#### Response to Anonymous Referee #2

#### **Summary:**

This study assessed how geographic controls (elevation, temperature, and slope) and % anthropogenic land cover (urban/agriculture) influence DOC export and DOM composition from mountainous rivers. The data presented shows that increased %urban/agriculture cover in lower reaches (and shallower gradients) of these catchments results in higher DOC concentrations, where carbon isotopic signatures (13 and 14C-DOC) of DOC are more deplete, and DOM is less aromatic. I believe these findings are of interest to a broad community. However, I have a number of concerns that I would like the authors to address and some suggestions to improve their manuscript.

**Our response:** We thank the reviewer for the time and effort spent reviewing our paper. We also appreciate your valuable remarks and constructive suggestions, which will much improve the manuscript.

# Major:

1. The SUVA254 values presented in this paper are typically >5 L mg C<sup>-1</sup>m<sup>-1</sup>. These values are extremely high when compared to blackwater riverine systems that typically have high aromaticity DOM (e.g., Holt et al., 2021; Spencer et al., 2010; Weishaar et al., 2003 and references there in). Please can you confirm how SUVA values were calculated (and include this information in text and/or DOC and absorbance data in a table). Were decadic or napierian absorbance measurements used for calculations? SUVA values should be calculated from decadic absorbance and if naperian values were used instead this may explain why the values here are so high (Hu et al., 2002; Spencer et al., 2014). Some justification of why SUVA values are seemingly so high would be useful here, and it would be good if these values were contextualized in relation to past work for mountainous rivers (and anthropogenically impacted catchments).

Our response: Thanks. The previous SUVA values were calculated from naperian values and thus why they are so high. We have re-calculated them from decadic absorbance. The related information has been added in the manuscript as follows: "Decadic absorbance values were used to calculate absorption coefficients as below (Poulin et al., 2014):  $a_{254} = Abs_{254}/L$ . Where,  $a_{254}$  is the absorption coefficients (cm<sup>-1</sup>),  $Abs_{254}$  is the absorbance at 254 nm, and L is the path length (cm). Specific UV absorbance at 254 nm (SUVA<sub>254</sub>) was determined according to Weishaar et al. (2003; Table 1):  $SUVA_{254} = a_{254}/DOC$ ." (P6 Line 159-164). The data on how SUVA values were calculated are listed in the table below:

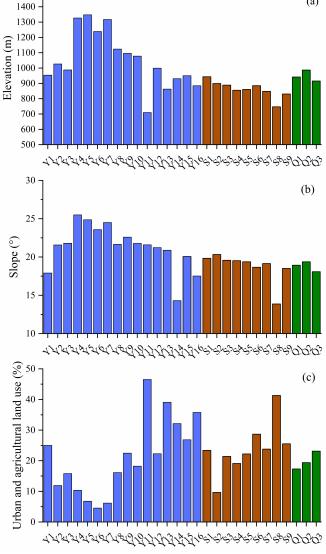
Site ID	DOC (mg L-1)	Abs254	a254 (cm <sup>-1</sup> )	SUVA254 (L mg <sup>-1</sup> m <sup>-1</sup> )
Y1	1.2	0.041	0.041	3.4
Y2	0.9	0.040	0.040	4.3
Y3	0.9	0.035	0.035	3.8
Y4	0.9	0.034	0.034	3.8
Y5	0.7	0.031	0.031	4.4
Y6	0.4	0.019	0.019	5.2
Y7	0.6	0.030	0.030	4.7
Y8	0.7	0.022	0.022	3.1
Y9	1.3	0.037	0.037	2.8
Y10	1.2	0.042	0.042	3.6
Y11	1.5	0.037	0.037	2.5
Y12	2.2	0.066	0.066	3.0
Y13	2.0	0.040	0.040	2.0
Y14	2.9	0.028	0.028	1.0
Y15	1.1	0.040	0.040	3.5
Y16	1.7	0.032	0.032	1.9
<b>S</b> 1	1.7	n.a.	n.a.	n.a.
S2	1.1	0.041	0.041	3.8
S3	1.4	0.101	0.101	7.0
S4	1.9	0.055	0.055	3.0
S5	1.7	0.047	0.047	2.7
<b>S</b> 6	1.4	0.020	0.020	1.4
S7	1.5	0.041	0.041	2.8
<b>S</b> 8	1.5	0.022	0.022	1.4
<b>S</b> 9	1.4	0.040	0.040	2.8
Q1	1.7	0.055	0.055	3.2
Q2	1.7	0.047	0.047	2.8
Q3	1.7	0.043	0.043	2.5

Note: n.a. means the data are not available.

In addition, these values were contextualized in relation to past work in the manuscript as follows: " $SUVA_{254}$  values for the three study rivers were comparable with those reported in coastal glacier mountainous streams with late succession in southeast Alaska (3.4 ± 0.5 L mg<sup>-1</sup> m<sup>-1</sup>, n = 5; Holt et al. 2021) and in the anthropogenic influenced downstream of the Yangtze River (3.4 ± 1.1 L mg<sup>-1</sup> m<sup>-1</sup>, n = 82; Zhou et al. 2021)." (P17 Line 388-391).

2. There are details missing from the methods in relation to how % land use, slope and elevation was calculated or whether this data is from the author's previous study. This information should be included in this paper since many of the figures and findings are reliant on this data. I would also recommend data from this analysis is presented and described within the site description before it is used to inform your analyses with DOM composition.

Our response: Thanks for your suggestions. We have added details on related information in "2.1 Study area" and figure about sites specific distribution of land use, slope and elevation in the SI as follows: "Catchment characteristics (e.g., mean drainage elevation, annual air temperature, mean channel slope, and proportion of urban and agricultural land uses) for the subcatchments were determined using ArcGIS (version 10.2). The mean drainage elevation of these three catchments ranges from 340 m to 2424 m with the lowest and highest elevation both reported in the Yinjiang River catchment, showing a great change in relief (Figs. 1a and S1a). Similar to elevation, the Yinjiang River catchment has a greater variation in mean channel slope (from 14.3° to 25.5°), while the channels in the Shiqian and Yuqing river catchments have a mean channel slope of approximately 20°, except the segment above site S8 (13.9°; Figs. 1b and S1b). Carbonate rock is widely distributed in the three catchments, accounting for a large proportion of the exposed strata (Han and Liu, 2004). The remaining areas are mainly covered by clastic rocks, igneous rocks, and low-grade metamorphic rocks. Forest, agriculture, and urban areas are the three dominant land uses in these studied catchments (Fig. 1c). Forest is generally distributed in high-elevation regions, while urban and agricultural land uses are mainly located in low-elevation regions. The proportion of urban and agricultural land uses in the Yinjiang and Shiqian river catchments is from 4.5% to 46.5% and from 9.6% to 41.3%, respectively, showing a large variation relative to the Yuqing River catchment (from 17.3% to 23.1%; Figs. 1c and S1c)." (P4 Line 101-113).



