Dear Reviewer 1,

we very much appreciate your thoughtful review of our manuscript. Thank you for your time and your valuable ideas. We absolutely agree that taking up your suggestions greatly improved our manuscript and we did so as described below.

* This manuscript reports an interesting dataset on C cycling at a temperate peatland, affected by increased nutrient input from a nearby reservoir. Carbon dioxide and methane fluxes were measured over a period of 1.5 years from four sites representing variable wetness, vegetation type and distance from the reservoir, and the flux measurements were accompanied by detailed soil profile measurements of CH4 and DIC concentration. Carbon stable isotopes were used in order to gain more information about CH4 production, oxidation and transport. The paper is well written and logically structured, and the appearance of the figures C1 BGD Interactive comment Printer-friendly version Discussion paper is very good. The methods are described in great detail, and the authors are clearly experts in selection and implementation of their field and analytical methods. The value of this work is in the high quality and completeness of the paper could be greatly improved, by taking the following comments into consideration.

***Major comments

1. The match between the content of the manuscript and the title is not ideal at the moment. The title and especially the starting sentence of the abstract make one expect a comparison of carbon cycling between anthropogenically altered vs. natural sites. If this is the focus, it would be important to describe the transect better in the abstract and also in the methods section (page 4, lines 2-3 & from lines 16 onwards): How much did the hydrological condition change along the transect, and was the human impact related to drying or wetting or to fluctuating water table? And, in the abstract, how much did the nutrient infiltration change along the transect (data in Table 1)?

– We understand your concern.

This present study is following up on a study, which was very recently published in SBB (114 (2017) 131-144; <u>http://dx.doi.org/10.1016/j.soilbio.2017.07.011</u>). While writing the current paper, this other study had not been published so we could not well refer to it. The study site was described in detail in this SBB-paper. As elaborated there, the water reservoir affects the entire northern tip of our peatland site. We find in our data that areas further away from the reservoir are likely less affected, but it is not entirely possible to distinguish 'natural' from 'anthropogenically altered' sites among the sites 1-4 (800-200 meters distance from the reservoir). Only areas further away from the reservoir could likely be regarded as pristine. So, our transect of study sites as presented in this present study rather assesses 'strongly altered' sites vs. 'less altered' sites with respect to the investigated features. Unfortunately, this entire gradient was not as obvious in the beginning of the study, so this part is indeed missing. So, our objective for this paper was to investigate carbon cycling along this transect and to identify effects of altered conditions along the transect on carbon cycling and fluxes.

In the revised version of our manuscript we described the transect better in the abstract (page 1, lines 14-17) and in the method section (page 4, line 31 to page 6, line 2), providing the information you requested.

With respect to the hydrological impact, particular information was added (page 4, line 31 to page 5, line 3): "Through flooding of the reservoir, Wylde Lake peatland has been exposed to altered hydrological conditions in a way that the water reservoir enhanced water level fluctuations in a large part of the site: in summer or under dry conditions, water is released from the reservoir, thereby draining water out of the peatland; under wet conditions, water table levels of the reservoir increase and water is pushed into the peatland. Those sites in closer vicinity to the reservoir are presumably more affected than sites further away from the reservoir (Berger et al., 2017)".

The title was changed to: "Differential response of carbon cycling to long-term nutrient input and altered hydrological conditions in a continental Canadian peatland". We believe the new title better reflects the contents of our manuscript.

* Further, instead of reporting just the results from the two highly affected sites 3&4 in the abstract, you should compare the results between anthropogenically altered vs. natural sites. This would justify the last sentence which claims clear anthropogenic effects on C cycling.

We agree. The abstract was rewritten, however, still the focus is on the results from the sites 3 and
 This is because the two sites were mostly altered and here we also found most significant differences. The last sentence of the previous abstract was deleted.

* This comparison between affected and unaffected sites should be the view-point throughout the MS. For example:

By rearranging Fig. 3 & 4 so that instead of ' showing various parameters from the same site in the same figure you would show a single parameter from all of the sites in the same figure.
By adding here some ' indication of the reservoir effect: Distance of the reservoir in the table itself, or descriptive sentence in the table caption.

