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Dear Editor and Reviewers,

We would like to thank you for the time and effort you put into the revision of this submission. We appreciate the opportunity to revise the paper for publication in Biogeosciences, and we are grateful for all the insightful suggestions. All the comments have been carefully considered and the manuscript has been revised accordingly. Please see our responses and detailed explanations of the subsequent modifications (marked in red) below:

**EDITORIAL DECISION**

Thank you very much for preparing your answers to the reviewer critique. I have in the meantime acquired a third reviewer opinion that arrived after the reviewing window was closed yet is rich in very constructive comments that I urge you to address. I believe addressing these comments will significantly improve your paper. I would like to specifically comment the following points, which were raised in this third review and also bear some resemblance with other points of critique by other reviewers:

**Answer:** Thank you for summarising the selection of the most important points for improvement. Taking the third review into account did indeed improve the clarity of the paper. As we have received the full version of the third review from the authors, we have provided answers to all the issues they raised.

1) Reviewers share a general feeling that geomorphological factors and hydrology were incompletely treated. This starts with the lack of a control stream. Please prepare more background information on the various ponds in this regard. Residence time is a variable (and concept) that must be discussed, ideally you actually find a way to explicitly include it in your analysis. The last reviewer suggests using proxies for residence time and the manuscript actually included a "pond-to-channel" ratio that may be exactly that.

**Answer:** We agree with the Reviewers that the information on the hydrogeomorphic background of the study was insufficient and slightly disorganised. We have improved this aspect in several ways following suggestions received.

Firstly, we have included a clear description of the geomorphic classification and expanded both the textual and tabular descriptions of the study sites with additional information on the character of beaver modifications (i.e. connection between ponds within cascades, pond and dam size, ratio of the pond sequence area to the area of the river channel). The relevant paragraph is now as follows:

'...The height of the beaver dams studied did not exceed 1.5m. In most dam sequences, the ponds were directly connected. The only exception was site 7, where short sections of free-flowing reaches were observed between the ponds. For further analyses, the study sites were classified based on the beaver pond characteristics. Two geomorphic types were distinguished considering the extent of a beaver dam: in-channel (dams within river banks) or overflowing (dams extending over river banks and leading to inundation of adjacent parts of river valley). The area covered by the overflowing ponds was two to seven times larger than their respective river channels which indicates a substantial proportion of flooded riparian areas. The age of beaver ponds was assessed based on the archival orthophotos and classified into three categories: young (≤3 years old), moderate (4-9 years old), and old (≥10 years old) (Table 1)...'.

Moreover, we clarified the hydrogeomorphic setting of the samples representing pond conditions by specifying the sampling method. It is now as follows:

'…The 'pond' sample was taken from the main channel section of the pond, i.e. excluding channel margins and backwaters. It was collected approximately 1m above the dam, from the middle of a water column. For sequences of multiple ponds, the representative 'pond' sample was collected from the biggest reservoir in the system…'.

Finally, the water residence time has been introduced at the beginning of the manuscript and discussed more closely as an important factor affecting the processes observed in beaver ponds. For example, it was included in the revised introduction to justify our approach to assessing the impact of pond geomorphic types and to set out the base for further discussion:

'…Spatially expansive beaver ponds are expected to facilitate a higher proportion of low velocity areas, increase in water residence time, extent of a hyporheic zone, overbank sedimentation and growth of aquatic vegetation, compared to the ponds constrained to the river channel. Both beaver pond age and its geomorphic type may correspond with the prolonged sediment-water interactions resulting in the enhanced biogeochemical processes…'.

The role of prolonged water residence time was particularly emphasised in the context of the effect of overflowing ponds, e.g.:

'...The rate of biogeochemical processes is controlled by residence time, as it determines the duration of contact between solutes and microbial communities (Briggs et al., 2012)....',

'…The inundation of the area surrounding the river channel (the riparian zone) reduces flow velocity and increases water residence time (Larsen et al., 2021). This in turn supports the development of aquatic vegetation which facilitates the attenuation of nitrogen (Murray et al., 2021)…'.

We agree that it would be highly beneficial to use control data collected before the beaver dams were built. However, as obtaining this kind of information was not feasible, we took a different approach and used upstream river sections as references. The study was based on percentage changes to enable comparisons between rivers despite the differences in their initial (upstream) water chemistry. Even though it limited some of the methodological constraints, we elaborated on the identified shortcomings resulting from our approach in the revised Study limitations.

2) Analytical precision and raw data: This point raised by reviewer 3 seems very important to me. Please answer this point of critique with great care and provide information that lets readers trust your physico-chemical data. The important issue is that some differences among sites may be within the uncertainty range of the used methods. With regard to data, I urge you to provide the original data, likely in a supplement, besides the relative differences caused by beaver ponds. Unreliable differences may be flagged as such in graphs and tables (note that this quality of a measurement gets lost in the "percentage change"). Also, the issue of data quality is connected to how sampling was actually done, in particular the accompanying flow conditions and day time of sampling. Note that the fact that water remains in reservoirs behind beaver dams for quite a while translates to an averaging effect, thus upstream vs. downstream comparisons must be assessed under consideration of eventually dynamic situations upstream or in the pond (probable less so downstream).

**Answer:** We have improved the manuscript in terms of methodological clarity and uploaded the database to an open access data repository (doi: 10.5281/zenodo.17937225) - the data availability statement has been updated accordingly. The uploaded datafile is supplemented with additional information regarding measuring devices, including detection limits.

However, the raw data has already been presented in the form of a graph in both the first and revised versions of the manuscript (Figure 4; now Figure 3). This graph illustrates the relationship between the physico-chemical properties of water such as pH, SEC [$\mu S \cdot cm^{-1}$], major ions, $NH_4^+$, $NO_3^-$, and DO concentrations [$mg \cdot L^{-1}$] in the beaver ponds vs. streams above the ponds and in the streams below the beaver ponds vs. streams above the ponds.

Several clarifications were made in the description of the sampling and computation process. That includes indication of the duration of each sampling session, part of the day and flow conditions when the sampling was conducted.

We described the treatment applied to values below the detection limit as follows:

'…The detection limits were 0.4 $mg \cdot L^{-1}$ for Ca and 0.1 $mg \cdot L^{-1}$ for the other ions. Any ion concentrations below the detection limit were replaced with 75% of the detection limit, which is a common procedure in chemistry (Palarea-Albaladejo and Martín-Fernández, 2013). Concentrations below the detection limit were found only for the $NO_3^-$ and $NH_4^+$ ions. Of all the analysed water samples (n=102), the $NO_3^-$ and $NH_4^+$ concentrations were below the detection limit in 17 and 20 samples, respectively…'.

We were aware that the differences we obtained could be within the measurement uncertainty range. Therefore, we limited our statistical analyses to those parameters for which the average differences between upstream and pond/downstream values were greater than the measurement errors. For most parameters, these errors did not exceed 5%, so we chose this threshold for these parameters. We also considered parameters with errors slightly higher than 5% if the difference in concentration between upstream and downstream was many times greater than 5%. We have described this procedure in detail in the revised methodology section:

'...The percent relative error, as determined by the data quality assessment based on CRM (KEIJM-02, GAW/WMO) was less than 5.0% for all parameters except $K^+$ and $NH_4^+$. The respective values were 5.8% for $K^+$ and 7.0% for $NH_4^+$...',

'...Physico-chemical water parameters were included in the PCA if their mean absolute difference $|Dc|$ (for all sampling sites combined) exceeded 5%. This procedure was used to account for the accuracy of analytical measurements defined as percent relative errors. The differences in parameters, which relative percent errors slightly exceeded 5% (i.e. $NH_4^+$ and $K^+$), were many times greater than their respective errors, which allowed for their inclusion in the analysis…',

'...Seasonal and site-to-site variations in *Dc were* also assessed for T and parameters that met the same criteria as those used for PCA…'.

We removed ions for which the observed changes were generally smaller than 5% ($Mg^{2+}$, $Na^+$) from Figure 4 and Table 3 to reduce the insignificant content. Moreover, to increase transparency of the summary presented in Figure 12 (now Figure 8), the average magnitudes of the observed changes are now presented in their original units of measurement alongside the percentage change. Both the changes relative to upstream river sections and differences between the studied groups in most cases markedly exceed the analytical errors.

3) Hyporheic exchange should be discussed also in the light of potentially being promoted by higher water pressures in the beaver ponds. The downstream water chemistry may thus be different not because of beaver pond-specific processes but because of forced increased hyporheic exchange flow (which of course in itself is a consequence of beaver activity).

**Answer:** Thank you very much for this comment. The influence of biogeochemical processes occurring in the hyporheic zone is indeed important in determining the chemical composition of water in beaver ponds. We have added this information to the Introduction and Discussion sections of the manuscript.