(a)

1500

**Figure S1** Distribution of (a) mean drainage elevation, (b) mean drainage slope, and (c) urban and agricultural land use in the Yinjiang (Y), Shiqian (S), and Yuqing (Q) catchments.

3. Findings of figure 5,6,7 are discussed without an initial description of the data/trends and thus much of the discussion comes across as a little abrupt. I would recommend you restructure and describe these figures/trends in the results section, then explain what these trends (either individually or collectively) may mean in the discussion. As it stands the discussion and results are a little brief and findings are not really discussed in detail (especially section 4.1). Given the structure it is also difficult to follow the primary reasons for the trends you observe. I wonder if you could examine trends within figures 5-7 collectively (e.g., through a PC analysis) so connections between land use and slope can be made and discussed in tandem rather than separately?

Our response: Thanks. We have restructured and described these figures/trends in the results section as you suggested. Details are in the section "3 result" of the revised manuscript (please see section 3.2 as an example shown below). We have tried to examine the trends through a PCA analysis, but it failed due to the limited number of <sup>14</sup>C data. Instead, we examine the trends in a correlation plot (Fig. S3) as shown below to facilitate the further discussion on the trends.

# 3.2. Riverine DOM Optical Properties

The average SUVA<sub>254</sub> were  $3.3 \pm 1.1$ ,  $3.1 \pm 1.8$ , and  $2.8 \pm 0.3$  L mg<sup>-1</sup> m<sup>-1</sup> in the Yinjiang, Shiqian, and Yuqing rivers, respectively. Although no spatial differences in SUVA<sub>254</sub> were found across the three rivers (Fig. S4a), SUVA<sub>254</sub> showed an increasing trend with increasing mean channel slope (Fig. 3e), which indicated that DOM in the lower reaches with a gentle channel slope was less aromatic. Furthermore, there was a significant negative correlation between SUVA<sub>254</sub> and proportion of urban and agricultural land uses (Fig. 6a).

Two humic-like fluorescence components (C1 and C3) and one protein-like fluorescence component (C2) were identified by PARAFAC model in these three rivers (Fig. 7; Table S1). Component C1 is similar to traditionally defined peak M and sourced form microbial processes or autochthonous production (Kim et al., 2022; Li et al., 2016; Walker et al., 2009). Component C2 was previously related to recent biological production (DeFrancesco and Guéguen, 2021; Du et al., 2019; Lambert et al., 2017). C3 was the most widely found component in previous research among three fluorescence components, and was identified as traditional fulvic-like peaks A and C, representing terrestrial delivered OM or authochthonous microbial sourced OM (Amaral et al., 2016; Ryan et al., 2022; Shutova et al., 2014). Although C1 and C2 varied more widely in the Yinjiang River compared with the Shiqian and Yuqing rivers, the two fluorescence components did not show a statistical difference among the three rivers (Figs.

7a and b). However, a greater proportion of C3 was found in the Shiqian and Yuqing rivers, exhibiting a distinctive signature compared with the Yinjiang River (Fig. 7c). The fluorescence components did not exhibit any significant variations with changing channel slope (Fig. S3), but C1 and C2 were positively or negatively related to proportion of urban and agricultural land uses (Figs. 6b and c).

For the fluorescence indexes, the overall fluorescence property did not vary significantly among the three rivers (Figs. S4b, c, and d). FI varied in a narrow range compared with  $\beta/\alpha$  and HIX. FI of DOM ranged from 1.66 to 1.94, averaging 1.78 (Fig. S4d), indicating a mixture of DOM of terrestrial and microbial origins. In comparison,  $\beta/\alpha$  varied from 0.70 to 1.22 (Fig. S4b) and HIX varied from 0.33 to 0.65 (Fig. S4c), with greater variability among the three rivers. Although no significant correlation was observed between channel slope and the fluorescence indexes, they (except for FI) were found to be closely related to land use pattern (Figs. 6d, e, f, and S3). The less recently produced DOM ( $\beta/\alpha$ ) in the urban and agricultural streams was also characterized by a higher proportion of C1 and lower proportion of C2 (Fig. S3).

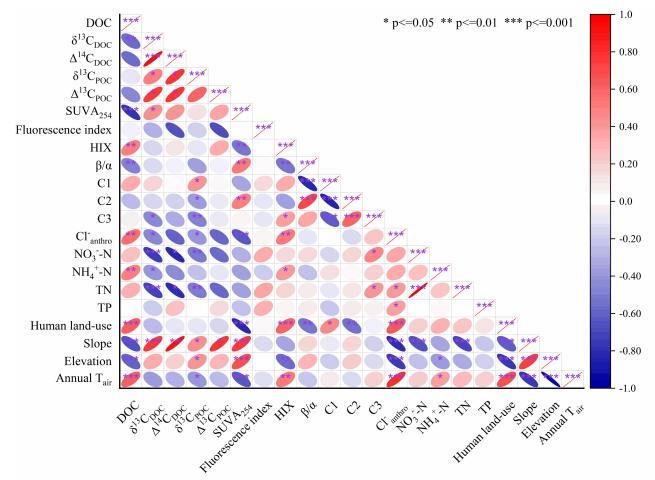


Figure S3 Correlation plot of the selected water chemistry and catchment characteristics. The colors represent the degree of pairwise correlation regarding Pearson's correlation coefficient.  $\delta^{13}C_{DOC}$  and  $\Delta^{14}C_{DOC}$  at site Y12 was excluded from analyses as the sample was collected after a rainfall event. In addition, SUVA<sub>254</sub> at site S3 was excluded from analyses as the sample was strongly influenced by the road construction, which was evidenced by high POC and TSM concentration (Chen et al., 2021). Human land use used here represents proportion of urban and agricultural land use. Elevation and Annual  $T_{air}$  represent mean drainage elevation and annual air temperature, respectively.

4. L189-190 it is unclear how 'enhanced' biodegradation of DOC would increase DOC concentrations. I would have thought that biodegradation would remove DOC. Also, how can you be sure that this trend in 13C-DOC is microbially driven, rather than an increased input from aged soil/C3 plants in lower elevation stream reaches? You note in your site description that there is C3/C4 agriculture across the catchments. Is this coverage variable and could this in part drive the trend in 13C-DOC? Additionally, wouldn't removal of 12C by microbes lead to enrichment in 13C-DOC not depletion as you describe?

