Investigating Stable Water Isotopes Across Beaver-Dammed Mountain Streams

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Abstract

Beaver dams increase surface-water residence time and storage, creating conditions that can enhance evaporative water loss and alter the stable isotope composition of stream water. During the anomalously low-snowpack 2026 season, I investigated natural variation in stable water isotopes ($\delta^{18}O$ and $\delta^{2}H$) across three headwater streams in Gunnison County, Colorado to evaluate evaporative fractionation associated with beaver activity with the purpose of identifying signals of evaporative water loss. The three streams occupy climatically similar subalpine watersheds but differ in bedrock geology, restoration history, and degree of beaver influence: Trail Creek, a young beaver system re-established through a 2021 restoration effort; Coal Creek, an older, naturally established beaver system; and Italian Creek, an undammed control. Water samples were collected from upstream and downstream reaches of beaver dammed creeks across the sampling season and analyzed for $\delta^{18}O$ and $\delta^{2}H$ using cavity ring-down spectroscopy, and continuous gauge records at Trail Creek were used to develop a stage-discharge rating curve. Lc-excess declined significantly downstream at Trail Creek across all sampling dates, tracking cumulative beaver dam count and dam length, while Coal Creek showed a weaker, directionally consistent pattern. The undammed Italian Creek control showed little spatial change. A linear mixed-effects model confirmed a significant upstream-downstream enrichment effect that varied in magnitude across the three watersheds, indicating that beaver damming, rather than distance alone, drives a negative drift in lc-excess, though its strength differs by site. At Trail Creek, lc-excess was strongly correlated with discharge, particularly downstream of beaver influence, and became significantly more enriched as streamflow declined. These results indicate that beaver ponds measurably increase evaporative water loss, with the strongest effect where dam density and residence time are greatest. Evaporative loss to streams is relevant to beaver dam analog (BDA) restoration because their structures do more than store water. They create a complex balance of net water loss and gains across the watershed.

Introduction

As ecosystem engineers, beavers modify floodplain exchange by diverting large amounts of channel water onto the floodplain, which enhances lateral hydrologic connectivity (Wegener et al., 2017), and impacts the hydrogeochemical properties of groundwater flowpaths (Briggs et al., 2019). Beaver dams hold large volumes of water in ponds and expand riparian wetlands (Wegener et al., 2017). Dams slow water flow, allowing floodplains to become saturated and extending the residence time of water in the river corridor, exposing it to greater potential evaporative loss (Sprenger et al., 2017).

The historical decline of North American beavers (*Castor canadensis*) is marked by the arrival of a substantial number of Europeans in the 1600s, resulting in extreme trapping that brought the species to near extinction on the continent by the 1900s (Naiman et al., 1986). Since the end of the fur trapping era, beavers have begun to repopulate their natural habitats across North America (Burchsted et al., 2010).

Beavers in the western United States have been expanding back into their historic range, mediated by warming temperatures and aridification (Pilliod et al., 2018). Understanding the functions of beaver dams therefore plays a vital role as global warming leads to hydrological extremes. Crested Butte, Colorado, a water-limited and snowmelt-dependent region, experienced a historically low snowpack in 2026, raising questions about how such anomalous conditions alter aquatic and water-dependent ecosystems and whether beaver-mediated systems are equipped to buffer against them (Johnson et al., 2025). Because beaver ponds slow water flow and extend surface water exposure, they create conditions that can drive the potential for physical loss of water from the landscape— a process measurable through stable water isotope ratios (Craig, 1961; Sprenger et al., 2017).

Stable water isotopes ($\delta^{18}O$ and $\delta^{2}H$) vary naturally across stream water, groundwater, and precipitation, with variation among these bodies serving as tracers of key hydrological processes. Enrichment of heavy isotopes can be a signature for evaporation, making them a powerful tool for characterizing how water moves and transforms within a watershed (Burns & McDonnell, 1998; Carroll et al., 2022). A water isotope is a slightly heavier or lighter form of a standard water molecule (H_2O). Water isotopes carry different atomic weights depending on the number of neutrons in their hydrogen or oxygen atoms (Craig, 1961). As water evaporates, lighter isotopes preferentially escape to the atmosphere, leaving the remaining water isotopically enriched in heavier molecules (Craig, 1961). By tracking this fractionation signal across upstream and downstream reaches of beaver-influenced streams, we can infer how beaver ponds modify the water balance through evaporation (Sprenger et al., 2024).

Previous stable isotope studies in this region have characterized snowmelt contributions to streamflow and beaver impacts on water chemistry at the watershed and reach scale (Briggs et al., 2019; Carroll et al., 2022; Dewey et al., 2022; Sprenger et al., 2024), but the extent to which beaver dams influence watershed hydrology through surface water storage, groundwater interactions, and evaporation remains poorly understood. Specifically their application to fine-scale evaporative fractionation within beaver pond systems during record low snowpack conditions remains limited. Determining whether beaver dams produce a consistent and detectable evaporative isotopic signal during years of exceptionally low snowpack has important implications for water resource management and ecological restoration. It is important to enhance our understanding of how beaver-modified watersheds respond to increasing aridity across the western United States, and specifically how much evaporative water loss occurs within beaver pond systems.

The objectives of the study was (i) to characterize whether beaver damming drives evaporative fractionation of stable water isotopes between upstream and downstream reaches, and (ii) to determine whether this evaporative isotopic enrichment is consistent across geographically distinct beaver-influenced sites in a subalpine-to-alpine region.

Methods

Site Description

This study investigated three sites located in subalpine watersheds of Gunnison County, Colorado. Trail Creek (TC; 38.92469° N, 106.60094° W) is a tributary of the Taylor River within Taylor Park, situated at an elevation of 9,681 feet. TC has been an established study site since 2024 through work on floodplain hydrology and biogeochemistry in beaver-impacted river systems. The beaver population at Trail Creek was reestablished following a major restoration effort initiated in 2021, during which over 300 structures were installed over a two year restoration period — including beaver dam analogs (BDAs), sod speedbumps, and woody material structures — were installed through partnerships at the federal, state, and local level. TC represents a relatively young beaver system.

Coal Creek (CC; 38.8564° N, 107.0559° W) is located west of the town of Crested Butte at an elevation of 9,500 feet. Unlike Trail Creek, Coal Creek supports an older, naturally established beaver dam system with no history of restoration.

Italian Creek (IT; 38.95166° N, 106.62920° W) is located directly north of Trail Creek within the Taylor Park area. Italian Creek was selected as an undammed control site because of its similar geology, climate, and elevation to TC, while lacking beaver damming activity.

Beaver Dam Characterization

Beaver dams at Trail Creek were identified and measured using existing drone imagery and processed in QGIS (Beardsley, 2026). Structures were classified as beaver dams if they met two criteria: visible construction material forming a dam structure, and evidence of ponded water or overflow creating a whitewater cue downstream indicating a difference in water height caused by dam structure. Dam width was measured in meters for all qualifying structures. At Coal Creek, beaver dams were classified during an on-site walking survey of the full study reach, with dam lengths measured manually using a handheld GPS device (Trimble DA2) with the same criteria.

Field

Water sampling: Seven sampling sites have been established across the TC stream corridor from downstream to upstream. River reach sampling captured a spatial gradient of undammed and dammed locations along the study area. In addition, two Teledyne ISCO autosamplers were installed at the downstream and upstream reaches of the sampling sites. Autosamplers were deployed for the purpose of investigating the effects with and without beaver treatment, using continuous data. The upstream Trail Creek sampling and ISCO autosampler functions as a control with no beaver damming. Six sampling sites were established at CC and five for IT from downstream to upstream. IT allows us to isolate the effect of beaver damming on evaporative fractionation of stable water isotopes by comparing isotopic signatures in an unimpacted stream to those observed at beaver-influenced sites. Water samples were collected throughout the upstream and downstream of beaver wetlands on CC and TC, and from upper and lower sections of IT. Samples were then filtered at 0.45 µm using syringe filters to remove particulates. 7.5 mL glass vials were used to minimize gas exchange, as plastic vials are permeable to gases and could promote evaporative fractionation during storage. Each vial was filled completely to create a convex meniscus at the opening so that no air remained inside. An additional wrap of Parafilm was applied over the cap to further prevent evaporation. Samples were stored on ice and in the dark until laboratory analysis.

Streamflow: Streamflow was measured manually using a SonTek FlowTracker handheld Acoustic Doppler Velocimeter (ADV). A measuring tape was first laid across the stream to define a cross-section, and velocity and depth measurements were taken at regular intervals across the section. Measurements were collected above and below the furthest upstream and downstream locations at each sampling location.

At Trail Creek, continuous discharge records were also collected using two ISCO automatic water samplers — one upstream and one downstream each paired with a stream gauge consisting of a pressure transducer and data logger (Onset HOBO MX2001 Bluetooth water level data logger) suspended within a slotted PVC pipe anchored in the streambed. These gauges provided continuous stage/discharge records throughout the sampling season.