Introduction: '…Spatially expansive beaver ponds are expected to facilitate a higher proportion of low velocity areas, increase in water residence time, extent of a hyporheic zone, overbank sedimentation and growth of aquatic vegetation, compared to the ponds constrained to the river channel…'.

Discussion: '...Briggs et al. (2012, 2013) found that the accumulation of organic matter in beaver ponds generates specific streambed morphologies, which enhance the exchange of stream water and groundwater within the hyporheic zone behind beaver dams. Furthermore, the high water pressure at the bottom of the ponds causes a downwelling flow over the dam structures. This promotes the transport of organic carbon and DO from the water column to the hyporheic zone, fuelling biogeochemical processes…',

‘...Under the anoxic conditions that prevail at the bottom of ponds (as measured at site 3 in July 2024; see Fig. 7), as well as in the hyporheic zone, $SO_4^{2-}$ ions are likely to be reduced to $H_2S$…’,

‘...The longer residence time of water in the overflowing ponds may also explain the higher denitrification rates in the water of both the pond and the hyporheic zone where the bottom pond water and groundwater mix (Merrill and Tonjes, 2014)...’,

‘...Extended hyporheic zones, which are associated with the formation of overflowing ponds, are likely areas of $NO_3^-$ removal due to denitrification…’.

**REVIEW 1**

GENERAL COMMENTS

1. The abstract currently cannot stand on its own, i.e., it is not understandable on its own without having read more text of the manuscript. Needs to be rewritten.

**Answer:** Some parts of the abstract were previously omitted to make it more concise, however, we agree that this reduced the overall clarity of the study and led to the omission of some important information. We have integrated additional details, such as a clearer description of the methods, and rewritten the abstract into more structured paragraphs.

2. In the introduction please be much clearer about the actual research questions/hypotheses of your study. For instance, it is unclear what is “an environmental response to beaver dams”, meaning a change in upstream-downstream gradients in physicochemistry? Or do you mean a change after-before (in a temporal sense!) at larger spatial scale?

**Answer:** We removed this general sentence and replaced it with a paragraph describing the idea in more detail. This includes a description of the role of specific beaver pond characteristics (age and geomorphic type) on the physico-chemical parameters, from which we further derived our study questions. The questions were also reformulated with more straightforward wording. It is now:

‘...The objective of this study was to determine the effects of beaver dams on physico-chemical water properties in mountain streams of the Western Carpathians (Poland and Slovakia), with regard to seasonality and pond characteristics. The paper focuses on answering the following three questions:

1) How do physico-chemical water parameters change between river sections located upstream of, within, and downstream of beaver ponds, and what biogeochemical processes might be responsible for these differences?
2) How do these differences vary seasonally?
3) How are these differences affected by the geomorphic type and age of beaver ponds?...’

3. Take care to robustly derive your study´s questions from past work AND explain them in an understandable way. The three questions are currently a slightly loose list without much connection and with poor derivation.

**Answer:** Thank you for pointing out this aspect that needed improvement. Some information was missing, while other (e.g. brief reasoning behind the pond classification) were scattered in the manuscript. We reorganized part of the introduction into three paragraphs that correspond more clearly with the research questions. We took special care to explain potential and already documented impacts of the age and geomorphic character of beaver ponds.

4. A weakness of the study is its focus on upstream-downstream comparisons without any controls. Water chemical conditions may change a lot between two sites along a stream continuum, independent of beaver dams. The study investigates only stream sites with beaver dams and compares data from above and below. There was no investigation before beaver dams were constructed, nor are there any control sites where no dams exist. I acknowledge that a full before-after-control-impact (BACI) study design was deemed infeasible,

but such shortcomings of the study design must at least be discussed. Typically, in studies lacking controls, a higher level of causality is needed for hypothesized processes, here in this case all processes that may affect physicochemical conditions. For ions without much explanation for change in beaver ponds, the current design does not allow to strongly pin down beavers as being the origin for any change, especially if the direction of change was not consistent. This should be reflected in the discussion.

**Answer:** We agree that the lack of data from before the construction of the beaver dams limits our ability to clearly determine the cause of changes in the chemical composition of the water. However, this study used a comparative ‘upper river – lower river’ method, where the upper river sections were used as reference sections. This research approach is commonly used in field studies where it is impossible to obtain data 'before' the construction of beaver dams for logistical or environmental reasons (e.g. Green and Westbrook, 2009; Puttock et al., 2017). The revised manuscript has been supplemented with a discussion of these limitations, indicating that the presented results are correlational in nature and should be interpreted as indicating potential links between the presence of beaver dams and changes in water physicochemical properties. Additionally, the ‘Study limitations’ section highlights that a complete Before–After–Control–Impact (BACI) design would be the most appropriate approach to testing the causal effects of beaver activity (Puttock et al., 2020; Dittbrenner et al., 2022), but its application to natural river ecosystems is often impractical. It also highlights directions for future research that combine a spatial approach with a temporal component, which would allow for a more definitive assessment of the processes occurring in beaver-modified systems:

‘...Although a Before–After–Control–Impact (BACI) model would be the most appropriate approach for studying the impact of beavers on the chemical composition of water, it was not possible to use this method in this area due to the lack of investigations prior to dam construction. Instead, the study focused on spatial and temporal contrasts throughout the year and used a comparative ‘upper-lower river’ method, where upstream sections were used as reference sections. This research approach is commonly used in field studies to assess hydrochemical gradients generated by beaver dams when it is logistically or environmentally impossible to obtain ‘before’ data on dam construction (Green and Westbrook, 2009; Klotz, 2010; Fuller and Peckarsky, 2011; Puttock et al., 2017; Stevenson et al., 2022). It enables the identification of local hydrological and chemical effects caused by beaver damming while minimising the confounding influence of catchment heterogeneity. Although this approach lacks independent control sections, as in a full BACI model, it provides robust comparative evidence for evaluating the magnitude and spatial extent of changes induced by beaver activity under natural field conditions. Future research should be more detailed and extensive to enable the application of the BACI model…’.

5. The paper also suffers from a relatively inefficient presentation of results. There are too many figures. Consider moving some figures to the Supplement and look for ways to more efficiently present data (e.g. do not use separate tables for PCA variable loadings, but rather include these as a separate panel in the figure). Consider removing, shortening the more detailed results on dam cascades or vertical profiles or move to supplement to give the paper more focus. Some plots are simply superfluous and don´t present much exciting (e.g. SEC in Fig 10).

**Answer:** We minimized the number of figures presented in the main text and relocated a significant part of the graphs to the appendices. At the same time, we reduced the size of the figures by removing superfluous content and combining sites and variables on the plots. We shorten and focused the text by removing part of section 3.1. concerning PCA analyses of the physico-chemical background, which included removing one figure. The two remaining PCA results (based on the changes observed in the ponds and downstream to the ponds) were combined into one figure (now Figure 5). We limited the redundancy by removing parts of the plots presenting the combined classification of points based on already presented classifications (seasons and sites). We also took your advice and incorporated information from the tables into the figures. The most relevant variables were selected for presentation in the form of longitudinal and vertical profiles in the main text (now Figures 6 and 7). Moreover, we presented only those parameters for which the average differences were greater than 5% on the revised Figure 4.

6. The discussion of the various anaerobic processes should be organized along the redox ladder.

**Answer:** We agree with this suggestion. Despite the lack of redox potential measurements, we attempted to organise the discussion of the aerobic/anaerobic processes along the redox ladder based on changes in the physico-chemical properties of the water, which indicate the occurrence of these processes, e.g.:

'...As a result, there are intensive physico-chemical transformations in the stream water flowing into the ponds in the warm seasons. The increase in $NH_4^+$ concentrations and the decrease in $NO_3^-$ concentrations in the studied beaver ponds were particularly prominent in the warm period of the year due to the intensive ammonification ($NH_4^+$ production) and denitrification processes ($NO_3^-$ depletion). This is consistent with the findings of Margolis et al. (2001) and Law et al. (2016): the greatest $NO_3^-$ decrease downstream of beaver ponds occurs in the summer. $NO_3^-$ ion is one of the most effective electron acceptors (oxidants) in the process of anaerobic oxidation of organic matter; therefore, $NO_3$ is the preferred oxidising agent for anaerobic microbes. The process of $NO_3^-$ reduction (denitrification) under anaerobic oxidation of organic matter occurs at a relatively high redox potential level (Anderson and Fidel, 2025). However, it should be noted that the decrease in $NO_3^-$ concentrations in the beaver ponds and streams below the ponds in the summer is partly related to the nitrogen fixation process, which is associated with increased microbial activity in sediments mediated by anaerobic conditions, as well as uptake by aquatic vegetation (Naiman and Melillo, 1984; Maret et al., 1987; Songster-Alpin and Klotz, 1995). Furthermore, the decrease in $SO_4^{2-}$ concentrations in the studied ponds and downstream of the beaver dams was mainly observed during the warm period. This is also likely due to the high rate of anaerobic oxidation of organic matter during that time. According to Smith et al. (1991), Cirmo and Driscoll (1993), and Margolis et al. (2001), the intensive anaerobic oxidation of organic matter can occur despite the relatively low redox potential of the water during the low-flow conditions of summer. During that time anaerobic microbes used even relatively inefficient oxidants, such as $SO_4^{2-}$, in the anaerobic oxidation of organic matter due to the low concentration of $NO_3^-$ (a more effective oxidant) in the studied waters. The average concentration of $NO_3^-$ in ponds and streams below them was very low during the summer, the average was up to 1.5 $mg{\cdot}L^{-1}$ …'.