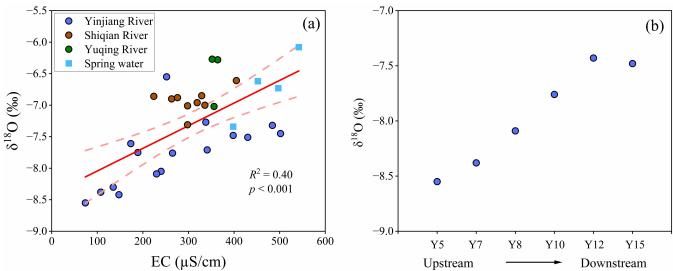
Our response: Thanks. Here, the enhanced biodegradation of DOC with the preferential removal of  $^{12}$ C is the process happen in the soil layers or spring water. Thus, the remaining DOC would be  $^{13}$ C-enriched and input into the rivers. We have rewritten this part in the manuscript as follow: "Previous studies have reported a decreasing  $\delta^{13}C_{DOC}$  with a corresponding increase in DOC concentrations (Fig. 5a) in spring water (Nkoue Ndondo et al., 2020) and for TOC in soil profiles (Lloret et al., 2016; Nkoue Ndondo et al., 2020). This can be explained by the lateral transport of DOC from microbial active soil horizons into rivers (Lambert et al., 2011), resulting in the enhanced biodegradation of DOC with the preferential removal of  $^{12}$ C. As a result, the remaining DOC of lower concentrations is typically characterized by a heavier  $\delta^{13}C_{DOC}$  (Nkoue Ndondo et al., 2020; Opsahl and Zepp, 2001)." (P15 Line 329-334).

5. I wonder if the authors could further discuss how they arrived at the conclusion that groundwater was a significant source of DOC to these rivers, and that this groundwater played an important role in diluting DOC concentrations during base flow (1)

given that statistical analysis was not performed between springs and river DOC samples; (2) that samples were taken during heavy rainfall periods (i.e., monsoon; September) and thus baseflow conditions were unlikely to have been examined; (3) optical and isotopic data was not used to inform this discussion.

**Our response:** Thanks. We have further discussed this point and arrived at the new conclusion that groundwater is an important but not a crucial source of riverine DOC.

(1) Statistical analysis was added in the "2.4 Statistical analysis" and in the results as follow: "The DOC concentrations in spring water were significantly lower than that in the surface water of the Shiqian and Yuqing rivers (Fig. 2d), and the average DOC concentration in spring water  $(0.74 \pm 0.30 \text{ mg L}^{-1})$  was also lower than the average DOC concentration in the Yinjiang River  $(1.27 \pm 0.66 \text{ mg } L^{-1})$ , indicating there must be other sources of DOC besides groundwater." (P9 Line 222-225). For question (2) and (3), we have rewritten this part and added information on  $\delta^{18}$ O as evidence to further support our conclusion in the manuscript as follow: "Groundwater with large SOC inputs due to highly active microbial activities has long been recognized as a significant source of DOC, especially under warm and wet conditions (McDonough et al., 2020; Shen et al., 2014). Several studies have reported increased groundwater contributions with distance downstream at the watershed scale (Asano et al., 2020; Cowie et al., 2017; Iwasaki et al., 2021), suggesting the important role of groundwater in mountainous rivers, though this is not a general phenomenon due to great differences in catchment characteristics (e.g., bedrock and topography) and climate (e.g., precipitation; Somers and McKenzie, 2020). The positive relationship between conductivity and  $\delta^{18}O$  are due largely to the mixing of two end-members for river water (Fig. S2a), though it may also indicate the evaporation processes in the catchment (Zhong et al., 2020). The trend showed that one of the end-members was characterized by high-conductivity with <sup>18</sup>Oenriched (groundwater), and the other was characterized by low-conductivity with  $\delta^{18}$ O-depleted headstream water. Similar relationships were also identified in other mountainous rivers (Lambs, 2004). In addition,  $\delta^{18}O$  values increased progressively from upstream to downstream (Fig. S2b), which also validates the two sources (i.e., headstream water, and groundwater) of downstream river water, indicating that groundwater was likely an important contributor to downstream river water. Despite that groundwater inflow seemed to be of great importance to river water, groundwater DOC was likely not the most important source of riverine DOC due to the relatively low groundwater DOC concentrations as compared with riverine DOC concentrations (Fig. 2d). Moreover, the groundwater contribution was probably much less significant in the wet season, even in catchments where DOC is mainly derived from groundwater (Lloret et al., 2016). Thus, we infer that groundwater is an important but not a crucial



source of riverine DOC in the three study rivers." (P15-16 Line 335-351).

Figure S2 Implications for water from deeper flow paths. (a) Relationship between EC and  $\delta^{18}O$  for the river water and spring water. This relationship represents the mixing of the two reference waters: groundwater and upstream river water. (b) Variations of  $\delta^{18}O$  for water in the mainstream of the Yinjiang River. The  $\delta^{18}O$  value used in panel b for site Y12 is from the sample we collected before rainfall. The  $\delta^{18}O$  value for site Y15 was influenced by rainfall. The statistical test used a significance level of 0.05.

6. Fluorescence properties of DOM are presented but not related to geographic or anthropogenic features of the catchments. It is unclear why this isn't considered in the manuscript, and I wonder if there are any trends observed? At least, the information gained from optical analyses should be explained and contextualized within the discussion.

Our response: Thanks for your suggestions. Fluorescence properties of DOM were related to anthropogenic features of the catchments in the SI of our original manuscript. We have restructured the related information and added it in the manuscript as shown in the Figure 3, 6 and S3. In addition, the information has been explained and contextualized in the manuscript as follow: "For the fluorescence indexes, the overall fluorescence property did not vary significantly among the three rivers (Figs. S4b, c, and d). FI varied in a narrow range compared with  $\beta/\alpha$  and HIX. FI of DOM ranged from 1.66 to 1.94, averaging 1.78 (Fig. S4d), indicating a mixture of DOM of terrestrial and microbial origins. In comparison,  $\beta/\alpha$  varied from 0.70 to 1.22 (Fig. S4b) and HIX varied from 0.33 to 0.65 (Fig. S4c), with greater variability among the three rivers. Although no significant correlation was observed between channel slope and the fluorescence indexes, they (except for FI) were found to be closely related to land use pattern (Figs. 6d, e, f, and S3). The less recently produced DOM ( $\beta/\alpha$ ) in the urban and agricultural streams was also characterized by a higher proportion of C1 and lower proportion of C2 (Fig. S3)." (P14 Line 296-302) and "However, DOM characters were found to be closely related to land use pattern (Fig. 6). Although significant relationships with urban and

agricultural land uses were found for C1 and C2 (Figs. 6b and c), it remains unclear how the autochthonous contribution to riverine DOC pool varied with land use change because C1 and C2 are both likely derived from autochthonous production but exhibit opposing trends with increasing urban and agricultural land uses. Overall DOM in catchments with a higher proportion of urban and agricultural land use area was distinct from other catchments as it was less aromatic (SUVA254, Fig. 6a), less recently produced ( $\beta/\alpha$ , Fig. 6d), and had a higher degree of humification (HIX, Fig. 6e). SUVA254 values for the three study rivers were comparable with those reported in coastal glacier mountainous streams with late succession in southeast Alaska (3.4 ± 0.5 L mg<sup>-1</sup> m<sup>-1</sup>, n = 5; Holt et al. 2021) and in the anthropogenic influenced downstream of the Yangtze River (3.4 ± 1.1 L mg<sup>-1</sup> m<sup>-1</sup>, n = 82; Zhou et al. 2021). Lower DOM aromaticity in the urban and agricultural streams and rivers was consistent with previous studies (Hosen et al., 2014; Kadjeski et al., 2020), though it was not a universal phenomenon (Zhou et al., 2021). Furthermore, the less aromatic and recently produced DOM could be due to soil organic materials from deep soil profiles most aged DOC because of the increased soil erosion rates associated with anthropogenic activities (Inamdar et al., 2011; Stanley et al., 2012)." (P17 Line 382-395).

