slower flows found in every sample except WC0600 and WC1950DUP at abundances ranging from 1-275 individuals.

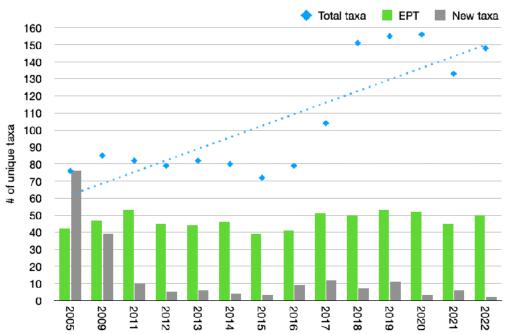
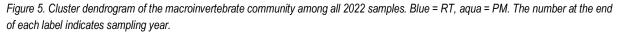


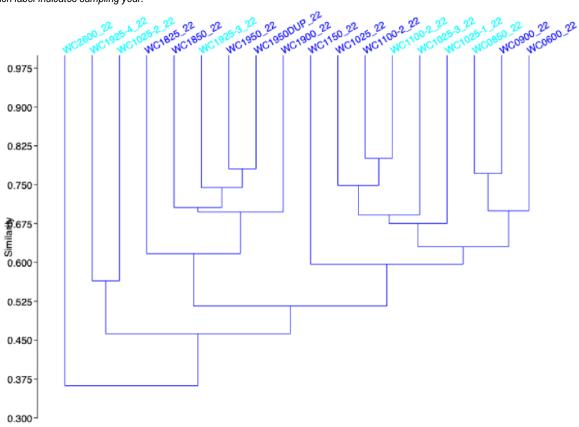
Figure 4. Taxa accumulation in Whychus Creek samples. Total taxa = # of unique taxa taken among all samples in each year; EPT = number of unique Ephemeroptera (mayfly), Plecoptera (stonefly), and Trichoptera (caddisfly) taxa; new taxa = # of taxa taken for the first time at any Whychus site in each year.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

Because PM samples are taken in all types of substrate in a reach, they contain more net sets from slower flows and/or softer substrates (i.e., sand/silt, vegetation, algae). However, all PM samples except the WC1025-2 side channel included net sets taken in at least one riffle (Figure 3). Despite inclusion of still water habitat, PM samples consistently capture sensitive organisms, and this more heterogenous habitat array often results in greater sample richness. The target sub-sampling number of 500 individuals was attained in every sample except WC1150 RT (267 organisms total), with 15-97% of the sample picked. Sample richness ranged from 30 to 67 taxa (mean 49, SD 8.7); this metric in the ORDEQ IBI receives the highest scaled score at >35 taxa, and only WC1150 RT was below this threshold (30 taxa). PM samples had significantly more total taxa and higher chironomid midge abundance as well as

more organisms that are multivoltine, tolerate a variety of flow types, and move as sprawlers. RT samples had significantly more organisms associated with faster (erosional) flows. No other calculated metrics differed significantly between sampling methods.

Macroinvertebrate community composition continues to be strongly influenced by reach location (Figure 5). Communities at most of the downstream sites (WC0600 through WC1150) were about 60% similar overall and clustered separately from most upstream sites (WC1825 through WC1950), regardless of the sampling method used. The WC2600, WC1925-4, and WC1025-2 communities were outliers, with overall similarity of less than 50% to all other 2022 samples; all were taken in reaches with some degree of side channel characteristics, and despite being more spatially separated, communities in the WC1925-4 and WC1025-2 side channels were more similar to each other (Bray-Curtis similarity index = 0.56) than to any other 2022 sample. The paired RT samples taken in the WC1950 reach were most similar to each other (Bray-Curtis similarity index = 0.78; Figure 2), confirming consistency and robustness of the sampling technique.





Differences between means of calculated trait values among all downstream (WC0600-WC1150) and upstream (WC1950-WC2600) sites were significant for only a subset of traits. Downstream reach samples had significantly more mayfly taxa, higher community temperature optima, and more scrapers and semivoltine organisms; upstream site samples had significantly more stonefly taxa, predators, and organisms associated with cool/cold water. Taxa that contributed the most to overall differences between downstream vs. upstream samples included *Optioservus* and *Zaitzevia*, riffle beetles associated with faster flows and more stable habitat, which were more abundant in downstream samples; and *Simulium*, a black fly associated with flowing water and recent habitat disturbance, which was more abundant in upstream reaches (Figure 6).

Differences in traits calculated as relative abundances primarily distinguished between the three PM samples that differed most from other 2022 samples (i.e., WC1025-2, WC1925-4, and WC2600; Figure 7), and included relative abundance of non-biting midges (%ab Chironomidae), organisms that tolerate a variety of flow types (%ab mixed), and multivoltine organisms (%MV), which were greater in those outlier samples. These traits are all associated with more disturbed conditions.

Figure 6. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all 2022 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 30% of total sample variation; axis 2 explains an additional 24% of variation.

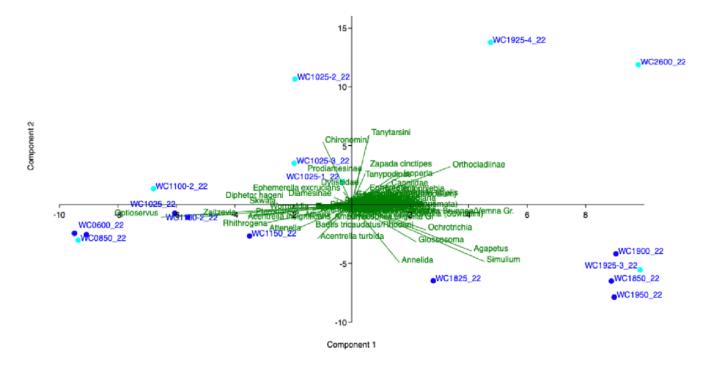
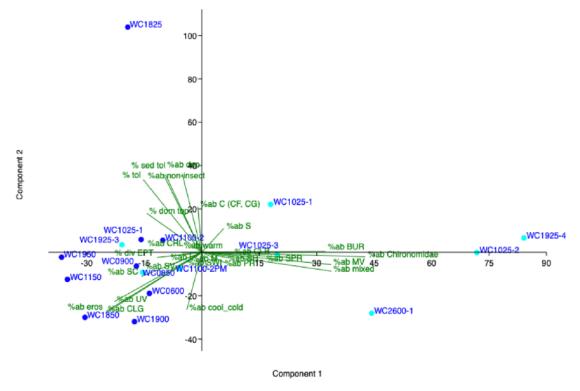


Figure 7. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances in all 2022 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 38% of total sample variation; axis 2 explains an additional 27% of variation.



Macroinvertebrate community characteristics at sampling sites

## WC0600 (Road 6360)

RT samples were taken in the primary channel at WC0600 in every sampling year from 2005-2021; in 2018 and 2019, a PM sample was taken in the same reach. WC0600 is a longterm 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 2022 sample was taken primarily in cobble substrate (Figure 3) and the macroinvertebrate community was most similar to those at the nearby downstream sites WC0850 and WC0900 (Figure 5). The community was dominated at 18.5% relative abundance by *Optioservus*, a tolerant riffle beetle associated with clear water in a variety of flows and temperatures whose presence suggests stable habitat conditions. *Optioservus* dominated the 2021 sample at similar abundance, but this site has been dominated at moderate abundance by a variety of taxa (Figure 8), mostly associated with faster flows. The majority of the 2022 community consisted of small, univoltine taxa that feed as collectors and move as clingers in faster, cooler flows. Only one taxon collected in 2022 was not found here in any prior sampling year: *Protoptila* (1 individual), a saddlecase-maker caddisfly associated with larger warmer rivers in both clear and sedimented waters.

Trends analysis found significant increases across all sampling years in IBI score, taxa richness, number of sensitive and sediment-sensitive taxa, and abundances of semivoltine, medium-length, and climber organisms; and a

significant decrease in crawler abundance (Appendix B). The target sub-sampling number of 500 organisms was attained in 11 of 14 sampling years, including every year since 2015 (Figure 9). Taxa richness was significantly greater in 2018-2022 (41-47) compared to earlier years (24-37; p = 0.0001). There were more EPT taxa (20) in 2022 than any prior year (Figure 10), and relative diversity of EPT (48.8%) was greater at this site than any other 2022 sample. IBI scores consistently reflect better biological condition (slight to minimal impairment) than O/E scores (moderately to most disturbed; Figure 11). IBI scores in 2018-2022 were significantly greater (36-40) than earlier sampling years (24-38; p = 0.0075), but the 2022 O/E score was the lowest at this site. However, there were more sensitive taxa (4) in 2022 than in any prior year (Figure 12), and fewer sediment-tolerant organisms (2% abundance; Figure 13) than any 2022 sample. The community fine sediment optima was in the range of recent years (Figure 14) and at the lower end of the 2022 samples. However, community temperature optima was the highest since sampling began and the highest of all 2022 samples. Samples in recent years contained more organisms associated with warmer temperatures and fewer with cool/cold temperature associations (Figure 15), and this site had the most warm-associated organisms (20%) of any 2022 sample.

Macroinvertebrate community similarity was greater among recent samples (2018-2022), and the 2022 community was most similar to the 2018-2021 samples (Bray-Curtis similarity = 0.50 to 0.70; Figure 16). Community composition differed between early and later sampling years (Figure 17), with later years having more *Rhithrogena*, a sediment-sensitive flatheaded mayfly found stones in faster flows; *Acentrella insignificans*, a small minnow mayfly found in warmer streams; and *Optioservus*, a tolerant riffle beetle found in clear water and a variety of flows and temperatures. Differences in traits calculated as relative abundances did not distinguish consistently between clusters of years (Figure 18). All factors that accounted most for between-sample differences, such as relative abundance of

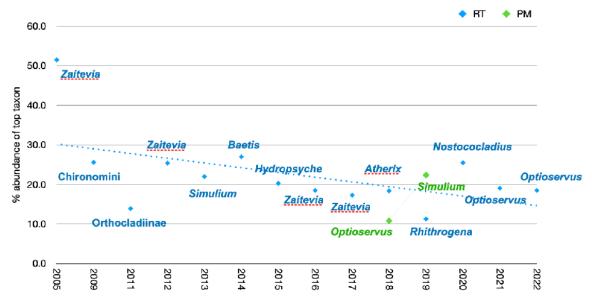


Figure 8. Relative abundance of the numerically dominant taxon at WC0600 in all sampling years. RT samples were taken every year; PM samples were taken in 2018 and 2019. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance.

organisms associated with cooler temperatures, scrapers, multivoltine organisms, and tolerant organisms varied annually with no consistent pattern or trends.

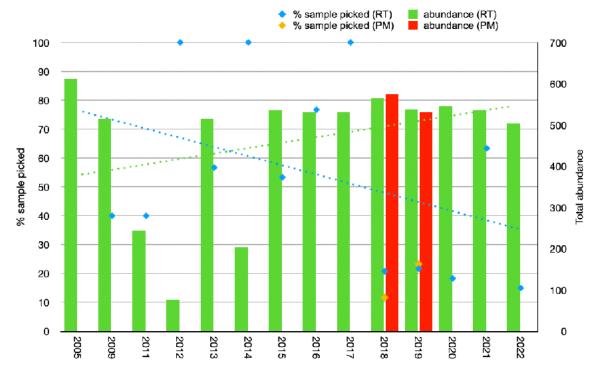
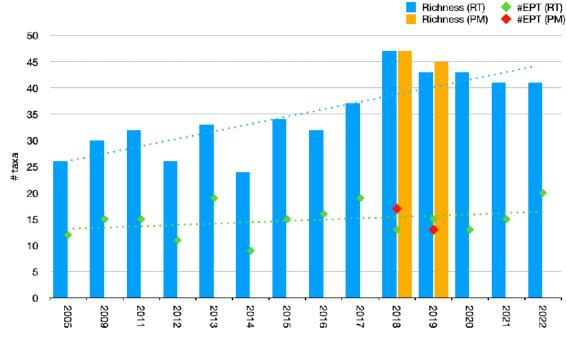


Figure 9. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC0600 in all sampling years. RT samples were taken every year; PM samples were taken in the same reach in 2018-2019. Target sub-sampling number is 500.

Figure 10. Sample richness and number of EPT taxa at WC0600 in all sampling years. RT samples were taken every year; a PM 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.



CASM Environmental 2022 UDWC Whychus

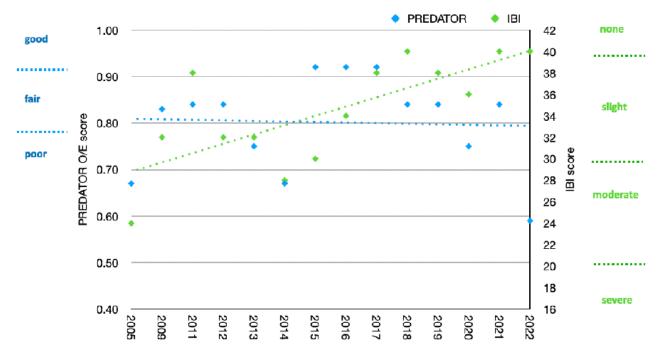


Figure 11. PREDATOR O/E and ORDEQ IBI scores at WC0600 in all sampling years. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

Figure 12. Numbers of sensitive and sediment-sensitive taxa at WC0600 in all sampling years. RT samples were taken every year; PM 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 >2 sediment-sensitive taxa.

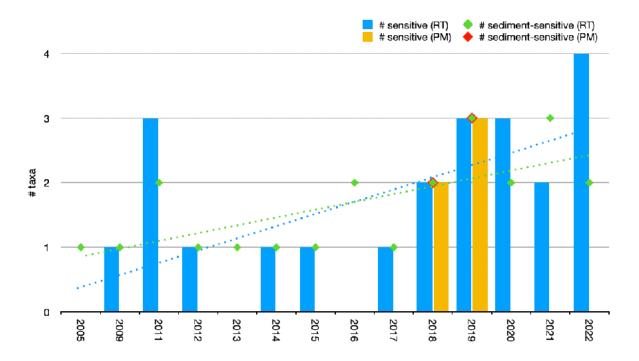


Figure 13. Relative abundance of tolerant and sediment-tolerant organisms at WC0600 in all sampling years. RT samples were taken every year; PM samples were also taken 2018-2019. Values were averaged for the years in which both sample types were taken. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

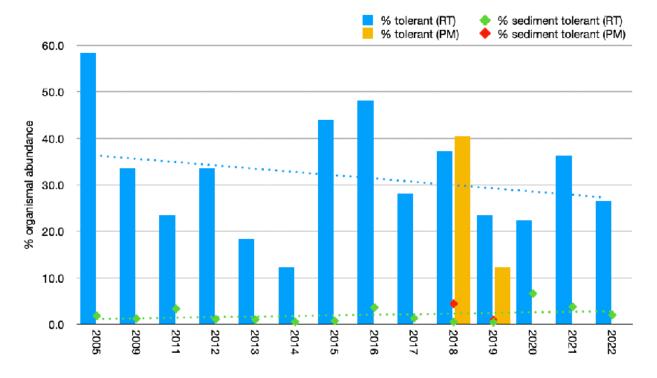
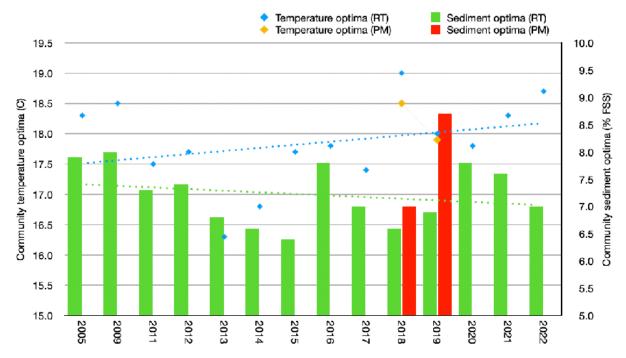


Figure 14. Temperature and fine sediment optima of the community (weighted means) at WC0600 in all sampling years. RT samples were taken every year; PM samples were also taken in 2018-2019. Values for years in which both sample types were taken were averaged. Linear trendlines are shown.



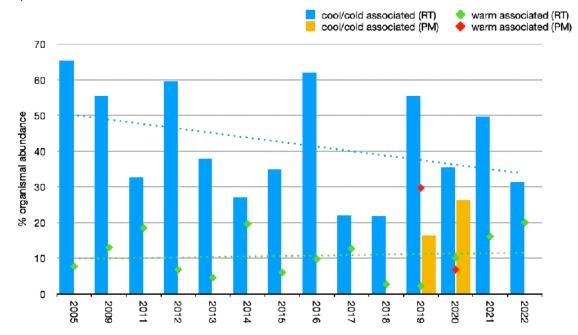


Figure 15. Temperature associations of the macroinvertebrate community at WC0600 in all sampling years. RT samples were taken every year; PM samples were also taken in 2018-2019. Linear trendlines are shown.

Figure 16. Cluster dendrogram of the WC0600 macroinvertebrate community in all sampling years. Blue = RT, aqua = PM. The number at the end of each label indicates sampling year.

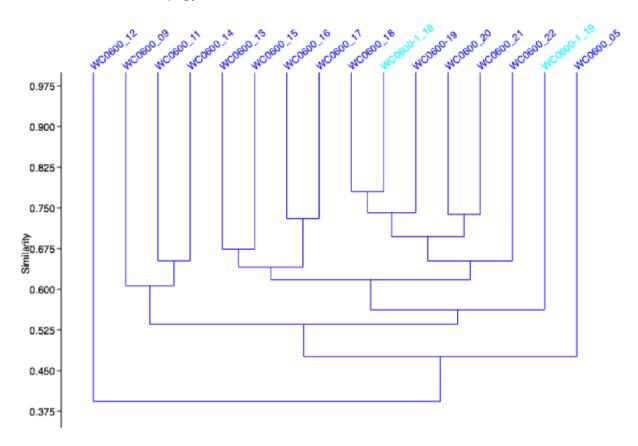


Figure 17. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0600 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 24% of total sample variation; axis 2 explains an additional 16% of variation.

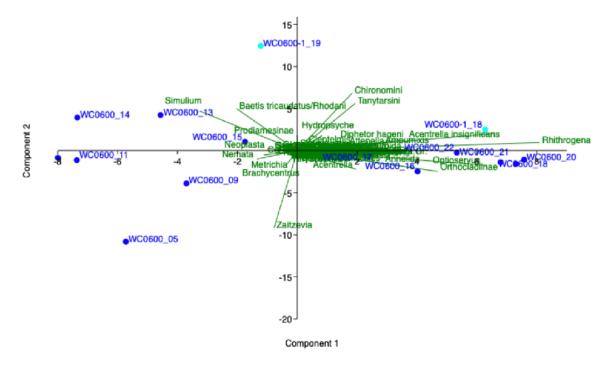
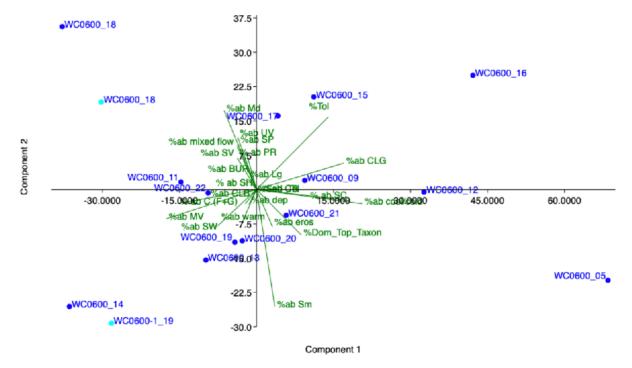


Figure 18. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0600 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 41% of total sample variation; axis 2 explains an additional 17% of variation.



#### WC0850 (Rimrock Ranch)

RT samples were taken in the primary channel of Whychus Creek around RM 8.5 in 2011-2017; PM samples were taken in 2020-2022 to provide additional baseline data for planned restoration actions. The 2022 PM sample was taken primarily in cobble/gravel substrate in moving water (Figure 1 & 2). The 2022 community was most similar to the adjacent WC0900 (Bray-Curtis similarity index = 0.77; Figure 5) and was dominated at 25.5% relative abundance by *Optioservus*, a tolerant long-lived riffle beetle associated with clear waters in a variety of flows and temperatures whose presence suggests stable habitat conditions. *Optioservus* also dominated the 2021 sample; this is a recent shift, as the dominant taxon in five of the eight sampling years between 2011 and 2020 was *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water that can increase in abundance with habitat disturbance (Figure 19). The 2022 community consisted mainly of small, tolerant, univoltine organisms that feed as collectors and move as clingers in faster, cooler flows. Two taxa collected in 2022 were not found at this site in any prior sampling year: *Anafroptilum* (1 individual), a tolerant small minnow mayfly associated with sandy sediments in cool waters; and *Protoptila* (1 individual), a saddlecase-maker caddisfly associated with larger, somewhat warmer streams in both clear and silted water.

A trends analysis revealed significant increases across time in relative abundance of scrapers, univoltine organisms, and organisms that tolerate a variety of flows; and a significant decrease in abundance of swimmers and organisms associated with faster flows (Appendix B). The target sub-sampling number of 500 organisms was attained in seven of 10 sampling years, including every year since 2016 (Figure 20). Taxa richness received the highest scaled score in the ORDEQ IBI (>35) for the last three years, and mean richness in 2020-2022 was significantly greater than all earlier sampling years (p = 0.0061). There were more EPT (22) in 2022 than in any prior year (Figure 21) as well as in any other 2022 sample except for WC2600. There were more sensitive and sediment-sensitive taxa in recent sampling years (Figure 22), and consistently very few sediment-tolerant organisms (Figure 23). However, the abundance of tolerant organisms is increasing, especially since 2017. Community temperature and sediment optima are increasing (Figure 24), and relative abundance of cool/cold-associated organisms has been lower in two of the last three sampling years, including 2022, although numbers of warm-associated organisms are consistently low (Figure 25). However, none of these trends were statistically significant.