Laboratory

Isotope samples were analyzed at Western Colorado University using a Picarro L2130-i, which operates on the principle of laser absorbance spectroscopy with a cavity ringdown detector. A 1500 µL aliquot of each filtered sample was transferred to glass autosampler vials fitted with septa-sealed caps. PPrecision for $\delta^{18}O$ and $\delta^{2}H$ abundance are 0.025‰ and 0.1‰,respectively. Results were reported relative to Vienna Standard Mean Ocean Water (VSMOW). Raw Picarro outputs were processed using ChemCorrect software to apply drift corrections and flag spectral contamination prior to final isotope value extraction.

Data Analysis

Stable isotope ratios ($\delta^{18}O$ and $\delta^{2}H$) were plotted in dual-isotope space and compared to a site-specific Local Meteoric Water Line (LMWL) derived from the Online Isotopes in Precipitation Calculator (Bowen, 2026) to visualize the degree of evaporative signal. Lc-excess values were calculated for all samples relative to the OIPC-derived LMWL using the specified coordinates and elevations from the site description above.

Spatial gradients of lc-excess were visualized by plotting values against cumulative stream length from upstream to downstream at each site. Gradients were used to assess whether evaporative enrichment accumulated progressively along the stream corridor as a function of beaver pond density and water residence time. Cross-site comparisons of mean lc-excess values among TC, CC, and IT were used to evaluate whether the evaporative signal driven by beaver damming was consistent across sites differing in restoration history and degree of beaver influence. To account for differences in reach length among sampling locations, changes in lc-excess were normalized by stream length by calculating the change in lc-excess between consecutive sampling points relative to the corresponding change in stream length between sampling locations. This allowed the magnitude of isotopic change to be compared across reaches with different physical distances.

A power-law regression was used to develop a rating curve relating water level to discharge at Trail Creek. Manual discharge measurements were paired with continuous water level measurements recorded at 10-minute intervals, and the resulting relationship was used to estimate discharge throughout the sampling season.

Results

Hydrology

The 2025-2026 water year was a low snowpack year, and this was reflected physically in the discharge record at the Trail Creek ISCO stations (Figure 7). Downstream discharge exceeded upstream discharge throughout the snowmelt in April and May. This pattern reversed on June 8, 2026 when the two hydrographs crossed, and from that point forward downstream discharge remained consistently lower than upstream through the end of the sampling period – apart from the brief, temporary reversal during a late-June precipitation event. This crossover marks a fundamental shift in the river reach behavior: a stream that had been gaining water through peak snowmelt began losing water as the summer progressed. At the manually sampled Coal Creek and Italian Creek sites, by contrast, downstream discharge exceeded upstream discharge on every sampling date (Table 8), indicating these creeks remained net gaining even as flow declined seasonally.

Relationship between discharge and lc-excess

This shift in relative discharge tracked closely with the isotopic pattern. Lc-excess was positively correlated with discharge at both stations, more strongly downstream ($r = 0.965$, CI 0.938-0.981, $n = 46$) than upstream ($r = 0.609$, CI 0.368-0.774, $n = 40$; Table 4; Figure 6), meaning that as downstream flow dropped below the upstream flow, the downstream lc-excess became more negative. The reach that lost the most water also carried the heaviest isotopic signature, directly consistent with evaporative loss. At maximum discharge (May 14, 2026), upstream and downstream lc-excess differed by just 0.555‰; as snowmelt inputs declined and downstream flow fell below upstream flow, the two stations diverged sharply, most visibly from early July onward. The late-June precipitation event that briefly reversed the discharge crossover (Figure 7) also interrupted the isotopic trend. Lc-excess became less negative at both stations before resuming its decline - illustrating that the isotopic signal is responding directly to discharge.

At Coal Creek and Italian Creek, however, this net-gaining (Table 8) did not translate to the same isotopic magnitude seen at Trail Creek. Both creeks still showed a downstream decline in lc-excess (Table 3; Figure 2). At CC this suggests that evaporative enrichment in the beaver-influenced ponds outpaces the effects of increasing discharge. At IT, the undammed control, lc-excess remained much closer to the LMWL (Table 7), and its downstream decline was weaker. In contrast to the Trail Creek ISCO stations, where declining downstream discharge coincided with progressively more negative lc-excess points.

Seasonal patterns of lc-excess

Lc-excess declined significantly across the sampling season at all the locations (fixed effect: -0.047‰/day, accounting for location and site-level random effects; Table 2; N = 115 observations, 20 sites). This seasonal decline was most pronounced at the Trail Creek ISCO autosamplers (Figure 1). At the upstream station, mean lc-excess dropped from −0.727‰ before peak flow ($n = 24$, $SD = 0.352$) to −2.980‰ after peak flow ($n = 62$, $SD = 1.739$; Table 5a). The difference was significant by both Welch's two-sample t-test ($t = -9.70$, $df = 72.46$, $p < 0.001$) and Mann-Whitney U test ($U = 9.00$, $p < 0.001$; Table 5b).

Spatial changes along each stream

Trail Creek showed a consistent significant downstream decline in lc-excess across all three sampling dates (Figure 2, middle panel), with the largest single-reach drop at TC-BDA1, the first beaver dam analog (Figure 3b), and significant relationship with cumulative dam count and cumulative dam length (Table 3). Coal Creek's spatial spatial pattern was weaker and less consistent – downstream declines on both sampling dates were non-significant (Figure 2, right panel), and dam metric relationship varied by date reaching significance for dam length on 6/23 (Table 3). The CC-3 tributary (between CC-2 and

CC-4) carried a lighter isotopic signature than the surrounding main channel sites, but downstream sites remained negative despite this input. Italian Creek, the undammed control, showed the weakest downstream pattern: non-significant on 7/1 ($R^2 = 0.278$, $p = 0.3617$), but significant on 7/16 ($R^2 = 0.837$, $p = 0.0295$) and 7/23 ($R^2 = 0.804$, $p = 0.0391$; Table 3; Figure 2, left panel; Figure 3a). Across the sampling period, Italian Creek averaged 1.648‰ versus -3.513‰ at Trail Creek (Table 7). Italian Creek samples (IT-1-4) were flagged by ChemCorret for elevated C2+ alcohol but were retained because its isotope values were not anomalous relative to surrounding observations.

Comparison among watersheds

Lc-excess differed significantly among the sampling locations, establishing unique baseline isotopic signatures across the watersheds (one-way ANOVA: $F(3,111) = 36.53$, $p < 0.001$; Table 1; Figure 4). Tukey HSD confirming all pairwise differences except upstream TC and CC (−0.75‰, $p = 0.3520$; Table 1) – likely reflecting distinct bedrock and landscape among watersheds. These differences remained significant in the mixed model after accounting for the date and site ($F(3,15.08) = 43.10$, $p < 0.001$; Table 2). Italian Creek's lc-excess was 2.917‰ higher than the reference location, while Trail Creek 2.887‰ lower and upstream Trail Creek 1.552‰ lower, with an overall seasonal decline of 0.047‰/day (SE = 0.005, $p < 0.001$). Beyond these baselines, upstream sites were consistently less evaporatively enriched than downstream sites across all three watersheds, but the rate of that enrichment varied significantly by watershed (reach x creek interaction: $F(2, 5.00) = 17.38$, $p = 0.0056$; Table 6b). Relative to the CC baseline (1.901‰ enrichment from upstream to downstream, $p = 0.0017$; Table 6a), the undammed control at IT showed a significantantly weaker gradient (-1.108‰ interaction, $p = 0.041$), while TC trended toward a steeper gradient (+1.022‰ interaction, $p = 0.0526$; Table 6a).

Dual-isotope evidence of evaporative enrichment

Site-specific local meteoric water lines (LMWLs) were slightly shallower slopes than the Global Meteoric Water Line (GMWL; slope = 8, intercept = 10) at all three watersheds: 7.94 (TC), 7.93 (CC), and 7.89 (IT), with intercepts of 9.37, 8.97, and 8.67 respectively (Figure 5a). Samples fell below and to the right of their LMWL with increasing enrichment. Trail Creek and upstream Trail Creek showed the widest spread below the line, while Italian Creek clustered tightly around it – a pattern consistent with the lc-excess results above.

Discussion

Beaver-modified reaches exhibit stronger evaporative enrichment

Across the three watersheds, the spatial gradients provide stronger evidence for beaver-associated evaporative water loss than absolute differences in isotope composition among watersheds. Trail Creek showed the most consistent downstream decline in lc-excess, Coal Creek showed a weaker but generally negative pattern, and Italian Creek showed the least consistent spatial response. Although the magnitude of the gradient differed among streams, the repeated negative direction of change at the beaver-influenced sites is consistent with increasing evaporative enrichment downstream. The difference in gradient strength among sites may reflect variation in dam density, pond structure, restoration history, floodplain morphology, and underlying hydrology.