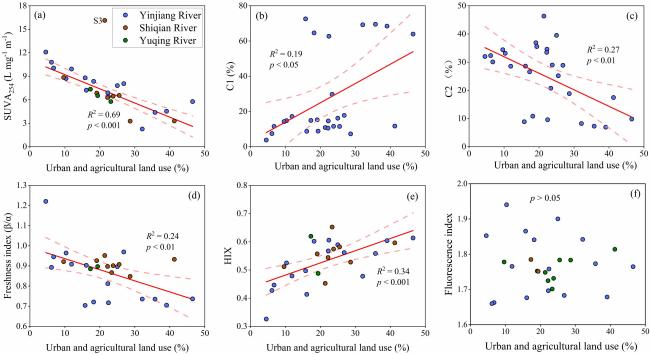


Figure 6. Land use pattern impacts on DOM character. (a) SUVA254, (c) C2, and (d) freshness index  $(\beta/\alpha)$  decreased with increasing proportion of urban and agricultural land use area in the studied catchments. Outlier (site S3) was excluded from analyses in panel a as the sample was strongly influenced by the road construction, which was evidenced by high POC and TSM concentration (Chen et al., 2021). (b) C1 and (e) humification index (HIX) positively related to the increasing proportion of urban and agricultural land use area.

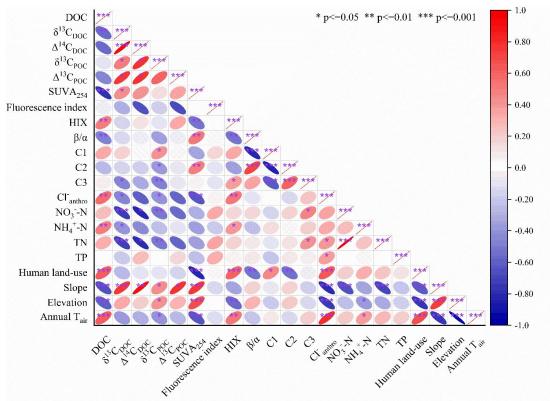


Figure S3 Correlation plot of the selected water chemistry and catchment characteristics.  $\delta^{13}C_{DOC}$  and  $\Delta^{14}C_{DOC}$  at site Y12 was excluded from analyses as the sample was collected after a rainfall event. In addition, SUVA<sub>254</sub> at site S3 was excluded from analyses as the sample was strongly influenced by the road construction, which was evidenced by high POC and TSM concentration (Chen et al., 2021). Human land use used here represents proportion of urban and agricultural land use. Elevation and Annual  $T_{air}$  represent mean drainage elevation and annual air temperature, respectively.

7. Similarly, it is unclear in section 4.3 why anthropogenic impacts are only discussed in relation to DOC concentration. I wonder if you can draw a connection with carbon isotopes (and optical properties) and if this could help you understand the primary drivers of variability within your dataset.

**Our response:** Same to last question, anthropogenic impacts on carbon isotopes and optical properties have been provided in Figure 6 and S3 (see above) and related discussion in "4.2 anthropogenic impacts on DOC" (please see the details in the revised manuscript or last question).

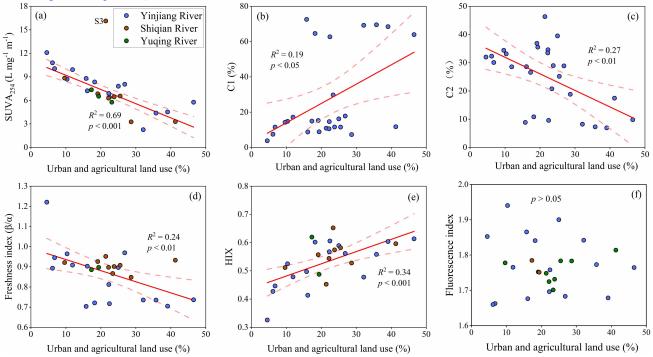


Figure 6. Land use pattern impacts on DOM character. (a) SUVA254, (c) C2, and (d) freshness index  $(\beta/\alpha)$  decreased with increasing proportion of urban and agricultural land use area in the studied catchments. Outlier (site S3) was excluded from analyses in panel a as the sample was strongly influenced by the road construction, which was evidenced by high POC and TSM concentration (Chen et al., 2021). (b) C1 and (e) humification index (HIX) positively related to the increasing proportion of urban and agricultural land use area.

## Minor comments and technical/typographical corrections:

#### Abstract:

L19 'of DOC' seems a little awkward here – consider rewording

**Our response:** Thanks. We have reworded it as: "Water chemistry, stable and radioactive carbon isotopes of DOC ( $\delta^{13}C_{DOC}$  and  $\Delta^{14}C_{DOC}$ ) and optical properties (UV absorbance and fluorescence spectra) were employed to assess the biogeochemical processes and controlling factors on riverine DOC." (P1 Line 17-19).

L21 POC is not defined. It is not clear how instream processing of POC is a source of DOC in this sentence. I would also make it clearer that you are using POC values from past work within the abstract and aims/objectives.

**Our response:** Thanks. POC has been defined in the manuscript as follow: "Our prior observations from these catchments showed that particulate organic carbon (POC) dynamics were highly affected by in-stream photosynthesis." (P3 Line 86-88). The point that "instream processing of POC is an important source of DOC" has been discarded in the new manuscript as you and other reviewers discussed. Therefore, we have removed this sentence.

L23-24 consider making the distinction between DOC and DOM here as I think it would help with sentence flow. It is also unclear how this was 'distinct' from those catchments with lower slopes/higher temperatures.

Our response: Thanks. The sentence is rephrased in the revised manuscript as: "Catchments with higher channel slope gradients and lower annual air temperature were characterized by lower DOC concentrations, enriched  $\delta^{I3}C_{DOC}$  and  $\Delta^{I4}C_{DOC}$ , and more aromatic dissolved organic matter (DOM), which were opposite to those with gentle channel slopes and higher temperature." (P1 Line 20-22).

#### L25 DOM is not defined

Our response: Thanks. We have added definition of DOM (dissolved organic matter) in the revised manuscript (P1 Line 21). L28 I think you could make the significance more specific/explicit here in relation to your findings.