7.The lower differences from upstream to downstream compared to upstream-pond may to some extent be related to the fact that downstream samples are from a well-mixed turbulent stream but pond sampling was likely not done in a spatially "integrative" way. Thus, pond samples may not represent average pond conditions. This needs discussion, also in light of anyway existing vertical profile data.

**Answer:** We agree that due to the spatial complexity of beaver ponds it's difficult to derive general conclusions concerning the whole pond without comprehensive sampling across the system. The regional scope of this research didn't allow for more detailed sampling at every pond each season. However, we applied consistent criteria for the location of the 'pond' sampling point across the whole study area and time, to increase their comparability. The detailed description that would help interpret the data was missing in the methodology section, therefore we clarified the sampling locations in the revised manuscript:

'...The 'pond' sample was taken from the main channel section of the pond, i.e. excluding channel margins and backwaters. It was collected approximately 1m above the dam, from the middle of a water column….'.

We have complemented the discussion with a description of the identified constraints:

'...Although the sample locations were standardised to a certain extent, they may not reflect typical stream or pond conditions. Stream samples are more likely to be well-mixed and representative due to turbulent flow conditions. This is also evidenced by the homogeneity of most longitudinal profiles upstream and downstream of beaver ponds. However, the spatial distribution of the pond samples may be more biased as it did not cover the full spectrum of hydrological units observed within the ponds (Majerova et al., 2020). Additionally, as the samples were taken from the middle of the water column, they do not represent the surface and bottom water layers, which according to the vertical sampling scheme could substantially affect observed physico-chemical parameters...'.

8. I strongly recommend authors to discuss the observed beaver pond effects in the light of residence time of water in the ponds. Doing this may offer a chance to integrate pond/stream size effects and seasons (differences in stream discharge). It may even be able to integrate geomorphic pond type (for which I find any observed

differences inadequately derived as well as discussed anyway). Residence time in concert with temperature may be able to explain a lot.

**Answer:** Thank you for this valuable suggestion. Although water residence time plays an important role in shaping the observed processes in beaver ponds, it was only briefly mentioned in the text. While modifying the introduction, we included it as one of the pond characteristics that may differ between the two geomorphic types, instead of the description previously made in the methodology section. The 'age' and 'geomorphic type' are not the direct causes of the difference, but this classification is likely connected to different hydrological and thermal conditions that influence biogeochemical processes. Much of the discussion on the impact of beaver pond age and geomorphological type was reorganised and supplemented with information regarding effects of water residence time and temperature, to clarify processes that may be responsible for the observed differences between the ponds.

9. Overall, I urge authors to shorten the paper and find more efficient wording in particular in the discussion. This will make the paper more accessible to a larger audience.

**Answer:** After reading the manuscript with a fresh perspective, we agree that it was somewhat overloaded with information. We revised the text and figures carefully. We removed part of section 3.1 from the results completely, because it didn't carry information that would be necessary for understanding the impact of beavers, which is the main focus of the paper. We also removed or rephrased some sentences that were redundant or purely structural, especially in the discussion and conclusions. Moreover, the combination of several variables and study sites on the plots decreased the size of the figures. An incorporation of the PCA loadings into the figures decreased an overall number of figures and tables. All the changes resulted in the shortening of the main body of the manuscript by a few pages.

SPECIFIC COMMENTS:

55: "attenuation of downstream …. concentrations" – please express more clearly, do you mean compared to upstream? Then is it measured relative to a control? Or does this imply some before-after comparison as well? The same problem applies to the text in the abstract.

**Answer:** We clarified the upstream-downstream relation in this fragment of the text, as well as in the description of our research objectives in the abstract.

58: maybe two sentences instead of semicolon?

**Answer:** Thank you, we changed this sentence.

60-65: Could be worthwhile to dive deeper into whether these studies aiming at investigating "reductions" and thus working with upstream-downstream differences actually follow BACI-study designs to statistically robust conclusions.

**Answer:** We used the word 'reduction' incorrectly. It should have been 'decrease' - we meant a decrease in concentration. This error appeared in several other places in the text and has now been corrected.

In this study, it was not possible to use the BACI model due to, among other reasons, the lack of pre-dam surveys. The study focused on spatial contrasts and seasonal changes within a single beaver-affected streamway. In our study, we used a stream-scale gradient approach, with sampling points located upstream of the beaver pond, within the pond, and downstream of the dam. This spatial model, commonly referred to as an upstream-downstream or longitudinal comparison (e.g., Klotz 2010; Fuller and Peckarsky 2011; Stevenson et al. 2022), has been widely adopted to assess in situ hydrochemical gradients generated by beaver dams. It allows for the identification of local effects of dam-induced hydrological and chemical transformations while minimizing the confounding effects of catchment heterogeneity. Although this approach does not include independent control sections, as in the full BACI project, it provides robust comparative evidence for assessing the direction and magnitude of changes induced by beaver activity in natural field conditions. Therefore, our sampling framework is consistent with established methods for assessing changes in stream water chemistry and sediment dynamics induced by beavers.

66: maybe explain a bit better why and how geomorphic factors could actually have such an influence.

**Answer:** Thank you for this suggestion. We rewrote a part of the Introduction and included a more detailed explanation of the expected influence of beaver pond characteristics in this section:

'…The identification of factors reflecting the diversity of beaver ponds and their effects on water chemistry, would improve the understanding of the effects of beaver presence in river systems. The age of beaver ponds was commonly proposed as a such feature in previous studies on upstream-downstream gradients of physico-chemical parameters in beaver-occupied streams. For example, Bason et al. (2017) found a greater reduction of suspended solids in older dam sequences compared to younger ones. Metal sequestration rates and a sediment organic matter content inside beaver ponds also rise with the pond age according to the study of Murray et al. (2021). Hydrological and thermal differences were observed between various geomorphic units within beaver ponds, representing e.g. main channel, its margins and backwater areas (Majerova et al., 2020). The share of such units, especially related to the floodplain inundation, was then suggested as a potential geomorphic factor that could explain differences between study sites (Murray et al., 2021). Spatially expansive beaver ponds are expected to facilitate a higher proportion of low velocity areas, increase in water residence time, extent of a hyporheic zone, overbank sedimentation and growth of aquatic vegetation, compared to the ponds constrained to the river channel. Both beaver pond age and its geomorphic type may correspond with the prolonged sediment-water interactions resulting in the enhanced biogeochemical processes…'

115: explain pond-to-channel ratio

**Answer:** The term 'pond-to-channel ratio' was originally intended to describe the distinction between 'overflowing' and 'in-channel' beaver ponds. To improve clarity and ensure consistency with the terminology used throughout the manuscript, we have replaced it with 'type of beaver pond'.

155: were both the original data and the differences (percent change) used in the same PCA? Please provide justification why this procedure would make sense.

**Answer:** We used the original (raw) data of solute concentrations for PCA to determine the physico-chemical background of the studied streams. For this purpose, we used the raw concentrations of solutes in the stream water above the beaver ponds that were unaffected by beaver dams. These concentrations were standardised prior to analysis. However, to determine the factors influencing changes in solute concentrations in ponds and streams below due to beaver dams, we performed PCA using the percentage difference in concentrations between the stream water above the beaver dams and the pond water/stream water below the dams. In this case, we used concentration differences rather than raw concentrations because we focused not on the factors determining solute concentrations in pond and stream waters below the dams, but rather on the factors shaping the differences in these concentrations due to the presence of beaver dams. Ultimately, in light of suggestions to shorten the manuscript, we decided to remove the results of the factor analysis based on the raw data from Section 3.1. The physico-chemical background of the studied streams.

161:There is no N in this formula.

**Answer:** It was our mistake - N was included in the second formula, but not the first. We have corrected this.

171: Variables are all concentrations or differences thereoef. I suggest to not mix untransformed with log-transformed variables in the same PCA. Concentrations may just all be log-transformed, as they are often log-normal distributed anyway. I cannot see any step in PCA where significance tests would become relevant. And to use significance for normality tests is a questionable approach anyway, as normally one "hopes" for high P to identify a variable as "not identifiable as not normal". Try to work without those debated and criticized tests. Same applies to Shapiro-Wilk used prior to ANOVA.

