

Anonymous Referee #1

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Author Response: We sincerely thank Referee #1 for their thoughtful and thorough comments that would greatly improve the first version of our paper. We have added statements in **blue** below that detail our response to each comment. We feel that most of the comments were relatively minor in nature and we have addressed things below to the best of our ability.

This paper contains an interesting dataset of DOC in the Kolyma River watershed that follows closely from previous work by the authors in this system. In this paper, the authors build on their prior work on DOC and BDOC concentrations in the Kolyma River by also making these measurements on the soil waters draining into the small streams and eventually the main stem of the Kolyma. The data and methods are strong. A few ideas in the discussion and conclusions could be strengthened by comparing to similar findings in the arctic in other systems, and by broadening the interpretations as suggested below.

Methods:

It was not clear whether bioassay incubations were conducted on unfiltered water, or water with sediment? Also, bioassays were conducted at 15°C but no rationale was provided for this temperature. How representative is this temperature of the soil waters, streams and the river studied here?

Waters used in the bioassays were filtered and treated the same as the DOC samples. We can make this more explicit in the Methods section. The bioassays were conducted at 15°C to mimic the standard BOD method, which was also the temperature of the laboratory the samples were handled in. A space heater in the laboratory was utilized occasionally to maintain this temperature on cooler days and/or overnight (which was the best that could be done given the available laboratory space). This temperature was only slightly warmer than environmental sampling conditions (i.e., the Kolyma River mainstem samples ranged from 11.40–13.90°C, river samples ranged from 10.70–14.20°C, and stream samples ranged from 4.40–13.80°C). However, again, we kept things at 15°C as is standard in the BOD method. This further allowed samples to be treated identically in the controlled experiment (as temperatures varied depending upon location as well as date/time of day, etc.). These further details can be added in the Methods section if necessary.

Were the bioassays started right after sample collection, or were samples allowed to equilibrate with the atmosphere prior to incubation? The authors should provide information on whether any samples were sub or anoxic at the start of the experiment?

The samples were allowed to equilibrate via filtering in a controlled laboratory environment, such that all experiments were able to be carried out as close to 15°C as possible (and t=0 was the start time of the incubation, with temperatures at 15°C). Bottles were wrapped tightly with paraffin and laboratory temperatures were as close to 15°C as possible, so physical degassing should have been minimal. We can add this to the Methods section if necessary as well.

Incubations were conducted at 15°C, but there was no information on how well the temperature was controlled over the course of the incubation, which is helpful to rule out influences from gas exchange (such as bubble formation if initial sample temperature at T0 in the BOD bottles is different than 15°C).

Again, the samples were allowed to equilibrate via filtering in a controlled laboratory environment, such that all experiments were able to be carried out as close to 15°C as possible (and t=0 was the

start time of the incubation, with temperatures at 15°C). Bottles were wrapped tightly with paraffin and laboratory temperatures were as close to 15°C as possible, so physical degassing should have been minimal. There was great attention to detail in terms of potential degassing and temperature maintenance, so we do feel quite confident in our experimental results. We can add this to the Methods section if necessary as well.

The extra information on Winkler titrations in this paragraph (lines 5-12) is out of place given that Winkler titrations are standard methods for O₂ consumption; no further examples needed that were not used in this study.

We can certainly remove these lines of text (lines 5–12 on page 12327) if they seem out of place or unnecessary. We included this information in the paragraph initially to provide further information on the Winkler method for those that may be unfamiliar with the method.

Results and discussion:

The authors mention relationship of DOC and BDOC concentrations with water residence time “in the system”, is this the water residence time in soils, streams or the river? What are the residence times of water in these different systems (none were provided directly or in citations to previous work).

Unfortunately we did not determine residence times directly for our sampled sites. Accurate discharge/flow rate data for streams and tributaries throughout the region are scarce if not nonexistent and tracer experiments (i.e., to directly determine residence times from soil pore waters downstream) have not been performed at these sites. However, we can refer to some previous studies that include information regarding residence times in the region. For instance, Vonk et al. (2013) estimate that in higher relief areas near Duvannyi Yar (adjacent to the Kolyma River mainstem), the transport time from permafrost thaw to entry into the Kolyma River may be less than one hour. Our study sites with lower relief may of course have longer transport times to adjacent streams/ivers. Furthermore, with respect to the mainstem, it has been estimated that water residence times in the Kolyma River from Duvannyi Yar to the river mouth may be ~3–7 days, assuming average mainstem velocities of 0.5–1.5 m/s (Holmes et al., 2012; Vonk et al., 2013). As such, permafrost-derived C may not be easily detectable at the river mouth, as this time is likely comparable to the rapid removal rates of highly labile permafrost C determined through incubation experiments (e.g., Holmes et al., 2012; Vonk et al., 2013). These examples give a range of possible residence times that may be experienced at our sampling sites as well. We can add these types of examples to the text to give additional context to the potential residence times of waters in this study.