- By focus the introduction better from general ' description of factors affecting in peatland C cycling towards a description of the effects of anthropogenic activities on it. It should be stated clearly, citing the relevant literature, why is it important to understand effects of increased nutrient inputs and changed water level on carbon cycling. This topic it touched on the paragraph starting on page 2, line 24, but also there anthropogenic effects are not sufficiently discussed to match with the title of the paper. Also, the motivation statement on page 3, lines 4-8 is very general. Could you develop a more specific research question that suits for this particular study?

- By rewriting the Concluding remarks section to answer the questions 'posed by the title and the introduction section.

Unfortunately, the manuscript cannot provide a comparison between "affected" and "unaffected" sites (see also above). However, it can provide a comparison of carbon cycling of *strongly altered* and *less altered* sites. We have clarified this point to avoid misunderstandings here.
 Please see:

-page 1, lines 16-17 -page 5, line 8 to page 6 line 2 -page 14, lines 16-17

We re-arranged figures 3 & 4 as suggested. We agree that this very much improves clarity. The development of pore gas DIC and CH_4 concentrations over time could, however, not be re-arranged as the 3D-plots would get too complicated.

Having re-arranged the figures as you suggested, differences between the sites in terms of CO_2 and CH_4 fluxes become indeed much more obvious.

Table 1 in its previous form was thought to summarize some results from the SBB-paper, but it turned out that presenting only that little information about the sites was quite misleading. Peat ages, pH and stochiometric ratios from table 1 were removed (as the information can be found in the SBB paper), instead, information on plant species abundances at each site were added in order to better illustrate the vegetation gradient. Moreover, the reader was oftentimes referred to the SBB-paper.

The introduction was rewritten (page 2, line 17 to page 4, line 16) according to your suggestion. The paragraph (starting on page 2, line 24 of the previous manuscript version) dealing with factors affecting peatland C cycling was extended towards a description of the effects of anthropogenic activities. In this regard, results from previous long-term peatland fertilization experiments (e.g. Mer Bleue, Whim Bog, Degerö Stormyr) as well as impacts of inundation on neighboring peatland ecosystems (e.g. Kim

et al., 2015; Ballantyne et al., 2014) were better summarized and we also provided a more adequate summary of what is known about gaseous carbon fluxes in relation to an altered plant community (e.g. Robroek et al., 2015) to set the picture for our study. Taking also into account our own recently published paper (Berger et al., 2017) it is now much more obvious why there still is a need to study changes in peatland C-cycling after increased nutrient inputs and changed water levels.

As far as a more specific research question is concerned, we developed new hypotheses which better meet the focus of our study (page 4, lines 17-21):

1) hydrologically altered and nutrient enriched peripheral sites feature accelerated C cycling, reflected in more decomposed peat,

2) increased abundance of vascular plants can increase CO_2 uptake but also change patterns of CH_4 production and emission, in particular if graminoids dominate, and

3) long-term nutrient enrichment in combination with hydrologically altered conditions may therefore cause differential responses of carbon cycling and does not necessarily cause a loss of the C-sink function of peatland ecosystems.

The concluding remarks section was adjusted accordingly. Please see page 19, line 18 to page 20, line 6.

* As you state in the discussion section (page 14, lines 13-15), it is hard if not impossible to separate the wetness effects from the nutrient infiltration effects. Thus, to draw any conclusions about anthropogenic effects on C cycling, it should be carefully considered how the data presentation is organized to serve that purpose.

– We agree. Please see our explanations above on how we improved our manuscript with respect to this aspect. Moreover, the discussion was in large parts completely rewritten.

* 2. The MS includes interesting isotopic data of the CH4 emission and porewater DIC and CH4. A better explanation of how the stable isotopic data can be interpreted would be very much needed already in the introduction section.

– A better explanation was added, which covers the interpretation regarding underlying pathways and methanotrophic activity. Please see page 2, lines 17-32.