**Answer:** We have removed the results of the PCA analysis in which both non-logarithmic and logarithmic variables ($Ca^{2+}$) were used (see section 3.1. The physico-chemical background of the studied streams). Therefore, we removed the information about PCA analysis on raw data from 2.3. Statistical analysis section. The information about significance threshold applies to all the other analyses, so we moved it to a different part of this section. To our knowledge, the use of the normality test remains a standard procedure, as it allows to select parametric or nonparametric tests for further analyses based on the data distribution. Both Shapiro-Wilk and Kolmogorov-Smirnov tests have been used prior to ANOVA in previous studies on beaver impacts on water chemistry (Błędzki et al., 2011; Bylak et al., 2024).

178-180: Before doing hard-core hypothesis tests, please cleanly derive why these hypotheses would anyway make sense to be tested in the intro. The derivation of your questions and subsequent hypotheses needs to be clearer. Currently it seems that the factor “pond type” was defined after the study was done.

**Answer:** The indicated fragment has been removed from this part of the text. We have clarified our underlying assumptions in the introduction instead. Please see point ‘66:’ above.

199: Reword “controls the values”, not appropriate wording to describe PCA results.

**Answer:** We agree with this comment. We have corrected this throughout the text.

Fig. 3: What are the various regions indicating? Information for A,A/B, etc. missing from the legend. I suggest you also present factor loadings as arrows, could be done in a separate plot (panel).

**Answer:** The regions A/B represented geological locations (corresponding with dominance of shale or sandstone in the catchments), while C/D represented sampling seasons. However, we removed Figure 3 in order to shorten the article and focus the result section on the impact of beavers.

Fig. 4: This figure does not allow to recognize which points belong to the same pond and date, i.e. the paired nature of the data is not shown (or “used” because in fact its consideration could make data analysis stronger). Consider working with differences or connections between points.

**Answer:** The purpose of this figure (now Figure 3) was not to distinguish solute concentrations in detail at each location and sampling time, but rather to illustrate a general pattern. The figure allows to distinguish between the solute concentrations in ponds and streams below the beaver dams (represented by squares and crosses, respectively) in relation to the stream above the pond, and categorises them according to the age of the beaver dam: less than three years old, four to nine years old, and more than ten years old (represented by small red signatures, larger yellow signatures, the largest green signatures, respectively). Inclusion of the season as another variable or links between the points would make the figure illegible. However, in most cases the relations between paired data (pond and downstream differences at particular location and season) can be distinguished on the figure. The points representing the same sampling site at the same time are located at the same position on the horizontal axis (which results from the same inlet value), therefore vertical distance between both points illustrates the difference.

Table 4:This table presents A LOT OF P-values. Since the same dataset was used multiple times, some P-value adjustment (e.g. Bonferroni) should be done. Certainly, P-values >0.05 (<0.1) should not be reported. I fear that not too many P-values will remain significant then, making clear why this strategy of extensive dataset testing may not be the best. Also, stronger analysis may take into account multiple factors (pond type, age, seasons) at once.

**Answer:** We didn’t consider results with p<0.1 statistically significant. They served as additional information and were presented in the table in regard to a scientific discussion on the arbitrariness of significance thresholds. However, we agree that their inclusion was unnecessary and could distract from the interpretation of truly significant results. Therefore, only results meeting the conventional significance threshold (p<0.05) were retained in the revised table. Moreover, in response to the very appreciated comment concerning multiple analyses on the same dataset we applied recommended correction to improve the interpretation of the results and updated methodology section:

‘...The significance level of p<0.05 was used for all the calculations. Additionally, to reduce the risk of family-wise error for multiple comparisons based on the same dataset, we applied a Bonferroni correction to the significance threshold (p<0.017) (Armstrong, 2014)...’.

269, 285: Was this PCA entirely done with "differences"? A graph of variable scores as arrows as a panel in Fig 6 may be more efficient than Table 5. I suggest to not speak of "water samples" in the legend of Fig 6 as two samples were needed to create one point in these graphs, as I understand. The site codes in fig 6 are hard to distinguish.

**Answer:** The results of the PCA analysis that were based on raw data in Section 3.1 were removed to shorten the text. The remaining PCA analyses are both based only on concentration differences (%), so subsequent update of the methodology description made it less confusing. Exact information on the type of data used can be found in Section 2.3. Statistical analyses.

As suggested, information on the factor loadings of F1 and F2 from Table 5 and Table 6 has been incorporated into Figure 5 in graphical form. Tables 5 and 6 have been combined and relocated to the appendix (Table A2) because, in addition to the factor loadings of F1 and F2, they also contain information about the factor loadings of F3. In the legend for Figure 6 (now Figure 5), we wrote 'water samples' because each point represents a single water sample, and the factor scores for factors 1 and 2 are marked for each sample. We have corrected the site codes in the figures relating to PCA.

Chapter 3.4: needs to be incorporated in the Discussion.

**Answer:** You are right. It became Chapter 4.4.

354: pointing out high TOC only makes sense in comparison to upstream.

**Answer:** We agree with this comment. We have corrected the text as follows:

'...Detailed investigations carried out in July 2024 at sites 3 and 6 showed that beaver pond water contains high concentrations of total organic carbon (TOC) compared to the stream water upstream. For example, the TOC concentration in the beaver pond at site 3 was found to be more than 13 $mg \cdot L^{-1}$, which was almost three times higher than in the stream flowing into the pond...'.

365: any idea about CH4 in these ponds? It would make sense to organize the discussion of the various anaerobic processes along the redox ladder.

**Answer:** Unfortunately, we did not examine the concentrations of $CO_2$ and $CH_4$ in the stream water and pond water, so we cannot address this issue in the discussion. However, we realise that it is essential to take this into account when studying the impact of beaver ponds on the chemistry of the stream below in future research.

366: ammonification is a fairly specific process and not a synonym for OM oxidation.

**Answer:** We agree with this comment. The sentence has been revised as follows:

'...The decomposition and mineralization of organic matter in the process of ammonification, under both aerobic and anaerobic conditions, leads to the production of $NH_4^+$ ions in water…'

370: Prefer "aerobic/anaerobic" for microbial processes, but "oxic/anoxic" or low/high redox potential for environmental conditions.

**Answer:** We agree with this suggestion. We have corrected the terminology throughout the text.

Fig. 12: Very appreciated conceptual figure. Consider using coloring the pond sequence in a color gradient instead of arrows?

**Answer:** Thank you. As the original figure was found to be difficult to understand, we considered various ways of transforming it to improve its clarity. The use of arrows has been modified to indicate direction and the average magnitude of change observed in each period, type and age of beaver pond, instead of the simple indication of one difference resulting from a comparison. In this way, even though there is more information presented in the figure, the content of the image should be more clear.

**REVIEW 2**

The study examines the impact of beaver ponds on biogeochemical processes and stream water chemistry. For testing, 9 mountain streams in Poland and Slovakia were selected. The study focuses on the physico-chemical parameters with a detailed sampling design and focuses on pond control effects, seasonality and typology. Based on the presented result, it is necessary to state that the manuscript is well written and structured. However, in several places (methodology and description of results), it contains a significant amount of detail that distracts from the main idea of the article. Conversely, some details and method explanations are missing. I also recommend revising several diagrams and charts, which would significantly improve the readability of the article. The manuscript requires small reconfiguration and improvement in readability. Hopefully, my comments will be helpful in that process.

**Answer:** Thank you for the positive feedback and all the constructive remarks. We agree that the original manuscript was not very clear due to the amount of content and inefficient presentation of the results. However, we managed to shorten the text and reduce the amount of distracting or redundant information. The reduction included, e.g., removal of the part of Section 3.1. from the Results as it was found to be not relevant enough for the understanding of the effects of beaver activity. Section 2 Methods was slightly modified to eliminate repeated and include missing information, and to direct the reader to the information already present in the table. The overall readability was also enhanced by modifying the graphic materials, i.e., by combining several variables and the two study sites in the plots, and by presenting the PCA results in one comprehensive figure. The size and number of the figures was therefore reduced, while retaining the same amount of information and enhancing comparability and visual appeal.

Study area (85-120): Here, I recommend the description of 2 factors, which are used for the interpretation of results in the discussion, and these are: geology and agricultural activities. Geology is completely missing and is described in the discussion that different structures affected stream chemistry. From human activities, it is important to add more information on how intensive agricultural activities are preserved in upstream conditions. Pastures, fertilisation or direct soil cultivation can affect chemical composition. The second important parameter is human presence in the upstream catchment area (population, number of buildings) that can affect a lot of selected chemical parameters. At the same time, it should be noted that the selected design of sampling strategy minimises ambiguities, but a broader description of these limits should be part of the discussion section.