Our response: Thanks. We have modified this sentence to make the significance more specific in the revised manuscript as: "This research highlights the significance of incorporating geographical controls on DOC sources and anthropogenic impacts on DOM composition into the understanding of DOC dynamics and quality of DOM in mountainous rivers which are globally abundant." (P1 Line 25-27).

## Introduction:

1. Generally, within the introduction I think it would be useful to provide more specific details in terms of DOM compositional shifts that have been noted with warming and land use changes as well as across geographic gradients (i.e., elevation and slope). Similar to that in line 63. Much of your description only specifies that there are 'changes' but doesn't note the typical direction of change. I think this would make it easier on the reader later when reading the results/discussion as many of the changes would be somewhat familiar and would also situate your study more firmly in relation to past work.

Our response: Thanks. We have provided more specific details on DOM compositional shifts with warming and land use changes as well as across geographic gradients in the manuscript as follow: "A recent global study found that increasing elevation was associated with greater protein-like fluorescence DOM and lower specific ultraviolet absorbance at 254 nm (SUVA254), which indicate the effect of enhanced UV radiation and accumulation of autochthonous DOM at higher elevation areas with low temperatures (Zhou et al., 2018)." (P2 Line 51-53) and "On the scale of decades to centuries, however, anthropogenic impacts would shift natural DOM to forms of low-molecular weight, enhanced redox state with potentially increased lability, or increased aromaticity due to warmer climate and altered hydrology (Stanley et al., 2012; Xenopoulos et al., 2021). In addition, a warmer climate can enhance microbial activity and in-stream production of DOM (He et al., 2016), and may simultaneously increase the microbial degradation rate of soil DOM, thus reducing the potential input of DOM into rivers (Voss et al., 2015). Consequently, this will increase the relative contribution of autochthonous DOM. Nevertheless, elevated temperature can also enhance photochemical degradation and reduce autochthonous microbial humic-like DOM (Henderson et al., 2009; Zhou et al., 2018), thus potentially limiting the accumulation of autochthonous DOM in inland waters. These two seemingly contradictory findings are due to the complex effect of temperature on organic matter." (P3 Line 73-81).

In addition, we have specified the typical direction of many changes in the manuscript, such as: "For example, elevated temperature has a dominant effect on DOC concentration and dissolved organic matter composition by enhancing decomposition and photochemical degradation rates of DOM (Zhou et al., 2018), contributing to significant CO<sub>2</sub> emissions from inland waters (Raymond et al., 2013)." (P2 Line 33-35) and "Compared with high-relief catchments, low relief regions with longer water residence time, stronger hydrologic connectivity to rivers, and greater development of wetlands are typically characterized by greater releases of DOC (Harms et al., 2016; McGuire et al., 2005)." (P2 Line 48-50).

2. I would also suggest you integrate the points made in lines 68-72 into the previous paragraphs. This section appears a little obvious and doesn't really make it clear why there is utility in using these techniques within your study.

Our response: Thanks. We have integrated the points into the previous paragraphs in the manuscript as you suggested: "Recent advances in spectroscopic techniques, especially the UV-visible spectrophotometry and fluorescence spectroscopy, and widespread application of stable and radiocarbon isotopes on bulk DOC have provided insights into the composition, source, and age of DOM in freshwater ecosystems (Coble, 1996; Fellman et al., 2010; Marwick et al., 2015; Minor et al., 2014; Sanderman et al., 2009). These new techniques have led to significant improvements in our understanding of the biogeochemical processes of DOC in river systems, which will continue to be effective tools for researchers to gain deeper insights into the riverine carbon

#### cycle" (P1 Line 40-46).

3. Finally, within the aims and objective paragraph it would be useful to be more specific of the techniques you are using (e.g., DOM quality assessed through optical metric) and the geographic/land use parameters use are assessing against DOM quality/DOC concentration.

Our response: Thanks. We have added related information to make it more specific in the revised manuscript as: "In this study, we evaluated how geographical controls (i.e., mean channel slope, mean drainage elevation, and annual air temperature) and anthropogenic impacts (i.e., land use patterns) affect the DOC dynamics and DOM characteristics in three subtropical catchments that contain many small to medium mountainous rivers in southwest China." (P3 Line 84-86) and "Relationships of DOC concentrations, stable and radiocarbon isotopic values of DOC (radiocarbon isotopes were only available for nine sampling sites in the Yinjiang River), DOM quality assessed through optical metric, nutrient concentrations, and land use patterns versus geographical characteristics (i.e., mean channel slope, mean drainage elevation, and annual air temperature) were examined." (P3-4 Line 91-95).

L33 given that your study has a large land use component, I'd suggest broadening this sentence out to encompass this. **Our response:** Thanks. We have broadened this sentence in the manuscript as: "Owing to continued climate warming and rapid land use changes, it is important to gain a better understanding of the spatial and temporal dynamics of DOC transport in river systems (Butman et al., 2014; Fasching et al., 2016; Zhong et al., 2021)." (P1 Line 31-33).

# L43 can you explain the 'difference' more specifically?

Our response: Thanks. We have modified this sentence as: "Compared with high-relief catchments, low relief regions with longer water residence time, stronger hydrologic connectivity to rivers, and greater development of wetlands are typically characterized by greater releases of DOC (Harms et al., 2016; McGuire et al., 2005)." (P2 Line 48-50).

## L53 this sentence is a little unclear to me. Consider rewording.

**Our response:** Thanks. We have reworded this sentence as: "Subtropical small mountainous rivers are characterized by steep channel slopes, high erosion rates, frequent rainfall events in wet seasons, and rapid change in hydrology during these rainfall events (Lee et al., 2019; Leithold et al., 2006; Qiao et al., 2019), however, the DOC dynamics in these rivers remain poorly studied." (P1 Line 60-62).

L65 consider rephrasing/reordering the sentence – 'recent pursuit' is a little awkward.

Our response: Thanks. We have rewritten this sentence as: "These two seemingly contradictory findings are due to the complex effect of temperature on organic matter. Clearly, it remains largely unknown how these impacts have regulated riverine DOC dynamics due to their complex regulating mechanisms and changing influencing factors." (P3 Line 80-83).

## L73 please specify the geographic and anthropogenic factors you are assessing

**Our response:** Thanks. The sentence was modified to specify the geographic and anthropogenic factors in the manuscript as follow: "In this study, we evaluated how geographical controls (i.e., mean channel slope, mean drainage elevation, and annual air temperature) and anthropogenic impacts (i.e., land use patterns) affect the DOC dynamics and DOM characteristics in three subtropical catchments that contain many small to medium mountainous rivers in southwest China." (P3 Line 84-86).

#### L75 remove 'their'.

Our response: Thanks. We have removed 'their' in this sentence from the revised manuscript: "Our prior observations from these catchments showed that particulate organic carbon (POC) dynamics were highly affected by in-stream photosynthesis." (P3 Line 86-88).

L76 add 'here'. So, it reads 'Here, we investigate...'