Macroinvertebrate community composition is more similar overall among PM samples, which differed more from all RT samples except 2016 (Figure 26). The 2022 PM community was most similar to 2021 (Bray-Curtis similarity index 0.734), despite some differences between the two years in metrics such as relative abundance of warm-associated organisms and taxa richness. Community differences were largely driven by the dominant taxa, such as *Simulium* and *Optioservus*, as well as the sediment-sensitive flatheaded mayfly *Rhithrogena*, which was more abundant in the PM and 2016 RT samples; and *Baetis tricaudatus*, which was more abundant in RT samples in earlier years (Figure 27). Differences in traits calculated as relative abundances also distinguished between the PM and 2016 RT samples and prior sampling years (Figure 28), with more collectors in earlier sampling years and more scrapers in later years, and few to no climbers except in PM samples.

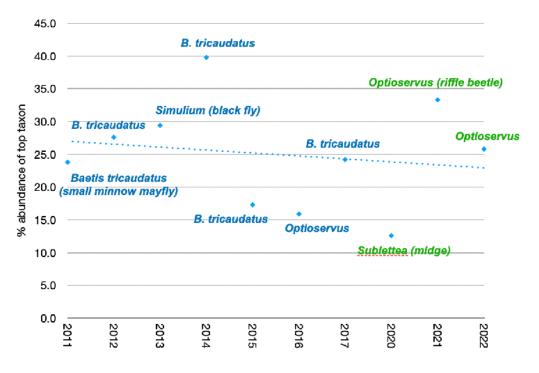
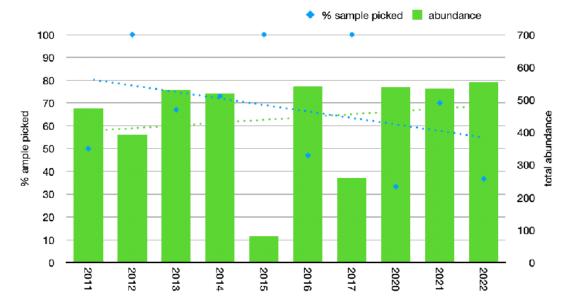


Figure 19. Relative abundance of the numerically dominant taxon at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken in 2020-2022. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

Figure 20. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken in 2020-2022. Target sub-sampling number is 500.



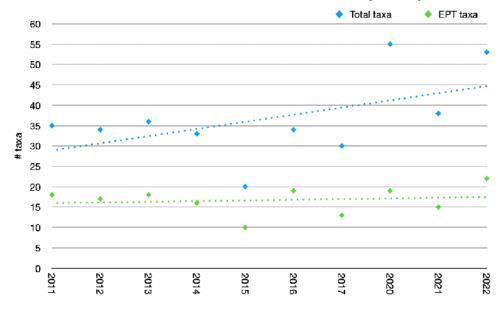
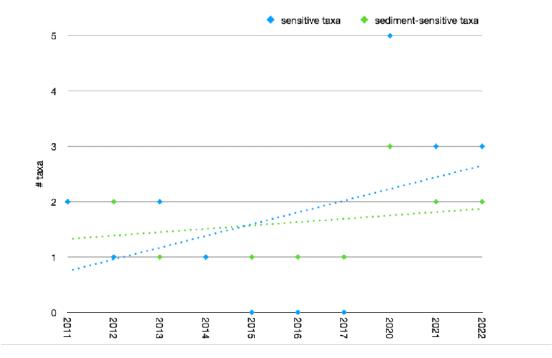


Figure 21. Sample richness and number of EPT taxa at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2020-2022. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

Figure 22. Numbers of sensitive and sediment-sensitive taxa at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2020-2022. 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.



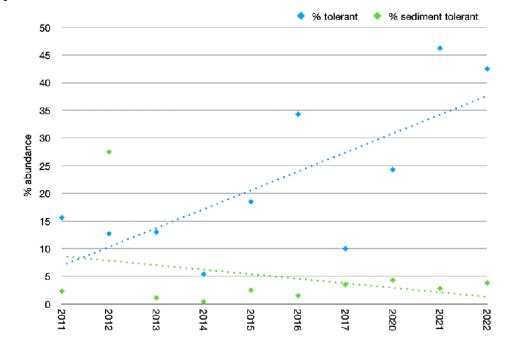


Figure 23. Relative abundance of tolerant and sediment-tolerant organisms at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2020-2022. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

Figure 24. Temperature and fine sediment optima of the community (weighted means) at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2020-2022. Linear trendlines are shown.



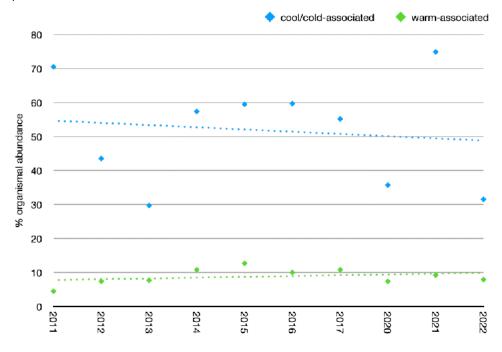


Figure 25. Temperature associations of the macroinvertebrate community at WC0850 in all sampling years. RT samples were taken 2011-2017; PM samples were taken in 2020-2022. Linear trendlines are shown.

Figure 26. Cluster dendrogram of the WC0850 macroinvertebrate community in all sampling years. Blue = RT, aqua = PM. The number at the end of each label indicates sampling year.

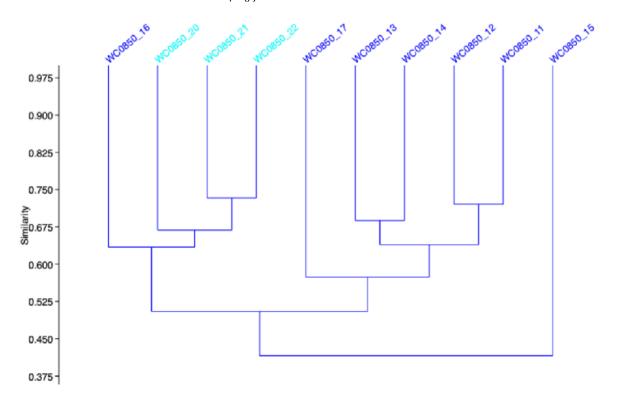


Figure 27. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0850 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 44% of total sample variation; axis 2 explains an additional 15% of variation.

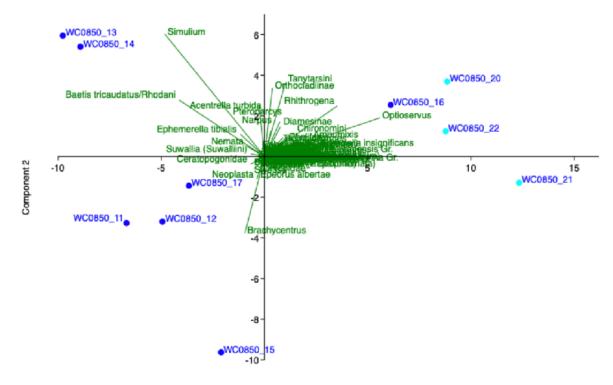
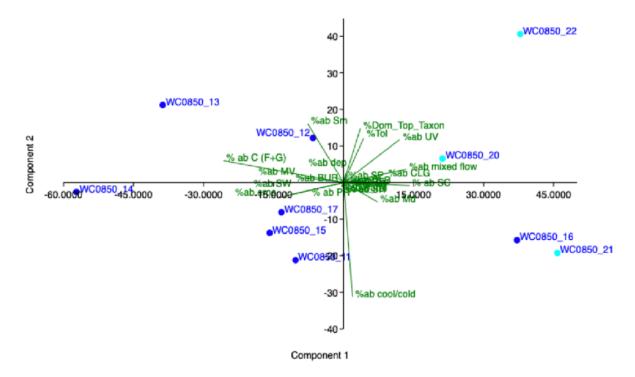


Figure 28. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0850 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 48% of total sample variation; axis 2 explains an additional 16% of variation.



#### WC0900 (Rimrock Ranch)

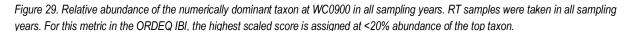
RT samples were taken in the primary channel of Whychus Creek at WC0900 in 2005-2017 and 2020-2022 to provide baseline data for future restoration. The 2022 sample was taken primarily in gravel substrate (Figure 3) and the macroinvertebrate community was most similar to those in nearby reaches (Figure 5), i.e., WC0850 PM and WC0600 RT (Bray-Curtis similarity index 0.771 and 0.664, respectively). The sample community was dominated at 18% abundance by *Optioservus*, a tolerant riffle beetle associated with clear waters in a variety of flows and temperatures whose presence suggests stable habitat conditions. This metric decreased since sampling began (Figure 29); relative abundance of the top taxon was significantly lower in 2015-2022 (mean 18.3%) compared to 2011-2014 (mean 26.7%; p = 0.0075), and samples scored in the highest range of the ORDEQ IBI for this metric in four of the last six years. The 2022 community consisted mainly of small, univoltine organisms that feed as collectors and move as clingers in faster flows, and all taxa were taken in at least one prior sampling year.

A trends analysis revealed significant increases across time in Ephemeroptera taxa and relative abundance of organisms that are scrapers, univoltine, medium-bodied, and climbers; and a significant decrease in relative abundance of the top taxon and of sprawlers (Appendix B). The target sub-sampling number of 500 organisms was attained in nine of 12 sampling years (Figure 30). Total and EPT taxa numbers are generally high at this site and there were more taxa in 2022 than any prior year (Figure 31). Taxa richness was also significantly higher in 2018-2022 (mean = 39 taxa) compared to all prior years (mean = 32 taxa, p = 0.0087). IBI scores consistently indicate slight to no impairment, and the 2022 score was at the top end of the range among all samples. PREDATOR scores reflect poor to fair condition, with the 2022 sample scoring lower than any since 2014 (Figure 32). Despite the lower O/E score, there were more sensitive taxa (4) in 2022 than in any prior year (Figure 33), and the number of sediment-sensitive taxa was consistent with the prior two years. Abundance of sediment-tolerant organisms was low enough to score in the top range of the ORDEQ IBI in nine of 12 sampling years including 2022, while relative abundance of tolerant organisms is consistently greater and increased in the last two years (Figure 34), though not significantly.

Community temperature optima were higher in 2020-2022 than in prior sampling years (Figure 35), though not significantly, and the 2022 sample (18.1°C) was at the upper end of the range for the year. Community fine sediment optima increased over time and was significantly higher in 2020-2022 (mean 8.2%) compared to all prior years (mean 7.0%; p = 0.0099; Figure 35). There are consistently fewer warm-associated than cool/cold-associated organisms and their abundance is low overall (Figure 36), and while there were only half as many cool/cold-associated organisms in 2022 compared to the prior year, their abundance was more than twice that of warm-associated organisms in the sample.

The 2022 macroinvertebrate community was most similar to the prior year (Bray-Curtis similarity index 0.786) and both were more similar to the 2005 community than to any other year (Figure 37). Community differences were largely driven by changes in dominant taxa, with more *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water that can also increase in abundance following habitat disturbance in 2013-2014; and more *Rhithrogena* (sediment-sensitive flatheaded mayfly found on the tops of stones in faster flows) and *Optioservus* 

(tolerant riffle beetle associated with clear waters and stable habitat in a variety of flows and temperatures) in 2016-2022 (Figure 38). Additional community variance was explained by *Attenella*, a spiny crawler mayfly that was more abundant in 2022 than any prior year; and *Zaitzevia*, a riffle beetle associated with faster flows and more stable habitat that was less abundant in 2014-2020. Differences in traits calculated as relative abundances also distinguished between later (2016-2022) and earlier sampling years (Figure 39), with more organisms associated with cool/cold flows and more scrapers and clingers in most recent sampling years.



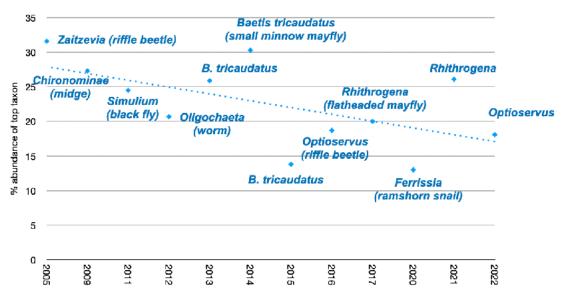
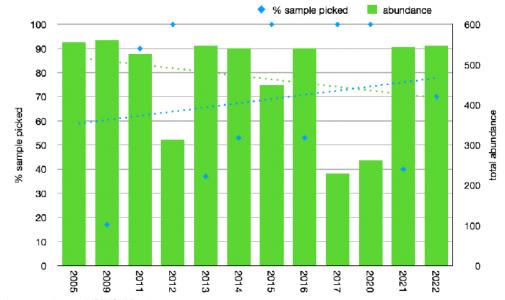


Figure 30. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC0090 in all sampling years. RT samples were taken 2in each year. Target sub-sampling number is 500.



CASM Environmental 2022 UDWC Whychus

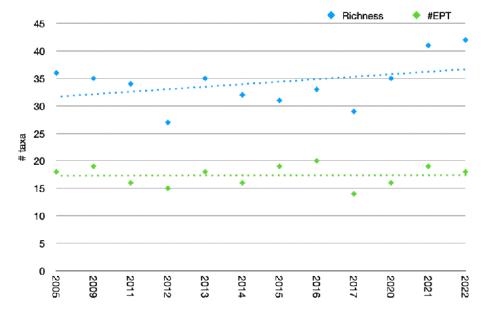


Figure 31. Sample richness and number of EPT taxa at WC00900 in all sampling years. RT samples were taken each year. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

Figure 32. PREDATOR O/E and ORDEQ IBI scores at WC0900 in all sampling years. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

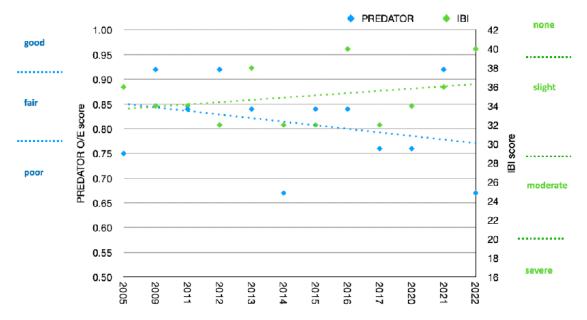


Figure 33. Numbers of sensitive and sediment-sensitive taxa at WC0900 in all sampling years. RT samples were taken all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.

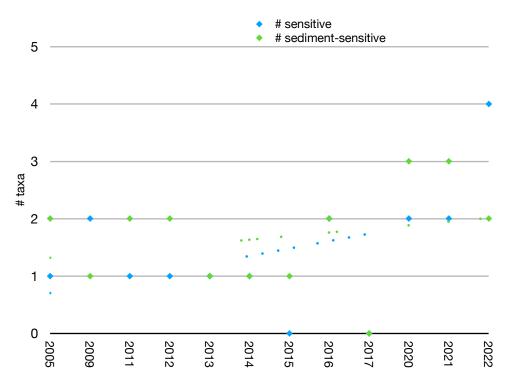


Figure 34. Relative abundance of tolerant and sediment-tolerant organisms at WC00980 in all sampling years. RT samples were taken in all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

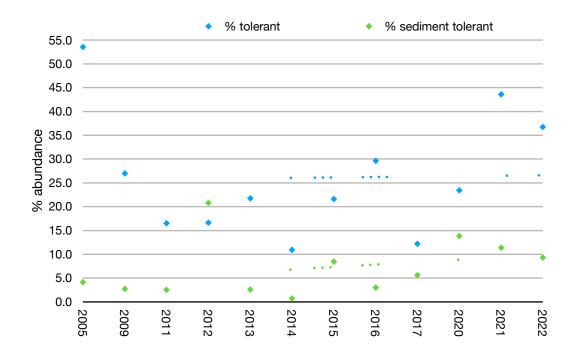


Figure 35. Temperature and fine sediment optima of the community (weighted means) at WC0900 in all sampling years. RT samples were taken in all years. Linear trendlines are shown.

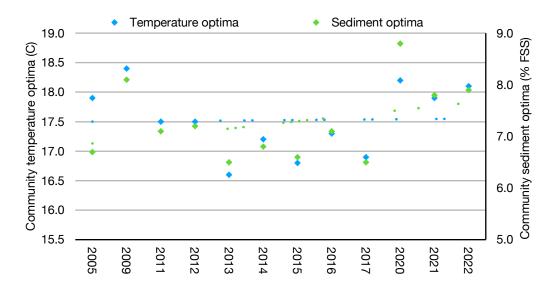


Figure 36. Temperature associations of the macroinvertebrate community at WC0090 in all sampling years. RT samples were taken in each year. Linear trendlines are shown.

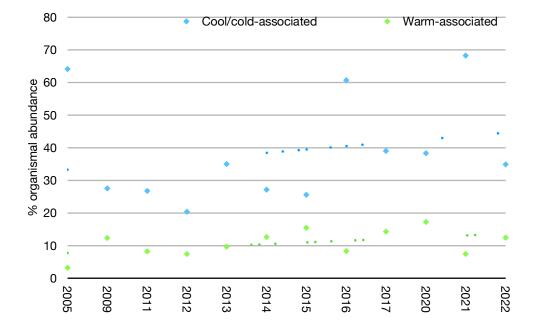


Figure 37. Cluster dendrogram of the WC0090 macroinvertebrate community in all sampling years. RT samples were taken in each year. The number at the end of each label indicates sampling year.

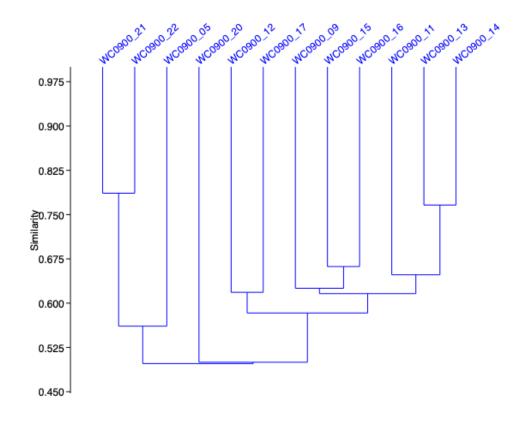
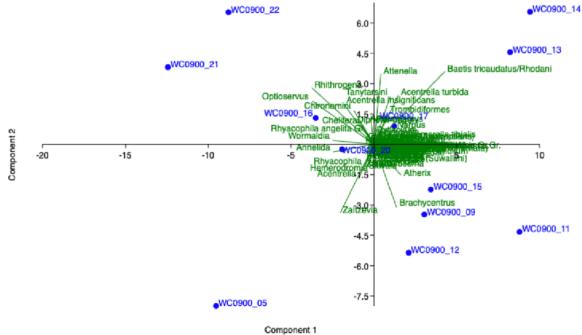


Figure 38. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0090 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. RT samples were taken each year. Axis 1 explains 36% of total sample variation; axis 2 explains an additional 15% of variation.



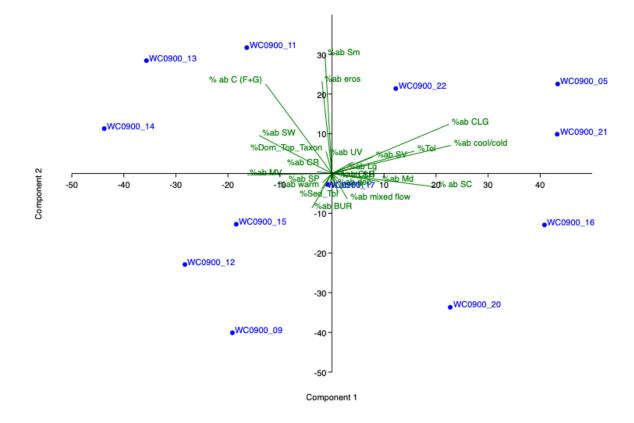


Figure 39. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC0900 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. RT samples were taken each year. Axis 1 explains 43% of total sample variation; axis 2 explains an additional 26% of variation.

## WC1025 (Rimrock Ranch)

RT samples were taken in the primary channel of Whychus Creek around RM 10.25 from 2011-2017; PM samples were taken 2019-2020. The reach was restored in 2021 as part of the Rimrock Phase II restoration project and no sampling was done. In 2022, PM and RT samples were taken in the primary channel (WC1025-1) and PM samples were taken in two new side channels (WC1025-2, WC1025-3). Substrate in the RT sample was a mixture of gravel, sand, silt, and bedrock (Figure 1). The WC1025-1 and WC1025-2 PM samples were taken primarily in sand/silt substrate and faster flows (Figure 2). The WC1025-3 PM sample was taken in roughly equal proportions of gravel, sand/silt, and wood and a variety of flows (Figure 2). More taxa new to the site (8 total) were taken here than in any other 2022 sample, with most found in side channels at abundances ranging from one to four individuals: *Clinocera*, a meniscus midge found in cooler flowing water (WC1025-2 SC); *Procloeon*, a small minnow mayfly associated with colder streams (WC1025-2 SC); *Dicosmoecus*, a Northern caddisfly associated with faster flows (WC1025-2 SC); *Dicosmoecus*, a Northern caddisfly associated with faster flows (WC1025-2 SC); *Peraleptophlebia*, a prong-gilled mayfly associated with sediments and detritus in fast cool flows (primary channel); and *Rhyacophila angelita*, a sediment-sensitive green rockworm caddisfly associated with clear flows (primary channel).