Of the three watersheds, Trail Creek provides the clearest example of this pattern. The consistent downstream decline in lc-excess at Trail Creek across all three sampling dates provides strong evidence that conditions associated with beaver dams are related to evaporative enrichment (Figure 2; Table 3). The same negative pattern was also observed with increasing cumulative dam count and dam length (Table 3). This is consistent with the mechanism proposed by (Wegener et al., 2017) and (Briggs et al., 2019), in which beaver dams slow channel flow, expand the wetted and ponded surface area, and extend water residence time in the river corridor (Schultz et al., 2026), all of which increase the opportunity for evaporative loss before water moves downstream. Sprenger et al.documented a comparable evaporation-driven isotope enrichment signal along a peatland drainage network with similarly slowed flow, supporting the interpretation that residence time, rather than simply channel length, may be an important factor influencing the isotope signal.

This interpretation is further supported by the dual-isotope pattern where a greater deviation observed at Trail Creek may reflect greater exposure of stream water to ponded conditions created by its dense network of restoration structures. Increased surface-water residence time and ponded surface area at beaver-influenced sites likely drive evaporative fractionation, consistent with Trail Creek samples deviating further from the LMWL than the undammed Italian Creek control (Figure 5a).

Italian Creek served as the undammed control, providing a comparison for evaluating whether downstream changes observed at Trail Creek and Coal Creek were associated with beaver influence rather than simply reflecting spatial or seasonal variation. The weaker and less consistent response at Italian Creek suggests that downstream isotopic enrichment is not simply an inherent characteristic of these stream systems. Unlike Trail Creek, where lc-excess consistently declined downstream across sampling dates. The downstream relationship was not significant on 7/1 but became significant on 7/16 and 7/23, although the fitted slopes were relatively small (Table 3; Figure 2; Figure 3a). This interpretation should be considered alongside a limitation in the final Italian Creek sampling date. The samples from IT-1-4, were flagged as potentially contaminated after analysis on the Picarro instrument and in ChemCorrect due to elevated C2+ alcohol values. Alcohol is an interference in the Picarro instrument because it can mimic the target gas signatures and can create false readings. The sample was rerun, but the flagged values were retained. However, it did not appear anomalous relative to the surrounding observations and there was no clear indication that the contamination shifted the isotope values in a particular direction.

Through time, lc-excess at Italian Creek remained relatively positive and did not show the strong progressive enrichment observed at the beaver-influenced sites. Although some temporal change occurred, the magnitude of this change was comparatively small. This suggests that downstream isotopic shifts are not an inevitable feature of the stream system and provides support for interpreting the stronger gradients at Trail Creek as being associated with differences in channel structure and water storage.

The reach × creek interaction provides the strongest direct test of whether the magnitude of upstream-to-downstream isotopic change was consistent among watersheds. The interaction was significant ($F(2,5) = 17.38$, $p = 0.0056$), while both reach ($F(1,5) = 138.08$, $p = 0.0001$) and creek ($F(2,5) = 15.43$, $p = 0.0073$) also had significant effects (Table 6b). Thus, lc-excess differed between upstream and downstream of the river reaches, but the magnitude of that difference depended on the creek. The results therefore support a generally consistent downstream enrichment pattern at the beaver-influenced sites while indicating that the magnitude of the response was not uniform among watersheds. The presence of beaver-associated structures appears to be associated with downstream enrichment, but the strength of that enrichment depends on characteristics of the individual watershed.

Evaporative enrichment increases under low-flow conditions

Having established that the magnitude of downstream enrichment differs among watersheds, the relationship between lc-excess and discharge provides insights into the conditions under which enrichment is strongest. The discharge relationship provides a hydrologic context for the isotope signal. At Trail Creek, lc-excess was positively correlated with discharge at both ISCO stations, with a substantially stronger relationship downstream ($r = 0.965$, 95% CI = 0.938–0.981, $n = 46$) than upstream ($r = 0.609$, 95% CI = 0.368–0.774, $n = 40$; Table 4; Figure 6). This stronger downstream relationship may reflect the greater influence of ponded conditions and altered residence time within the beaver-modified reach. More negative lc-excess indicates greater evaporative enrichment; therefore, the stronger downstream relationship suggests that isotope composition was more closely coupled to flow conditions within the beaver-influenced reach.

This relationship was also evident through time (Figure 7). During the period of highest discharge, lc-excess was also relatively high at both the upstream and downstream Trail Creek ISCO stations. As discharge declined following the seasonal peak, lc-excess became progressively more negative, with the downstream station showing a stronger shift than the upstream station. The two stations began to diverge more clearly in early July as discharge declined. This pattern suggests that lower-flow conditions may increase the opportunity for evaporative enrichment within the beaver-influenced reach. Conversely, evaporative water loss from the expanded pond surface area may itself contribute to reduced downstream discharge, suggesting a potential feedback between flow and evaporation rather than a strictly one-directional relationship. During the later portion of the sampling period, lc-excess became less negative as discharge increased again, coinciding with the late June precipitation event. The precipitation event provides a short-term counterexample to the declining seasonal pattern.

The pre-versus post-peak comparison provides an additional indication of the seasonal shift. The mean lc-excess at the upstream and downstream TC ISCO autosamplers declined from −0.727‰ before the May 14, 2026 peak-flow date ($n = 24$, $SD = 0.352$) to −2.980‰ after peak flow ($n = 62$, $SD = 1.739$; Table 5a). This difference was significant by both Welch's t-test ($t = -9.70$, $df = 72.46$, $p < 0.001$) and the Mann-Whitney U test ($U = 9.00$, $p < 0.001$; Table 5b). The seasonal shift therefore extended beyond a single sampling date, although it should not be attributed to beaver activity alone because it coincided with the seasonal decline in discharge.

The magnitude of the response to the beaver damming depends on local watershed characteristics

Coal Creek showed the same downstream decline in lc-excess as Trail Creek, though with weaker statistical support. This difference may partly reflect lower statistical power, as Coal Creek had fewer sampling locations and lacked the upstream control reach that is present at Trail Creek. However, differences in the structure and hydrology of the two watersheds may also contribute to the weaker gradient at Coal Creek.

Trail Creek's dams are the product of a two-year restoration effort using beaver dam analogs, sod speedbumps, and woody structures, whereas Coal Creek's dams are older and naturally established. Differences in dam spacing, age, and structural integrity could alter the amount of surface area and water residence time within each reach, potentially influencing the magnitude of evaporation enrichment.Thus, the weaker Coal Creek gradient does not necessarily contradict the broader relationship between beaver modification and evaporation enrichment. Instead, it suggests that the hydrologic effect of beaver structures may depend on how those structures modify water storage and movement within a particular watershed.

Differences in underlying geology may provide another possible explanation for the contrast between Coal Creek and Trail Creek. Coal Creek's shale bedrock, in contrast to Trail Creek's granitic bedrock, may contribute to differences in groundwater contribution or subsurface exchange that are not captured by the surface-water isotope gradient alone. The layered and fractured nature of shale may facilitate greater lateral inputs from hillslope subsurface flow into Coal Creek, potentially introducing water with a different isotopic composition and weakening the downstream evaporation signal. Conversely, the dense granitic bedrock at TC may restrict lateral inputs, allowing the main-channel evaporative signal to become more pronounced downstream. While differences in bedrock geology may contribute to differences in hydrologic connectivity or groundwater contribution (Johnson et al., 2025), these potential differences in hydrologic connectivity cannot be distinguished from the present isotope data alone and should be treated as a hypothesis requiring additional hydrologic evidence.

The particularly large decline at the first beaver dam analog, TC-BDA1 (Figure 3b), further illustrates how local floodplain conditions may amplify the broader evaporation pattern. The large pond surrounding this reach may increase surface water storage and residence time, while the presence of clays and organic matter creates a low permeability later that may influence infiltration, water storage, or exchange between surface water and the surrounding floodplain. A small drainage entering the main channel between P3-DS and TC-BDA1 may also contribute water with a different lighter isotopic composition, potentially affecting the observed change in lc-excess. Together, these features could explain why the largest isotopic shift occurs at this particular transition rather than being distributed evenly among dams.

Another example of hydrologic complexity occurs at CC-3, a tributary that enters the main Coal Creek channel between CC-2 and CC-4. The isotopic signature at CC-3 was lighter than that of the main channels, indicating that this tributary introduced water with a different isotopic composition. Although the source of this isotopic difference cannot be determined from the present data alone, the additional input provides an example of how tributary inflows can modify the downstream isotope signal. Despite the lighter input at CC-3, lc-excess remained negative at CC-4 and farther downstream, suggesting that the downstream influence associated with the beaver-affected channel continued after the tributary input. Thus, CC-3 may partially modify the isotopic signal without eliminating the broader downstream pattern.