**Answer:** The geology of the study area was presented briefly in the first paragraph of section 2.1, including an explanation of its regional diversity. As part of the discussion, we refer to the geology at a similar regional level of detail. Unfortunately, based on the available data we could not provide a more detailed description of each river channel. However, we found that the reference made in the discussion could be confusing, as the phrase ‘correspond to various geological features’ implies that we are referring to more detailed components instead of the differences in general geological structure. We have modified this fragment to: ‘corresponded to the geological structure.’ Additional information on the water chemistry was also included:

‘...Streams draining areas underlain by shales and mudstones are typically characterised by higher concentrations of bicarbonates, calcium and magnesium ions due to increased weathering of carbonates and clay minerals, while streams in areas composed mainly of sandstone are usually more diluted and characterised by lower specific conductivity (e.g. Siwek, 2021; Solár and Tomaškovič, 2023)...’.

As suggested, we added a paragraph in the Section 2.1 Study area describing land use and some of the anthropogenic impacts:

‘...All of the catchments consisted of a mixture of forest and agricultural areas, i.e. arable lands and meadows. Despite the widespread abandonment of agricultural land and the generally low-to-moderate intensity of agricultural practices in the mountainous catchments (Pazúr et al., 2014; Kolecka et al., 2017), the impact of these activities may be significant locally, as evidenced by the increase in nitrate concentrations observed in the Ondava catchment due to the use of fertilisers (Pekarova et al., 2006; Balejčíková et al., 2020). Overall, agricultural practices in the Polish and Slovakian Carpathians are mostly extensive, characterised by low–input management and mowing and grazing on a small-scale (Pazúr et al., 2014; Kolecka et al., 2017; Affek et al., 2023). According to national agricultural statistics (Central Statistical Office of Poland, 2025), the average consumption of mineral fertilisers (NPK) in the Małopolskie and Podkarpackie Voivodeships remains one of the lowest in the country (80–95 kg NPK $ha^{-1}$ $year^{-1}$ compared to the national average of ~125 kg $ha^{-1}$) and has shown little change over the last two decades. Similar situation is observed in the northern part of Slovakia where the average use of industrial fertilisers is less than 60 kg NPK $ha^{-1}$ $year^{-1}$ (Siman and Velísková, 2018). Consequently, the direct inflow of nutrients from fertilisers into watercourses is minimal, although local enrichment may occur near farms or areas of intensive grazing. In contrast, domestic wastewater was identified as the main source of stream water pollution in the rural areas of the Polish Carpathians (Siwek, 2021). The intensity of housing development across the study area ranged from negligible levels to artificial surfaces covering up to 4% of the area and containing a few hundred buildings…'.

Table 1 was also supplemented with information on the number of buildings in the studied catchments.

The potential impact of catchment diversity on the obtained results was described in the discussion:

'...As there was no identified relationship between the initial values and the percentage change in the physico-chemical parameters, it was assumed that the observed changes would not differ significantly based on upstream conditions. This allowed various catchments with different land uses and levels of anthropogenic influence to be included in the study. However, it should be noted that the inclusion of the streams characterised by particularly high concentrations of certain components, e.g. highly anthropogenically modified, may have resulted in different observations...'.

Methods (145 - 150): For increasing the readability of the paper, I recommend adding an exact complex statement of all physico-chemical parameters with a description and interpretation of what the source is and how it is theoretically interpreted when its values change.

**Answer:** The complex list of the measured parameters was previously presented on the bottom of Figure 2. However, we agree that listing those parameters in the text makes them more clearly visible. Therefore, we included the list at the beginning of the Methods section, before the detailed description of the sampling:

'..The research followed a three-dimensional sampling scheme: seasonal sampling, longitudinal and vertical profile sampling (Fig. 2). The following water parameters were measured: temperature (T), dissolved oxygen (DO), specific conductivity (SEC), pH and the ionic composition ($Ca^{2+}$, $Mg^{2+}$, $Na^{+}$, $K^{+}$, $NO_3^{-}$, $NH_4^{+}$, $HCO_3^{-}$, $SO_4^{2-}$, $Cl^{-}$)...'.

Although we agree that a description of each water parameter would be valuable, we are concerned that it would take up too much space and make the paper even longer. We did not make any assumptions about the source of each component in advance. Instead, we tried to identify patterns relating inlet values to land use and other catchment characteristics, such as size and slope. The PCA analyses indicated that the inlet values differed based on the location (geological setting). Additionally, we observed correlation between $Cl^{-}$, $Na^{+}$, $K^{+}$ and land use. Those informations were collected in the revised Results section '3.1 Physico-chemical background of the studied streams' as follows:

'...Significantly higher SEC and concentrations of major ions were found in streams that drained areas with a high proportion of shales in the bedrock (sites 1, 2, 4, and 6). Lower SEC and concentrations of major ions were found in streams draining catchments dominated by sandstones (sites 3, 7–9). Concentrations of $Cl^{-}$, $Na^{+}$ and $K^{+}$ showed high correlations with the share of artificial surfaces in the studied catchments (r=0.85, 0.78 and 0.61 respectively). Additionally, concentration of $Cl^{-}$ correlated with the number of buildings in the catchments (r=0.61)...'.

We modified the first paragraph of the discussion by adding a brief interpretation of those results:

'...The extensive distribution of the study sites resulted in a diverse initial physico-chemical water properties, which corresponded mainly to the geological structure. Higher SEC and higher concentrations of most of the major ions were found in streams that drain catchments dominated by shales in their bedrock, and they were lower in streams that drain catchments dominated by sandstones. This pattern is characteristic of the Outer Carpathians and it is likely associated with a different resistance to weathering (Siwek et al., 2011; Siwek, 2021;

Solár and Tomaškovič, 2023). The relation between $Cl^-$, $Na^+$, $K^+$ and anthropogenic modification of the catchment suggests human activity as an important factor for concentration of these ions in the study area...'.

Also, the exact morphological description of pond type, age, and other typologies that are used in interpretation should be stated here. For example, in the methods section, the type is defined as overflowing and in-channel, but in the Results section, it is interpreted as channel and overbank flow. Is it considered the same type of beaver pond?

**Answer:** Following this suggestion, we have supplemented the Methods section with this clarification:

'...For further analyses, the study sites were classified based on the beaver pond characteristics. Two geomorphic types were distinguished considering the extent of a beaver dam: in-channel (dams within river banks) or overflowing (dams extending over river banks and leading to inundation of adjacent parts of river valley). The area covered by the overflowing ponds was two to seven times larger than their respective river channels which indicates a substantial proportion of flooded riparian areas. The age of beaver ponds was assessed based on the archival orthophotos and classified into three categories: young (≤3 years old), moderate (4-9 years old), and old (≥10 years old) (Table 1)...'.

We have unified the references to the pond types throughout the manuscript.

Statistical analyses (170-180): Please add more information on how the clustering of the PCA analysis was performed, as is presented in Fig.3.

**Answer:** The part of the text referring to Figure 3 has been removed, as has the Figure 3 itself. However, we have clarified the methodological section, which included adding more information on PCA analysis.

Discussion section (Fig. 12): I am not sure if I can understand the scheme in Fig. 12. Complex and comprehensive information output, what image (period, age, geomorphology type) represents and how this typology can affect chemical composition is missing. From the image, it is not clear how different periods, pond types and age are related to chemical water properties (which typology affects the increase or decrease of chemical composition) and what changes are related to which presented type.

**Answer:** We modified this figure (now Figure 8) completely to make its content clearer and more comprehensive. Previously it was intended to emphasize the difference between periods, types or age of the beaver ponds by describing the direction of the difference alone. The revised figure presents the average magnitude and direction of change observed in each period, type, and age of beaver pond. We believe that enriched content of the figure makes it more informative, and at the same time more understandable.

Discussion: Even though there is a chapter in the Results section devoted to the limitations of the study, I would recommend moving it and expanding it in the Discussion section as a separate chapter.

**Answer:** As suggested, we moved this chapter to the Discussion section (chapter 4.4.). It has been substantially revised and complemented with more content.

**REVIEW 3**

This manuscript examines the influence of Eurasian beaver (Castor fiber) ponds on the physico-chemical composition of stream water in two Carpathian regions spanning Poland and Slovakia. The authors compare in-channel and overflow-type ponds across different seasonal and hydrological conditions. The topic is both timely and relevant given the widespread recolonization of Eurasian beavers and the increasing emphasis on nature-based water quality management in Europe. The Carpathian setting adds important biogeographical context.

The manuscript is generally well written and logically organized, and the authors have assembled a valuable dataset. The introduction provides appropriate background, and the discussion connects findings to the broader literature on beaver-mediated ecosystem processes.

