Author's response to comments by anonymous referee #2

We would like to thank referee #2 for taking the time to read our manuscript carefully and for providing constructive feedback. The helpful suggestions will improve the quality of our manuscript. Our responses to the individual comments are shown in blue below.

Summary

Greiwe et al. collected diel nitrate data from three locations in a stream over multiple months to determine the controls of diel nitrate signals. They used cross correlation to show that diel signals were controlled by local in-stream processes rather than from upstream. Next, they used cluster analyses to identify consistent patterns in the diel signals. This is a novel and interesting approach. Finally, they relate the clusters with light and discharge to tease apart what is controlling each cluster. I think this is an interesting and worthwhile paper. I particularly like the use of cluster analyses on the diel data to identify common trends in the diel cycle. However, I believe major revisions are necessary before publication. The biggest issue I have is the attempt to explain diel patterns based on unmeasured microbial processes. This is especially complicated given that many of these processes can cancel each other out (e.g., nitrification and denitrification) and we do not have easily measured proxies (like we have light for photosynthesis). Thus, I suggest that the authors tone down much of the speculation about microbial pathways, and instead focus on what they can show with data.

> Reply: We appreciate the positive attitude of referee #2. We agree that our statements on the relative importance of processes are too speculative and should not be presented as key findings and will significantly shorten this section in a revised version of this manuscript. This has also been suggested by referee #1.

I also have some concerns with the methods. The cross-correlation approach could be described in more detail. Most importantly, there should be more detail about how the cluster analysis was performed. I am not an expert on cluster analyses and found it confusing how diel curves with multiple data points were put into a cluster analysis. As I mentioned above, I really liked this novel approach and I think it could be used for other constituents (DO, CO2, etc.). A better description of the methods would make it easier for others to replicate the analysis.

> Reply: We will add information on the methodology to make our analysis more understandable. The description of the cluster method was intentionally kept short as it was applied before by Aubert and Breuer (2016) (L. 117).

Title: I would remove the reference to microbial pathways. This paper has no data to back up the suggested trends in microbial processes.

> Reply: The title will be changed to "Diel patterns in nitrate concentration produced by in-stream processes".

Line 15: What is plug-flow?

> Reply: The concept of plug flow originates from fluid mechanics where it describes uniform velocity profiles in pipes. In this concept zero dispersion and zero storage zone exchange are assumed. We will introduce the concept in a revised manuscript.

Line 25: A key part of the spiral is that the nutrients are then mineralized to the water column to be taken up again downstream. This should be added here.

> Reply: We will add this aspect to the description of 'nutrient spiraling'.

Line 31: Can you better describe the link between climate change and nutrient retention? What role does drought play?

> Reply: Climate change increases the probability of drought (at least these are the predictions for Southwest Germany) causing low flow conditions or zero flow (as in the reference) to occur more frequently. This in turn may influence stream temperature, light penetration and

other aspects that are important for biochemical in-stream processes. We will explain this in more detail in a revised manuscript.

Line 46: Denitrification is a heterotrophic process. This line implies that denit could occur via autotrophic processes. Please revise.

Reply: We will revise this phrase to prevent misunderstanding.

Line 93: Please provide more information about the periodical movement of the sensors. Were the sensors moved at equal intervals? Is the data available from each sub-reach stratified across the sample period?

Reply: In this figure it will be visible which sensor was placed where for how long.

Lines 106-108: Could you provide more info on interpreting the cross-correlation data? What does a low and high correlation mean? How does this help better elucidate N transported from upstream vs. from in stream processes? A few lines here will help the reader going forward, especially to understand figure 2.

Reply: We thank referee #2 for pointing out that we provided insufficient information on our methods. We will provide more background information in a revised manuscript.

Line 110: What travel time distribution is this referring to? You only conducted one tracer release (I think).

Reply: Exactly, that is the one we are referring to here. So this only gave us one data point (black cross in Fig. 2). In order to visualize how travel time would change when discharge changes we additionally estimated a range of nominal travel time estimates based on the local channel geometry.

Line 120: I believe that residual should be added earlier in the sentence. ": : :was done on the residuals of the diel solute concentration signal."

Reply: Thank you. This will in fact improve readability and will be changed accordingly.

Lines 115-127: I am having troubles understanding how the clusters were determined, or in other words, how the k-means approach turned diel data into clusters. Could that be described more? I am used to clusters being used with single values (i.e., animal abundance data), so how can multiple points be used (i.e., from a diel curve). I do not have much experience with clustering, but that will be true for many readers as well. More detail would be helpful.