The communities in both primary channel samples and the WC1025-3 side channel were most similar to each other and to adjacent WC1100-2 and WC1150 samples (Figure 5). The WC1025-2 side channel community was an outlier and was more related to a side channel sample at a site restored in 2012 (WC1925-4). Despite recent restoration at WC1025, the community in all samples was well-balanced, with relative abundance of the top taxon ranging from 13-21% (Figure 40). Primary channel RT and WC1025-3 side channel samples were dominated by *Optioservus*, a tolerant riffle beetle associated with clear waters and stable habitat in a variety of flows and temperatures. The other samples were dominated by more tolerant taxa, including Naidinae, a tolerant, sediment-tolerant sludge worm found in softer sediments and slower flows, which dominated abundance in the primary channel PM sample; and *Tanytarsus*, a non-biting midge that tolerates a wide range of habitats and ecological conditions, which dominated the WC1025-2 side channel community. This site generally supported a more balanced community, scoring below or near the range for the highest scaled ORDEQ IBI score every year except 2017 (Figure 40).

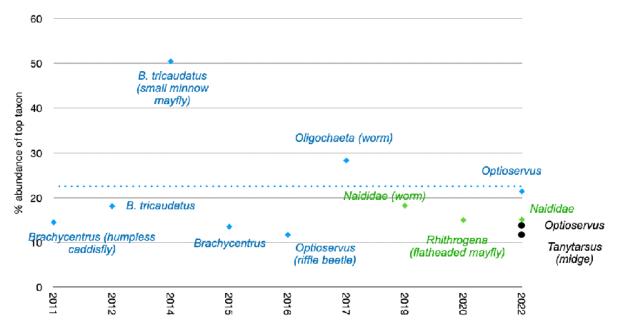
A trends analysis of primary channel samples revealed significant increase across time in total and Ephemeroptera taxa, community sediment optima, and abundance of tolerant and predator organisms (Appendix B). The target subsampling number of 500 organisms was attained in all samples except the 2017 RT sample (Figure 41). Total and EPT taxa numbers are generally high and both increased, although unidirectional trends were not significant. However, there were significantly more total taxa (mean = 55) in primary channel samples in 2019-2022 compared to 2011-2017 (mean = 32, p = 0.0001) and significantly more EPT taxa (mean 2019-2022 = 20 EPT, mean 2011-2017) = 16 EPT, p = 0.0255). Despite recent restoration, total and EPT richness in the primary and side channel samples was in the range of the most recent sampling years (Figure 42), and WC1025-2 had more mayfly taxa than any other 2022 sample. PREDATOR scores varied and reflected a variety of conditions (Figure 43), but the 2022 score (0.93; good) was the highest of any year at this site and at the top of the 2022 sample range. The IBI score indicated slight disturbance and was the highest since 2012 (Figure 43). There were more sensitive taxa in recent sampling years in primary channel samples (Figure 44), while the number of sediment-sensitive taxa, which increased in the two years prior to restoration, was lower (Figure 44). The WC1025-2 side channel community, which more closely resembled primary channel samples, had the most sensitive taxa (5) of any year at this site, although the WC1025-3 side channel sample had only two. Both side channels had one sediment-sensitive taxon, similar to the primary channel samples. Abundance of tolerant and sediment-tolerant organisms in the primary channel increased over time (Figure 45), though the increase was significant only for tolerant organisms. Primary and side channel samples in 2022 had similar abundance of tolerant organisms, but side channels had fewer sediment-tolerant organisms (3% abundance in WC1025-2, 7.4% abundance in WC1025-3, 16-21% in primary channel samples).

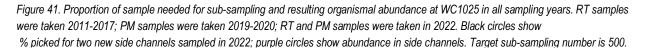
Community temperature and sediment optima increased over time, though the increase was significant only for sediment optima (Figure 46). Side channel community optima (temperature optima 17.8-17.9°F, sediment optima 9-10% FSS) were similar to the primary channel (temperature optima 17.3-17.9°F, sediment optima 8%). There is consistently low abundance of organisms associated with warmer water, and all 2022 samples had fewer warm-associated organisms than any prior year (<4% in primary channel samples, <3% in side channels; Figure 47). Cool/cold-associated organisms were 2-14 times more abundant than warm-associated organisms in the same

sampling year (Figure 47). Values for this metric in side channel samples (1.1-2.5% warm-associated, 40-43% cool/cold-associated) were similar to the primary channel (3.7-3.9% warm-associated, 32.8-42.1% cold-associated).

Macroinvertebrate community similarity was greater among recent (2019-2022) samples compared to earlier years (Figure 48), and all 2022 samples were more similar to each other than to 2019-2020 samples. Differences between recent and early sampling years were driven by *Baetis tricaudatus*, a small minnow mayfly associated with faster flows, clearer water, and recent habitat disturbance that was more abundant in early sampling years; and the midge tribe Chironomini and Annelida segmented worms, both of which are tolerant, burrowing taxa that were more abundant in recent years (Figure 49). Additional differences included a greater abundance in 2022 of *Ephemerella tibialis*, a spiny crawler mayfly associated with cooler water; and greater abundances in 2019-2020 of *Hydropsyche* (a tolerant netspinning caddisfly) and Tanytarsini (non-biting midge tribe that builds tubes in soft sediments). Differences in traits calculated as relative abundances also distinguished between most of the recent (2019-2022) vs. earlier sampling years, with recent years having more univoltine organisms and organisms that tolerate a variety of flow types (Figure 50).

Figure 40. Relative abundance of the numerically dominant taxon at WC1025 in all sampling years. RT samples (blue) were taken 2011-2017; PM samples (green) were taken 2019-2020; RT and PM samples were taken in 2022 after restoration. Black circles indicate two new side channels sampled in 2022. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.





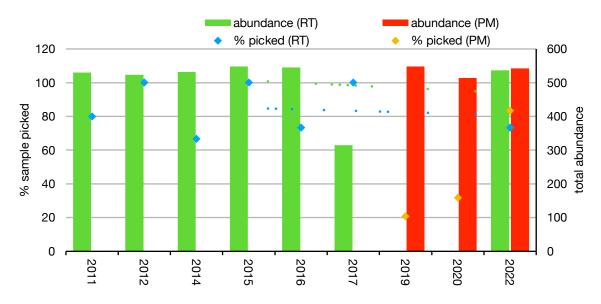
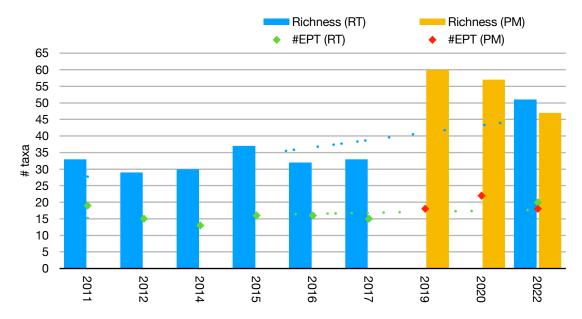


Figure 42. Sample richness and number of EPT taxa at WC1025 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2019-2020; RT and PM samples were taken in 2022 after restoration. Black circles indicate richness in two new side channels sampled in 2022; purple circles indicate side channel EPT taxa. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.



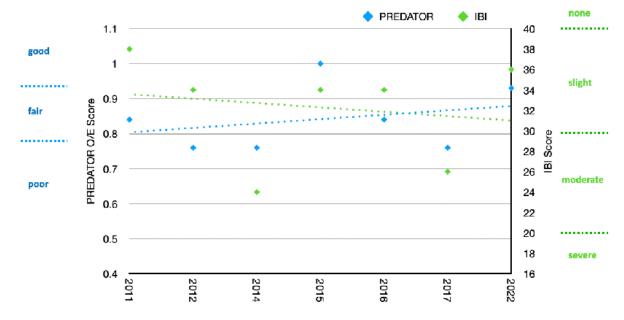


Figure 43. PREDATOR O/E and ORDEQ IBI scores at WC1025 in all years RT samples were taken. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

Figure 44. Numbers of sensitive and sediment-sensitive taxa at WC1025 in all sampling years. RT samples were taken 2011-2017; PM samples were taken in 2019-2020. RT and PM samples were taken in 2022. 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.

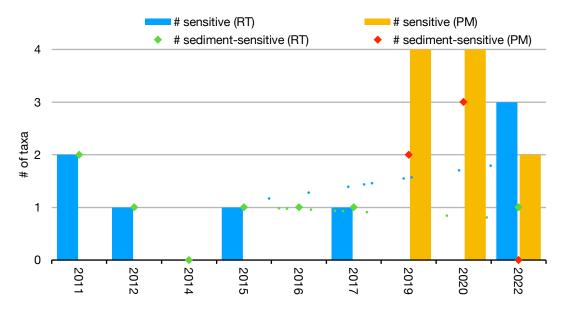


Figure 45. Relative abundance of tolerant and sediment-tolerant organisms at WC1025 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2019-2020; RT and PM samples were taken in 2022. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

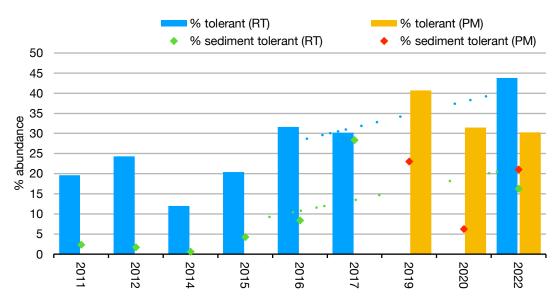
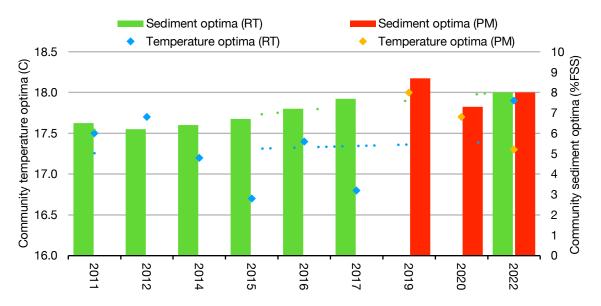


Figure 46. Temperature and fine sediment optima of the community (weighted means) at WC1025 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2019-2020; RT and PM samples were taken in 2022. Linear trendlines are shown.



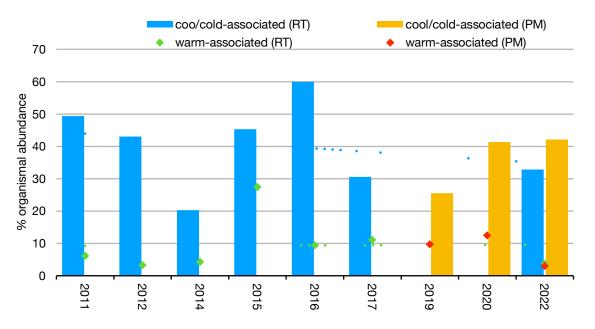


Figure 47. Temperature associations of the macroinvertebrate community at WC1025 in all sampling years. RT samples were taken 2011-2017; PM samples were taken 2019-2020; RT and PM samples were taken in 2022. Linear trendlines are shown.

Figure 48. Cluster dendrogram of the WC1025 macroinvertebrate community in all sampling years. RT samples (blue) were taken 2011-2017; PM samples (aqua) were taken in 2019-2020. RT and PM samples were taken in 2022. The number at the end of each label indicates sampling year.

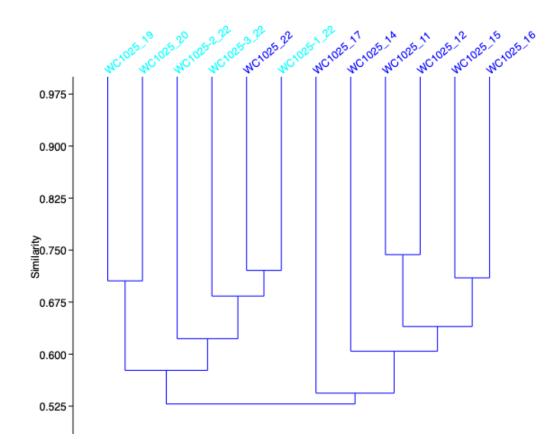
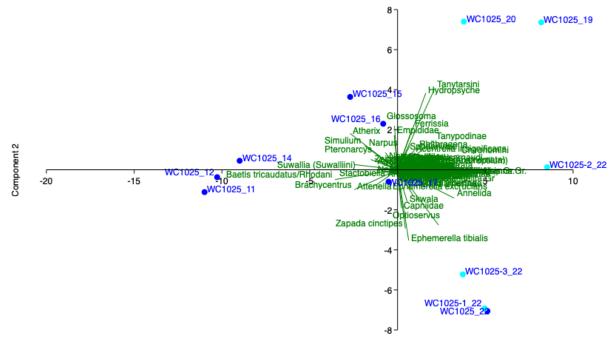
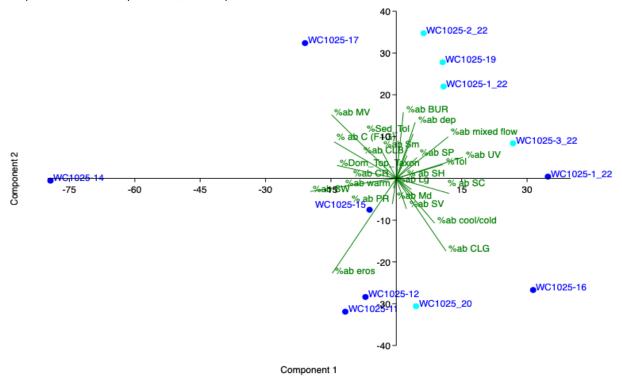


Figure 49. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1025 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 31% of total sample variation: axis 2 explains an additional 15% of variation.



Component 1

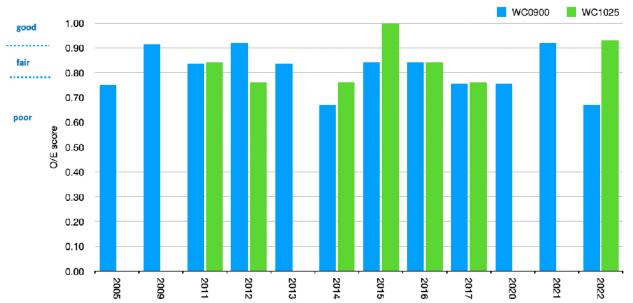
Figure 50. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1025 samples. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 36% of total sample variation; axis 2 explains an additional 25% of variation.



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### Comparison to adjacent unrestored reach

The reach immediately upstream of WC1025 is not suitable for comparison, as WC1100 was restored in 2016 and is now highly braided. However, the downstream WC0900 site has not been actively restored, and based on the fact that many metric values at WC0900 such as total, EPT, and sensitive taxa and IBI score in 2022 were similar to or greater than previous years, the reach does not seem to have been substantially impacted by restoration at WC1025. PREDATOR scores at WC1025 were similar to or greater than WC0900 in almost every year that both sites were sampled including 2022 (Figure 51), when the newly-restored reach scored higher (0.93 vs. 0.67 at WC0900). IBI scores varied more and were generally similar or lower at WC1025 (Figure 52). The 2022 scores were similar (40 at WC0900 vs. 36 at WC1025) but corresponded to different biological condition (no disturbance vs. slightly disturbed). Taxa richness at WC1025 was consistently similar to or greater than richness at WC0900 (Figure 53), including immediately post-restoration. There were generally fewer EPT at WC1025 than at WC0900 pre-restoration, but the magnitude of difference in each year was small and the numbers were virtually identical in 2022 (18 EPT at WC0900 vs. 19 at WC1025; Figure 53). Prior to restoration, there were more years in which cool/cold-associated organisms were more abundant at WC1025 than WC0900, and in 2022 abundances were similar (Figure 54). Warm-associated organisms were never very abundant at either site, but there were fewer at WC1025 than WC0900 in most sampling years, including 2022, and this metric did not increase post-restoration (Figure 54). Based on these comparisons and the differences in 2022 metric values at WC1025 pre- and post-restoration, restoration activities appeared to cause less disruption in the invertebrate community than was seen at other sites.



# Figure 51. PREDATOR O/E scores at WC1025 and WC0900. WC1025 was restored in 2021; WC0900 has not undergone active restoration.

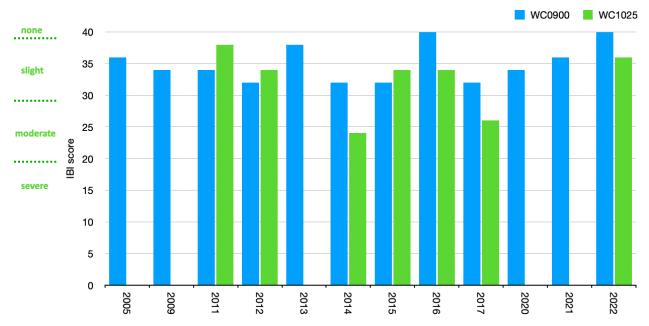
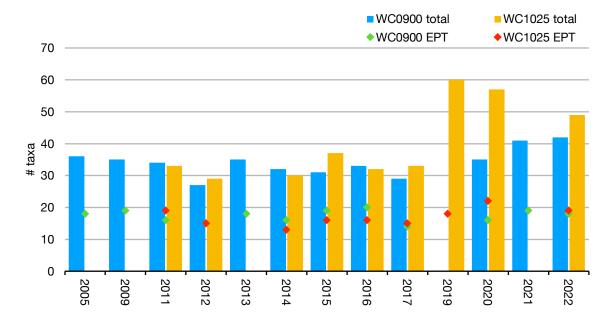
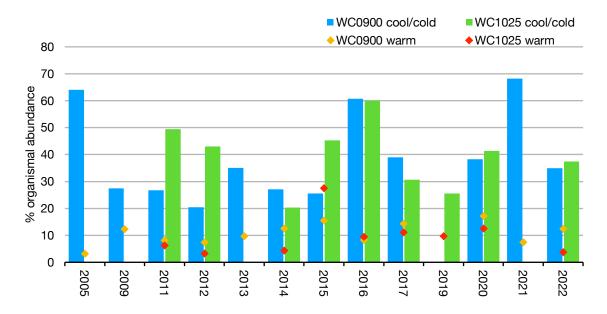
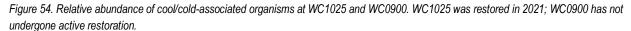


Figure 52. ORDEQ IBI scores at WC1025 and WC0900. WC1025 was restored in 2021; WC0900 has not undergone active restoration.

Figure 53. Total taxa and EPT richness at WC1025 and WC0900. WC1025 was restored in 2021; WC0900 has not undergone active restoration.







### WC1100 (Whychus Canyon)

Riffle-targeted samples were taken in the primary channel of Whychus Creek 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 reach of the primary channel (PC; 2017-2022), although extensive channel braiding post-restoration resulted in the original primary channel also having secondary channel characteristics. Multihabitat side channel (SC) samples were also taken (SC; 2017-2021). In 2022, only the primary channel was sampled; an RT sample was taken in primarily cobble/gravel substrate (Figure 1), and a PM sample was taken primarily in cobble and sand/silt (Figure 2) and in moving water (Figure 3).

The RT and PM sample communities were most similar to each other (Bray Curtis similarity index 0.801) and to samples from the WC1025 primary and side channels (Figure 5). The 2022 community in both samples consisted mainly of small, univoltine organisms that feed as collectors and move as clingers in faster flows. All taxa taken in 2022 were found in at least one previous year at this site. RT and PM samples were dominated at similar relative abundance (26.4% and 31.2%, respectively) by *Optioservus*, a tolerant riffle beetle associated with clear waters in a variety of flows and temperatures whose presence suggests stable habitat conditions. This was also the dominant taxon in both 2021 samples (Figure 55). This metric decreased overall since sampling began, though not significantly, and while abundance of the dominant taxon increased since 2020, the mean in 2021-2022 samples was not significantly higher than the 2014-2020 sample mean. The primary channel was dominated by taxa associated with faster flows in all years except the two immediately after restoration.

A trends analysis of primary channel samples revealed significant increases across time in IBI score, number of Ephemeroptera, Trichoptera, and sediment-sensitive taxa, and abundance of tolerant, large, semivoltine, clinger, and cool/cold-associated organisms; and significant decrease in collector abundance (Appendix B). The target sub-sampling number of 500 organisms was attained in all RT samples except immediately after restoration (Figure 56) and in three of the five years that PM samples were taken, including 2021 and 2022. The community immediately following restoration (2017) was severely disturbed, with all metric values lower than pre- or later post-restoration years, but recovery and improvement was evident from 2018 onwards. Richness was significantly greater in RT samples taken 2018-2022 (mean = 49 taxa) compared to pre-restoration (mean 2014-2015 = 29.5 taxa; p = 0.0006; Figure 57), as was the number of EPT taxa (mean 2014-2015 = 13 EPT; mean 2018-2022 = 20 EPT; p = 0.0003). Total and EPT taxa numbers in PM samples and RT samples were similar. IBI and PREDATOR scores show a pattern of post-restoration = 41.2; p = 0.0047). Both were slightly lower in 2022 compared to the previous year (Figure 58); the O/E score still corresponded to good condition while the IBI score reflected slight disturbance as opposed to no disturbance in 2018-2021.

Prior to restoration no sensitive and few or no sediment-sensitive taxa were taken (Figure 59), but since 2018 numbers of both were at or near the range assigned the highest scaled scores in the ORDEQ IBI. Tolerant and sediment-tolerant organisms were most abundant immediately after restoration; both were lower in 2018 but increased steadily since then (Figure 60), although sediment-tolerant organismal abundance was low enough to receive the highest scaled score in the ORDEQ IBI in all but one year from 2018-2022. Community temperature and sediment optima were similar in RT and PM samples (Figure 61); both increased post-restoration (2018-2022), though not significantly, and the 2022 samples had the highest temperature and sediment optima of any year. However, cool/cold-associated organisms consistently occur at much higher abundance than warm-associated organisms since restoration was done (Figure 62).

Both 2022 sample communities were most similar to samples from more recent years (2019-2021). Pre-restoration communities were less similar overall to post-restoration (Figure 63) and the disturbed community in 2017 was an outlier from all other years. Community differences were largely driven by *Optioservus*, a tolerant riffle beetle associated with clear waters and stable habitat in a variety of flows and temperatures, which dominated samples in 2021-2022; and *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water that can increase in abundance following habitat disturbance and was more abundant for a few years post-restoration (2019-2020; Figure 64). Differences in traits calculated as relative abundances distinguished between pre- and post-restoration communities, with more clingers (generally found in faster flows) and more organisms associated with cool/cold temperatures from 2020-2022 (Figure 65).