Implications for beaver restoration and future water-balance research

Collectively, these results establish that beaver damming exerts a measurable influence on stable water isotope composition in subalpine headwater streams through evaporative fractionation, with effect magnitude amplified by dam density, water residence time, and low-flow conditions. The site-to-site variation in response strength underscores that the isotopic consequences of beaver activity are not uniform but are instead shaped by the interacting influences of restoration history, floodplain geometry, and bedrock geology. These findings contribute to a growing body of evidence that beaver-modified reaches function as hydrologically distinct units within their watersheds, and highlight the need to account for evaporative water loss when assessing the net water balance benefits of beaver dam restoration in an increasingly arid and snow-limited West. Future work should quantify the volume of water lost through evaporation within beaver-modified reaches.

Figures

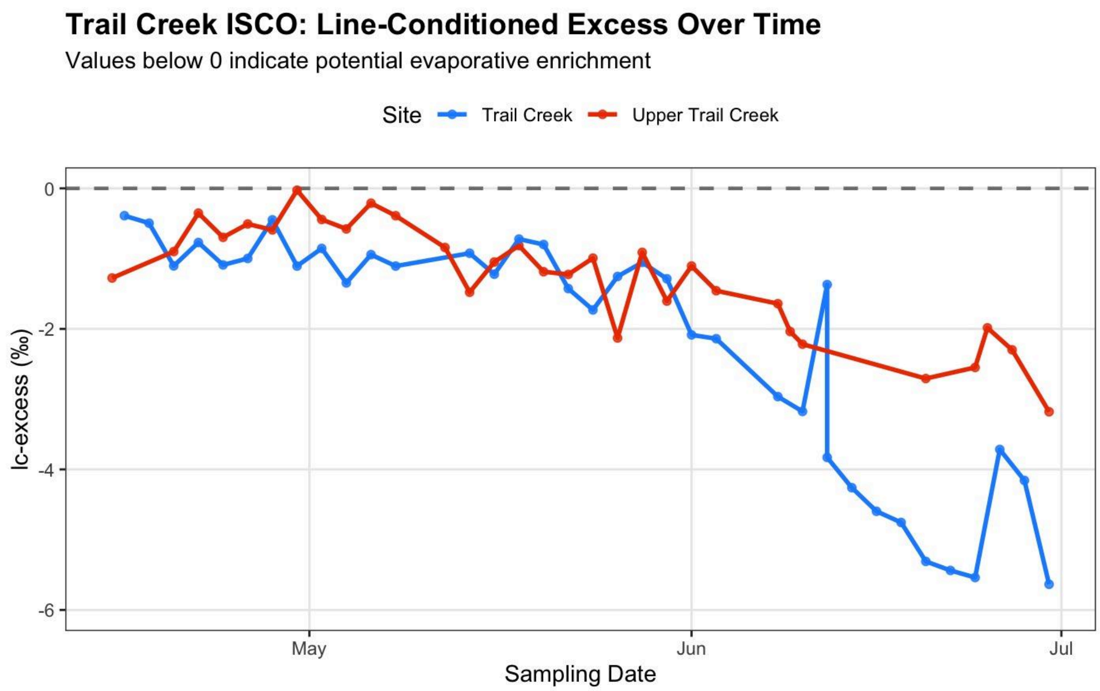

Figure 1. Line-conditioned excess over time at the Trail Creek ISCO auto-samplers.

Lc-excess (‰) is plotted against sampling date for the downstream Trail Creek ISCO (blue) and upstream Trail Creek ISCO (red/orange). Values below the dashed line at 0 indicate potential evaporative enrichment.

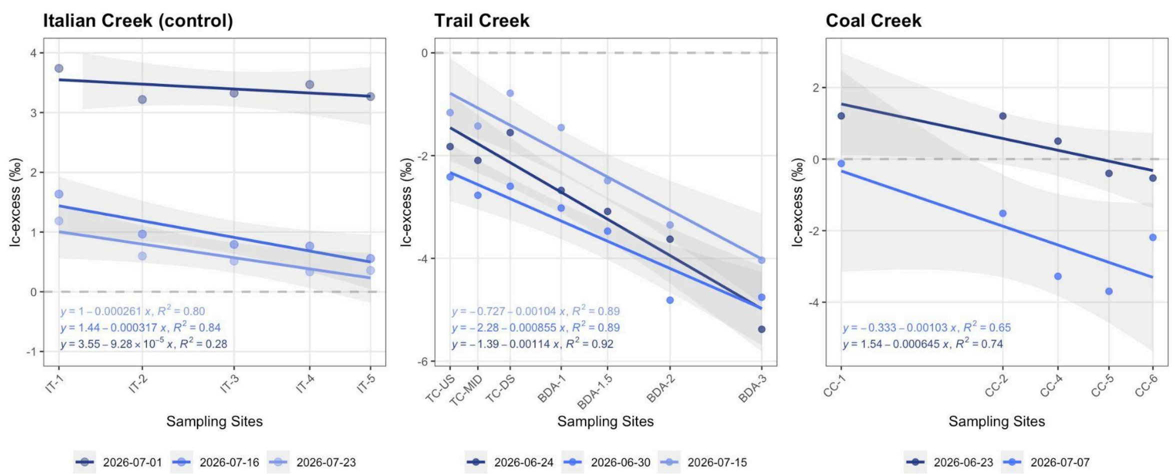

Figure 2. Spatial patterns in lc-excess across the three study streams.

Lc-excess (‰) is plotted by sampling sites for Italian Creek (control), Trail Creek, and Coal Creek. Line color and slope equations correspond to individual sampling dates; site spacing on the x-axis reflects normalized stream distance between sites, not equal intervals.

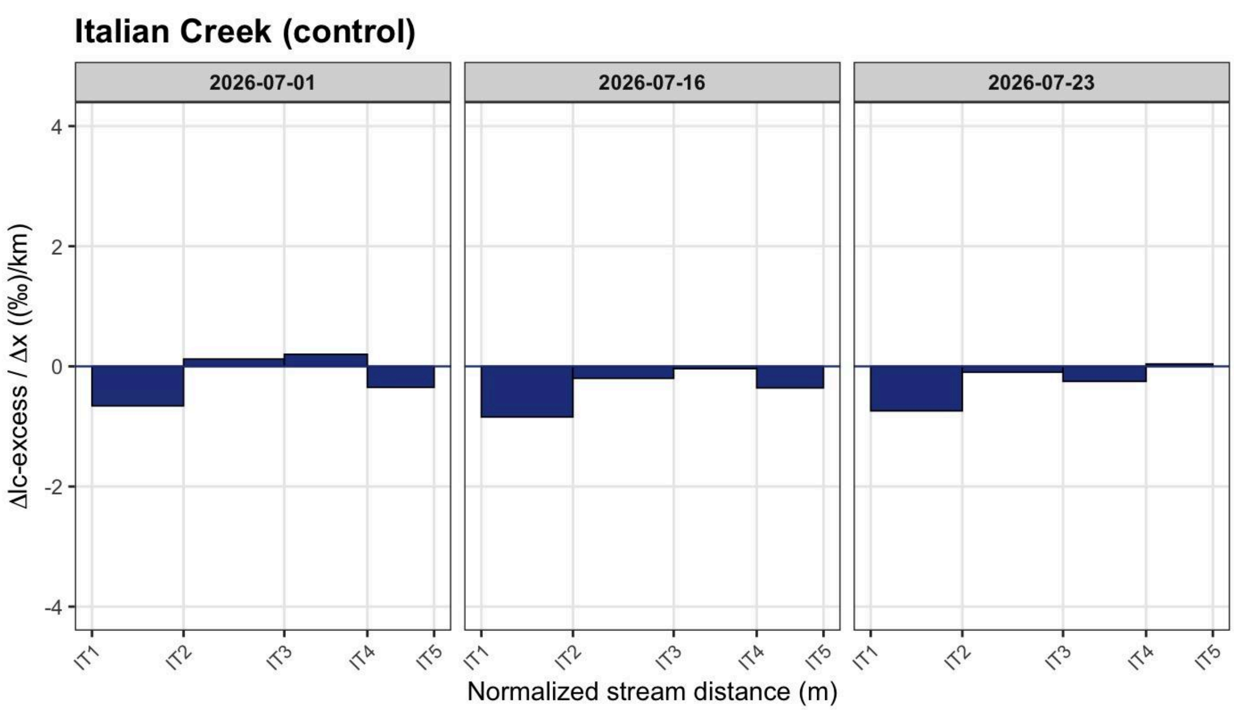

Figure 3. Line-Conditioned Excess gradient at Italian Creek (control).

Change in lc-excess per unit stream distance (Δlc-excess/Δx, (‰/km)) is by site for three sampling dates. The site spacing reflects the normalized stream distance between sites.