* In the discussion section (page 15, line 24) you mention that the isotopic signature in methane is affected by methane production, oxidation and transport but you do not explain anywhere why and how the isotopic composition is affected by these processes.

– Such explanations were added to the introduction (page 2, lines 23-25; lines 27-32) and to the discussion (page 17, lines 7-22; page 18, lines 33-34).

Further, the discussion of isotopic data is related to methane oxidation. Could the dominant methane production pathway (acetoclastic, hydrogenotrophic) or transport pathway have caused differences in isotopic signatures and how? At the moment, the discussion on isotopic signatures is related to methane oxidation only.

– We understand your concern. Of course, the dominant CH₄ production and transport pathway cause differences in isotopic signatures. This aspect is now included in the revised version of our manuscript. According to Whiticar et al. (1986) isotope fractionation factors α_c were calculated for depths below 35 cm as for such depths we could assume water-saturated, anoxic conditions. We think distinguishing

methane production pathways for the upper depths with our data would be critical. That is why we would prefer to refrain from it.
Explanations were added:
-methods: page 10, lines 20-23
-results: page 13, line 29 to page 14, line 3
-discussion: page 17, lines 7-22; page 18, lines 13-29; page 19, lines 28-32.

* 3. In many occasions, you refer often to your own, yet unpublished work (Berger et al., submitted). Since that work seems to contain information quite crucial for the present paper, it is somewhat problematic that the paper is not available for the reader. If the submitted paper has not been published meanwhile, you should consider elaborate those results in more detail when necessary, e.g., in the methods section, page 4, line 2-3 about the hydrological changes caused by the reservoirs and page 4, line 23.

The paper is published now in Soil Biology & Biochemistry 114 (2017) 131-144
 <u>http://dx.doi.org/10.1016/j.soilbio.2017.07.011</u>
 We apologize that this information could not be presented earlier.

***Minor comments

* page 1, lines 18-19 & page 11, lines 19-21: The study period includes a full year of measurements. It would be good to give values also on cumulative annual fluxes. This was enable using this carefully collected data in flux syntheses, and facilitate comparison with literature values.

Done. Please see:
-page 1, line 23, 24
-page 10, lines 9-12; lines 22-24
-page 15, line 26 to page 16, line 4

* page 2, lines 3-5: One-sentence paragraphs should be avoided. I suggest combining this sentence with the next paragraph. See also page 4, line 29; and page 17, lines 16-19.

- Done. Please see: page 2, lines 9-11; page 5, lines 6-7; page 19, lines 29-31.

* page 2, lines 26-28: Reference missing.

- The respective sentence was removed.

* page 3, lines 12-15 & page 4, line 2: Also here, I would like to see a mentioning about how the hydrology is altered – drying or wetting, or more variable in the course of the year?

– Information was added. Please see page 4, line 31 to page 5, line 3.

* page 4, line 13: microtopography is a single word

- This sentence was removed.

* page 4, lines 18-19: Listing the sites starting from number 4 is counterintuitive. Would it be possible to change the order in which you mention the sites, or simply change the numbering? You indicate that site 2 was further away from the reservoir than site 3, but it would be better to describe the whole transect, e.g., that the distance from reservoir decreases with growing number.

– We are sorry, but we don't think that would be possible. As the site names were established in the SBB paper, changing the names now would be a bit confusing...

* page 4, lines 25-28: It is not clear if and how this is related to the vicinity of reservoir?

- You are right. This sentence was removed.

* page 5, line 8: At the first appearance, write the complete instrument type instead of the abbreviation FTIR.

– Done. Please see page 6, line 13.

* page 5, line 18: Regarding UV-VIS, see the previous comment.

– Done. Please see page 6, lines 24-25.

* page 6, line 12: Was the image analysis based on satellite/aerial or other imagery? Please specify!

- It was done via aerial imagery with images obtained from UAV flights. This information was added. Please see: page 7, lines 15-16.