Figure 55. Relative abundance of the numerically dominant taxon at WC1100 in all sampling years. RT samples (blue) were taken 2014-2015; RT and PM (green) samples were taken in the primary channel 2017-2022. Side channels are not shown. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

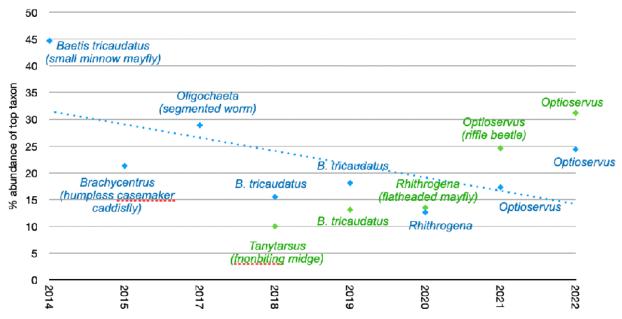


Figure 56. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1100 in all sampling years. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022. Only primary channel data are shown. Target sub-sampling number is 500.

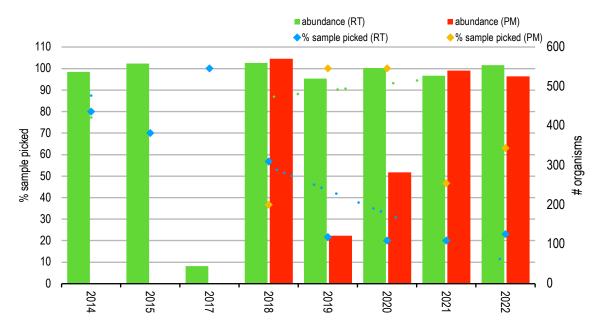


Figure 57. Sample richness and number of EPT taxa at WC1100 in all sampling years. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

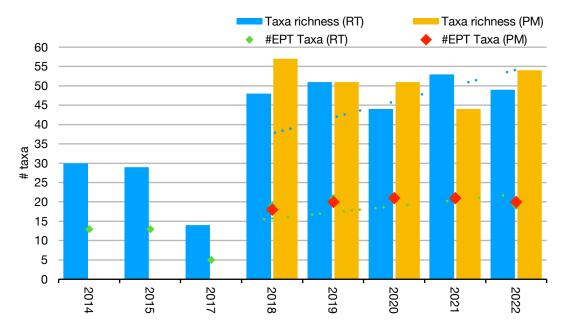


Figure 58. PREDATOR O/E and ORDEQ IBI scores at WC1100. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

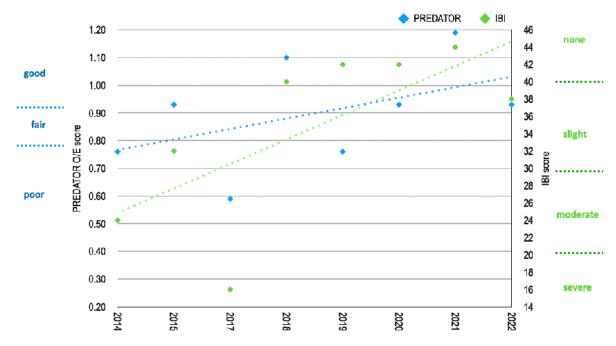


Figure 59. Numbers of sensitive and sediment-sensitive taxa at WC1100 in all sampling years. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and ≥2 sediment-sensitive taxa.

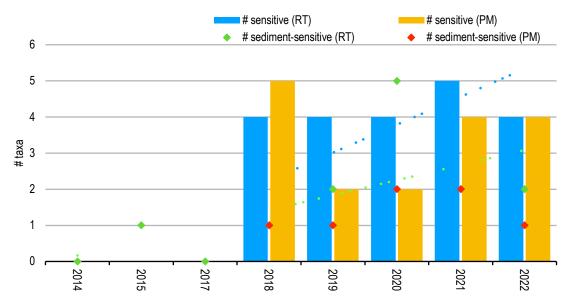


Figure 60. Relative abundance of tolerant and sediment-tolerant organisms at WC1100 in all sampling years. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

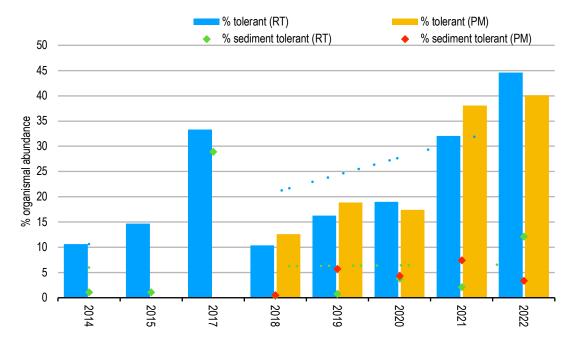


Figure 61. Temperature and fine sediment optima of the community (weighted means) at WC1100. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. Linear trendlines are shown.

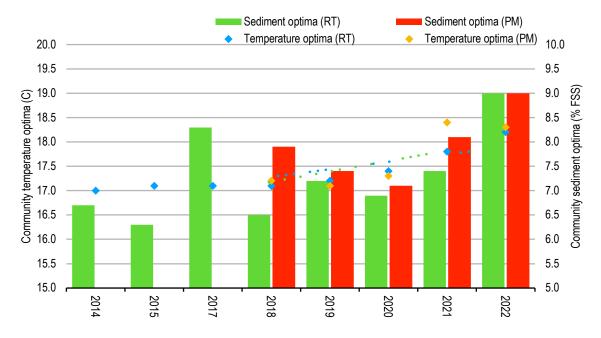


Figure 62. Temperature associations of the macroinvertebrate community at WC1100 in all sampling years. RT samples were taken 2014-2015; RT and PM samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. Linear trendlines are shown.

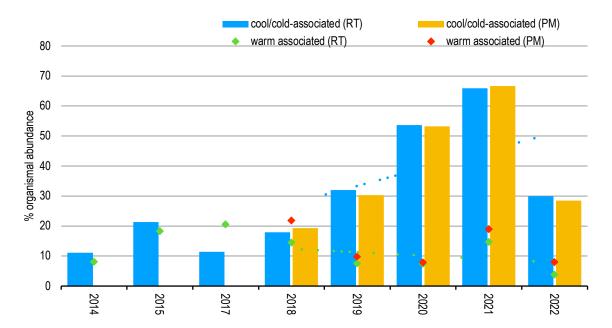


Figure 63. Cluster dendrogram of the WC1100 macroinvertebrate community in all sampling years. RT samples (blue) were taken 2014-2015; RT and PM (aqua) samples were taken in the primary channel 2017-2022 and PM samples were taken in side channels 2017-2021. Only primary channel data are shown. The number at the end of each label indicates sampling year.

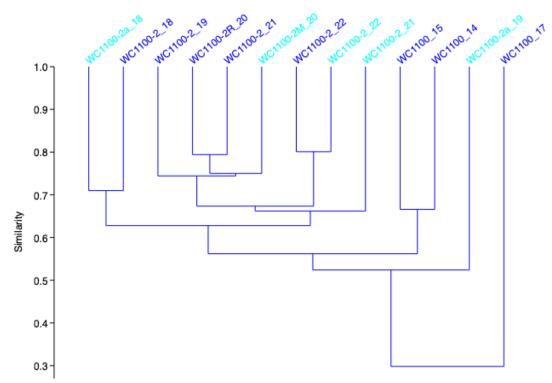
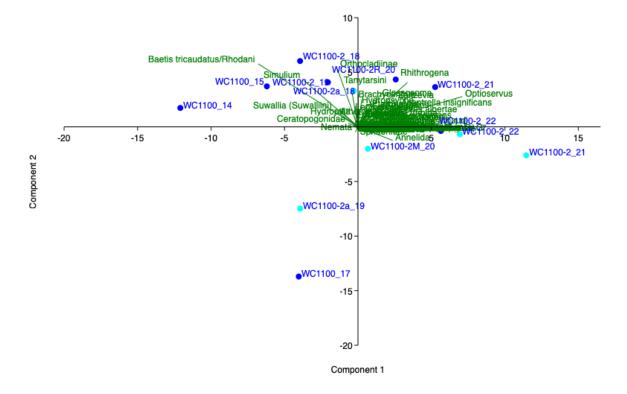


Figure 64. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1100 samples. Only samples taken in the primary channel are shown. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 29% of total sample variation; axis 2 explains an additional 22% of variation.



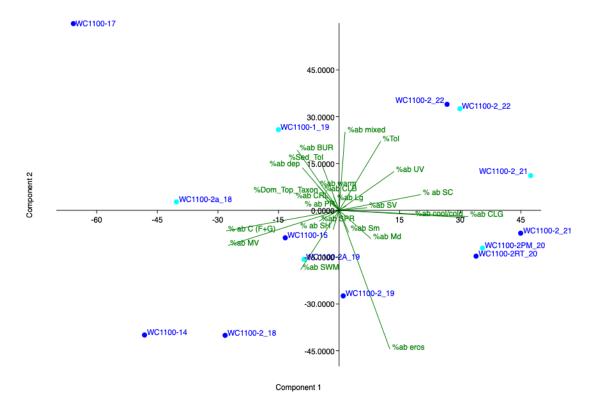


Figure 65. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1100 samples. Only samples taken in the primary channel are shown. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 44% of total sample variation; axis 2 explains an additional 28% 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. O/E scores were higher at WC1100 except in 2017, when the WC1100 community was impacted by restoration-related disturbance (Figure 66). In contrast, although ORDEQ IBI scores were higher at WC1150 between 2014 and 2017, increased IBI scores at WC1100 post-restoration were similar to WC1150 (Figure 67). There were more total taxa in WC1150 samples compared to WC1100 prior to and immediately following restoration (Figure 68). However, richness in the restored reach was almost twice as high postrestoration and similar to, though generally lower than, richness at WC1150 except in 2022, when WC1100 RT and PM samples had substantially more total taxa than the WC1150 RT sample. There were consistently more EPT taxa at WC1150 than WC1100 through 2017 but as recovery continued, EPT richness at WC1100 was similar to or slightly exceeded WC1150 (Figure 68). Cool/cold-associated organisms were more abundant at WC1150 in 2014-2018 (Figure 69), but the difference was not quite significant (WC1100 mean 15.9%; WC1150 mean 34.5%; p =0.0577). However, abundances were similar in 2019 and significantly greater at WC1100 (mean 59.7%) than at WC1150 (mean 25.2%; p = 0.0092) in 2020-2021, although numbers were again similar among all samples in 2022. Relative abundance of warm-associated organisms was generally low in both reaches, but since restoration there were generally fewer at WC1100, especially in RT samples (Figure 69), and both reaches had low values in 2022.

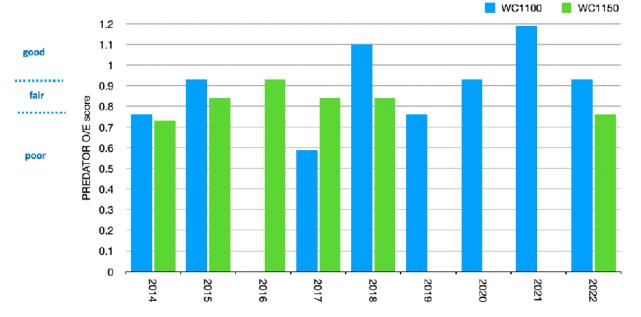
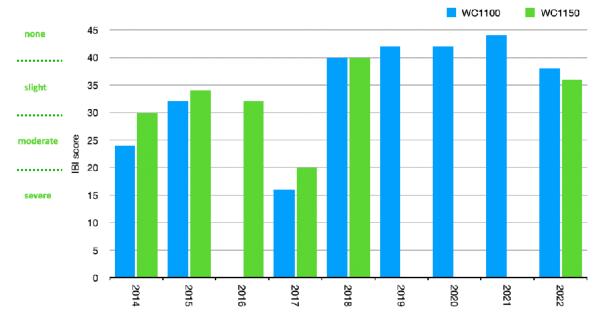


Figure 66. PREDATOR O/E scores at WC1100 and WC1150. WC1100 was restored in 2016; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.

Figure 67. ORDEQ IBI scores at WC1025 and WC0900. WC1100 was restored in 2016 and was not sampled that year; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.



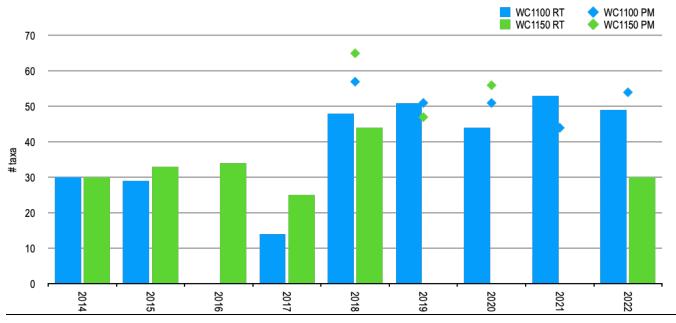
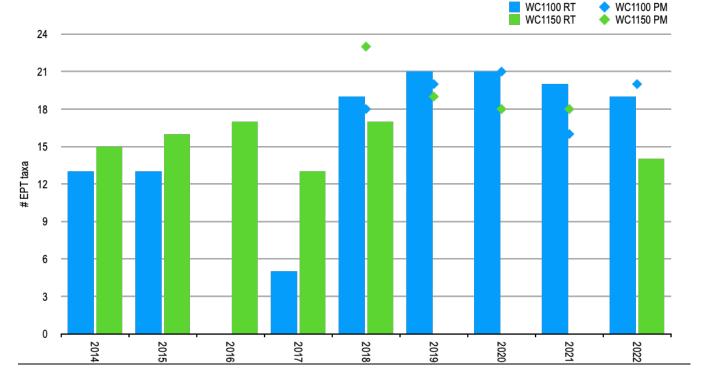


Figure 68A. Total taxa at WC1100 and W1150. WC1100 was restored in 2016 and was not sampled in that year; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.

Figure 68B. EPT richness at WC1100 and W1150. WC1100 was restored in 2016 and was not sampled in that year; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.



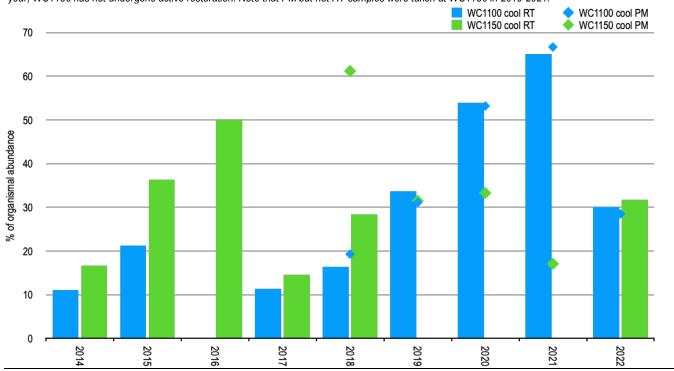
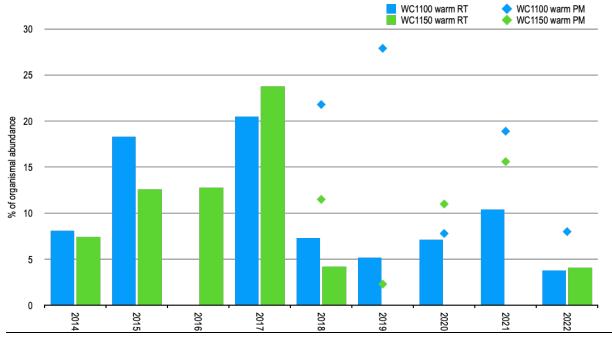


Figure 69A. Relative abundance of cool/cold-associated organisms at WC1100 and W1150. WC1100 was restored in 2016 and was not sampled that year; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.

Figure 69B. Relative abundance of warm-associated organisms at WC1100 and W1150. WC1100 was restored in 2016 and was not sampled that year; WC1150 has not undergone active restoration. Note that PM but not RT samples were taken at WC1150 in 2019-2021.



### WC1150 (Whychus Canyon)

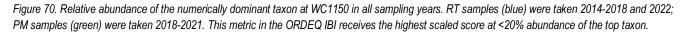
Sampling began at WC1150 in 2014 to provide an upstream reference for WC1100, as no active restoration was done here. RT samples were taken in 2014-2017; in 2018, PM and RT samples were taken in the same reach; from 2019-2021, only PM samples were collected; and in 2022, an RT sample was collected. The 2022 sample was taken primarily in cobble substrate (Figure 1). The community was most similar to sites further downstream (WC0600-WC1100-2; Figure 5), and two taxa were new for the site: *Drunella flavilinea*, a sensitive, sediment-sensitive spiny crawler mayfly associated with cool, swift mountain streams (1 individual); and *Epeorus longimanus*, a flatheaded mayfly found on rocks in cooler streams (1 individual). The community consisted mainly of small univoltine organisms that feed as collectors and move as clingers in faster, cooler flows. The sample community was dominated at relatively low abundance (19.1%) by *Optioservus*, a tolerant riffle beetle associated with clear waters in a variety of flows and temperatures whose presence suggests stable habitat conditions. This reach was dominated in most years by taxa associated with cool flowing water (Figure 70), including *Glossosoma* (saddlecase caddisfly), *Ampumixis* (riffle beetle), *Rhithrogena* (flatheaded mayfly), and *Nostoccoladius* (nonbiting midge that mines in *Nostoc* algae).

A trends analysis showed significant increases across time in Ephemeroptera, Plecoptera, and sediment-sensitive taxa and in relative abundance of tolerant, large, and clinger organisms; and significant decreases in predator, collector, multivoltine, crawler, and swimmer organisms (Appendix B). The target sub-sampling number of 500 organisms was attained in all PM samples and in four of the six RT samples; this did not include the 2022 RT sample, which contained only 267 organisms (Figure 71). Total and EPT taxa richness was greater overall in 2018-2022 compared to 2014-2017, though the difference was significant only for total taxa (mean 2014-2017 = 30.5 taxa, mean 2018-2022 = 46.3, p = 0.0251). However, both were lower in 2022 compared to the prior four sampling years (Figure 72). IBI scores for RT samples generally reflected better biological condition (slight to no disturbance) with the exception of 2017 (moderate disturbance), while PREDATOR O/E scores ranged from poor to good (Figure 73). In 2022, the IBI score corresponded to slight disturbance while the O/E score was in the upper end of the poor range.

There were significantly more sensitive taxa in 2018-2022 (mean = 4 taxa) compared to 2014-2017 (mean = 1 taxon; p = 0.0012; Figure 74) and significantly more sediment-sensitive taxa (mean 2014-2017 = 1 taxon; mean 2018-2022 = 3 taxa; p = 0.0007). Relative abundance of tolerant and sediment-tolerant organisms changed similarly in that same span, with fewer in 2018-2021; both were higher in 2022 (Figure 75), but sediment-tolerant organisms have never accounted for more than 10% of total sample abundance except in 2017. Community temperature and sediment optima increased overall since sampling began (Figure 76) but not significantly. Abundance of organisms associated with cooler temperatures greatly exceeded those associated with warmer water in every sampling year except 2017 and abundance of warm-associated organisms was consistently low ( $\leq$ 15%; Figure 77).

The 2022 community was most similar to the 2021 PM sample (Bray-Curtis similarity index = 0.62; Figure 78), and the community in the earliest sampling years (2014-2017) differed more from recent years. Community differences were largely driven by *Baetis tricaudatus* (small minnow mayfly associated with faster flows and clearer water that can increase in abundance following habitat disturbance), *Simulium* (black fly associated with flowing water and

habitat disturbance), and Annelida (tolerant segmented worms), which were all more abundant in early sampling years; and *Rhithrogena* (sediment-sensitive flatheaded mayfly found on the tops of stones in faster flows), which was more abundant in later years (Figure 79). Differences in traits calculated as relative abundances also distinguished between recent (2019-2022) and early sampling years (Figure 80), with more clingers and scrapers and fewer multivoltine organisms in recent years. Taxonomic and trait differences suggest a shift towards faster flows, increased clear cobble (i.e., less sediment), and greater habitat stability.



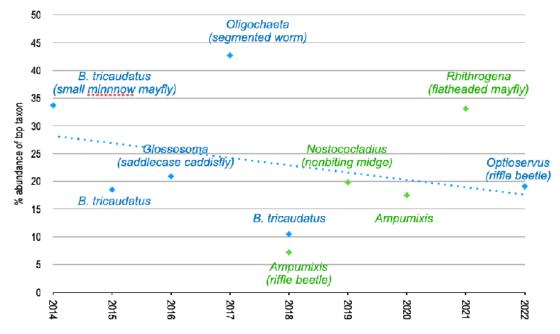
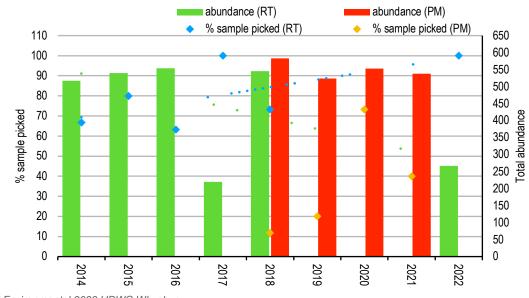


Figure 71. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1150 in all sampling years. RT samples were taken 2014-2018 and in 2022: PM samples were taken 2018-2021. Target sub-sampling number is 500.