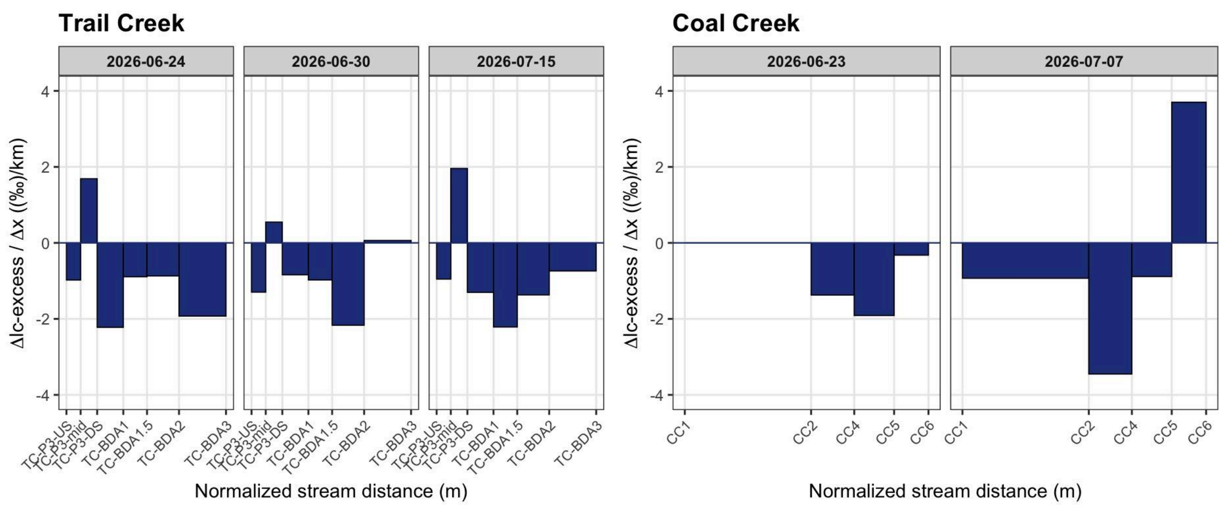

Figure 3a. Lc-excess gradient by stream distance at Trail Creek and Coal Creek.

Change in lc-excess per unit stream distance (Δlc-excess/Δx, (‰/km)) is shown by site for each creek across their sampling dates. Site spacing reflects the normalized stream distance between sites.

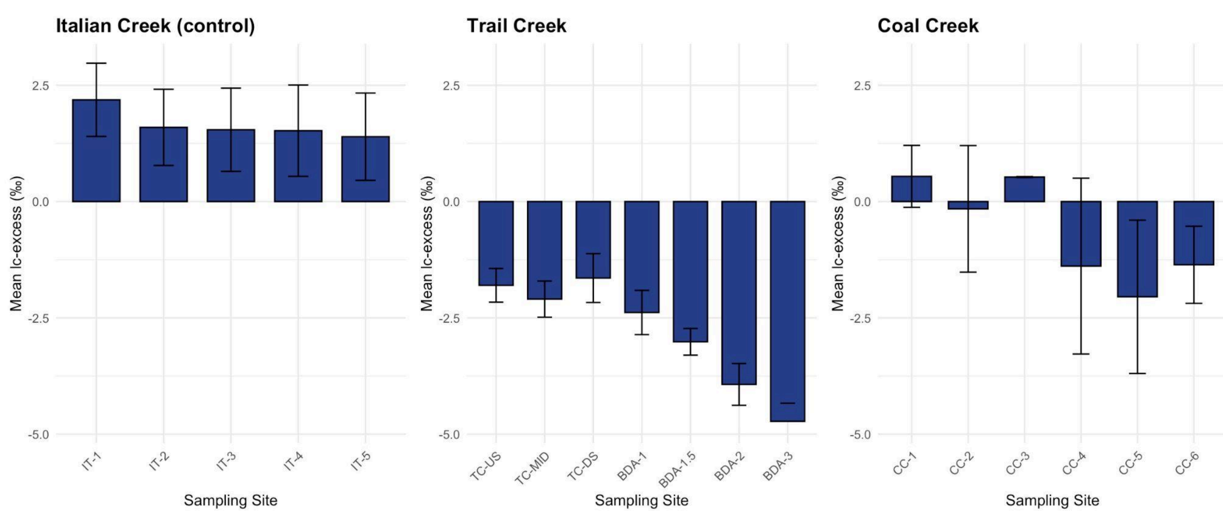

Figure 4. Mean lc-excess by sampling sites across the three study creeks.

The bars show mean lc-excess (‰), with error bars (SD) for each site at Italian Creek (control), Trail Creek and Coal Creek.

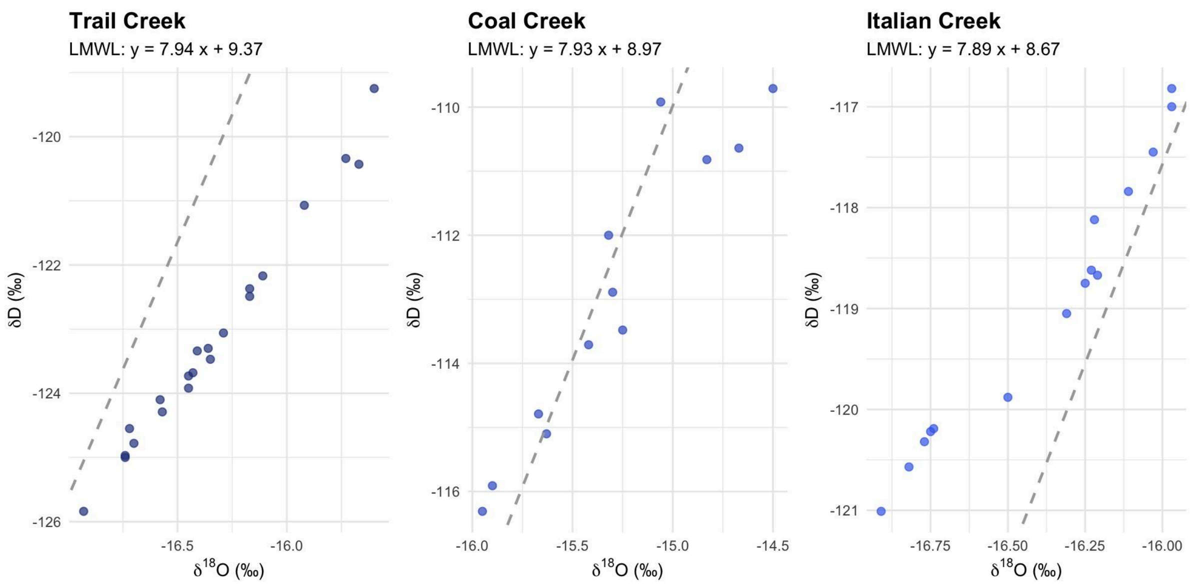

Figure 5. Dual-Isotope plots for Trail Creek, Coal Creek, and Italian Creek.

Each panel shows stream water isotope samples relative to the site-specific local meteoric water line (LMWL, dashed line), with the equation of the LWML above the plot.

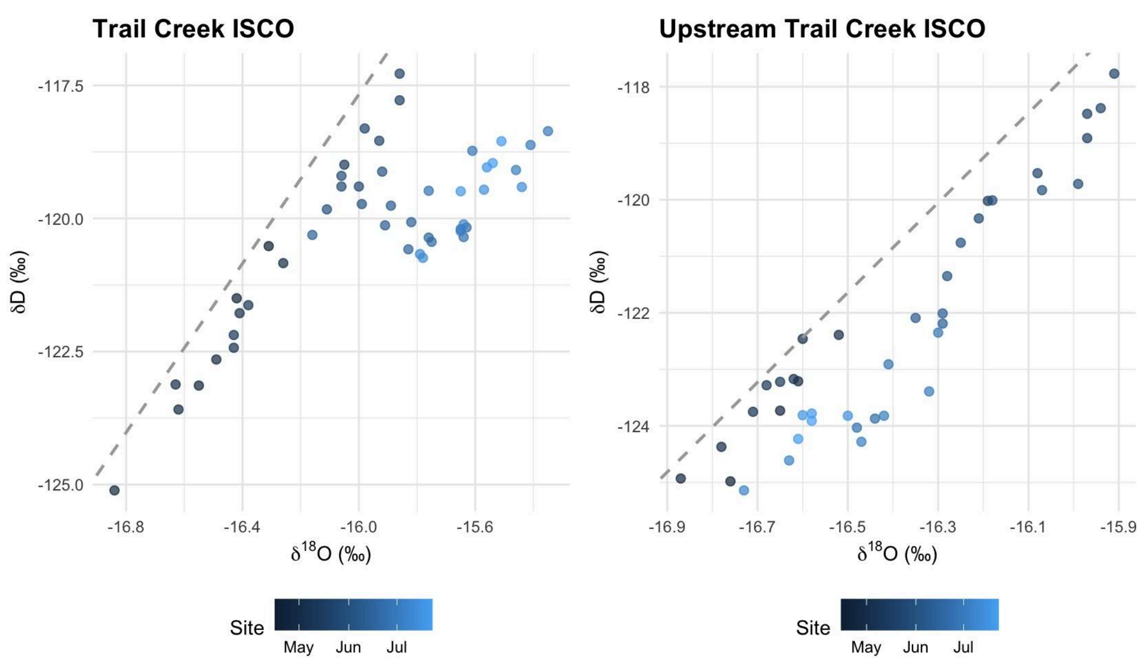

Figure 5a. Dual-Isotope plots for the Trail Creek ISCO auto-samplers.

