

## ***Interactive comment on* “Stable isotopes of nitrate reveal different nitrogen processing mechanisms in streams across a land use gradient during wet and dry periods” by Wei Wen Wong et al.**

### **Anonymous Referee #3**

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In the manuscript *Stable isotopes of nitrate reveal different nitrogen processing mechanisms in streams across a land use gradient during wet and dry periods*, Wong et al present natural abundance nitrate isotopes from five streams across a land use gradient during wet and dry seasons, allowing them to elucidate the controls on sources and transformations of nitrate. This is an interesting dataset and the authors have been resourceful and knowledgeable in their presentation and interpretation of the data. However, prior to publication the manuscript would benefit from a clear and concise definition of terms, and a clearer explanation of the isotope effects and their subsequent implications, as currently it seems hard to follow in places for the none expert reader.

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An important aspect of the interpretation of this dataset is that there is a tight coupling of mineralisation and nitrification, resulting in no isotope effect being expressed and hence the  $^{15}\text{N}$  of organic matter / ammonium and nitrate are similar. Currently this is not fully explained until Page 9 Line 24, making it difficult to understand the authors interpretation of the data prior to this, explaining this earlier on in the discussion will enable the reader to follow your thoughts / interpretation. A good example of this is Page 6 Line 15/16, break this thought down and explain to the reader here the tight coupling between mineralization and nitrification and hence no isotope effect being observed.

### Specific comments

Page 2 Line 10: it would be valuable here to state that you are talking about kinetic isotope effects and not equilibrium.

Page 2 Line 23 to 25: for the none expert, please explain why rainfall patterns are different in the southern hemisphere and its subsequent effects.

Page 2 Line 32 to 33: start preparing your reader here, why are denitrification and assimilation more prevalent in wet periods.

Study area: throughout this section you refer to the gradient in land use across the catchment, a map of this would be a great addition to the manuscript (or could maybe be added to Figure 1).

Page 3 Line 32: how do the authors think using an integrated signal could of biased their interpretation of the results?

Page 5 Lines 9 to 10: the term total nitrogen needs to be defined here, as it is important for the mineralization discussion later on. Would particulate organic nitrogen not be a more suitable term? Also, please add in that the values are relative to AIR and the precision of the measurements.

Page 5 Lines 24: Please add in nitrite concentrations, to confirm for the reader that the

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values are less than 1% of the nitrate (as stated in the methods).

Page 6 Line 3: Be clear that you are talking about 15N values here.

Page 6 Line 4: The enriched 15N-nitrate values seem to be constrained to a thin band between 70 to 85% agriculture, but then drop away again at higher percentage agriculture, do the authors have any hypotheses for this?

Page 6 Line 5: Surely the same is true for the Bass.

Page 6 Line 16 to 28 and Equation 1: It would be valuable to explain to the readers the value of using both N and O isotopes i.e. N is recycled between fixed N pools and the O atoms are removed and then replaced by nitrification and thereby sensitive to internal processing (this could come here or in the introduction). The authors need to discuss the more recent literature when introducing and determining the oxygen isotope signal imparted by nitrification, the work of Carly Buchwald is particularly pertinent here.

Page 6 Line 29: Is this value for cow manure similar to the literature to date?

Page 7 Line 14: 'terrestrial' what are the authors referring to here, fertilizer, manure, leaf litter, please be consistent with the use of terms throughout.

Page 7 Lines 15 onwards: a slight restructure here would be beneficial, you are presenting your conclusions before the evidence, discussing your isotope data first in this section would make it easier to follow.

Page 7 Line 21: I think the authors are referring to Table 2 here.

Page 8 Line 15 and 32: I strongly suggest the authors cite and discuss the implications of the outcomes from the work of Granger and Wankel, 2016 (Isotopic overprinting of nitrification on denitrification as a ubiquitous and unifying feature of environmental nitrogen cycling; PNAS) and how this may influence your interpretation of N turnover in your catchment.

Page 10 section (3): an earlier introduction of the different behaviors of N and O iso-

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topes during internal processing of nitrate will make this section easier to understand for the none expert reader. I would not put denitrification under the heading recycling, if the authors are referring to nitrate reduction, followed by reoxidation please say so.

Page 10 Line 11: Do the authors know when fertilizer is applied in this catchment, how does this align with your runoff / turnover hypotheses?

Figure 1: Please mark on the map of Australia where southern Victoria is.

Figure 3: Mark on upper / lower Bunyip.

Figure 5: Where have the authors taken these isotope effects from? Please cite the relevant literature in the caption. A positive / inverse isotope effect for nitrification?

Figure 7: More details are needed in the figure caption, what do the crosses and dashed line mean? I also assume that it is the y intercept values determined in Figure 6 that have been plotted.

Figure 9: define what starting values you have used and where they have come from, particularly for the oxygen isotopes.

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