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Figure 72. Sample richness and number of EPT taxa at WC1150 in all sampling years. RT samples were taken 2014-2018 and in 2022; PM samples were taken 2018-2021. In the year where both sample types were taken (2018), values were averaged. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

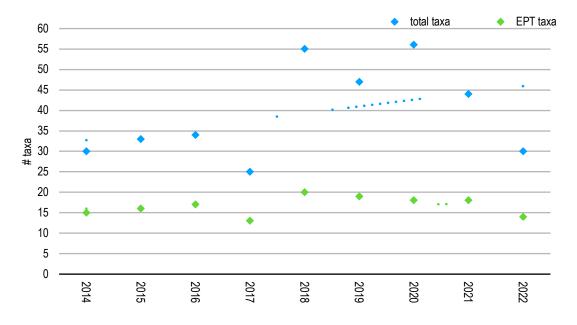


Figure 73. PREDATOR O/E and ORDEQ IBI scores at WC1150. Riffle samples were taken 2014-2018 and in 2022; PM samples were taken in the intervening years. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

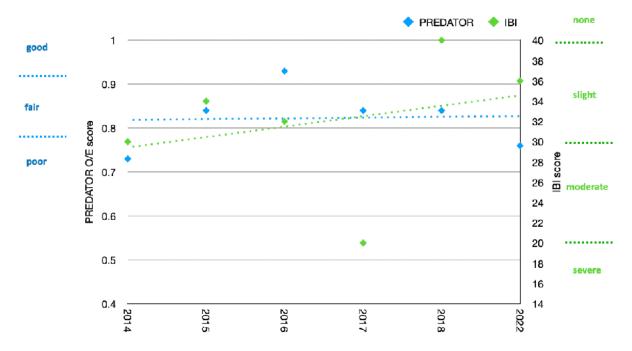


Figure 74. Numbers of sensitive and sediment-sensitive taxa at WC1150. RT samples were taken 2014-2018 and 2022; PM samples were taken 2018-2021. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and  $\geq$ 2 sediment-sensitive taxa.

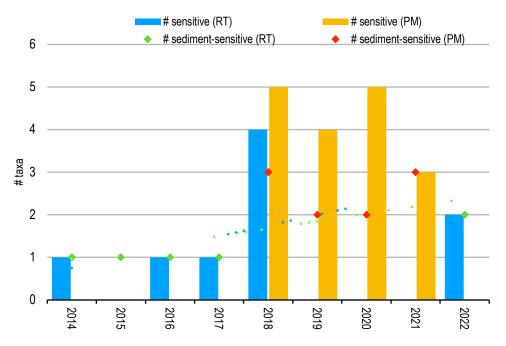
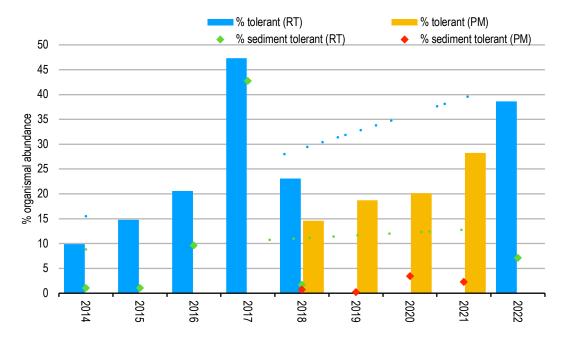


Figure 75. Relative abundance of tolerant and sediment-tolerant organisms at WC1150 in all sampling years. RT samples were taken 2014-2018 and 2022; PM samples were taken 2018-2021. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.



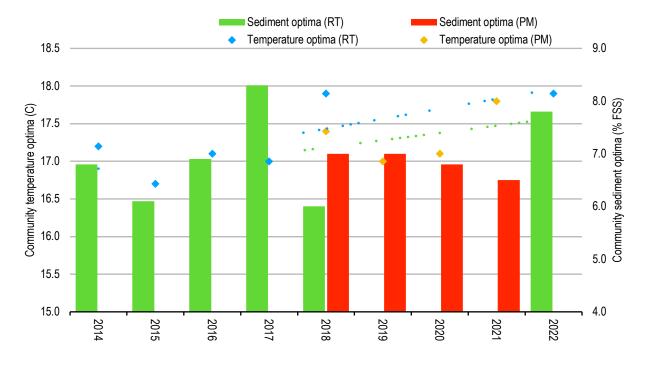


Figure 76. Temperature and fine sediment optima of the community (weighted means) at WC1150 in all sampling years. RT samples were taken 2014-2018 and 2022; PM samples were taken 2018-2021. Linear trendlines are shown.

Figure 77. Temperature associations of the macroinvertebrate community at WC1150 in all sampling years. RT samples were taken 2014-2018 and 2022: PM samples were taken 2018-2021. Linear trendlines are shown.

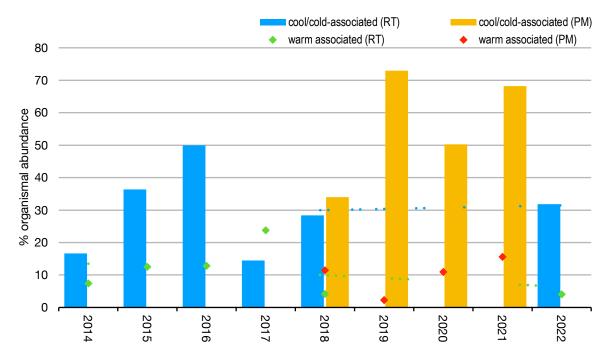


Figure 78. Cluster dendrogram of the WC1150 community in all sampling years. RT samples (blue) were taken 2014-2018 and 2022; PM samples (aqua) were taken 2018-2021. The number at the end of each label indicates sampling year.

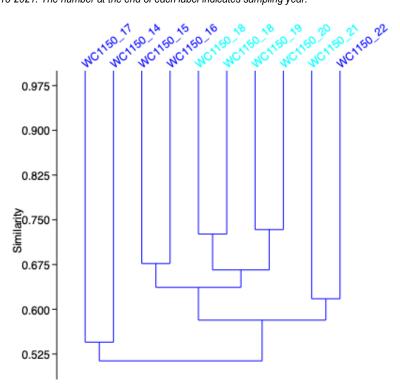


Figure 79. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1150 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 26% of total sample variation; axis 2 explains an additional 19% of variation.

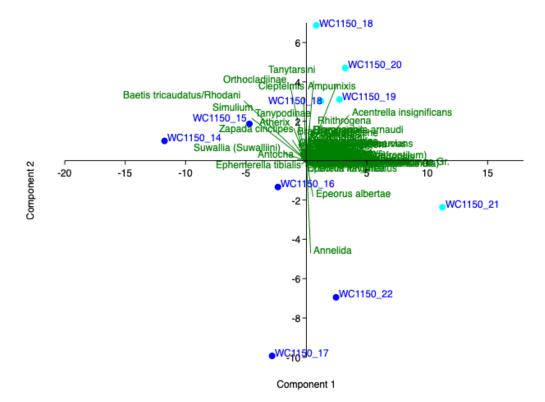
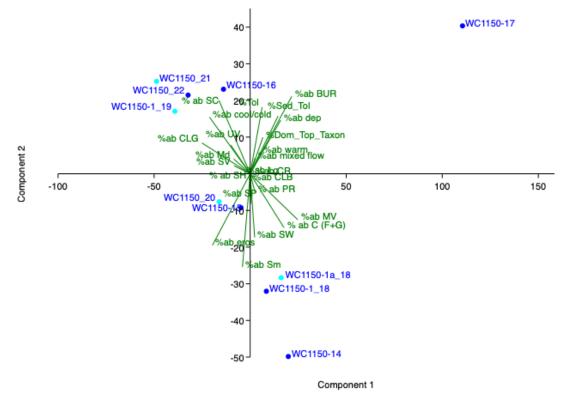


Figure 80. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1150 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 55% of total sample variation; axis 2 explains an additional 24% of variation.



# WC1825 (Camp Polk)

The sample reach at WC1825 is one of five directly affected by restoration at Camp Polk completed in 2012; WC1825 was directly downstream of the project reach and it is hypothesized that the macroinvertebrate community would change in conjunction with changes in the project reach. RT samples were taken in 2009-2017 and 2022. The substrate in the 2022 sample was primarily cobble/gravel (Figure 1), and the community was similar to that at nearby upstream reaches (WC1850-WC1950; Figure 5). All taxa in 2022 were found in at least one previous year at this site. The community was dominated by tolerant Naidinae sludge worms at high relative abundance (52.5%; Figure 81), and consisted mainly of small, univoltine organisms that feed as collectors and move as burrowers in slower flows. This is the most unbalanced community among all 2022 samples and the first year that the community was this unbalanced since 2013, when most of the sample consisted of *Simulium* black flies that were likely early post-restoration colonizers.

A trends analysis revealed significant decreases across time in IBI score and abundance of swimmer organisms. The target sub-sampling number of 500 organisms was attained in seven of nine sampling years, including 2022 (Figure 82). Sample richness increased over time (Figure 83) and the 2022 sample had more taxa than any prior year, but pre- and post-restoration means did not differ significantly and the number of EPT taxa changed little over time (Figure 83). PREDATOR O/E scores were higher in recent years (Figure 84), reflecting fair to good conditions in most

years from 2015-2022, but there is no significant difference in pre- and post-restoration scores. However, IBI scores decreased significantly overall (Figure 84), with the lowest scores in 2015 and 2022 (20 and 24 respectively). Samples consistently have more sensitive than sediment-sensitive taxa (Figure 85) and numbers of both stabilized in the last three sampling years. Abundance of tolerant and sediment-tolerant organisms was significantly greater in 2015-2022 compared to all earlier years (Figure 86), and both were more abundant in 2022 than any prior year. Community sediment optima changed little across time but community temperature optima increased every year since 2014 and was higher in 2022 than any prior year (Figure 87), though the increase was not significant. There were fewer cool/cold-associated organisms in 2016-2022, though not significantly so, but abundance of warm-associated organisms was very low in all years (Figure 88). This site also had 2-25 times more non-insect organisms (54% abundance) than any other 2022 sample due largely to the abundance of naidid worms, suggesting recent habitat disturbance.

The 2022 community was most similar to the 2017 sample (Bray-Curtis similarity index = 0.53) and both differed more from all other sampling years (Figure 89). Community differences were largely driven by Annelida, tolerant segmented worms that dominated the 2022 sample; *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water that was abundant every year except 2017 and 2022; and *Simulium*, a black fly associated with flowing water that was more abundant prior to 2017 (Figure 90). Differences in traits calculated as relative abundances also distinguished the two most recent sampling years (Figure 91), with fewer collectors and fewer organisms associated with flows in 2017, and more organisms that are tolerant, sediment-tolerant, and associated with slow flows in 2022.

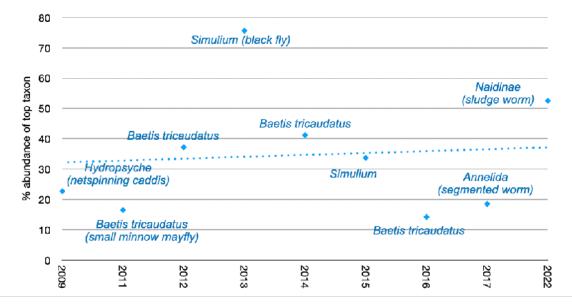


Figure 81. Relative abundance of the numerically dominant taxon at WC1825 in all sampling years. RT samples were taken in all years. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

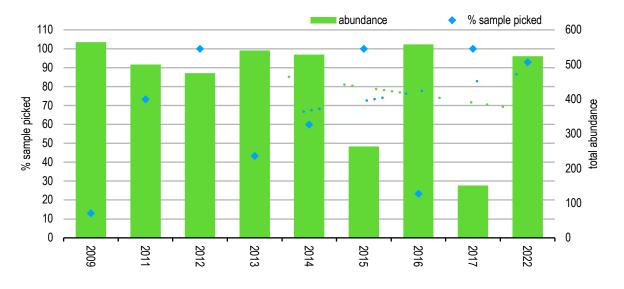
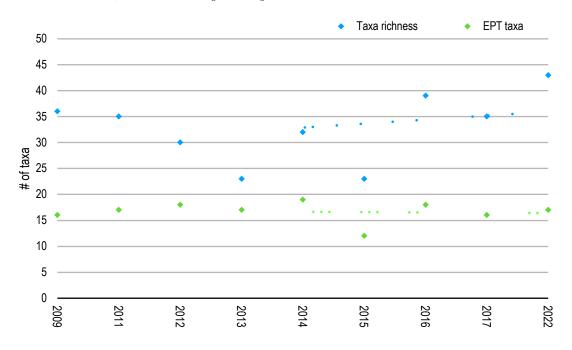


Figure 82. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1825 in all sampling years. Riffle samples were taken in all years. Target sub-sampling number is 500.

Figure 83. Sample richness and number of EPT taxa at WC1825 in all sampling years. RT samples were taken in all years. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.



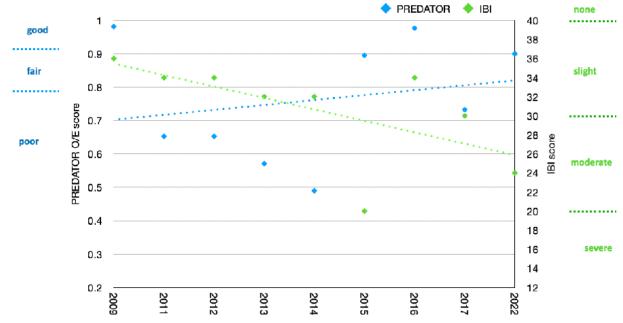
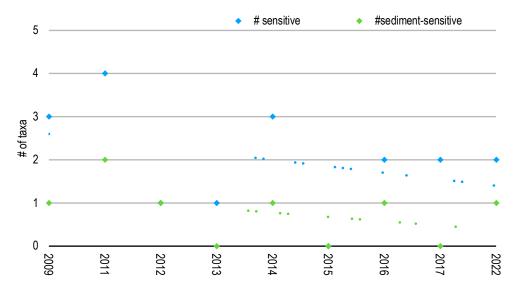


Figure 84. PREDATOR O/E and ORDEQ IBI scores at WC1825. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

Figure 85. Numbers of sensitive and sediment-sensitive taxa at WC1825. RT samples were taken in all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and >2 sediment-sensitive taxa.



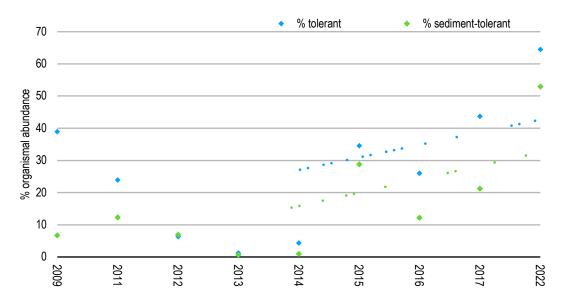
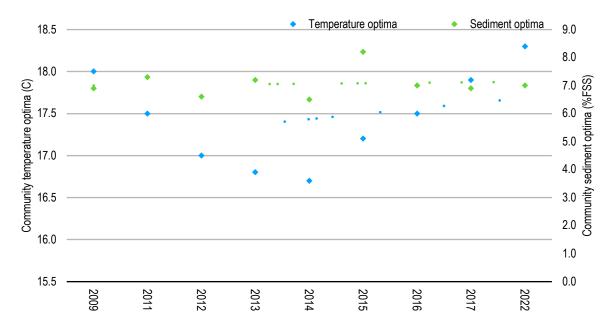
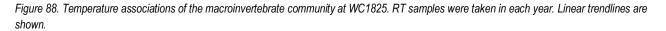


Figure 86. Relative abundance of tolerant and sediment-tolerant organisms at WC1825. RT samples were taken in all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.

Figure 87. Temperature and fine sediment optima of the community (weighted means) at WC1825. RT samples were taken in all years. Linear trendlines are shown.





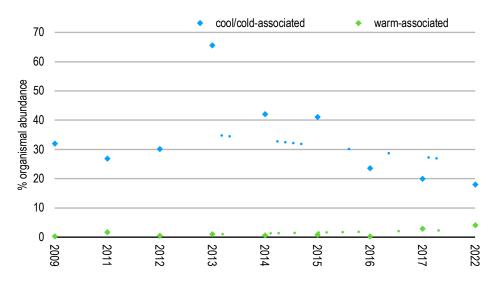


Figure 89. Cluster dendrogram of the WC1825 community in all sampling years. RT samples were taken in each year. The number at the end of each label indicates sampling year.

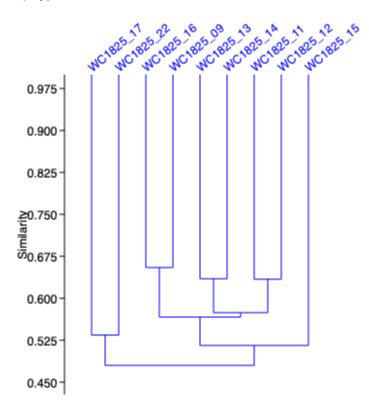
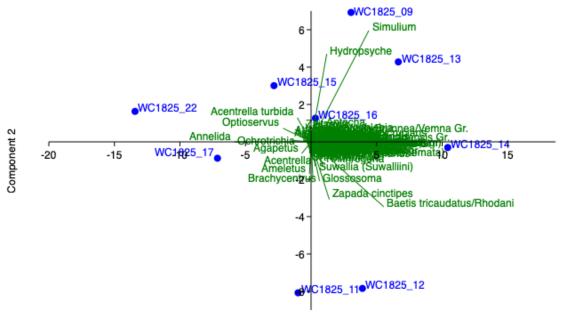
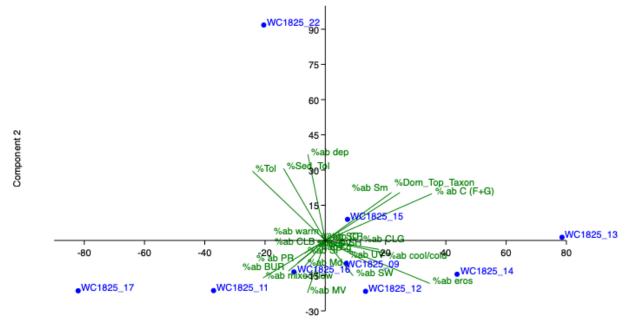


Figure 90. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1825 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 36% of total sample variation; axis 2 explains an additional 18% of variation.



Component 1

Figure 91. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1825 sampling years. 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 46% of total sample variation; axis 2 explains an additional 28% of variation.



Component 1

#### WC1850 (Camp Polk)

The sample reach at WC1850 was within the restoration project at Camp Polk that was completed in in 2012. RT samples were taken in 2009-2017 and 2022. The 2022 sample was taken in primarily cobble/gravel substrate (Figure 1), and the community was most similar to those at nearby upstream reaches (WC1900-WC1950; Figure 5). One taxon taken in 2022 was not found in any prior year in this reach: *Drunella flavilinea* (1 individual), a sensitive, sediment-sensitive spiny crawler mayfly associated with cool, swift mountain streams that was also new to the WC1150 site in 2022. The community was dominated by *Simulium* black flies, and consisted mainly of small, univoltine organisms that feed as collectors and move as clingers in faster flows. Abundance of the top taxon varied annually with no pattern (Figure 92), with dominance by taxa that are either tolerant (Annelida) or associated with flowing water and recent disturbance (*Baetis tricaudatus, Simulium*).

A trends analysis revealed significant increases across time in number of total, Trichoptera, and sediment-sensitive taxa (Appendix B). The target sub-sampling number of 500 organisms was attained in every sampling year (Figure 93). The number of total and EPT taxa increased over time (Figure 94), though pre-restoration (2009-2011) and postrestoration (2012-2022) means were not significantly different. PREDATOR and IBI scores increased overall (Figure 95) but not significantly, and there was no significant difference between pre- and post-restoration means. Both model scores were lower in 2022 (IBI = slight disturbance; O/E = poor) compared to the most recent sampling year (2017; IBI = no disturbance, O/E = fair). Equal numbers of sensitive and sediment-sensitive taxa were taken in 2015-2022 (Figure 96); fewer sediment-sensitive taxa were taken overall since 2012, with none in samples taken soon after restoration (2013-2014), but the samples scored in the highest range of the ORDEQ IBI for this metric in 2015-2022. Abundance of tolerant and sediment-tolerant organisms decreased over time, though not significantly, and both were low enough to receive the highest scaled score in the IBI in 2022 (Figure 97). Community temperature optima varied annually and were higher in the last three sampling years (17.2 - 17.4°F; Figure 98), but the increase was not significant; community sediment optima showed much less variation. Despite higher temperature optima in recent years, relative abundance of organisms associated with cool temperatures increased over time (Figure 99); the trend was not significant across all sampling years, but there were significantly more cool/cold-associated organisms in 2013-2022 (mean = 58.2%) than in 2009-2012 (mean = 26.3%, p = 0.036). This reach consistently has very low abundance (<2%) of organisms associated with warm water.

The 2022 community was most similar to the 2017 sample (Bray-Curtis similarity index = 0.69). Overall community similarity among all years was fairly high, with samples clustering in nearest-year pairs (Figure 100). Community differences were driven by *Simulium*, a black fly associated with flowing water that was much less abundant 2009-2012; *Baetis tricaudatus*, a small minnow mayfly associated with faster flows and clearer water that was more abundant in earlier years; and Annelida, tolerant segmented worms that were unusually abundant in 2011 (Figure 101). Differences in traits calculated as relative abundances also distinguished between recent and early sampling years, with greater abundance of univoltine, cool/cold-associated, and clinger organisms in post-restoration samples (2013-2022), and fewer swimmers from 2015-2022 (Figure 102).