However, the study has several methodological, analytical, and interpretive weaknesses that currently limit its robustness. Firstly, there is a notable lack of information regarding the quantification of detection limits and analytical uncertainties. This raises concern that the observed differences may be within the analytical uncertainty range of the employed methods, potentially rendering some of the reported changes statistically or analytically insignificant. Clarifying the magnitude of measurement precision and error propagation is therefore essential for assessing the robustness of the findings.

The absence of a control stream, incomplete treatment of hydrological and geomorphological variables (e.g., residence time, hyporheic exchange), and the reliance on PCA as the primary analytical tool weaken the causal inferences that can be made. In addition, the discussion sometimes over-interprets the observed data, especially regarding microbial processes, without corresponding measurements or mechanistic evidence.

Overall, I find the study promising and potentially suitable for publication after major revisions that fully address the issues outlined below

**Answer:** Thank you for all your comments and suggestions on how to improve the manuscript. We have responded to each comment below.

Major comments

The landscape and geomorphic context of the studied sites needs to be clarified. It is not entirely clear whether the overflow-type ponds are located within semi-confined valley settings and whether they inundate the entire valley bottom or only a portion of it. Similarly, please specify whether the studied ponds form cascading systems (i.e., multiple ponds connected in sequence) or if they are isolated structures separated by stream reaches. Providing a more detailed geomorphic and hydrological description of each pond type would greatly improve the reader's understanding of the environmental setting and the spatial variability in the measured parameters.

**Answer:** We expanded the description of study sites and geomorphic classification in the section Study area. It is now as follows:

'...The primary criterion for the selection of streams for the study was the presence of beaver dams that span the entire riverbed. The study sites were distributed randomly through the transect of the relatively low-elevation areas in the north to the southern slopes of the ridgeline close to the Poland–Slovakia border. In addition to varying topography, the sites differed in terms of human activity, such as catchment land use and presence of river control structures. The height of the beaver dams studied did not exceed 1.5m. In most dam sequences, the ponds were directly connected. The only exception was site 7, where short sections of free-flowing reaches were observed between the ponds. For further analyses, the study sites were classified based on the beaver pond characteristics. Two geomorphic types were distinguished considering the extent of a beaver dam: in-channel (dams within river banks) or overflowing (dams extending over river banks and leading to inundation of adjacent parts of river valley). The area covered by the overflowing ponds was two to seven times larger than their respective river channels which indicates a substantial proportion of flooded riparian areas. The age of beaver ponds was assessed based on the archival orthophotos and classified into three categories: young (≤3 years old), moderate (4-9 years old), and old (≥10 years old) (Table 1)...'.

Moreover, additional information, including pond to channel ratio which indicates the extent of overflow, were added to Table 1 presenting characteristics of the study sites.

Study design.

Firstly, there is a notable lack of information regarding the quantification of detection limits and analytical uncertainties. Most results are expressed as percentage changes, yet the underlying absolute concentrations and associated measurement errors are not reported. Table 2, for example, presents only upstream values—downstream concentrations should also be shown to allow direct evaluation of differences and variability. Moreover, Figure 4 (despite the very small and hard-to-read legend) appears to show input and output values that are strikingly similar. This raises concern that the observed differences may be within the analytical uncertainty range of the employed methods, potentially rendering some of the reported changes statistically or analytically insignificant. Clarifying the magnitude of measurement precision and error propagation is therefore essential for assessing the robustness of the findings.

**Answer:** We have improved the manuscript in terms of methodological clarity and uploaded the database (raw data) to an open access data repository (doi: 10.5281/zenodo.17937225).

The data in Table 2 only serves as background information on the hydrochemical properties of the studied streams. Therefore only data obtained upstream of beaver ponds is used to illustrate the baseline conditions. Presenting the data from the ponds or downstream of the ponds in the same way would not allow for direct comparisons. However, the raw data has already been presented in the form of a graph in both the first and revised versions of the manuscript (Figure 4; now Figure 3). This graph illustrates the relationship between the physico-chemical properties of the water such as pH, SEC [$\mu S \cdot cm^{-1}$], major ions, $NH_4^+$, $NO_3^-$, and DO concentrations [$mg \cdot L^{-1}$] in the beaver ponds vs. streams above the ponds and in the streams below the beaver ponds vs. streams above the ponds. The graph clearly illustrates the magnitude of the differences in cases of higher parameter values; in fact, these differences are more difficult to identify in cases of lower values. For example, changes are clearly visible for some parameters (e.g. pH, DO and $NH_4^+$), while for others they are very small and almost invisible (e.g. $Na^+$ and $Mg^{2+}$).

Several clarifications were made in the description of the computation process. For example, we included the detection limits of studied ions in the methodology section and described the treatment applied to values below the detection limit as follows:

'…The detection limits were 0.4 $mg \cdot L^{-1}$ for Ca and 0.1 $mg \cdot L^{-1}$ for the other ions. Any ion concentrations below the detection limit were replaced with 75% of the detection limit, which is a common procedure in chemistry (Palarea-Albaladejo and Martín-Fernández, 2013). Concentrations below the detection limit were found only for the $NO_3^-$ and $NH_4^+$ ions. Of all the analysed water samples (n=102), the $NO_3^-$ and $NH_4^+$ concentrations were below the detection limit in 17 and 20 samples, respectively…'.

We were aware that the differences in the physical-chemical parameter values that we obtained could be within the measurement uncertainty range. Therefore, we limited our statistical analyses to those parameters for which the average differences between upstream and pond/downstream physical-chemical parameter values were greater than the measurement errors. For most parameters, these errors did not exceed 5%, and this value was therefore used as a threshold for these parameters. We also considered parameters with errors slightly higher than 5% if the difference in concentration between upstream and downstream was many times greater than 5%. We have described this procedure in detail in the methodology section:

'...The percent relative error, as determined by the data quality assessment based on CRM (KEIJM-02, GAW/WMO) was less than 5.0% for all parameters except $K^+$ and $NH_4^+$. The respective values were 5.8% for $K^+$ and 7.0% for $NH_4^+$…',

'...Physico-chemical water parameters were included in the PCA if their mean absolute difference $|Dc|$ (for all sampling sites combined) exceeded 5%. This procedure was used to account for the accuracy of analytical measurements defined as percent relative errors. The differences in parameters, which relative percent errors slightly exceeded 5% (i.e. $NH_4^+$ and $K^+$), were many times greater than their respective errors, which allowed for their inclusion in the analysis…',

'...Seasonal and site-to-site variations in *Dc* were also assessed for T and parameters that met the same criteria as those used for PCA…'.

To increase the transparency of the summary presented in Figure 12 (now Figure 8), the average magnitudes of the observed changes in their original units of measurement are now presented alongside the percentage change. Both the changes relative to upstream river sections and differences between the studied groups in most cases markedly exceeded the analytical errors.

1. Statistical design and analytical approach

The use of Principal Component Analysis (PCA) as the primary method of data interpretation migt not optimal for addressing the research questions. PCA is a dimension-reduction tool used for exploring structure in multivariate datasets, not for hypothesis testing or identifying drivers of variability. The study's stated objectives—to assess the effects of beaver pond type, season, and age on water chemistry would hint towards inferential statistical approaches that explicitly test these relationships. For example:

- Multiple regression analysis (e.g., linear or generalized linear models) linking dependent variables (e.g., $NO_3^-$, $NH_4^+$, DO, pH) to independent predictors (pond type, age, season).
- Multivariate analysis of variance (MANOVA) or PERMANOVA, if multivariate relationships are of interest.

Including these analyses would allow the authors to quantify the strength and significance of each factor, rather than relying solely on visual clustering. The PCA can still be retained as a descriptive complement but should not serve as the basis for major conclusions.

**Answer:** We used PCA to provide an overview of the differences between the study sites and seasons, as well as the relationships between the measured parameters. A more quantitative evaluation was based on ANOVA and its non-parametric equivalents. The results of these analyses are presented in Table 3. We removed some references made in the discussion that could have suggested PCA as the main source of some conclusions. Furthermore, we reduced the number of plots and figures presenting the results of PCA in order to minimise the visual dominance of this analysis. Following the reviewer's suggestion to improve the interpretation of the results obtained from multiple analyses carried out on the same dataset we adjusted significance thresholds presented (Bonferroni correction).

Hydrological context: residence time and hyporheic exchange

The paper does not account for hydrological processes that are fundamental to understanding biogeochemical changes in beaver ponds.

Residence time: Water residence time controls oxygen availability, redox gradients, and microbial processing rates. Beaver ponds with longer residence times are expected to show greater reductions in dissolved oxygen and nitrate due to denitrification and organic matter decomposition. Conversely, short residence times may result in weaker effects. The manuscript should discuss residence time conceptually, even if it cannot be quantified directly. If possible, proxy indicators (e.g., pond size, depth, flow velocity) could be used to infer relative differences in residence time among sites.