Holmes, R. M., et al. (2012), Seasonal and annual fluxes of nutrients and organic matter from large rivers to the Arctic Ocean and surrounding seas, *Estuaries Coasts*, doi:10.1007/s12237-011-9386-6.

Vonk, J. E., et al. (2013), High biolability of ancient permafrost carbon upon thaw, *Geophys. Res. Lett.*, 40, 2689–2693, doi:10.1002/grl.50348.

The authors interpret decrease in DOC and BDOC concentrations from soil waters to small streams and rivers as evidence for rapid, in-stream processing, but the fraction of DOC that is labile is similar across all water types. If the most labile DOC is rapidly removed as water moves from small to large streams, how do the authors interpret that there is a consistent fraction of BDOC (% of DOC) in the waters studied? While the authors can't rule in or out the reasons for relatively consistent fraction of BDOC in their sites, they can do more to discuss alternative explanations within the context of the literature,

such as photodegradation, changes in microbial community structure, or inputs of labile DOC along stream and river channels from soil waters.

This is an excellent comment by the reviewer and we agree that we could elaborate on potential mechanisms behind the consistent % bioavailable DOC pattern we observed across all water types. As the reviewer highlights (and we can include as additional text in the manuscript), changes in DOM composition downstream may drive community structural differences leading to consistent microbially-driven DOC losses despite significant changes to DOM structure. An alternate (or additional) mechanism could be caused by continued sunlight exposure (and resulting photodegradation) acting to make a proportion of the DOM pool bioavailable during transit. A shifting microbial community downstream could also certainly lead to these types of results. As the reviewer suggests, including references to additional work (below) would strengthen this discussion of potential mechanisms for our observations of consistent % bioavailable DOC patterns across all water types.

For example, for photodegradation, the authors suggest that a decrease in CDOM slope ratio from streams to the river is consistent with photodegradation of DOC, which has previously been observed as a function of water residence times in arctic freshwaters (Cory et al. 2007 JGR-B, Merck et al. 2012 Hydrol. Proc.). The authors could strengthen their interpretation of CDOM and slope ration by comparing to these previously observed and similar patterns.

We appreciate the reviewer's suggestion for including references to these studies, as they are quite relevant to our work in the Kolyma River basin. We agree that we can refer to these important studies in the context of our work and can easily add sentences in our Discussion section to describe this previous work. Cory et al. (2007) and Merck et al. (2012) demonstrate the importance of residence times as well as a significant combined role for photo- and biological degradation along the flowpath in Arctic watersheds. These previous results show that the photochemical "pretreatment" of stream DOM that occurs during export into lakes and coastal zones may impact the ability of microorganisms to mineralize DOM. Therefore, the residence time and flowpath of waters should greatly influence the ultimate fate of DOM (organic matter versus carbon dioxide) exported to the adjacent ocean. As such, in our case, we find that our slope ratio (S_R) values suggest important photodegradation processes are occurring along the flowpath continuum, and these previous studies suggest that this photodegradation may potentially release significant quantities of labile DOM for "continued" microbial processing of DOM further downstream in these Arctic stream networks.

Cory, R. M., D. M. McKnight, Y.-P. Chin, P. Miller, and C. L. Jaros (2007), Chemical characteristics of fulvic acids from Arctic surface waters: Microbial contributions and photochemical transformations, *J. Geophys. Res.*, 112, G04S51, doi:10.1029/2006JG000343.

Merck M, B. Neilson, R. Cory, and G. Kling (2012), Variability of in-stream and riparian storage in a beaded arctic stream. *Hydrological Processes*, 26, 2938–2950.

In addition, the authors seem to be interpreting the decrease in slope ratio from the streams to the river as evidence that photodegradation is important in this system. How might photodegradation influence the fraction of BDOC with distance downstream, given that light exposure has a substantial effect on DOC lability in bacteria in arctic freshwaters (for example, Cory et al. 2013 PNAS; Mladenov & Laurion 2013 Env. Res. Let).

We thank the reviewer for pointing out these additional previous studies (Cory et al., 2013; Laurion and Mladenov, 2013) that are quite relevant to our results in the Kolyma River basin. These studies additionally highlight the importance of photodegradation for "pretreating" DOM for further

microbial degradation downstream in the system along the flowpath continuum. We certainly think it is important to highlight these additional studies in the context of our results. In particular, this previous work suggests that the consistent BDOC pattern we observed across all water types in our study is a result of photodegradation processes potentially releasing significant quantities of labile DOM for “continued” microbial processing of DOM further downstream in these Arctic stream networks. If this (or something similar) were not the case, we would expect to see declining % bioavailable DOC along the flowpath continuum.

Cory, R. M., B. C. Crump, J. A. Dobkowski, and G. W. Kling (2013), Surface exposure to sunlight stimulates CO₂ release from permafrost soil carbon in the Arctic, *Proceedings of the National Academy of Sciences*, 110(9), 3429-3434.

Laurion, I. and N. Mladenov (2013), Dissolved organic matter photolysis in Canadian arctic thaw ponds, *Environmental Research Letters*, 8, 035026.