Figure 92. Relative abundance of the numerically dominant taxon at WC1850 in all sampling years. RT samples were taken in all years. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

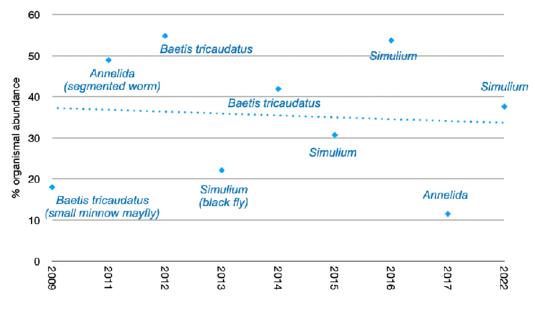
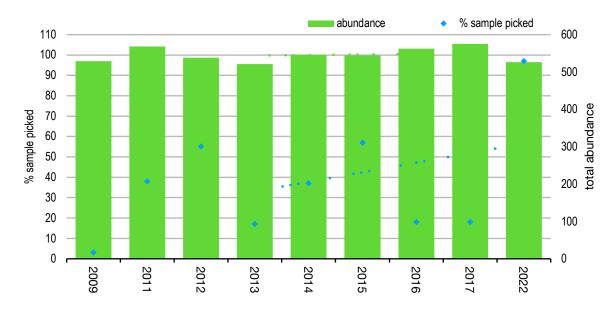


Figure 93. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1850 in all sampling years. RT samples were taken in all years. Target sub-sampling number is 500.



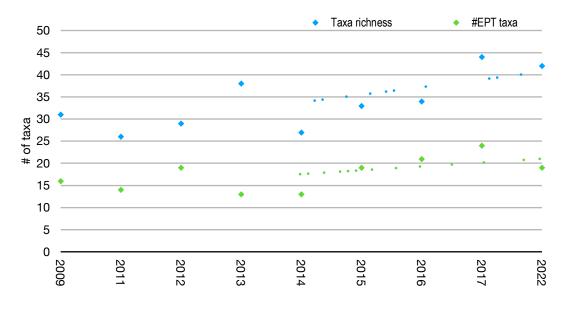


Figure 94. Sample richness and number of EPT taxa at WC1850 in all sampling years. RT samples were taken in all years. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

Figure 95. PREDATOR O/E and ORDEQ IBI scores at WC1850. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

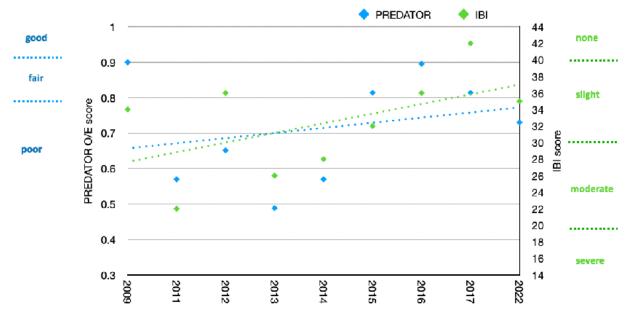


Figure 96. Numbers of sensitive and sediment-sensitive taxa at WC1850. RT samples were taken in all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and  $\geq$ 2 sediment-sensitive taxa.

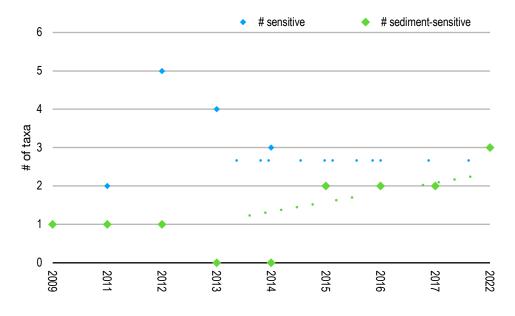
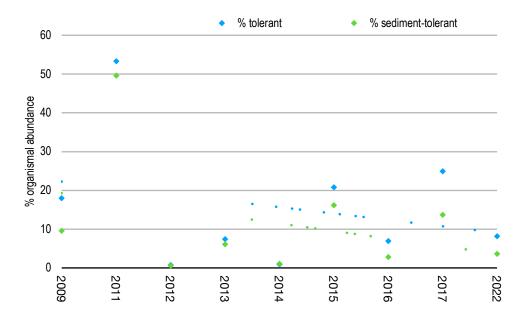
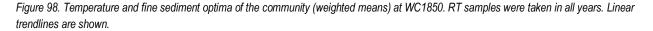


Figure 97. Relative abundance of tolerant and sediment-tolerant organisms at WC1850 RT samples were taken in all years. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.





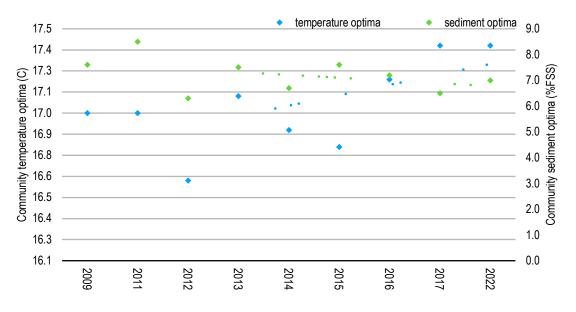


Figure 99. Temperature associations of the macroinvertebrate community at WC1850. RT samples were taken each year. Linear trendlines are shown.

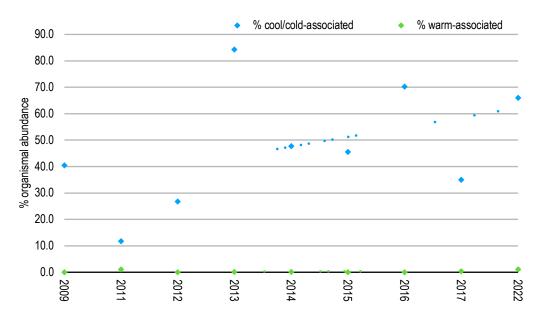


Figure 100. Cluster dendrogram of the WC1850 community in all sampling years. RT samples were taken in each year. The number at the end of each label indicates sampling year.

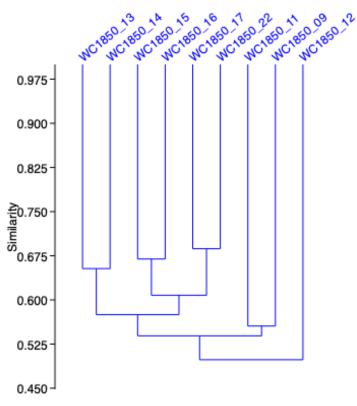


Figure 101. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1850 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = riffle-targeted, aqua = multihabitat. Axis 1 explains 36% of total sample variation; axis 2 explains an additional 22% of variation.

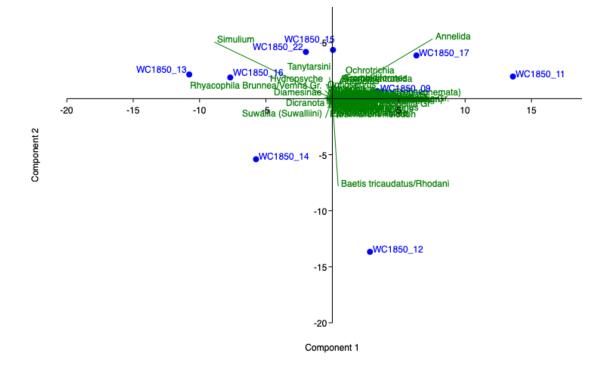
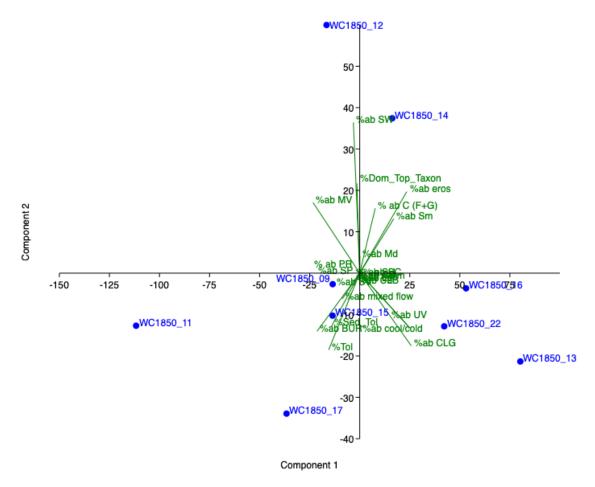


Figure 102. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1850 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 65% of total sample variation; axis 2 explains an additional 18% of variation.

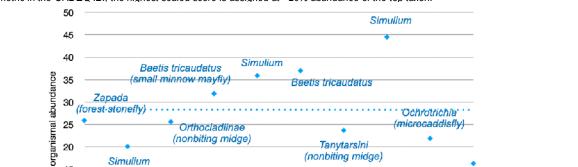


#### WC1900 (Camp Polk)

The WC1900 reach is one of four restored at Camp Polk in 2012. RT samples were taken in 2009-2017 and 2022. The 2022 sample was taken in primarily cobble/gravel substrate (Figure 1), and the community was most similar to those in adjacent reaches (WC1850-WC1950; Figure 5). Four taxa taken in 2022 were not found here in any prior year: *Lara*, a riffle beetle that feeds on submerged wood in cool, clear, flowing water (1 individual); *Acentrella insignificans*, a small minnow mayfly that tolerates warmer flows (1 individual); *Neoleptophlebia*, a prong-gilled mayfly associated with sediment and detritus in cooler flows (5 individual); and *Zapada oregonensis*, a forest stonefly associated with cooler, faster flows (1 individual). The community was dominated at low relative abundance (16%) by *Agapetus*, a saddlecase-maker caddisfly associated with cool flows, and consisted mainly of small, univoltine organisms that feed as collectors and move as clingers in colder, faster flows. The top taxon varied annually in relative abundance (Figure 103) and included more tolerant types (Orthocladiinae and Tanytarsini midges) as well as groups associated with faster and/or colder flows (*Agapetus, Simulium, Zapada*). This was the first year in which relative abundance of the top taxon was low enough to score in the top range of the ORDEQ IBI.

Unidirectional trends analysis showed significant increase across time in taxa richness and semivoltine organisms. The target sub-sampling number of 500 organisms was attained every year (Figure 104). Total and EPT taxa increased over time (Figure 105), though only significantly for taxa richness, and there were more taxa in 2022 than any prior year. O/E scores had a non-significant decrease (Figure 106) influenced by a drop from 2012-2014 likely due to restoration impacts, as scores were higher in subsequent years. IBI scores increased in recent years with the highest values in 2021 and 2022 (Figure 105). There are few sensitive or sediment-sensitive taxa (Figure 107), but the 2022 sample had more sensitive taxa (3) than any prior year. Abundance of sediment-tolerant organisms was low enough to receive the highest scaled score in the ORDEQ IBI in five sampling years, including 2022 (Figure 108); tolerant organism abundance varied with an overall increasing trend, but there were half as many in 2022 compared to 2021 (Figure 107). Community temperature optima decreased though 2013 and increased afterwards but with little change from 2017 to 2022 (Figure 109) and pre- and post-restoration means were not significantly different. Community sediment optima were similar pre- and post-restoration (Figure 108). There was a non-significant increase in abundance of cool/cold-associated organisms (Figure 110), and the 2022 sample had more than any prior year. As with other Camp Polk reaches, abundance of warm-associated organisms is very low (Figure 109).

The 2022 community was most similar to 2017 (Bray-Curtis similarity index = 0.62) but community similarity among all years was fairly high and most samples clustered in nearest-year pairs (Figure 111). Community differences seemed to reflect larger shifts in community composition after 2005 that occurred after perennial flow restoration (Figure 112), with high abundances of Zapada (a forest stonefly associated with cold water and a variety of flows) and Acentrella (a small minnow mayfly) in 2005 that dropped in subsequent years, as well as more Baetis tricaudatus, which can be abundant following a disturbance, in the years surrounding channel restoration (2012-2014). The most recent samples (2017, 2022) had more Agapetus, a saddlecase caddisfly associated with cold water, and Ochrotrichia, a microcaddisfly found in a variety of flows and temperatures. Differences in traits calculated as relative abundances distinguished between recent and early sampling years (Figure 113), with more univoltine and cool/coldassociated organisms and fewer swimmers in recent years (2016-2022).



Orthocladiinae (nonbiting midge)

2012

2013

2014

Figure 103. Relative abundance of the numerically dominant taxon at WC1900 in all sampling years. RT samples were taken in all years. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

Tanytarsini

(nonbiting midge)

2015

2016

Ochrotrichia (microcaddisfly)

2017

Agapetus (saddlecase\_caddisfly)

2022



Simulium

(black fly)

2008

201

25

20

\$ 15

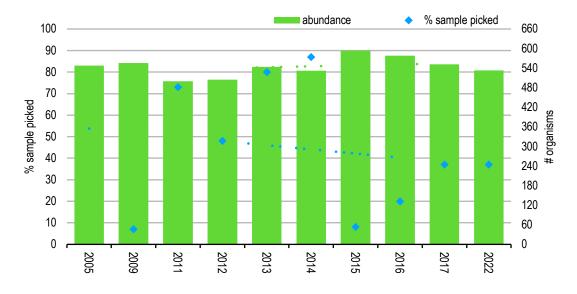
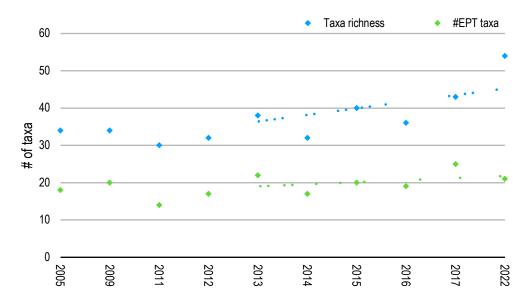


Figure 104. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1900 in all sampling years. RT samples were taken in all years. Target sub-sampling number is 500.

Figure 105. Sample richness and number of EPT taxa at WC1900 in all sampling years. RT samples were taken in all years. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.



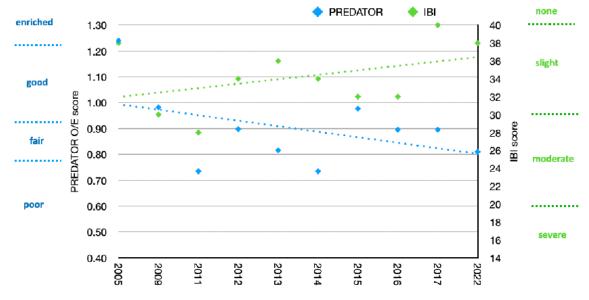
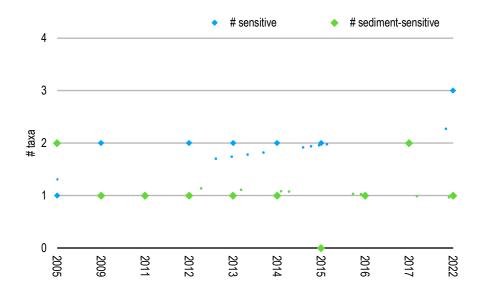
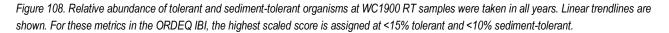


Figure 106. PREDATOR O/E and ORDEQ IBI scores at WC1900. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

Figure 107. Numbers of sensitive and sediment-sensitive taxa at WC1900. RT samples were taken in all years. 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.





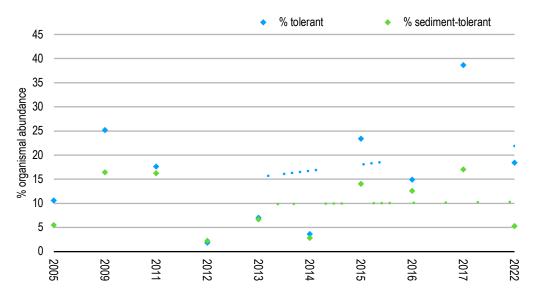
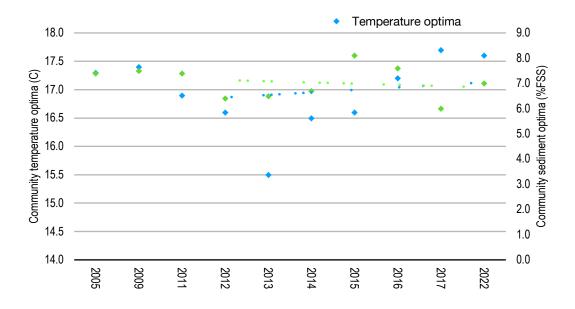


Figure 109. Temperature and fine sediment optima of the community (weighted means) at WC1900. RT samples were taken in all years. Linear trendlines are shown.



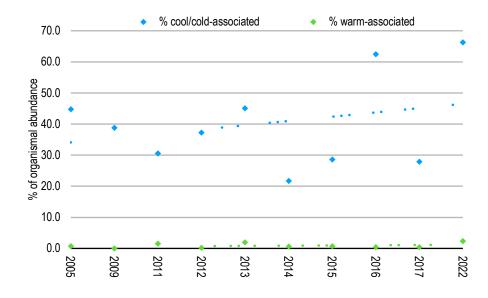


Figure 110. Temperature associations of the macroinvertebrate community at WC1900. RT samples were taken in each year. Linear trendlines are shown.

Figure 111. Cluster dendrogram of the WC1900 community in all sampling years. RT samples were taken in each year. The number at the end of each label indicates sampling year.

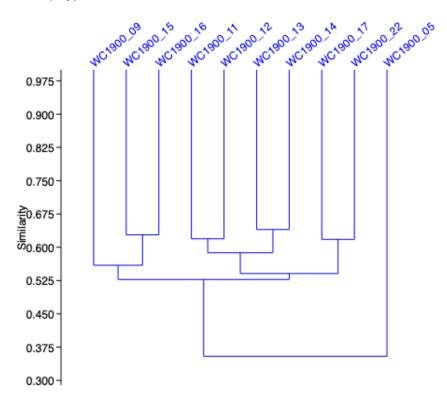


Figure 112. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1900 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 25% of total sample variation; axis 2 explains an additional 20% of variation.

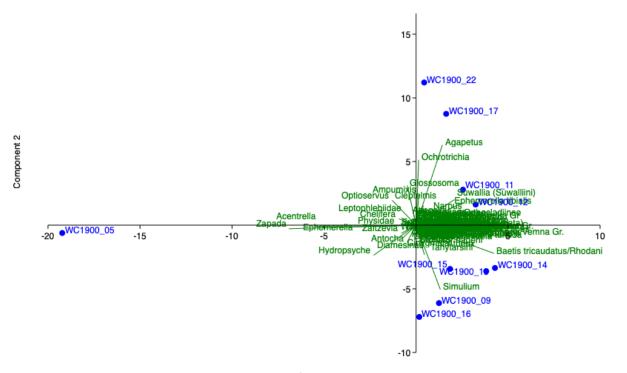
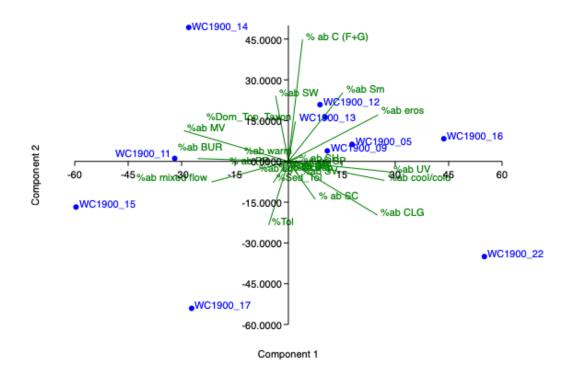


Figure 113. Ordination plot of a Principal Components Analysis (PCA) of texa anong all WC1900 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 44% of total sample variation: axis 2 explains an additional 29% of variation.



### WC1925 (Camp Polk)

The WC1925 reach is among those restored in 2012 at Camp Polk. PM samples were taken in 2019 in the primary channel (WC1925-3) and in several connected braided side channels (WC1925-1, WC1925-2, WC1925-4); in 2022, PM samples were taken in the primary WC1925-3 and WC1925-4 side channel. The primary channel (PC) sample was taken mainly in gravel/sand substrate and moving water and the side channel (SC) sample was taken in mostly sand/silt substrate and a variety of flows (Figure 2 & 3).

The PC sample community was most similar to RT samples in the adjacent WC1950 reach, while the SC community was most similar to the WC1025-2 side channel. Four taxa taken in 2022 were not found here in 2019: *Drunella flavilinea*, a sensitive, sediment-sensitive spiny crawler mayfly associated with cool, swift mountain streams (1 individual, PC); *Epeorus albertae*, a flatheaded mayfly found on the bottom of rocks in faster flows and a broad temperature range (1-2 individuals, PC and SC), *Neoleptophlebia*, a prong-gilled mayfly associated with sediment and detritus in cooler flows (7 individuals, SC); and *Rhyacophila atrata*, a green rockworm caddisfly associated with cold, clear, flowing water (3 individuals, PC). The PC community was dominated by Enchytraeidae (21.3%), a tolerant, sediment-tolerant pot worm found in slow current and shorelines of cold rivers and lakes. The SC community was dominated by *Polypedilum* (8.4%), a widespread non-biting midge found in sedimented substrate in a variety of flows and temperatures. In 2019, both sites were dominated by *Tanytarsus*, another widespread non-biting midge with broad ecological associations, at similar relative abundances (Table 2).

With two years of data, trends analysis could not be done but changes in metrics were summarized (Table 2). The target subsampling number of 500 organisms was attained in all samples, with less SC sample required in 2022. Total and EPT taxa numbers are high; both were lower in each reach in 2022, but values were still similar except for a decrease in total taxa in the PC sample from 67 to 52. The PC reach scored in the top range of the ORDEQ IBI for five metrics in 2019 and four in 2022; the SC sample scored in the top range for the same three metrics in both years. The PC reach had more sensitive and sediment-sensitive taxa than the SC, while abundance of sediment-tolerant organisms was lower in the SC sample. Community temperature optima were the same in each year in both reaches and were slightly higher in 2022. Community sediment optima was higher. There were more organisms associated with cooler water in 2022 in both samples, with nearly twice as many in the PC compared to 2019, and abundance of warm-associated organisms was very low in both reaches and decreased substantially from 2019.

Community similarity was greatest among most 2019 samples (Figure 114), and the 2022 communities were fairly similar to these (53-58% overall similarity). Community differences (Figure 115) reflected a shift in the dominant taxon from Tanytarsini, which dominated 2019 samples but was less abundant in 2022; and *Ochrotrichia*, a microcaddisfly found in a variety of flows and temperatures that was abundant only in the 2022 PC sample. Differences in traits calculated as relative abundances were driven by greater abundance of univoltine and cool/cold-associated organisms in the 2022 PC sample, and more burrowers and multivoltine organisms in the 2022 SC sample (Figure 116).