Reply: We will provide additional information on that to make the methods section more understandable.

Line 160/Figure 2: This took some time to determine what I am looking at. Is the main point that points with a high cross-correlation are typically between 0 and the nominal travel time (the shaded area)? Either way I would add a line or two describing the main result out of this figure. Also, how is it possible that a travel time is negative?

Reply: Thank you. We realize that this figure requires more explanation which will be added in a revised manuscript. Of course, travel time cannot be negative, but lags determined by cross-correlation can. In our study this occurred when cross-correlation between the two signals was weak, i.e. the signals were not very similar. We agree that these data points are confusing and distract from the key massage of this figure. As suggested by referee #1 we will therefore remove all data points representing non-similar signals ($r > 0.75$).

Line 175/Figure 3: Do the shaded areas represent a confidence interval? And what calculations were used to calculate the shaded area?

Reply: Shaded areas in figure 3 represent the range between the 5^{th} and 95^{th} percentile, i.e. 90% of the data and black dots represent medians. We will add this information to the figure.

Line 200/Figure 5: Would it be logical to make the y-axis a proportion? The ups

and downs are distracting. Making them a proportion would better show the seasonal trends.

Reply: We agree that the ups and downs are somewhat distracting. On the other hand, showing relative data may suggest that there is information where we actually have none. We therefore opt to keep the absolute values but represent months as bars (not areas) as suggested by referee #1.

Line 201: Something is missing here. Maybe, "Relation of nitrate clusters and reach balance"

Reply: Thank you. This will be corrected.

Line 220: Please define or further explain short-circuiting.

Reply: This phrasing will be revised.

Line 223: "stated"? Maybe observed?

Reply: This phrasing will be revised.

Line 226: This explanation of the Hensley and Cohen paper is confusing and hard to follow. Could you describe the point of the paper without getting into the details?

Reply: As referee #1 had similar concerns regarding this paragraph, we will do our best to simplify this section and make it more understandable.

Line 240: I don't believe the description of clusters E and F being influenced by discharge is in the results section. How did you come to this conclusion?

Reply: We indeed missed to state that in the results section. The fact that discharge during cluster F (n=5) was elevated can be seen in figure 4. We may have been a bit quick stating the same for cluster E (n=21). Anyway, we should be careful not to overinterpret clusters with only few data points.

Line 260: What is the relevance of the 0.5 mg/L SRP? What does this threshold indicate?

Reply: Around this threshold we would be able to detect SRP via ion chromatography. However, we are aware that such values would be quite high for surface waters in the study region. This threshold is not of any biological relevance and we will delete this sentence from the manuscript.

Line 287: This is also true for estimates of stream metabolism.

Reply: In fact, stream metabolism estimated from dissolved oxygen data relies on very similar assumptions.

Line 247-300: There is a lot of speculation on the drivers of diel patterns in here. It would be much more convincing to use a statistical analyses/models to make conclusions about what is controlling the diel trends rather than relying on the literature and instinct. The correlations with light are somewhat compelling for the first two clusters but it is still hard to disentangle the different microbial pathways relative to the autotrophic. For the other clusters it gets much more complicated and interpretation is pure speculation. That being said, I still think these data are useful and novel. But tying each cluster to a specific driver is for another paper in my opinion. I suggest that this part of the discussion be substantially shortened. I like how you first describe the strong evidence that in-stream, not upstream, processes are driving diel trends. Then go through the clusters or sets of clusters and do some light speculation on the drivers of the signals in relation to the literature. This is done quite well in lines 303-331.

Reply: We agree that parts of our discussion are very speculative and are not sufficiently backed up with data and our analysis. We will shorten corresponding parts of the discussion.

Line 353: Is there a citation for these data?

Reply: The corresponding groundwater data are publicly available. A and figure and references will be provided in the supplementary material.

Line 355: The topic of groundwater should be introduced and described much earlier in the methods section. Also, please address how groundwater might affect the diel curves? Groundwater is likely an important factor for diel curves during summer low flows.

Reply: We will introduce the potential influence of groundwater earlier in the manuscript.

Line 371: We know this already–In my opinion, this is not the strength of this paper. I would end here with a line noting how you were able to separate diel trends in NO3 concentrations into clear clusters with distinct diel patterns and probably different drivers.
These clusters can be used a blueprint for future efforts to model drivers of N cycling.
Likewise, using the cluster analyses on diel data is a novel approach and could be used for other measurements (e.g., DO, CO2, SRP, etc).

Reply: We appreciate these suggestions and we will revise the conclusion in accordance with the new focus of the paper.