Points are colored by the sampling date (May – July) for the downstream ISCO and upstream ISCO; the dashed line represents the LMWL.

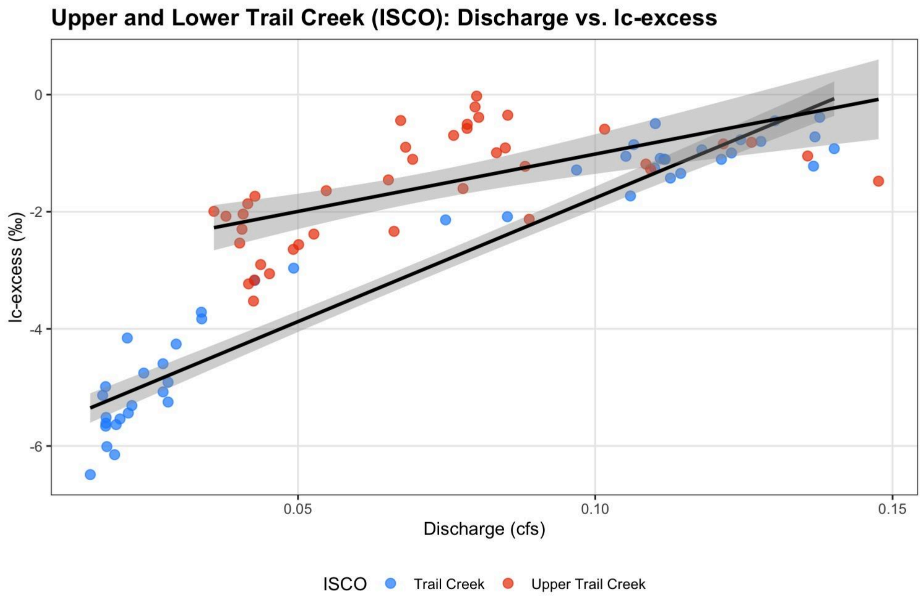

Figure 6. Relationship between discharge and lc-excess at the upstream and downstream Trail Creek ISCO stations.

Lc-excess (‰) is plotted against discharge (cfs) for the downstream TC ISCO (blue) and upstream TC ISCO (orange/red), with a linear regression and 95% confidence interval.

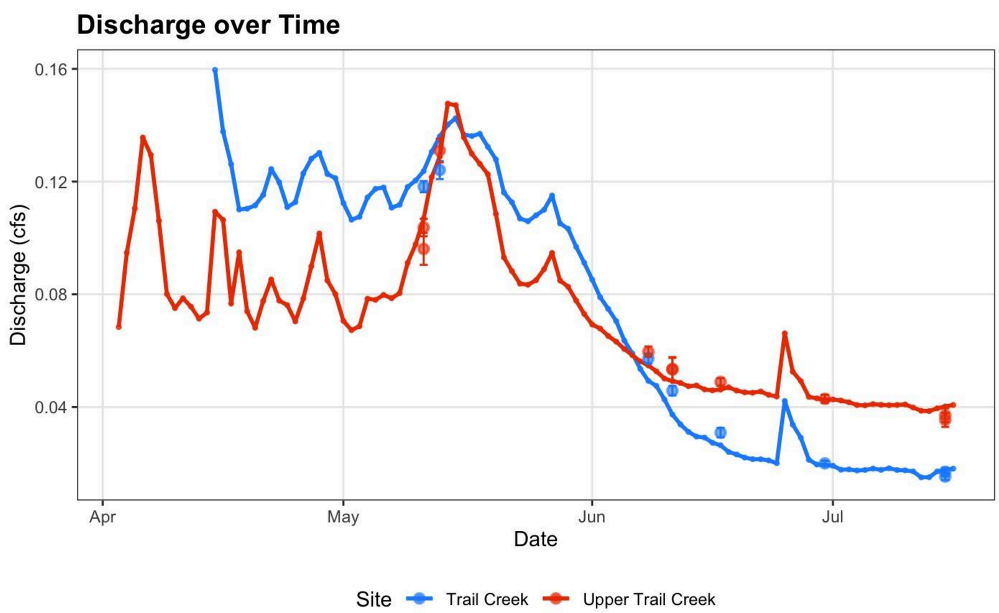

Figure 7. Discharge over time at the upstream and downstream Trail Creek ISCO stations.

Continuous discharge (cfs) at the downstream (blue) and upstream (orange) Trail Creek ISCO stations from April to July 2026. Points with error bars show manual discharge measurements (± % error) collected at Trail Creek over the same period. Discharge over time at the upstream and downstream Trail Creek ISCO stations.

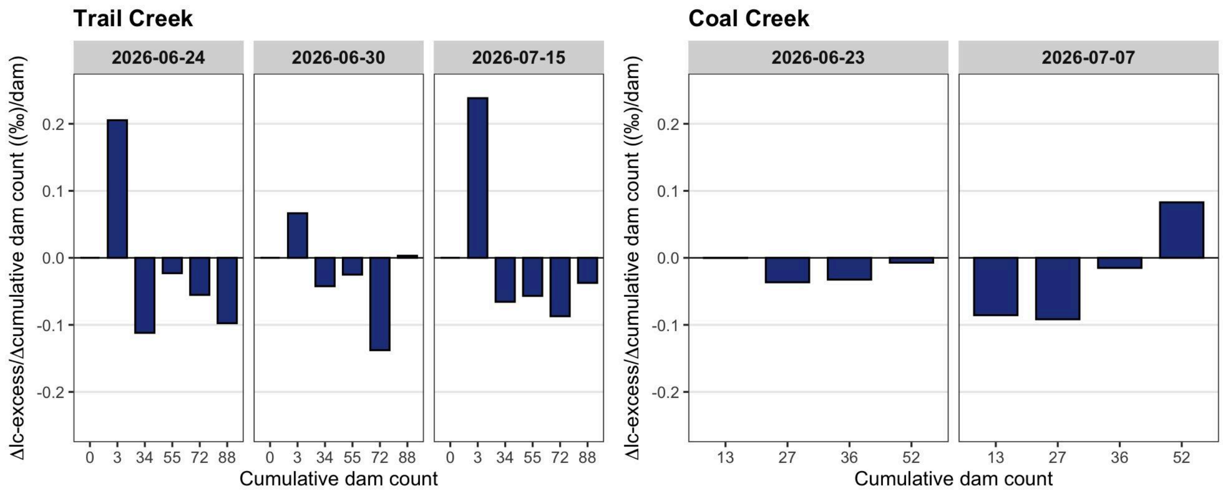

Figure 8. Lc-excess gradient by cumulative dam count at Trail Creek and Coal Creek.

Change in lc-excess per unit cumulative dam count (Δlc-excess/Δcumulative dam count, (‰/dam)) is shown by sampling date for each creek.

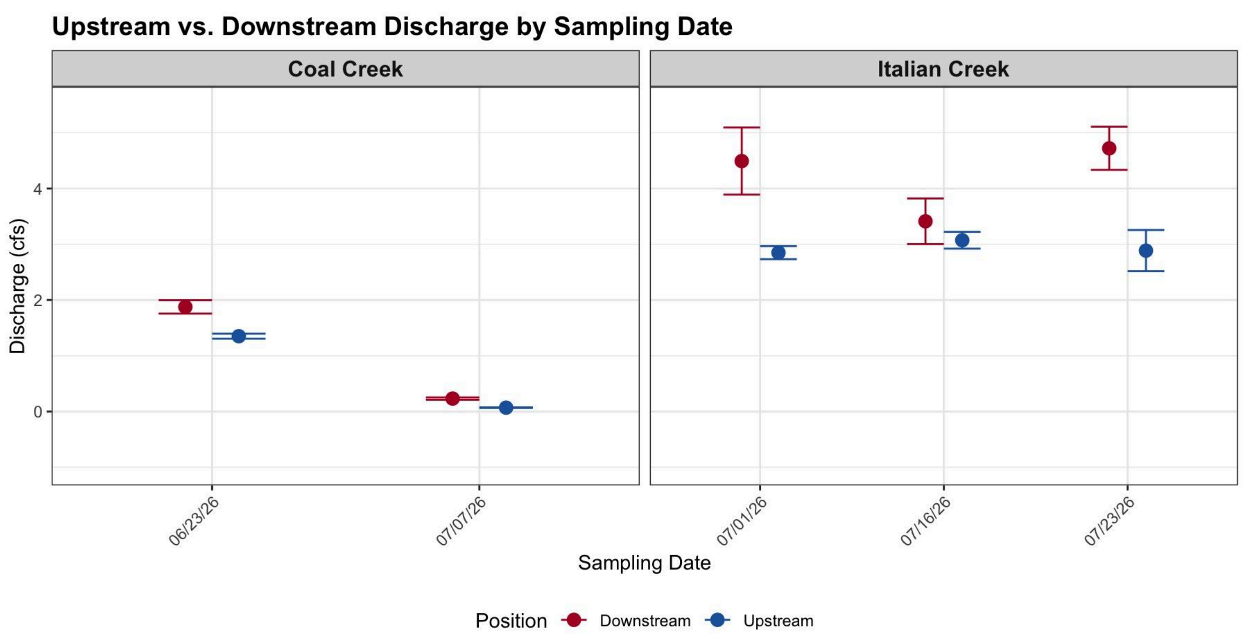

Figure 9. Manual discharge measurements at the upstream and downstream sites of Coal Creek (CC-1, CC-6) and Italian Creek (IT-1, IT-5) on each sampling date. Points show discharge (cfs) with error bars representing measurement error.