**Answer:** Thank you for this comment. The role of residence time in the various types of beaver pond that we studied may indeed be crucial in shaping biogeochemical processes. In the Discussion section, we considered the potential impact of residence time on biogeochemical processes in beaver ponds, in both the water column and the hyporheic zone:

'...The longer residence time of water in the overflowing ponds may also explain the higher denitrification rates in the water of both the pond and the hyporheic zone where the bottom pond water and groundwater mix (Merrill and Tonjes, 2014). The rate of biogeochemical processes is controlled by residence time, as it determines the duration of contact between solutes and microbial communities (Briggs et al., 2012)...'.

Hyporheic exchange: The study also omits consideration of hyporheic exchange, the mixing between surface water and groundwater within streambed sediments. This process significantly affects nutrient transformations, nitrification, and dissolved oxygen concentrations (Triska et al., 1990; Merrill & Tonjes, 2014; Dewey et al., 2022). Importantly, hyporheic processes occur irrespective of beaver activity and can confound interpretations if not acknowledged. Including this discussion would demonstrate awareness of key hydrological-biogeochemical linkages and strengthen the conceptual framework of the paper.

**Answer:** We appreciate this comment and agree with it. The processes occurring in the hyporheic zone of a newly formed beaver pond are undoubtedly different to those in a stream channel's hyporheic zone. One reason for this is the different directions and dynamics of water flow in the hyporheic zones of these two environments. We have supplemented the discussion on this issue, however we did not expand on it in great detail because we did not investigate these processes directly. The following statements were added to the Discussion:

'...Briggs et al. (2012, 2013) found that the accumulation of organic matter in beaver ponds generates specific streambed morphologies, which enhance the exchange of stream water and groundwater within the hyporheic zone behind beaver dams. Furthermore, the high water pressure at the bottom of the ponds causes a downwelling flow over the dam structures. This promotes the transport of organic carbon and DO from the water column to the hyporheic zone, fuelling biogeochemical processes…',

'...Under the anoxic conditions that prevail at the bottom of ponds (as measured at site 3 in July 2024; see Fig. 7), as well as in the hyporheic zone, $SO_4^{2-}$ ions are likely to be reduced to $H_2S$…',

'...Extended hyporheic zones, which are associated with the formation of overflowing ponds, are likely areas of $NO_3^-$ removal due to denitrification…',

'...Biogeochemical processes in beaver pond water and sediments of the hyporheic zone, such as organic matter decomposition, denitrification and microbial sulphate reduction, are very slow at low temperatures compared to the warm conditions (Qu et al., 2022)...'.

3. Interpretation of microbial and biogeochemical processes. The authors attribute seasonal changes in nitrogen species ($NO_3^-$, $NH_4^+$) and DO to microbial processes such as denitrification, nitrification, and ammonification. While these mechanisms are plausible, the current dataset does not include direct measurements of these processes (e.g., redox profiles, dissolved organic carbon, or isotopic composition). As such, these explanations remain hypothetical. This means wording should be cautious: "likely related to" or "consistent with".

**Answer:** We agree with this comment. We infer processes such as ammonification, denitrification and nitrification based on the ratios of ion concentrations and dissolved oxygen (DO) in water. However, we did not conduct direct research on these processes. In accordance with this comment, we have revised the entire text to make it clear that these interpretations should be viewed as indirect assessments rather than direct observations.

- Highlighting the need for further studies measuring microbial activity or redox potential directly.

**Answer:** We added the following section in the Study limitations:

'...One of the greatest limitations of the study was the absence of field measurements of redox potential in streams and vertical profiles of beaver ponds. Such measurements would have enabled the direct identification of redox conditions in the studied waters rather than relying on speculation based on the concentration of key redox reaction ions. Direct measurements of redox potential, microbial activity and vegetation cover in the future studies would enable empirical assessment of the role of the discussed processes…'.

- Discussing potential alternative explanations, such as dilution effects, sediment adsorption, or vegetation uptake.

**Answer:** We did not consider the influence of the dilution process because our research was conducted during baseflow, when the inflow of event water was negligible. However, the influence of sediment adsorption (cation exchange) and vegetation uptake is indicated in the following statements:

'...However, it should be noted that the decrease in $NO_3^-$ concentrations in the beaver ponds and streams below the ponds in the summer is partly related to the nitrogen fixation process, which is associated with increased microbial activity in sediments mediated by anaerobic conditions, as well as uptake by aquatic vegetation (Naiman and Melillo, 1984; Maret et al., 1987; Songster-Alpin and Klotz, 1995)...',

'...According to Cirmo and Driscoll (1993), beaver pond sediments and vegetation act as temporal sinks for $Ca^{2+}$ in the summer, presumably as a result of $H^+$ exchange and their enhanced biological uptake during the growing season…'.

Additionally, please consider that reduced DO levels in beaver ponds may not always signify enhanced denitrification; they may also reflect increased biological oxygen demand (BOD) from organic matter accumulation.

**Answer:** Although we agree with this comment, the original version of the manuscript contained the following statement:

'...The clear decrease in DO concentrations within the beaver ponds compared to the DO concentrations above the ponds (at all study sites) suggests that DO is consumed in the oxidation processes of organic matter under aerobic conditions in the ponds…'.

Methodological transparency. Several key methodological details are missing or unclear, which hinders reproducibility and assessment of data quality. Please expand the Materials and Methods section to include the following:

- Calibration and maintenance of probes: Specify the calibration, field recalibrations.

**Answer:** Thank you for this valuable comment. We expanded the Materials and Methods section to provide full methodological transparency regarding probe calibration and maintenance. We clarify that all pH, EC and dissolved-oxygen probes were calibrated before each field campaign in the laboratory of the Research Station in Szymbark, Institute of Geography and Spatial Organization Polish Academy of Sciences. Calibration was performed according to the manufacturer's guidelines using certified analytical standards: pH probe: three-point calibration (pH 4.00, 7.00, and 9.00 buffers), SEC probe: calibration with a 1,41 mS/cm standard solution, selected according to anticipated conductivity of the sampled waters. DO probe: air-saturation calibration (100%) followed by zero-oxygen verification using sodium sulfite solution. Electrodes were rinsed with deionised water before and after each measurement, stored in appropriate conditioning solutions (pH electrode), and periodically cleaned to remove biofilm or sediment residues.This approach ensured stable measurement conditions and reproducible data quality throughout the entire sampling period. Following statement was added in the manuscript:

'...All pH, SEC and DO probes were calibrated using certified standards before each field study at the laboratory of the Institute of Geography and Spatial Organization Polish Academy of Sciences. Calibration was performed according to the manufacturer's guidelines using certified analytical standards: pH probe: three-point calibration (pH 4.0, 7.0, and 9.0 buffers); SEC probe: calibration with a 1,41mS·cm$^{-1}$ standard solution, selected according to anticipated conductivity of the sampled waters; DO probe: air-saturation calibration (100%) followed by zero-oxygen verification using sodium sulfite solution…'.

- Sampling design: Clarify whether samples were collected under baseflow or post-rain conditions. Define how "normal water levels" were determined (e.g., relative to long-term discharge averages).

**Answer:** The research was conducted under baseflow conditions. For this reason, we collected spring water samples at the end of spring, in order to avoid the effects of snowmelt. We replaced 'during normal water levels (i.e., excluding periods of precipitation or snowmelt)' with 'during baseflow conditions (i.e., excluding periods of rainfall events or snowmelts)' to clarify the hydrologic context.

- Temporal resolution: Indicate how long each sampling campaign lasted and whether samples were taken at consistent times of day (important for DO and temperature).

**Answer:** We added information on temporal resolution in the description of methods:

'...Samples were collected between late morning and afternoon, with sampling at each site completed within about an hour…'.

- Analytical precision: Report detection limits and analytical errors for key parameters ($NO_3^-$, $NH_4^+$, $SO_4^{2-}$).

**Answer:** Before each analysis, the samples were filtered through a glass microfiber filter Whatman GF/D. The analysis of chemical composition (the main anions and cations) was carried out using an ion chromatograph (DIONEX ICS 3000) with analytical columns: IonPac CS16 3×250 mm (for cations) and IonPac AS18 2×250 mm (for anions). The calibration curve was plotted based on *Dionex six cation II standard* and *Dionex seven anion standard II*. Certified reference material KEIJM-02 from *Environment Canada* was used for the purpose of quality control of the obtained results. The accuracy of the analytical determinations was evaluated using the relative difference (Dev) between the measured and certified concentrations. For the majority of analysed ions, Dev values ranged from –3.9% to 2.1%, demonstrating very good agreement with the certified reference material. For $K^+$, the Dev value was slightly higher (–5.8%), yet still within an acceptable deviation range for this ion.