**Our response:** Thanks. We have deleted this sentence as we did not take autochthonous processes as a significant source of DOC in the revised manuscript.

L76 it is unclear from this sentence how you asses autochthonous processes.

**Our response:** Thanks. Same to the last question, we have deleted this sentence as autochthonous processes were not a significant source of DOC in the revised manuscript.

# L77 I feel this hypothesis could be more specific based on past literature.

Our response: Thanks. The hypothesis was modified to be more specific in the revised manuscript as: "We hypothesize that catchments with a higher proportion of agricultural and urban land use, more gentle channel slope, and lower elevation would exhibit higher riverine DOC concentrations. We further hypothesize that there will be a large difference on DOM quality and carbon isotopes between these catchments and those with less influences by agricultural and urban land uses but steeper channel slopes and higher elevation." (P3 Line 88-91).

L80 seems a little obvious, can you be more specific on the insight gained?

**Our response:** Thanks. We have modified this sentence as: "This study allows us to gain a deeper insight into the geographical controls and anthropogenic impacts on the DOC dynamics and DOM quality in the subtropical, anthropogenically influenced small mountainous rivers." (P4 Line 95-96).

#### Methods:

1. It appears from the results that 14C-DOC values only available for the Yinjiang River. This must be made clear in the aims and methods. Also, why is this the case?

Our response: Thanks. We have added related information in the aims and methods in the manuscript as follow: "Relationships of DOC concentrations, stable and radiocarbon isotopic values of DOC (radiocarbon isotopes were only available for nine sampling sites in the Yinjiang River), DOM quality assessed through optical metric, nutrient concentrations, and land use patterns versus geographical characteristics (i.e., mean channel slope, mean drainage elevation, and annual air temperature) were examined." (P3-4 Line 91-95) and "In this study, nine water samples collected from the Yinjiang River were selected for  $\Delta^{14}C_{DOC}$  analysis as the Yinjiang River catchment has the greatest change in geographical characteristics (i.e., elevation and channel slope) and the highest proportion of agricultural and urban land uses among the three catchments." (P6 Line 150-152). "The Yinjiang River catchment has the greatest change in geographical characteristics (i.e., elevation and channel slope) and the highest proportion of agricultural and urban land uses among the three catchments" is one of the reasons we only collect 14C-DOC data in the Yinjiang River. The other reason is the expensive analytical cost (McNichol and Aluwihare, 2007), which is about \$500 per sample.

Reference: McNichol A. P. and Aluwihare, L. I.: The power of radiocarbon in biogeochemical studies of the marine carbon cycle: Insights from studies of dissolved and particulate organic carbon (DOC and POC), Chem. rev., 107, 443-466, doi:10.1002/chin.200724246, 2007.

## L86 'of the' replace with 'in'

Our response: Thanks. 'Of the' has been replaced with 'in' in the manuscript as follow: "The mean drainage elevation of these three catchments ranges from 340 m to 2424 m with the lowest and highest elevation both reported in the Yinjiang River catchment, showing a great change in relief." (P4 Line 103-104).

## L89 rephrase so land use is not repeated

**Our response:** Thanks. We have rephrased the sentence as: "Forest, agriculture, and urban areas are the three dominant land uses in these studied catchments." (P4 Line 109-110)

## L104 there is a missing word here.

**Our response:** Thanks. We have reworded this sentence as: "water samples were filtered through 0.45 μm cellulose acetate membranes." (P5 Line 131).

L108 I think more information would be useful here, despite information being published in your previous work.

Our response: Thanks. We have added more details in the revised manuscript as: "Water samples were also filtered for determining dissolved inorganic carbon (DIC) through titration with hydrochloric acid and analyzing POC using retained suspended particles on the filter membranes. Moreover, the water samples filtered through 0.22  $\mu$ m cellulose-acetate filter membranes were used to determine water isotopes ( $\delta^{18}O$  and  $\delta D$ ). Detailed information on sampling methods was provided in Chen et al. (2021) and Zhong et al. (2020)." (P5 Line 135-138).

## L127 replace substances with fluorescence

**Our response:** Thanks. We have replaced it as you suggested: "The fate of humic-like fluorescences may be self-assembly particles or be adsorbed onto minerals, while protein-like fluorescences are tightly associated with biological processes, and biodegraded into inorganic matter" (P7 Line 167-169).

# L135 – double brackets to be replaced with ';' - please check throughout.

Our response: Thanks. We have replaced double brackets with ';' throughout the manuscript. Here are two examples: "Several common indices of DOM composition were determined from EEMs, including fluorescence index (FI; McKnight et al., 2001), humification index (HIX; Ohno, 2002), and freshness index ( $\beta/\alpha$ ; Parlanti et al., 2000; Table 1)." (P7 Line 175-177) and "Global average  $\Delta^{14}C_{DOC}$  is -11.5±134%, higher than that in the Yinjiang River (-54.7±39.9%; Tables 2 and 3; Marwick et al., 2015) while similar to many other mountainous rivers (e.g., the Mackenzie River; Campeau et al., 2020) and small mountainous rivers in Puerto Rico; Moyer et al., 2013).  $\Delta^{14}C_{DOC}$  values for the world's mountainous streams and rivers were shown by climate (according to the Köppen–Geiger climate classification (Beck et al., 2018; Table 3) and ranged from tropical monsoon climate (Marwick et al., 2015." (P19 Line 434-439).

L140 how was proportion of different land uses/elevation/slope calculated? Was this data previously published? I would recommend adding this information as a table to the text and how this data came about in the site

previously published? I would recommend adding this information as a table to the text and how this data came about in the site description.

Our response: Thanks. We have added details on related information in "2.1 Study area" in the manuscript as: "Catchment characteristics (e.g., mean drainage elevation, annual air temperature, mean channel slope, and proportion of urban and agricultural land uses) for the sub-catchments were determined using ArcGIS (version 10.2). The mean drainage elevation of these three catchments ranges from 340 m to 2424 m with the lowest and highest elevation both reported in the Yinjiang River catchment, showing a great change in relief (Figs. 1a and S1a). Similar to elevation, the Yinjiang River catchment has a greater variation in mean channel slope (from 14.3°to 25.5°), while the channels in the Shiqian and Yuqing river catchments have a mean channel slope of approximately 20°, except the segment above site S8 (13.9°; Figs. 1b and S1b). Carbonate rock is widely distributed in the three catchments, accounting for a large proportion of the exposed strata (Han and Liu, 2004). The remaining

areas are mainly covered by clastic rocks, igneous rocks, and low-grade metamorphic rocks. Forest, agriculture, and urban areas are the three dominant land uses in these studied catchments (Fig. 1c). Forest is generally distributed in high-elevation regions, while urban and agricultural land uses are mainly located in low-elevation regions. The proportion of urban and agricultural land uses in the Yinjiang and Shiqian river catchments is from 4.5% to 46.5% and from 9.6% to 41.3%, respectively, showing a large variation relative to the Yuqing River catchment (from 17.3% to 23.1%; Figs. 1c and S1c)." (P4 Line 101-113) and figure about sites specific distribution of land use, slope and elevation in the SI (please see the figure S1).