Table 2. Changes in metric values at WC1925. Multihabitat samples were taken in both years. PC = primary channel, SC = side channel. Numbers in bold indicate values that score in the top scaled range of the ORDEQ IBI.

	WC192	5-3 (PC)	I	WC1925-4 (SC)			
	2019	2022	change	2019	2022	change	
% of sample picked	10.8	27	+	100	15	-	
# of organisms in sample	546	539	-	541	536	-	
Taxa richness	67	52	-	64	61	-	
# Ephemeroptera	8	9	+	7	7	0	
# Plecoptera	4	3	-	4	4	0	
# Trichoptera	11	7	-	8	7	-	
#EPT taxa	23	19	-	19	18	-	
% dominance top taxon	22.7	21.3	-	10	8.4	-	
# sensitive taxa	4	5	+	4	3	-	
# sediment-sensitive taxa	3	2	-	1	1	0	
% tolerant	14.7	34.9	+	20.5	21.8	+	
% sediment-tolerant	4.8	21.9	+	8.7	6	-	
Community temperature optima (C)	17.1	17.7	+	17.2	17.7	+	
Community sediment optima (%FSS)	8.4	6	-	8.0	12	+	
% cool/cold-associated	33.2	66.2	+	29.6	36.8	+	
% warm-associated	27.3	2.8	-	14.0	7.6	-	

Figure 114. Cluster dendrogram of the WC1925 community in all sampling years. PM samples were taken in each year. WC1925-3 is the primary channel reach; the others are side channels. The number at the end of each label indicates sampling year.

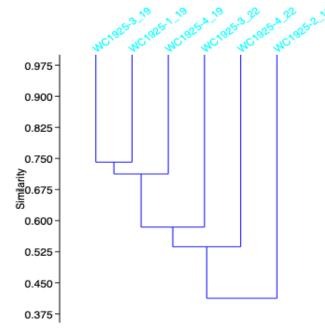


Figure 115. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1925 sampling years. PM samples were taken in each year. WC1925-3 is the primary channel; the others are side channels. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 48% of total sample variation; axis 2 explains an additional 22% of variation.

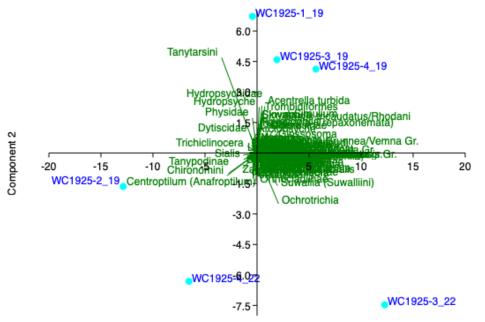
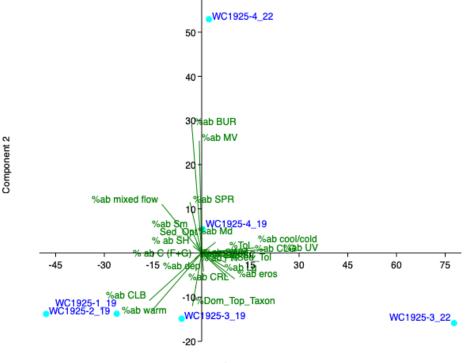




Figure 116. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1925 sampling years. WC1925-3 is the primary channel; the others are side channels. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 62% of total sample variation; axis 2 explains an additional 25% of variation.





#### WC1950 (Camp Polk)

The reach at WC1950 is among those restored in 2012 at Camp Polk. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken in 2019-2021. A duplicate sample was taken simultaneously in 2022 for quality assurance. The substrate in the 2022 samples was primarily cobble (Figure 1) and the communities paired in a CLUSTER dendrogram of all 2022 samples (Bray-Curtis similarity index = 0.78; Figure 5). Five taxa taken in 2022 were not found here in any prior year (one individual of each): *Pacifastacus*, our native western crayfish which tolerates a variety of flows and temperatures; *Hydra*, a tolerant sessile cnidarian that clings to the tops of rocks in slower flows; *Drunella flavilinea*, a sensitive, sediment-sensitive spiny crawler mayfly associated with cool, swift mountain streams; *Epeorus deceptivus*, a sensitive flatheaded mayfly found in fast cold flows; and *Epeorus longimanus*, a flatheaded mayfly associated with rocks in cool streams. The primary sample was dominated by Enchytraeidae (tolerant, sediment-tolerant pot worm associated with areas of slow current and shorelines of cold rivers and lakes) while the duplicate was dominated by *Simulium* (a black fly associated with flowing water), but the two occurred at similar relative abundance (22.4% and 23.3%, respectively). Top taxa at this site have been both tolerant and sensitive types, but relative abundances were low enough to score in the top range of the ORDEQ IBI in three of the five years since restoration (Figure 117). The target sub-sampling number of 500 organisms was attained in all but two sampling years, with much less of the total sample required since 2015 (Figure 118).

A unidirectional trends analysis found significant increases across time in total, EPT, and sediment-sensitive taxa and relative abundance of large organisms, and a significant decrease in multivoltine organisms. Taxa richness was significantly greater in recent sampling years (mean 2009-2017 = 32 taxa; mean 2019-2022 = 51 taxa; p = 0.0001), but the difference in mean EPT taxa in the same periods (18 and 20, respectively) was not significant (Figure 119). PREDATOR and IBI scores were not strongly influenced by restoration (Figure 120). O/E scores decreased from 2009 (good) to 2014 (poor) but rose into the zone between fair and poor in 2016-2022. IBI scores reflected slight disturbance except in 2016 (no disturbance) and varied little. Recent years had significantly more sensitive taxa (mean 2009-2017 = 2, mean 2019-2022 = 4, p = 0.0236) and sediment-sensitive taxa (mean 2009-2017 = 1, mean 2019-2022 = 2, p = 0.0119; Figure 121). There were also more tolerant and sediment-tolerant organisms in recent sampling years, but the differences were not significant (Figure 122), and both were more abundant in 2022 than the prior year. Mean community temperature optima was higher in 2019-2022 (17.4°F) compared to 2009-2017 (16.7°F; Figure 123) but not quite significantly (p = 0.054). However, mean fine sediment optima in recent years (7.3%) was significantly greater (mean 2009-2017 = 6.5%; p = 0.0491). Despite these trends, there were more cool/cold-associated organisms and fewer warm-associated organisms in 2022 than any prior year (Figure 124), though means before and after the 2018 sampling gap were not significantly different.

Communities in 2022 RT samples were most similar to the 2019 RT sample and the 2015 and 2016 samples (Bray-Curtis similarity index 0.61; Figure 125); all PM communities were more similar to each other than to any RT sample. Community differences were driven by Tanytarsini (non-biting midge tribe that builds tubes in soft sediments), which was more abundant following restoration (2015-2020); *Baetis tricaudatus* (a small minnow mayfly associated with faster flows and clearer water), which was more abundant in earlier sampling years; and Annelida (tolerant segmented worms) and *Simulium* (a black fly associated with flowing water), which were more abundant in 2022 samples (Figure 126). Differences in traits calculated as relative abundances reflected the greatly increased abundance of univoltine and cool/cold-associated organisms in 2022, as well as more organisms associated with faster flows and a more balanced community (lower top taxon abundance) in most years following the 2018 sampling gap (Figure 127).

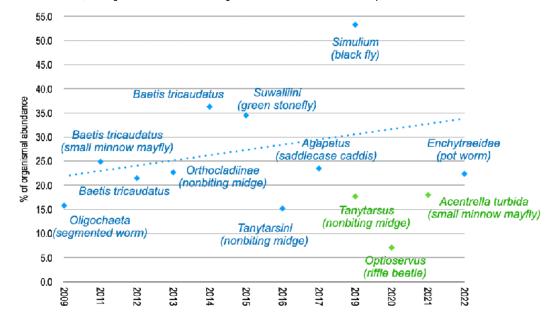
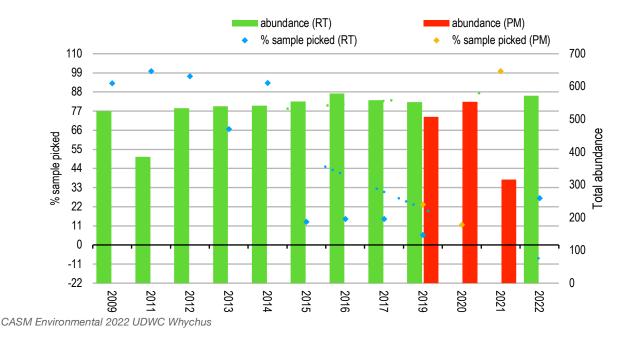


Figure 117. Relative abundance of the numerically dominant taxon at WC1950 in all sampling years. Blue = RT sample, green = PM sample. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

Figure 118. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC1950 in all sampling years. RT samples were taken 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Target sub-sampling number is 500.



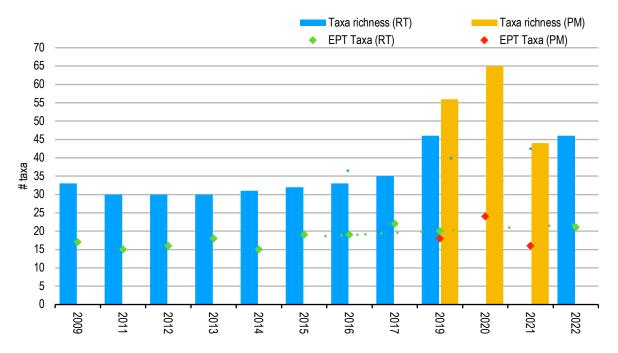


Figure 119. Sample richness and number of EPT taxa at WC1950 in all sampling years. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.

Figure 120. PREDATOR O/E and ORDEQ IBI scores at WC1950. Dotted lines outside the axes show cutoff values for different condition scores. Linear trendlines are shown.

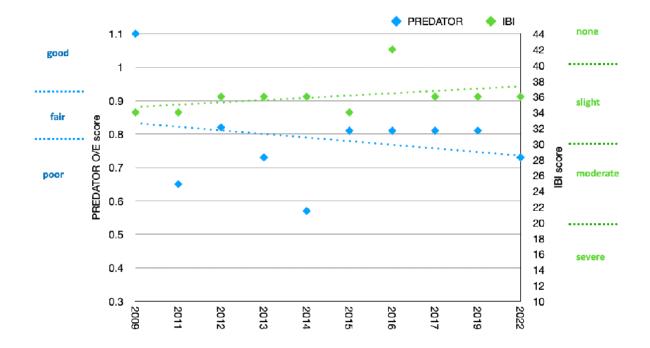


Figure 121. Numbers of sensitive and sediment-sensitive taxa at WC1950. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at >4 sensitive and  $\geq$ 2 sediment-sensitive taxa.

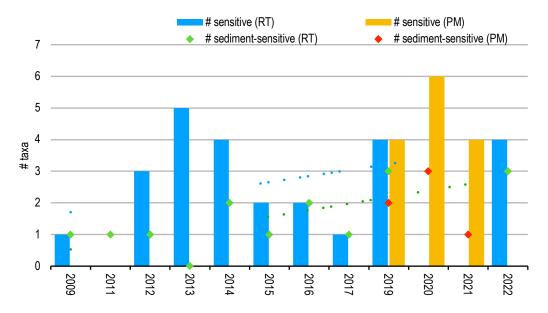
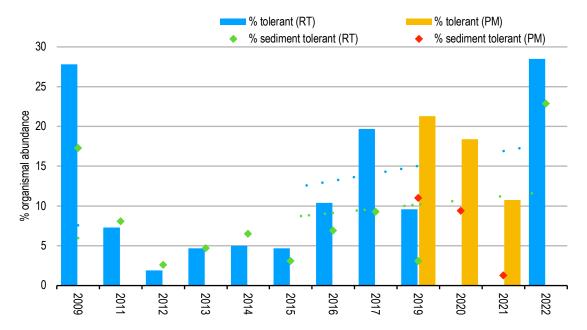


Figure 122. Relative abundance of tolerant and sediment-tolerant organisms at WC1950. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.



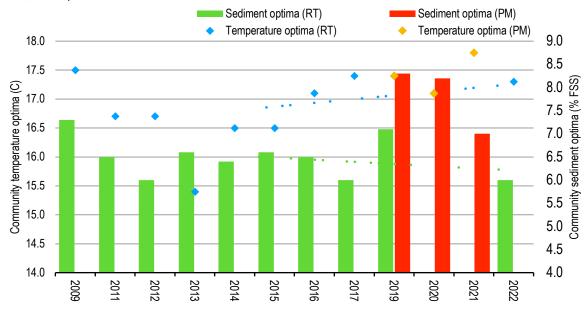


Figure 123. Temperature and fine sediment optima of the community (weighted means) at WC1950. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Linear trendlines are shown.

Figure 124. Temperature associations of the macroinvertebrate community at WC1950. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Linear trendlines are shown.

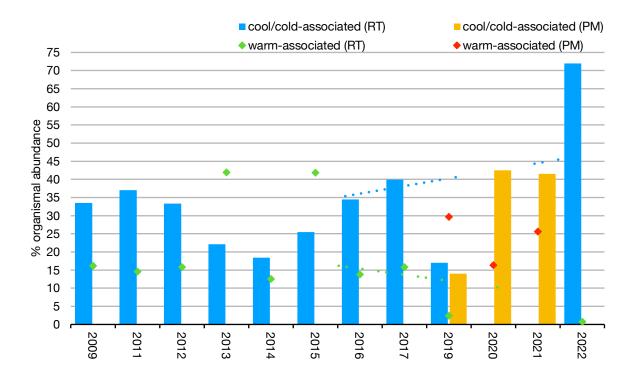


Figure 125. Cluster dendrogram of the WC1950 community in all sampling years. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Blue = RT, aqua = PM. The number at the end of each label indicates sampling year.

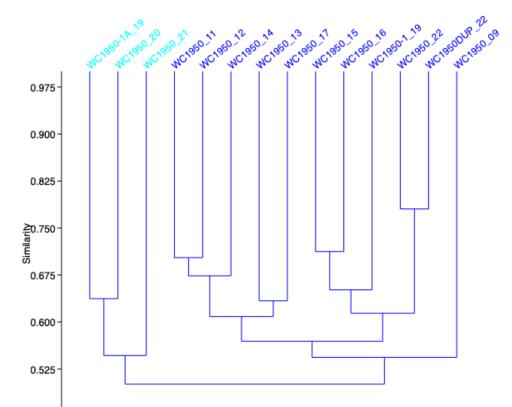


Figure 126. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC1950 sampling years. RT samples were taken in 2009-2017, 2019, and 2022; PM samples were taken 2019-2021. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 26% of total sample variation; axis 2 explains an additional 19% of variation.

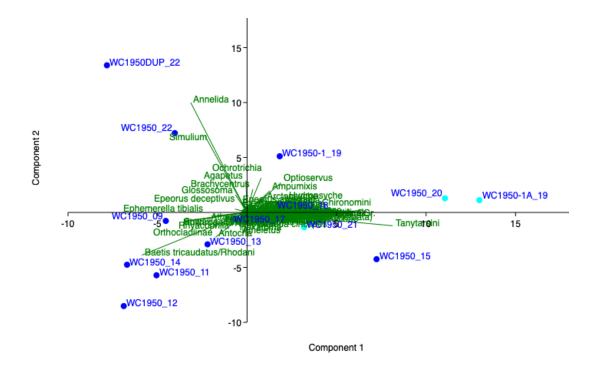
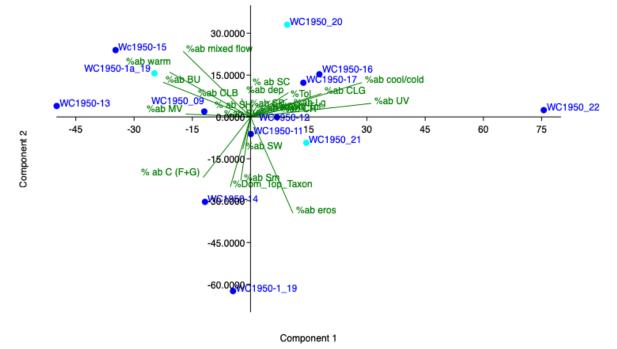


Figure 127. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC1950 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM. Axis 1 explains 35% of total sample variation; axis 2 explains an additional 23% of variation.



#### WC2600 (Whychus Floodplain)

The reach at WC2600 was sampled in 2005-2022, with RT samples taken through 2020 and PM samples taken 2018-2022. In 2015, the stream was directed into a new meandering channel, 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, the reach included elements of side channel habitat. The 2022 PM sample was taken in moving water (Figure 3) in a variety of mineral and organic substrates (Figure 2). The community was an outlier to all other 2022 samples (Figure 5), with an overall average similarity of less than 37%. Only one taxon taken in 2022 was not found here in any prior year: *Neoleptophlebia*, a prong-gilled mayfly associated with sediment and detritus in cool flows (11 individuals). The community was dominated at low abundance (17.3%) by *Isoperla*, a spring stonefly found in streams, springs, and lakes. The top taxon in most years was a non-biting midge, and this is the first year that a stonefly dominated abundance (Figure 128). The target sub-sampling number of 500 organisms was attained in 10 of the 12 sampling years, including every year since 2018 (Figure 129).

A unidirectional trends analysis revealed significant increase across time in total, Ephemeroptera, EPT, sensitive, and sediment-sensitive taxa, and in community sediment optima, abundance of shredder, sprawler, large, and cool/cold-associated organisms; and significant decreases in abundance of collectors and warm-associated organisms. There were also significantly more total and EPT taxa in 2018-2022 compared to all prior sampling years (mean total taxa = 27 vs. 52, p = 0.0001; mean EPT taxa = 15 vs. 21, p = 0.0084; Figure 130). O/E scores reflected poor condition in every year except 2018 and 2020, which scored as fair. However, ORDEQ IBI scores indicated no disturbance from 2017-2020, and the 2022 PM sample scored in the highest range of the ORDEQ IBI for every individual metric except

number of caddisfly taxa. This site also had the most total, EPT, stonefly, and sensitive taxa as well as the highest predator abundance, lowest community temperature optima, and fewest non-insect organisms of any 2022 sample, and more sediment-sensitive taxa than any sample except WC1850 and WC1950. However, it also had the greatest abundance of tolerant organisms among all 2022 samples.

There were significantly more sensitive taxa in 2018-2022 (mean = 6.8) than in prior years (2005-2017; mean = 3.4; p = 0.0059; Figure 131). There are fewer sediment-sensitive taxa overall but means were also significantly greater in 2018-2022 (mean = 3.1; mean 2005-2017 = 1.3; p = 0.0002). Relative abundance of tolerant and sediment-tolerant organisms varies but is generally low (Figure 132). Community temperature optima are lower overall compared to other sites and decreased since 2020 (Figure 133), though not significantly. Community sediment optima increased significantly over time and were significantly greater in 2018-2022 (mean = 8.0%) compared to all prior years (mean = 6.7%; p = 0.0018). Abundance of cool/cold-associated organisms increased over time, though not significantly, and there were more in the 2022 sample than in any prior year (Figure 134). The decrease in warm-associated organisms was also not significant, but there were fewer in the 2021 and 2022 PM samples than in prior years (Figure 135).

The 2022 community was most similar to PM samples taken in the primary and side channels in 2018-2020 (Figure 136), although the 2021 PM community was more similar to RT samples taken in other years. Community differences reflect the more heterogeneous flow and habitat in both side channel and primary channel samples in recent years (Figure 137); these had more Tanytarsini (non-biting midge tribe that builds tubes in soft sediments) and *Zapada cinctipes* (forest stonefly associated with a variety of cool waters whose presence indicates ongoing contributions from the riparian zone to the instream food base). Differences in traits calculated as relative abundances distinguished more between sample types, with greater abundance of clingers and organisms associated with faster (erosional) flows in RT samples, a less balanced community with more collector feeders in the earliest sampling years, and more fast-developing (multivoltine) organisms associated with a variety of flows in the side channels.

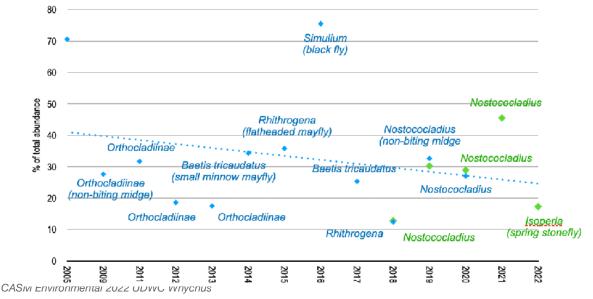


Figure 128. Relative abundance of the numerically dominant taxon at WC2600 in all sampling years. Blue = RT sample, green = PM sample. For this metric in the ORDEQ IBI, the highest scaled score is assigned at <20% abundance of the top taxon.

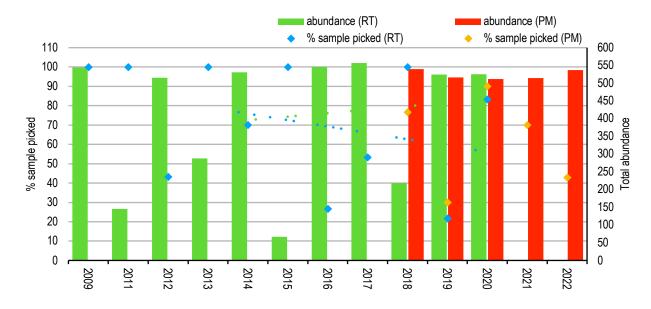
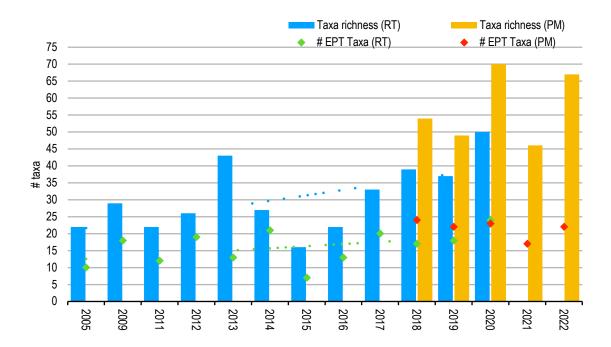
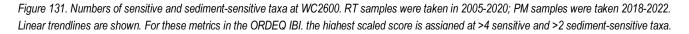


Figure 129. Proportion of sample needed for sub-sampling and resulting organismal abundance at WC2600 in all sampling years. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Target sub-sampling number is 500.