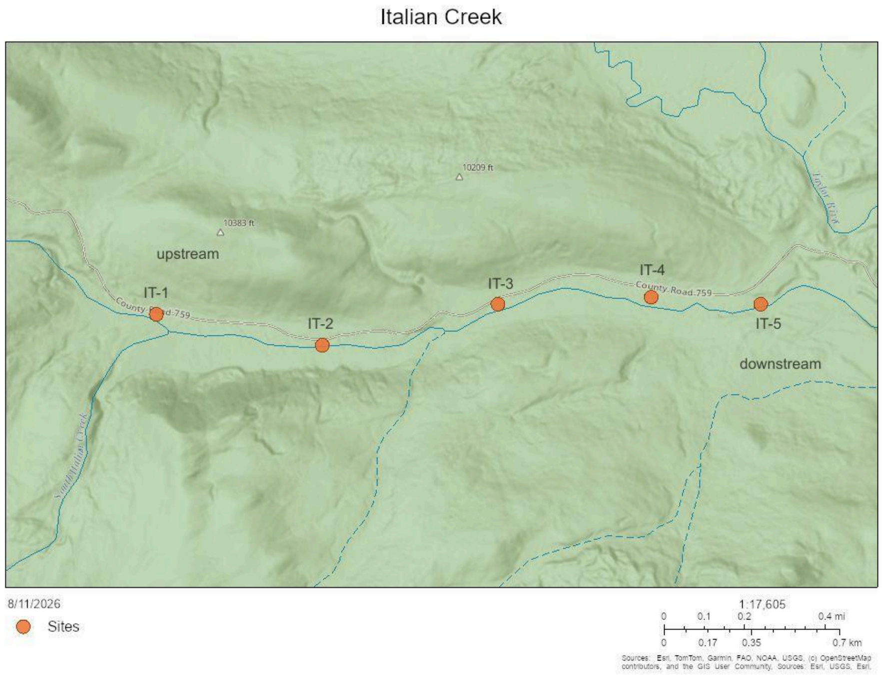

Figure 10. Map of sampling sites along Italian Creek (control).

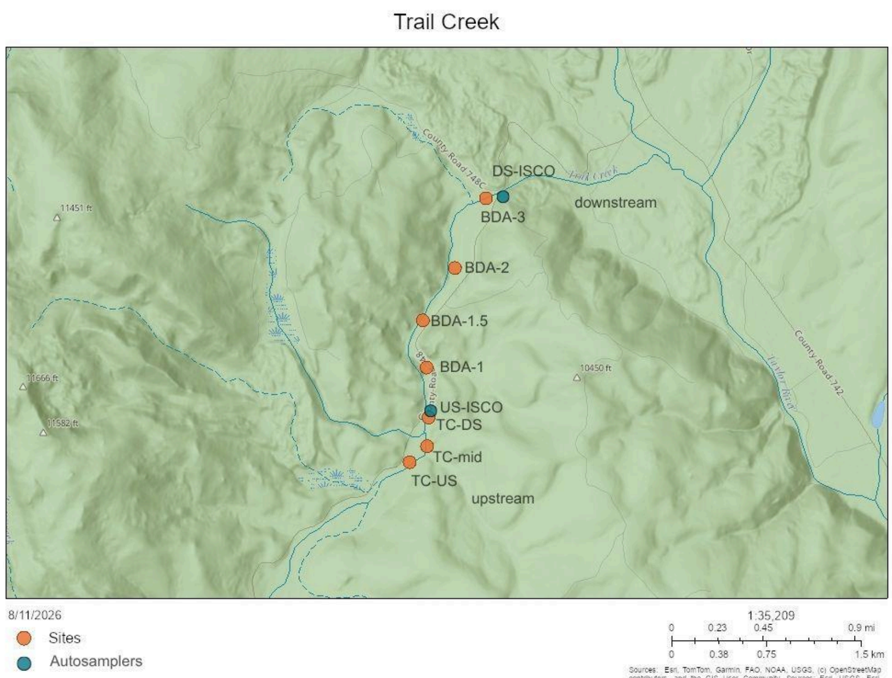

Figure 11. Map of sampling sites and ISCO auto-sampler locations along Trail Creek.

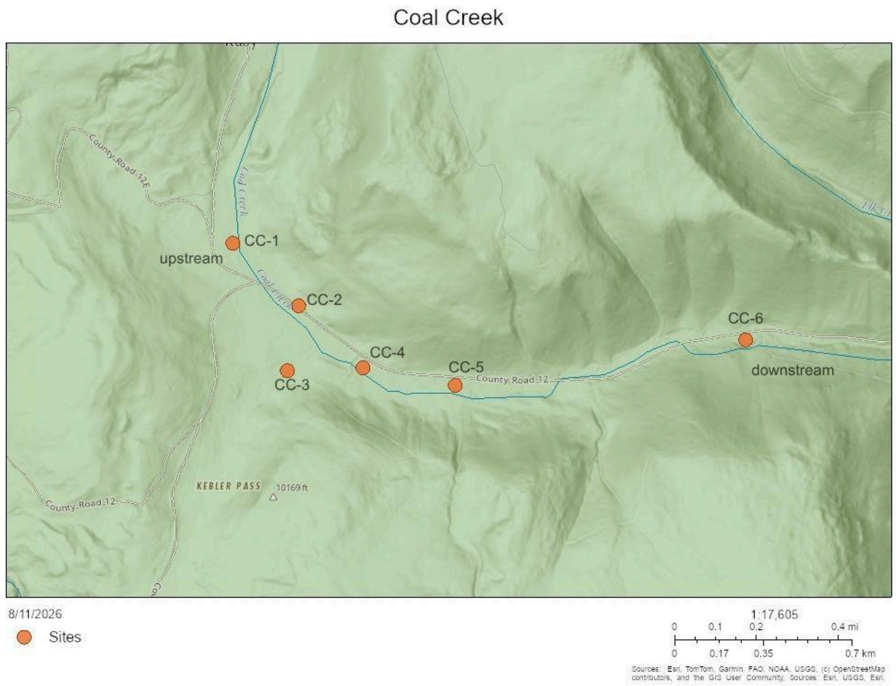

Figure 12. Map of sampling sites along Coal Creek.

Tables

Comparison	Mean diff.	95% CI (lower)	95% CI (upper)	Adj. p
it-cc	2.30	0.90	3.69	0.0002
tc-cc	-1.90	-3.06	-0.74	0.0003
utc-cc	-0.75	-1.94	0.43	0.3520
tc-it	-4.19	-5.26	-3.13	0.0000
utc-it	-3.05	-4.14	-1.96	0.0000
utc-tc	1.15	0.37	1.92	0.0010

Table 1. Pairwise comparisons of mean lc-excess among the four sampling locations (Italian Creek [it], Trail Creek [tc], and Coal Creek [cc]), based on Tukey HSD following one-way ANOVA ($F(3,111) = 36.53$, $p < 0.001$). Mean differences, 95% confidence intervals, and adjusted p-values are shown for each pairwise comparison.

Term	Estimate	SE	df	t	p
(Intercept)	0.286	0.389	25.191	0.735	0.4690
locationit	2.917	0.537	19.550	5.429	0.0000
locationtc	-2.887	0.521	16.754	-5.542	0.0000
locationutc	-1.552	0.549	15.908	-2.828	0.0122
date_c	-0.047	0.005	109.921	-10.179	0.0000

Table 2. Linear mixed-effects model of lc-excess as a function of sampling location and date, with a random intercept for site. Coefficient estimates, standard errors, degrees of freedom, t-values, and p-values are shown for each fixed effect (N = 115 observations across 20 sites).