L161 – the median value for river Y is not higher than the other rivers. Thus, it's unclear what you are referring to here. **Our response:** Thanks. Here, "DOC concentrations in the three study rivers varied from 0.36 to 2.85 mg  $L^{-1}$  with the highest average concentrations in the Yuqing River  $(1.70 \pm 0.04 \text{ mg } L^{-1}; Fig. 2d)$ " (P9 Line 221-222) means the average concentrations in the Yuqing River (Q) were the highest, not the Yinjiang River (Y).

# Results/Discussion:

- 1. Description of optical properties in the results is extremely brief and lacks quantitative details. E.g., what are the average SUVA values for each river? What is the % each component of fluorescence is explaining? Please include these details. Our response: Thanks for your suggestions. We have added these details in the manuscript as follow: "The average SUVA254 were  $3.3 \pm 1.1$ ,  $3.1 \pm 1.8$ , and  $2.8 \pm 0.3$  L mg<sup>-1</sup> m<sup>-1</sup> in the Yinjiang, Shiqian, and Yuqing rivers, respectively. Although no spatial differences in SUVA254 were found across the three rivers (Fig. S4a), SUVA254 showed an increasing trend with increasing mean channel slope (Fig. 3e), which indicated that DOM in the lower reaches with a gentle channel slope was less aromatic. Furthermore, there was a significant negative correlation between SUVA254 and proportion of urban and agricultural land uses (Fig. 6a)." (P12 Line 264-268) and "Two humic-like fluorescence components (C1 and C3) and one protein-like fluorescence component (C2) were identified by PARAFAC model in these three rivers (Fig. 7; Table S1). Component C1 is similar to traditionally defined peak M and sourced form microbial processes or autochthonous production (Kim et al., 2022; Li et al., 2016; Walker et al., 2009). Component C2 was previously related to recent biological production (DeFrancesco and Guéguen, 2021; Du et al., 2019; Lambert et al., 2017). C3 was the most widely found component in previous research among three fluorescence components, and was identified as traditional fulvic-like peaks A and C, representing terrestrial delivered OM or authochthonous microbial sourced OM (Amaral et al., 2016; Ryan et al., 2022; Shutova et al., 2014). Although C1 and C2 varied more widely in the Yinjiang River compared with the Shiqian and Yuqing rivers, the two fluorescence components did not show a statistical difference among the three rivers (Figs. 7a and b). However, a greater proportion of C3 was found in the Shigian and Yuqing rivers, exhibiting a distinctive signature comprred with the Yinjiang River (Fig. 7c). The fluorescence components did not exhibit any significant variations with changing channel slope (Fig. S3), but C1 and C2 were positively or negatively related to proportion of urban and agricultural land uses (Figs. 6b and c)." (P13 Line 277-288).
- 2. Generally, geographic and land use parameters are not discussed in the results. However, SUVA is briefly described and then related to slope. Given the structure of the results it would make sense to wait to draw the comparison with slope. Consider including a summary figure (e.g., boxplot for SUVA) and then including SUVA v slope analysis. Similarly, why is Figure 4 a boxplot whilst other relationships with slope conducted as linear regressions?

Our response: Thanks. We have added more details on geographic and land use parameters in the manuscript as follow: "Catchment characteristics (e.g., mean drainage elevation, annual air temperature, mean channel slope, and proportion of urban and agricultural land uses) for the sub-catchments were determined using ArcGIS (version 10.2). The mean drainage elevation of these three catchments ranges from 340 m to 2424 m with the lowest and highest elevation both reported in the Yinjiang River catchment, showing a great change in relief (Figs. 1a and S1a). Similar to elevation, the Yinjiang River catchment has a greater variation in mean channel slope (from 14.3° to 25.5°), while the channels in the Shiqian and Yuqing river catchments have a mean channel slope of approximately 20°, except the segment above site S8 (13.9°; Figs. 1b and S1b)." (P4 Line 101-107) and "The proportion of urban and agricultural land uses in the Yinjiang and Shiqian river catchments is from 4.5% to 46.5% and from 9.6% to 41.3%, respectively, showing a large variation relative to the Yuqing River catchment (from 17.3% to 23.1%; Figs. 1c and S1c)." (P4 Line 111-113)

In addition, "SUVA vs slope" has been included in Figure 3 (see below) in the revised manuscript and all the DOM property data were plot as a boxplot (Figure S4; see below).

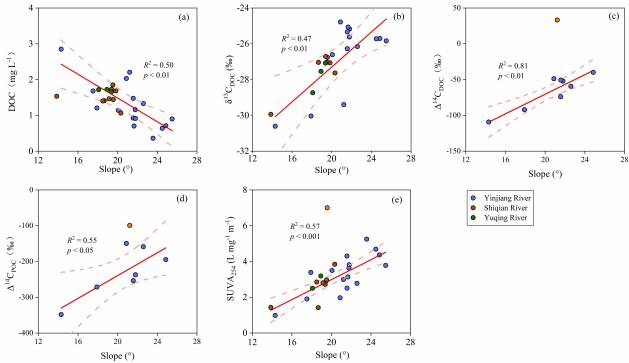


Figure 3. Mean drainage slope (°) controls on (a) DOC concentrations, (b) stable carbon isotopes of DOC ( $\delta^{13}C_{DOC}$ ), (c) radiocarbon isotope of DOC ( $\Delta^{14}C_{DOC}$ ), (d) radiocarbon isotope of POC ( $\Delta^{14}C_{POC}$ ) and (e) SUVA<sub>254</sub>. The  $\Delta^{14}C_{DOC}$  and  $\Delta^{14}C_{POC}$  are only available for the Yinjiang River. Outliers in orange were excluded from analyses as they were samples at site Y12 collected after a rainfall event in panels (c) and (d) and sample collected at site S3 due to the highly influence by the road construction, which was evidenced by high POC and TSM concentration (Chen et al., 2021). The statistical test used a significance level of 0.05.