Figure 130. Sample richness and number of EPT taxa at WC2600 in all sampling years. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Linear trendlines are shown. In the ORDEQ IBI, >35 total taxa is assigned the highest scaled score.





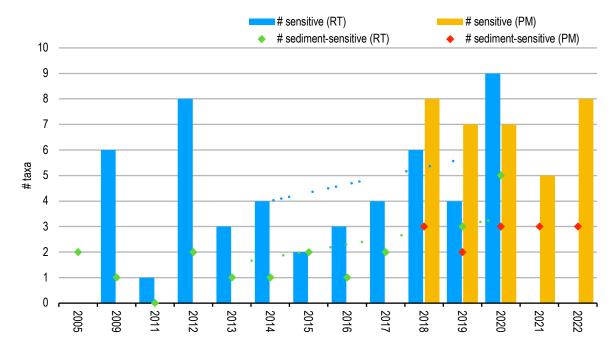
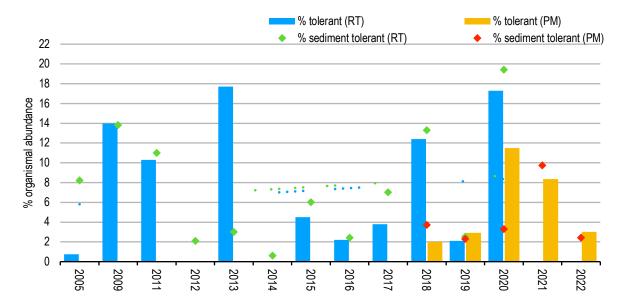


Figure 132. Relative abundance of tolerant and sediment-tolerant organisms at WC2600. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Linear trendlines are shown. For these metrics in the ORDEQ IBI, the highest scaled score is assigned at <15% tolerant and <10% sediment-tolerant.



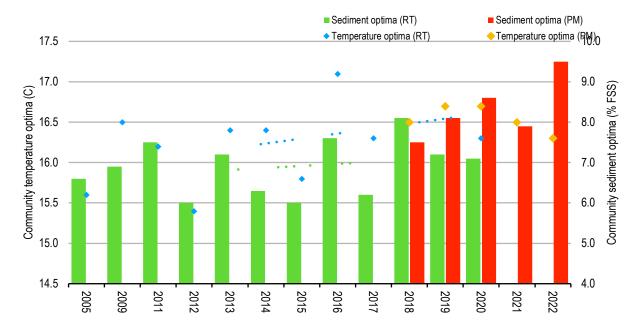


Figure 133. Temperature and fine sediment optima of the community (weighted means) at WC2600. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Linear trendlines are shown.

Figure 134. Temperature associations of the macroinvertebrate community at WC2600. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Linear trendlines are shown.

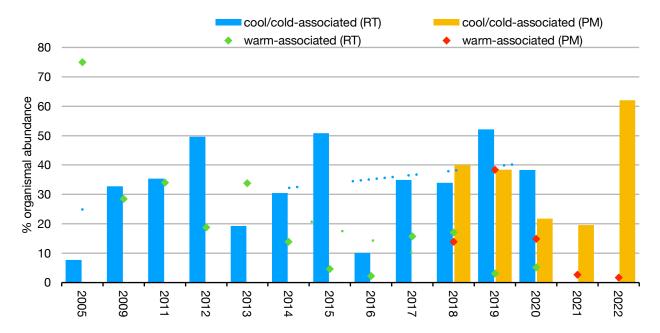


Figure 135. Cluster dendrogram of the WC2600 community in all sampling years. RT samples were taken in 2005-2020; PM samples were taken 2018-2022. Blue = RT, aqua = PM, green = side channel PM. The number at the end of each label indicates sampling year.

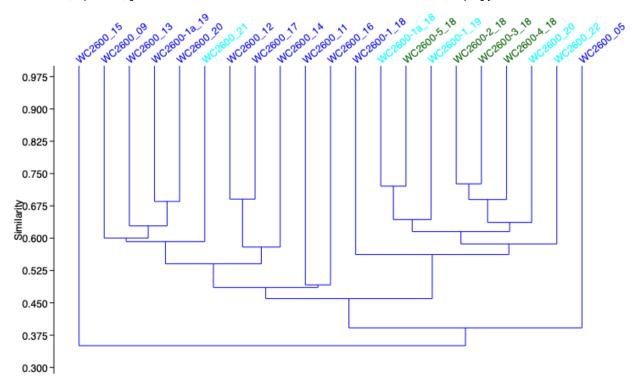


Figure 136. Ordination plot of a Principal Components Analysis (PCA) of taxa abundances among all WC2600 sampling years. Blue = RT, aqua = PM. areen = side channel PM. Axis 1 explains 26% of total sample variation: axis 2 explains an additional 19% of variation.

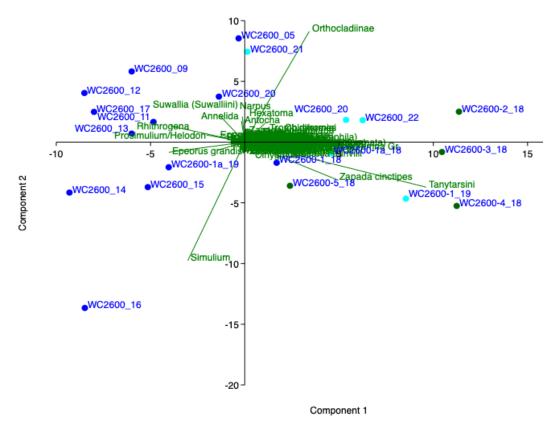
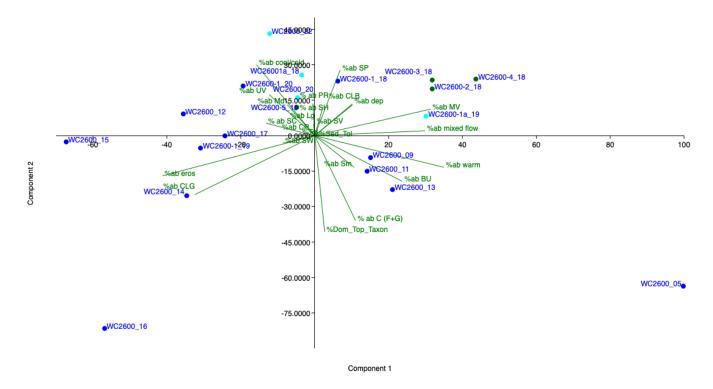


Figure 137. Ordination plot of a Principal Components Analysis (PCA) of traits calculated as relative abundances among all WC2600 sampling years. Eigenvectors show taxa contributions to between-sample variation, where vector length is related to the strength of the contribution. Blue = RT, aqua = PM, green = side channel PM. Axis 1 explains 38% of total sample variation; axis 2 explains an additional 24% 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, such as re-meandering mainstem channels, restoring floodplain connectivity, and creating new heterogeneous side channels and a more dynamic, mobile primary channel. Restoration activities drove changes in macroinvertebrate community composition, but this extended sampling period spans other changes that impact macroinvertebrates, such as altered precipitation patterns, more extreme heat, and widespread wildfires.

Samples were taken in Whychus Creek on 14 occasions spanning 16 years, and the new species accumulation curve should be approaching saturation. However, although only two taxa new to the complete Whychus dataset were taken in 2022, all but two reaches had taxa that were new to that site in 2022. The number of new taxa per site ranged from 1-8, with the greatest number among the side channel samples at the recently restored WC1025 reach.

Macroinvertebrate community composition is strongly influenced by reach location, with greater similarity among communities from upstream reaches (i.e., WC1950-WC2600) compared to those in downstream reaches (i.e., WC0600-WC1150), regardless of sampling technique or whether a primary or side channel reach was sampled. However, the macroinvertebrate community continues to change even at sites where no active restoration was done in ways that suggest an overall uplift. Site scores from PREDATOR and IBI models are generally less informative than

individual taxonomic and ecological metrics. There is often little agreement in biological condition scores between the two models, with only one of nine RT samples taken in 2022 scoring lower than slightly or not disturbed based on IBI score, while five of the nine sites scored as most disturbed (poor) based on O/E score. No significant unidirectional longterm trends were detected in O/E scores at any 2022 site that was sampled in multiple years, and significant trends in IBI scores were seen at only three sites: WC0600 (increasing; unrestored site), WC1100-2 (increasing; restored site), and WC1825 (decreasing; restored site).

However, more sites showed significant unidirectional trends over time in positive taxonomic and ecological traits, such as more total taxa (six sites), more mayfly taxa (five sites), and more sediment-sensitive taxa (six sites), and more semivoltine and large-bodied organisms, which are indicative of stable habitat conditions (four sites each). Only one to two sites showed significant trends in any metrics relating to temperature or flow associations, although one site had a significantly increasing trend for community temperature optima (WC1100-2) and two showed significant increases in community fine sediment optima (WC1025, WC2600). Abundance of tolerant organisms, a negative metric, increased significantly across time at three sites (WC1025, WC1100-2, WC1150). This may be influenced by the use of multihabitat sampling in recent years, as PM samples include slower flows, but PM samples in other reaches and years have had low abundances of tolerant organisms. These three sites span a longer reach where restoration was done (at WC1100-2), and restoration-related disturbance could be a factor.

### Unrestored reaches

The reaches at WC0600, WC0850, WC0900, and WC1150 have not undergone active restoration. However, as these sites are among the furthest downstream on the creek, they are likely to be impacted by habitat and macroinvertebrate community changes at multiple reaches upstream where extensive restoration was done, as well as the ongoing impacts of changing climate. WC0600 is the only reach sampled in every project year, and macroinvertebrate community composition and associated ecological metrics changed over time in ways that largely suggest improved conditions. This is one of only two sites sampled in 2022 to show a significant increase in IBI scores over time, as well as significantly more total, sensitive, and sediment-sensitive taxa. The significant increases in semivoltine (long-lived) and medium-bodied taxa also suggest greater habitat stability, as this supports a longer life span and growth to larger size. Abundance of crawler organisms decreased significantly, which could indicate fine sediment stress, but abundance of sediment-tolerant organisms is consistently very low and community sediment optima have not been increasing. However, changes in community temperature optima and organismal temperature association, although not significant, suggest that temperature could become a stressor at this site.

Significant changes in metrics at WC0850 suggest increased habitat stability and heterogeneity, with more organisms that tolerate a variety of flow types and fewer associated with faster (erosional) flows. The increase in scrapers, which feed on algae and biofilm on mineral surfaces that can be smothered by fine sediment, as well as a decrease in swimmers, which can more rapidly escape sedimented or disturbed habitat, suggest that fine sediment is not a stressor. Similar to the adjacent WC0850 reach, the WC0900 community showed a significant increase over time in scrapers and univoltine organisms. The significant increases in mayfly taxa and medium-bodied organisms along with

decreases in relative abundance of the top taxon and of sprawlers suggests more stable habitat without a strong sediment stressor signal.

WC1150 is immediately upstream of several reaches in Whychus Canyon that were restored in 2016 and 2021. Trends in metrics at this site also indicate uplift, with taxonomic and trait differences suggesting a shift towards faster flows, and increased clear cobble (i.e., less sediment), including significant increases over time in number of mayfly, stonefly, and sediment-sensitive taxa. The significant increase in large organisms and decrease in multivoltine organisms over time along with significantly more sensitive taxa suggest greater habitat stability. There are also consistently few sediment-tolerant or warm-associated organisms in each year. The 2022 sample was somewhat anomalous, as it had fewer organisms, total taxa, and EPT taxa compared to recent years, but it is not known if that is due to an issue with sampling procedure or if it reflects a real recent change.

### Restored reaches

Restoration at WC1025 occurred in 2021, but taxonomic and ecological metrics suggest less post-restoration disturbance compared to other restored reach along Whychus. All 2022 primary and side channel communities were most similar to communities in recent pre-restoration sampling years. The communities in primary and side channel samples were fairly well-balanced (<22% abundance of the top taxon), which indicates less habitat disruption, and the primary channel RT sample had higher O/E and IBI scores than the two years that preceded restoration. Total and EPT taxa richness in both new side channels was comparable to primary channel PM and RT samples, indicating rapid colonization of the new habitats, though with a greater abundance of more tolerant taxa in the side channels, which is expected. Surprisingly, the number of sensitive and sediment-sensitive taxa and abundance of tolerant organisms in 2022 did not differ greatly from the 2020 sample, although there were more sediment-tolerant organisms. More taxa new to the site (8 total) were taken here than in any other 2022 sample, with the majority in side channels, suggesting positive effects of increased habitat heterogeneity in supporting diversity. Comparison to the unrestored WC0900 reach downstream revealed little difference in most metrics regardless of restoration, and in 2022 the WC1025 sample had a higher O/E score (good vs. poor at WC0900), more total and EPT taxa, and more cold-associated and fewer warm-associated organisms than WC0900.

The WC1100 reach was restored in 2016, and a significant increase was seen across all years in IBI score, number of Ephemeroptera, Trichoptera, and sediment-sensitive taxa, and abundance of tolerant, large, semivoltine, clinger, and cool/cold-associated organisms; as well as a significant decrease in collector abundance. The 2017 community reflected substantial restoration-related disturbance that recovered through subsequent years, with significantly more total and EPT taxa and cool/cold-associated organisms as well as higher IBI scores post-restoration. In addition, many metric values post-restoration were comparable to or greater than those at the upstream WC1150 reference site. The increased abundance of tolerant organisms may be a result of greater overall diversity, though community temperature and sediment optima were also higher in 2022 than in any prior year.

The WC1825 reach is directly downstream of the Camp Polk Meadow restoration project completed in 2012 and had the fewest significant trends in metrics across time, although it was the only site at which IBI scores decreased significantly. There were more taxa and higher O/E scores in recent years, though the differences were not significant, but there were also significantly more tolerant and sediment-tolerant organisms in recent years, and abundance of both was greater in 2022 than in any prior year at the site. Community temperature optima increased steadily since 2014 while abundance of cool/cold-associated organisms decreased. Neither trend was significant, and abundance of warm-associated organisms was consistently very low but disturbance, temperature, and fine sediment may be more of a stressor here than in other reaches.

The adjacent WC1850 reach restored in 2012 also showed few trends in many pre- and post-restoration metrics. However, there were significant increases across all years in total, caddisfly, and sediment-sensitive taxa, and significantly more cool/cold-associated organisms post-restoration. This reach consistently has very few organisms associated with warm water (<2% abundance). Similarly, WC1925 showed significant increases over time in only two metrics, taxa richness and abundance of long-lived (semivoltine) organisms. However, there were more total taxa, sensitive taxa, and cool/cold-associated organisms in 2022 than any prior year, and fewer tolerant and sediment-tolerant organisms compared to the previous sampling year (2017). Similar to WC1825 and WC1850, relative abundance of warm-associated organisms is consistently very low in the reach.

Less can be said about restoration impacts or trends in metrics at WC1925, which was also restored in 2012 but was only sampled in 2019 and 2022. Between those years, values of positive metrics in primary channel samples such as number of mayfly taxa and abundance of cool/cold-associated organisms increased, while values of others, such as number of sensitive and sediment-sensitive taxa and abundance of the dominant taxon were relatively unchanged. Among the negative metrics, there were fewer warm-associated organisms in 2022 but more tolerant and sediment-tolerant organisms, which could reflect impacts of an LTPBR project implemented immediately upstream in 2022. More metrics in the WC1925-4 side channel were the same or only slightly different between the two years; the main differences included a higher community sediment optima, more cool/cold-associated organisms and fewer warm-associated. More years of sampling will be needed to reveal significant trends.

Restoration occurred at the WC1950 reach in 2012 and significant changes across time suggesting improved habitat quality and/or stability were seen in multiple metrics, including increased total, EPT, and sediment-sensitive taxa and greater abundance of large-bodied organisms, and a significant decrease in multivoltine organisms. There were also significantly more total, sensitive, and sediment-sensitive taxa in the most recent sampling years (2019-2022). Relative abundance of warm-associated organisms was generally greater than in other Camp Polk reaches, but the 2022 sample had fewer warm-associated and more cool/cold-associated organisms than any prior year at this site.

WC2600 was restored in 2014 and is the only reach except WC0600 that was sampled in every project year. This site also reflects uplift, with significant increases across time in the number of total, mayfly, EPT, sensitive, and sediment-sensitive taxa, and in the number of shredders, large-bodied, and cool/cold-associated organisms. Community sediment optima also increased significantly over time, which suggests sediment could be becoming a

stressor, but relative abundance of sprawlers, which can be smothered by fine sediment, increased significantly in the same span. In addition, there were significantly more total, EPT, sensitive, and sediment-sensitive taxa in 2018-2022 compared to all prior sampling years. In 2022, the sample scored in the highest range for all metrics in the ORDEQ IBI except number of mayfly taxa, and there were more total, stonefly, sensitive, and sediment-sensitive taxa at WC2600 than any other 2022 site. This sample also had the lowest community temperature optima of all 2022 samples and the fewest non-insect organisms. Overall, changes in metric values indicate a diverse community in stable, higher-quality habitat.

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	Minimum	Maximum	Mean	Std. Dev.
Taxa richness	30	67	48.9	8.7
Ephemeroptera taxa	7	13	8.9	1.6
Plecoptera taxa	2	8	4.2	1.3
Trichoptera taxa	3	8	6.1	1.6
# sensitive taxa	2	8	3.6	1.5
# sediment-sensitive taxa	0	3	1.7	0.8
% dominance top taxon	8.4	52.5	22.2	10.1
% tolerant organisms	3.0	64.5	30.5	15.1
% sediment-tolerant organisms	2.0	52.9	11.5	12.4
Community BI	3.0	5.1	3.9	0.6
EPT richness	14	22	19.1	2.1
% diversity EPT	29.5	48.8	39.8	5.5
community temperature optima	16.3	18.7	17.8	0.6
community sediment optima	6.0	12.0	8.0	1.6
% predators	2.2	25.9	8.0	5.4
% scrapers	4.3	37.6	24.5	10.2
% shredders	0.2	17.3	3.7	4.7
% collectors	40.9	79.2	53.6	9.7
% large	0.6	24.8	8.4	6.8
% medium	11.2	31.3	17.1	5.1
% small	54.2	83.8	69.6	7.8
% multivoltine	9.4	63.1	27.9	13.9
%ab univoltine	24.9	75.9	51.9	15.0
%ab semivoltine	3.3	39.7	11.7	9.2
% cool/cold-associated	17.9	76.9	45.0	18.0
% warm-associated	0.5	20.0	4.9	4.9
% depositional-associated	0.0	53.1	12.4	12.3
% erosional-associated	22.8	73.8	47.4	14.4
% mixed flows	11.2	65.5	38.1	16.1
% burrowers	0.3	46.7	10.1	13.1

# Appendix A. Range, mean, and SD of all 2022 community metrics.

	Minimum	Maximum	Mean	Std. Dev.
% climbers	0.2	11.6	3.6	3.5
% crawlers	0.0	22.7	3.9	6.9
% clingers	27.9	81.6	57.0	15.1
% sprawlers	3.0	41.0	13.6	11.4
% swimmers	4.3	28.8	9.9	5.9
% non-insect	2.2	54.0	13.0	12.2
% Chironomidae	2.6	57.3	17.0	16.4

# Appendix B. Site-level trends in metrics.

Trend analysis was done on all years of data at each individual sampling site. Metrics given as percentages indicate relative organismal abundance. Indicated trends are statistically significant (p < 0.05).

Metric	WC0600	WC0850	WC0900	WC1025	WC1100-2	WC1150	WC1825	WC1850	WC1900	WC1950	WC2600
PREDATOR	_								_		
IBI	inc				inc		dec				
Total taxa	inc			inc				inc	inc	inc	inc
Mayfly taxa	_		inc	inc	inc	inc					inc
Stonefly taxa		dec				inc					none
Caddisfly taxa	_				inc	—		inc			none
#EPT Taxa						—				inc	inc
# Sensitive	inc					—	—			_	inc
#Sediment- Intolerant	inc			_	inc	inc		inc		inc	inc
Temp. Optima					inc						
Sediment Optima				inc							inc
%Dom. Top Taxon			dec								
%Tolerant				inc	inc	inc					
%Sediment Tolerant											
% Predator				inc		dec					
% Scraper		inc	inc		inc	—					

Metric	WC0600	WC0850	WC0900	WC1025	WC1100-2	WC1150	WC1825	WC1850	WC1900	WC1950	WC2600
% Shredder	_										inc
% Collector					dec	dec					dec
% Large	_				inc	inc				inc	inc
% Medium	inc		inc								
% Small											
% Multivoltine						dec				dec	
% Univoltine		inc	inc								
% Semivoltine	inc				inc	inc			inc		
% Cool/cold					inc						inc
% Warm				_							dec
% Depositional											
% Erosional		dec									
% Mixed flow	_	inc									
% Burrower											
% Climber	inc		inc	_							
% Crawler	dec					dec					
% Clinger					inc	inc					
% Sprawler			dec								inc
% Swimmer		dec				dec	dec				

## Appendix C. Metric value ranges from downstream to upstream.

Boxplots show data from all sampling years at each site. Horizontal line in each box shows median value; filled box shows interquartile range; whiskers depict data range; dots show outlier values.