	Date	Intercept	Slope	SE (slope)	R^2	p
Stream distance (m)						
Trail Creek	6/24	-1.391	-0.001	0.000	0.917	0.0007
Trail Creek	6/30	-2.280	-0.001	0.000	0.893	0.0013
Trail Creek	7/15	-1.103	-0.001	0.000	0.892	0.0014
Coal Creek	6/23	1.539	-0.001	0.000	0.740	0.0614
Coal Creek	7/7	-0.333	-0.001	0.000	0.652	0.0986
Italian Creek	7/1	3.549	0.000	0.000	0.278	0.3617
Italian Creek	7/16	1.438	0.000	0.000	0.837	0.0295
Italian Creek	7/23	1.005	0.000	0.000	0.804	0.0391
Cumulative dams (n)						
Trail Creek	6/24	-1.613	-0.035	0.007	0.867	0.0069
Trail Creek	6/30	-2.501	-0.025	0.005	0.874	0.0062
Trail Creek	7/15	-1.241	-0.033	0.005	0.906	0.0035
Coal Creek	6/23	1.694	-0.047	0.012	0.885	0.0593
Coal Creek	7/7	-2.138	-0.017	0.041	0.074	0.7276
Cumulative dam length (m)						
Trail Creek	6/24	-1.716	-0.056	0.008	0.923	0.0023
Trail Creek	6/30	-2.602	-0.040	0.007	0.884	0.0053
Trail Creek	7/15	-1.349	-0.052	0.006	0.947	0.0011
Coal Creek	6/23	1.548	-0.028	0.004	0.958	0.0214
Coal Creek	7/7	-2.113	-0.011	0.023	0.108	0.6709

Table 3. Spatial gradient regressions of lc-excess against normalized stream distance, cumulative beaver dam count, and cumulative dam length, by creek and sampling date. Regression intercept, slopes, standard errors, R^2, and p-values are shown for creek-date combination.

Reach	Pearson r	95% CI (lower)	95% CI (upper)	p	n	Regression slope
Upstream	0.609	0.368	0.774	0.0000	40	19.597
Downstream	0.965	0.938	0.981	0.0000	46	42.209

Table 4. Relationship between discharge and lc-excess at the upstream and downstream Trail Creek ISCO stations. Pearson correlation coefficients (with 95% confidence intervals), p-values, sample sizes, and linear regression slopes for reach are shown.

Period	n	Mean	SD	Median
post-peak	62	-2.980	1.739	-2.458
pre-peak	24	-0.727	0.352	-0.734

Table 5a. Summary statistics of lc-excess before and after peak flow at the upstream and downstream Trail Creek ISCO stations (peak flow occurred 5/14/2026). Sample size, mean, standard deviation, and median are shown for the pre-peak and post-peak.

Test	Statistic	df	p
Welch two-sample t-test	-9.70	72.46	0.0000
Mann-Whitney U	9.00	NA	0.0000
F test (equality of variance)	5.35	27.00	0.0057

Table 5b. Statistical tests comparing pre- vs. post-peak lc-excess at the upstream and downstream Trail Creek ISCO stations, including Welch two sample t-test, Mann-Whitney U test and F-test for equality of variance.

Term	Estimate	SE	df	t	p
(Intercept)	-1.360	0.805	5.392	-1.689	0.1477
reach upstream	1.901	0.313	5.000	6.075	0.0017
creekItalian Creek	2.753	1.039	5.392	2.650	0.0422
creekTrail Creek	-3.364	1.039	5.392	-3.238	0.0206
reachupstream:creekItalian Creek	-1.108	0.404	5.000	-2.742	0.0407
reachupstream:creekTrail Creek	1.022	0.404	5.000	2.530	0.0526

Table 6a. Fixed effects of reach (upstream/downstream) and creek on lc-excess, with a random intercept for sampling date. Coefficient estimates, standard euros, degrees of freedom, t-values, and p-values are shown for each fixed effect and interaction.

Term	Num. df	Den. df	F	p
reach	1.00	5.00	138.08	0.0001
creek	2.00	5.00	15.43	0.0073
reach:creek	2.00	5.00	17.38	0.0056

Table 6b. Type III ANOVA testing the effects of reach, creek, and their interactions on lc-excess. Numerator and denominator degrees of freedom, F-statistic, and p-values are shown for each.

Location	N	Mean (‰)	SD (‰)	Median (‰)
cc	12	-0.647	1.669	-0.262
it	15	1.648	1.330	0.966
tc	12	-3.513	1.106	-3.410
utc	9	-1.847	0.676	-1.825

Table 7. Descriptive statistics of lc-excess by sampling location (cc = Coal Creek, it = Italian Creek, tc = Trail Creek, utc = upstream Trail Creek with no beaver impact). Sample size, mean standard deviation, and median are shown for each location.

Date	Creek	Upstream Q (cfs)	Downstream Q (cfs)	Difference (cfs)	% Change
6/23/26	CC	1.351	1.876	0.525	38.860
7/7/26	CC	0.069	0.232	0.163	236.232
7/1/26	IT	2.849	4.492	1.643	57.669
7/16/26	IT	3.072	3.412	0.340	11.068
7/23/26	IT	2.886	4.721	1.835	63.583

Table 8. Paired discharge comparison, upstream vs. downstream sites by creek (cc = Coal Creek, it = Italian Creek) Upstream Q, downstream Q, differences in Q, and percent of change.

Works Cited

Beardsley, M. (2026). *Trail Creek Drone Footage* [Video recording].

Bowen, G. J. (2026). *The Online Isotopes in Precipitation Calculator* (Version version 3.1.) [Computer software]. http://www.waterisotopes.org.

Briggs, M. A., Wang, C., Day-Lewis, F. D., Williams, K. H., Dong, W., & Lane, J. W. (2019). Return flows from beaver ponds enhance floodplain-to-river metals exchange in alluvial mountain catchments. *Science of The Total Environment*, *685*, 357–369. https://doi.org/10.1016/j.scitotenv.2019.05.371

Burchsted, D., Daniels, M., Thorson, R., & Vokoun, J. (2010). The river discontinuum: Applying beaver modifications to baseline conditions for restoration of forested headwaters. *BioScience*, *60*(11), 908-. Gale In Context: Biography.

Burns, D. A., & McDonnell, J. J. (1998). Effects of a beaver pond on runoff processes: Comparison of two headwater catchments. *Journal of Hydrology*, *205*(3–4), 248–264. https://doi.org/10.1016/S0022-1694(98)00081-X

Carroll, R. W. H., Deems, J., Maxwell, R., Sprenger, M., Brown, W., Newman, A., Beutler, C., Bill, M., Hubbard, S. S., & Williams, K. H. (2022). Variability in observed stable water isotopes in snowpack across a mountainous watershed in Colorado. *Hydrological Processes*, *36*(8), e14653. https://doi.org/10.1002/hyp.14653

Craig, H. (1961). Isotopic Variations in Meteoric Waters. *Science*, *133*(3465), 1702–1703.

Dewey, C., Fox, P. M., Bouskill, N. J., Dwivedi, D., Nico, P., & Fendorf, S. (2022). Beaver dams overshadow climate extremes in controlling riparian hydrology and water quality. *Nature Communications*, *13*(1), 6509. https://doi.org/10.1038/s41467-022-34022-0

Johnson, K., Williams, K. H., Christensen, J. N., Carroll, R. W. H., Li, L., Beutler, C., Bird, K. S., Dong, W., & Sullivan, P. L. (2025). Hidden Features: How Subsurface and Landscape Heterogeneity Govern Hydrologic Connectivity and Stream Chemistry in a Montane Watershed. *Hydrological Processes*, *39*(3), e70085. https://doi.org/10.1002/hyp.70085

Naiman, R. J., Melillo, J. M., & Hobbie, J. E. (1986). Ecosystem Alteration of Boreal Forest Streams by Beaver (Castor Canadensis). *Ecology*, *67*(5), 1254–1269. https://doi.org/10.2307/1938681

Pilliod, D. S., Rohde, A. T., Charnley, S., Davee, R. R., Dunham, J. B., Gosnell, H., Grant, G. E., Hausner, M. B., Huntington, J. L., & Nash, C. (2018). Survey of Beaver-related Restoration Practices in Rangeland Streams of the Western USA. *Environmental Management*, *61*(1), 58–68. https://doi.org/10.1007/s00267-017-0957-6

Schultz, K., Morrison, R. R., Rathburn, S., Mertz, C., & Christensen, N. (2026). *Forensic Hydraulic Analysis of Floodplain Connectivity Driven by Historical Beaver Dams in Colorado Headwater Streams*. https://doi.org/https://doi.org/10.1002/rra.70165

Sprenger, M., Carroll, R. W. H., Marchetti, D., Bern, C., Beria, H., Brown, W., Newman, A., Beutler, C., & Williams, K. H. (2024). Stream water sourcing from high-elevation snowpack inferred from stable isotopes of water: A novel application of d-excess values. *Hydrology and Earth System Sciences*, *28*(7), 1711–1723. https://doi.org/10.5194/hess-28-1711-2024

Sprenger, M., Tetzlaff, D., Tunaley, C., Dick, J., & Soulsby, C. (2017). Evaporation fractionation in a peatland drainage network affects stream water isotope composition. *Water Resources Research*, *53*(1), 851–866. https://doi.org/10.1002/2016WR019258

Wegener, P., Covino, T., & Wohl, E. (2017). Beaver-mediated lateral hydrologic connectivity, fluvial carbon and nutrient flux, and aquatic ecosystem metabolism. *Water Resources Research*, *53*(6), 4606–4623. https://doi.org/10.1002/2016WR019790