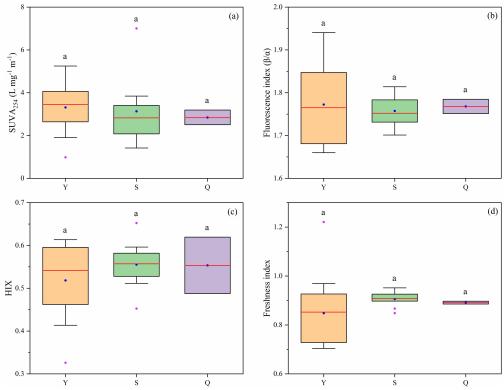


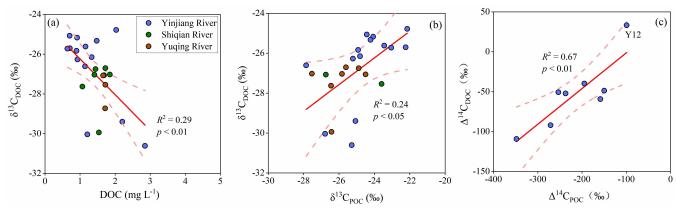
Figure S4 Spatial variations in DOM property in the Yinjiang (Y), Shiqian (S), and Yuqing (Q) catchments. (a) SUVA<sub>254</sub>, (b) freshness index ( $\beta/\alpha$ ), (c) HIX, and (d) fluorescence index. In each box plot, the end of the box represents the 25th and 75th percentiles, the blue solid dot represents the average, the horizontal red line represents the median, and whiskers represent 1.5 IQR. The magenta solid dot represents the outlier, which is outside of the 1.5 interquartile ranges. Different lowercase letters above the boxes denote significant differences across rivers based on statistical analysis with *p*<0.05.

Figure 4 – specify units of SUVA254 on axis

Our response: Thanks. We have added the unit of SUVA<sub>254</sub> on axis (please see the above Figure 3 and S4).

Figure 5 – place key at the bottom of the four panels. Dots in key maybe confused with datapoints.

Our response: Thanks. To avoid the confusion, we have added a box for the key in Figure 5 (see below).



**Figure 5.** Scatter plot showing (a)  $\delta^{13}C_{DOC}$  versus DOC in river water, (b)  $\delta^{13}C_{DOC}$  against  $\delta^{13}C_{POC}$  in the Yinjiang River (Y), Shiqian River (S), and Yuqing River (Q), and (c) relationship between  $\Delta^{14}C_{POC}$  and  $\Delta^{14}C_{DOC}$  in the Yinjiang River. For panel (c) the DOC with modern age at site Y12 was shown in the top-right corner. The statistical test used a significance level of 0.05.

Figure 5 – why are results in panel A reported as 1/DOC? Please just report as DOC since this leads to confusion in text e.g., line 188

Our response: Thanks. We have modified it to be reported as DOC in Figure 5 (please see figure above). The reason for using 1/DOC in the original manuscript is because previous studies have reported similar relationships (i.e.,  $\delta^{13}C_{DOC}$  vs 1/DOC) in "spring water (Nkoue Ndondo et al., 2020) and for TOC in soil profiles (Lloret et al., 2016; Nkoue Ndondo et al., 2020)" (P15 Line 329-332) to explain the lateral transportation of DOC from microbial active soil horizons into rivers (Lambert et al., 2011)."

L166 it is unclear why you contextualize this finding but not others. Please contextualize data throughout results or move this point to the discussion.

Our response: Thanks. We have moved this point to the discussion (P19 Line 434).

L225 If anthropogenic activities were not the primary source of aged DOC in your catchments (as you imply). What is the primary source of aged DOC? This point should be made clearer

Our response: Thanks. We have added related information in the revised manuscript as follow: "In comparison, when relatively  $^{14}$ C-depleted DIC and  $CO_2$  (aq) derived from carbonate weathering is incorporated into primary production in low relief regions, it would also produce aged organic carbon (Fig. 3d; Chen et al., 2021). Furthermore, aged DOC in river systems has been attributed to old soil organic matter in deeper layer input into rivers through deeper flow paths (Barnes et al., 2018; Masiello and Druffel, 2001). This also indicates that low relief regions with higher hydrologic connectivity in river network are likely the major source of riverine DOC (Connolly et al., 2018; Mzobe et al., 2020)." (P15 Line 309-314) and "Furthermore, the weak positive correlation between the  $\delta^{13}C_{DOC}$  and  $\delta^{13}C_{POC}$  of these three rivers (Fig. 5b) indicated that DOC and POC may derive from the same source. We also found a strong positive correlation between  $\Delta^{14}C_{DOC}$  and  $\Delta^{14}C_{POC}$  in the Yinjiang River ( $R^2 = 0.67$ , p < 0.01; Fig. 5c). The coupling between  $\Delta^{14}C_{DOC}$  and  $\Delta^{14}C_{POC}$  was an unusual relationship rarely found in other rivers (Campeau et al., 2020; Longworth et al., 2007). Campeau et al. (2020) have attributed this relationship to common controls of landscape and/or hydrology on the sources of organic carbon in rivers, and this correlation could be masked by the mixing of waters from other tributaries, which further support the combined geographical controls (e.g., as a driver of landscape) and anthropogenic impacts (e.g., dam construction, see discussion below) on DOC sources." (P18 Line 415-422).

## L230 it would be prudent to explain the two endmembers here in more detail.

Our response: Thanks. As we have modified the original view (two endmembers mixing) due to further discussion. We have deleted the figure which showed relationships between  $\delta^{13}C_{DOC}$  and  $\Delta^{14}C_{DOC}$ , and take it as common controls of landscape and/or hydrology on the sources of organic carbon. Please see related discussion in the revised manuscript as: "Furthermore, the weak positive correlation between the  $\delta^{13}C_{DOC}$  and  $\delta^{13}C_{POC}$  of these three rivers (Fig. 5b) indicated that DOC and POC may derive from the same source. We also found a strong positive correlation between  $\Delta^{14}C_{DOC}$  and  $\Delta^{14}C_{POC}$  in the Yinjiang River ( $R^2 = 0.67$ , p < 0.01; Fig. 5c). The coupling between  $\Delta^{14}C_{DOC}$  and  $\Delta^{14}C_{POC}$  was an unusual relationship rarely found in other rivers (Campeau et al., 2020; Longworth et al., 2007). Campeau et al. (2020) have attributed this relationship to common controls of landscape and/or hydrology on the sources of organic carbon in rivers, and this correlation could be masked by the mixing of waters from other tributaries, which further support the combined geographical controls (e.g., as a driver of landscape) and anthropogenic impacts (e.g., dam construction, see discussion below) on DOC sources." (P18 Line 415-422)

#### L255 is this supported by your data?

**Our response:** Thanks. We just compare it with our study, rather than showing the views from our data. In order to avoid the misunderstanding of the sentence, we have modified it as: "Channel slope in a number of Arctic watersheds has also been found to be positively associated with the FI, which was possibly due to the differences in soil characteristics (e.g., volumetric water content and soil temperature) and the resulting changing extent of microbial processing (Harms et al., 2016). Moreover, channel slope was negatively associated with terrestrial humic-like organic material due to the effects of climate and organic layer

thickness (Harms et al., 2016). However, there were no similar correlation between channel slope and fluorescence components/indexes in this study, demonstrating the likely complicated mechanisms (e.g., soil property, catchment characteristics, and anthropogenic activities) in regulating DOM. DOC in low relief regions was characterized by more <sup>13</sup>C depleted values, which may be due to the greater inputs of C3-derived organic carbon (e.g., from rice)." (P15 Line 319-327).