In addition to the routine use of certified reference materials (CRM) in every analytical batch, the laboratory participates annually in interlaboratory comparison exercises organized by the Global Atmosphere Watch/World Meteorological Organization (GAW/WMO) in cooperation with the Quality Assurance/Science Activity Centre – Americas (QA/SAC Americas). This programme provides an international framework for the external quality assessment of chemical data related to atmospheric deposition.The interlaboratory tests involve the analysis of a set of artificial rain samples containing deionised water spiked with known concentrations of major inorganic ions: $SO_4^{2-}$, $NO_3^-$, $NH_4^+$, $Na^+$, $Mg^{2+}$, $K^+$, and $Ca^{2+}$. These samples are analysed using the same analytical methods and procedures that are applied routinely in the laboratory for field samples. The results obtained are compared with: i) assigned reference values provided by the organizing laboratory, ii) the median values reported by all participating laboratories, and iii) the acceptable deviation thresholds defined by the GAW/WMO QA programme. According to GAW/WMO quality criteria, analytical results are considered satisfactory when the accuracy falls within: 5% for $SO_4^{2-}$ and $NO_3^-$, ±7% for $NH_4^+$. The laboratory meets these criteria annually, confirming the stability of the analytical method, the correctness of the calibration procedures used and the high reliability of the data generated. Regular participation in comparative studies is an additional element of external validation and quality assurance (QA/QC), in line with international standards for reporting atmospheric deposition chemistry. We have supplemented section 2.2 Water sampling and chemical analysis, with the necessary information about chemical analyses:

'...The ionic composition ($mg \cdot L^{-1}$) was analysed in the Dionex ICS3000 ion chromatograph with analytical columns: IonPac CS16 3×250 mm and IonPac AS18 2×250 mm. Each sample was previously filtered through a glass microfiber filter Whatman GF-D. The calibration curve was based on the certificate reference standard (CRM) Dionex Combined Six Cation Standard II and Seven Anion Standard II. All solutions were prepared with 1% nitric acid (Suprapur). The following concentrations were prepared for calibration: 0 (blank), 1, 10, 50, 100 and 1000 $\mu g \cdot dm^{-3}$. The percent relative error, as determined by the data quality assessment based on CRM (KEIJM-02, GAW/WMO) was less than 5.0% for all parameters except $K^+$ and $NH_4^+$. The respective values were 5.8% for $K^+$ and 7.0% for $NH_4^+$…'.

- Pond characteristics: Include basic information on pond area, depth, dam height, and vegetation cover. These features are crucial for interpreting variability among sites.

**Answer:** We complemented Table 1 with additional information on the pond area of the biggest pond in the sequence which was selected as a representative pond for the sampling, and the ratio of the pond sequence area to the area of the river channel which represents the intensity of overflowing. Additional textual description was also added:

'...The height of the beaver dams studied did not exceed 1.5 m. In most dam sequences, the ponds were directly connected. The only exception was site 7, where short sections of free-flowing reaches were observed between the ponds…'.

Unfortunately, quantitative data on vegetation cover were not collected during this study, which does not allow for any direct assessment of this biotic factor. We have taken into account the importance of vegetation cover measurements while revising the limitations and recommendations.

- Data processing: Explain any data transformations or normalization applied before PCA.

**Answer:** We used standardised data on solute concentrations in stream water above beaver dams, to determine the physico-chemical background of the studied streams. However, we removed this section (and so the information on standardization) in order to shorten the paper and focus on the results related to the effects of beaver ponds. We based the rest of our analyses on the percentage changes rather than raw data. Nevertheless, we supplemented and corrected the description of data processing and parameter selection (section 2.3 Statistical analysis).

Figures and tables

The figures contain useful information but are difficult to interpret in their current form. They all could benefit from more detailed figure captions. The following revisions are recommended:

- Figure 1: Add a scale bar and north arrow. Increase contrast for better readability.

**Answer:** We have revised this figure. The scale bar and north arrow have been moved to a white insert to increase their visibility. The grey colours were changed to black and white on the regional map, and a lighter background colour was used on the main map, to increase contrast.

- Figures 2–3: Clearly define all abbreviations and units. For seasonal plots, consider color-coding by season rather than by site to emphasize temporal patterns.

**Answer:** We removed the list of measured parameters from Figure 2. It is now presented in the beginning of the section '2.2 Water sampling and chemical analyses' and include a description of the abbreviations that are used further in the text:

'…The following water parameters were measured: temperature (T), dissolved oxygen (DO), specific conductivity (SEC), pH and the ionic composition ($Ca^{2+}$, $Mg^{2+}$, $Na^{+}$, $K^{+}$, $NO_3^{-}$, $NH_4^{+}$, $HCO_3^{-}$, $SO_4^{2-}$, $Cl^{-}$)…'.

Units were introduced in this section along the description of the detailed measurement methods.

Figure 3 has been removed from the text in order to shorten the paper, following reviewers' suggestions regarding focusing on the most relevant content.

- Figures 4–7: Reduce the number of variables plotted. Focus on the most ecologically relevant ones (e.g., pH, $NO_3^{-}$, $NH_4^{+}$, DO, $SO_4^{2-}$). Move less critical parameters (e.g., $Mg^{2+}$, $K^{+}$) to supplementary materials.

**Answer:** We have kept all the variables in Figure 4 (now Figure 3) as it was intended to present the general overview of the changes observed. This is important to set out the background for further discussion.

However, while revising Figure 5 (now Figure 4) we applied a similar approach to the presentation of the parameters, as to their inclusion in PCA analyses. This allowed for both reduction of the number of plots and increase in consistency. Only the parameters for which mean absolute difference is greater than 5% are now presented. This includes removal of less influential water parameters, such as $Mg^{2+}$ and $Na^{+}$.

Figures 6 and 7 (now Figure 5) present the combined changes in water chemistry based on selected parameters. Only the parameters for which mean absolute difference is greater than 5% are taken into account.

- Figures 8–11 (PCA): Improve label readability and explain the meaning of PC1 and PC2 in the caption. Include the percentage of variance explained by each component.

**Answer:** Figures 6 and 7 (now Figure 5), which present the results of PCA analyses, have been revised. We kept the plots showing the grouping by study sites and seasons, but removed the plot combining these two classifications, as it did not provide any new information and could confuse the reader. We included factor loadings and share of explained variance for F1 and F2 on the plot, instead. Additional information (e.g. regarding factor F3) can be found in a supplementary table. Figures 8-11 (now Figures 6, 7, B1, B2) have been changed substantially. Combining the sites and parameters improved efficiency and clarity.

- Tables: Ensure consistent unit formatting and significant figures.

**Answer:** We have unified units and significant figures for all the chemical components.

Minor comments

- Terminology: Use "physico-chemical properties" or "water chemistry" consistently throughout.

**Answer:** 'Water chemistry' was replaced with 'physico-chemical water parameters' in the core parts of the manuscript, such as the revised study aim and chapter titles, to make it clearer and ensure consistency with the rest of the text, in which particular water parameters are referred to. We also propose a slight change to the title of the manuscript, which includes replacing 'water physico-chemical composition' with 'physico-chemical properties of water'. The term 'water chemistry' was retained in the text when referring to general information on water chemistry.

- Units: Ensure all concentrations use consistent units ($mg\ L^{-1}$).

**Answer:** We have changed units to $mg \cdot L^{-1}$ and corrected their formatting throughout the manuscript.

- pH interpretation: A pH decrease is not inherently negative; interpret it relative to baseline conditions.

**Answer:** We agree that a pH decrease is not inherently negative. However, we did not write such a statement anywhere in the text. We agree that pH changes should be interpreted relative to baseline conditions. In our study, the baseline is defined as the upstream reference sections located above the ponds. To emphasize this, we added this statement in the following sentences in the Methodology and Results sections:

'...The section 'above' the beaver pond or pond sequence that reflected baseline conditions was defined as 50–100 m upstream from a backwater, and the section 'below' was 50–100 m downstream from the last dam…',

'...The physico-chemical properties of stream water at baseline conditions varied between the catchments studied (Table 2)...'.

- Seasonal terminology: Define "warm" and "intermediate" seasons clearly (e.g., May–September, October–April).

**Answer:** We specified the warm season clearly in the revised methods as spring-summer and provided the sampling dates

**OTHER CHANGES**

In response to the community comment posted during an interactive discussion and the third review, we would like to propose a slight change to the title, from 'The effect of beaver ponds on water physico-chemical composition in the Carpathians (Poland and Slovakia)' to 'The effect of beaver dams on physico-chemical properties of stream water in the Carpathians (Poland and Slovakia)'.

**Overall, we are grateful for all the thorough reviews that led to substantial improvements in the resubmitted text. We look forward to hearing from you regarding the revised manuscript.**

On behalf of all authors of the submitted manuscript, sincerely

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