* page 8, lines 5-6: Have you tested if there were any discrimination against the lighter isotope during diffusion into the silicon collectors?

- We are very sorry for confusing "silicone" (in German: "Silikon") with "silicon". This funny mistake was removed from the manuscript. Moreover, explanations on equilibrium times and fractionation at the silicone membrane were added. Please see page 9, lines 11-13.

* page 8, lines 13: Please check the sentence (some words missing/in a wrong order).

– Done. Please see page 9, lines 7-10.

"Silicone tubes for isotope sampling had an inner diameter of 1 or 0.5 cm, corresponding to a volume of 20 or 5 ml. The samplers with a volume of 20 ml were installed in 5 cm depth and the smaller samplers below, as close to surface larger volumes of samples were necessary in order to obtain sufficiently high concentrations (2.5 < x < 2000 ppm) for isotope analysis."

* page 9, lines 29-31: Please check the sentence (some words missing/in a wrong order).

- Done. Please see page 10, line 34 to page 11, line 2.

"Means were compared with t-Tests (if data was normally distributed) respectively Kruskal-Wallis and post hoc Wilcoxon-Mann-Whitney-Test (if data was not normally distributed). The confidence level for the statistical tests was $\alpha = 0.05$."

* page 11, lines 22-24: These are important results for this paper. But, can you really say that it is anthropogenic effect, or just a consequence of different location (edge effect, more mineral site?). It's interesting that the site receiving more nutrients is showing lower CO2 uptake.

- We agree that the observed effects can have different causes. However, looking at the entire transect the peat quality found at the sites suggests a quite similar history before dam construction at the site. So, based on our analysis the factor we can identify is the enrichment in nutrients and the concomitantly altered vegetation. Of course, site 4 thus had longest exposure to more minerotrophic conditions from intrusion of lake water. However, we would also consider this effect as anthropogenic then.

We provided statements in this regard in our methods (page 5, line 33 to page 6, line 2) and discussion (page 14, lines 18-22) sections.

* page 13, lines 19-21: Or, is the higher lability of organic matter caused by higher productivity and high input of labile compounds from vegetation? This site showed the highest C accumulation (page 11, lines 19-21). If it is a reservoir effect, should not the organic matter at site 4 be even more labile? Now, the site 4 was showing the highest proportion of aromatic compounds.

– That is a very interesting question! You are right, it could very well be the case that the higher lability of organic matter is caused by the vegetation (higher productivity \rightarrow higher input of labile compounds). That idea is now included in the discussion. (Please see page 15, lines 13-19.) We first came to our conclusions as DOM is usually a small but easily accessible pool and should thus reflect a residual enrichment of refractory compounds. Presence of labile DOM we thus attributed to external input. Indeed, this would then also apply to site 4. This discussion was not straightforward and was now modified considering your suggestion.

* page 13, lines 19-21: Please add references: In recent studies by Bragazza et al. ...

- Sentence was removed.

* page 14, line 21: Decrease in the CO2-sink strength in response to what?

- Sentence was removed.

* page 16, lines 3-4: You write about "deepening of soil oxygenation probably promoting a highly active methanotrophic bacteria community, which drew CH4 from the atmosphere down to that depth". Why do you think it was atmospheric and not peat-derived CH4 that was oxidized at 15 cm? Atmospheric methane cannot diffuse to the soil against the concentration gradient (when the porewater concentrations are above ambient).

– You are right. That is speculative. We thought it probably was atmospheric CH₄, which entered the soil because the CO₂ and CH₄ concentrations in the upper peat layers were particularly low on that day. The water table even was below -15 cm depth. Also, the surface peat looked quite dry during that summer period and the *Sphagnum* mosses were white and inactive (later in the year they recovered). So, our idea was that the CH₄ in 5 cm depth was of atmospheric origin. But we had indeed no proof for this interpretation. Therefore, we adjusted our discussion here. Please see page 17, line 31 to page 18, line 1.

* page 16, line 5: Why enriched signals would mean low CH4 production? Do you mean more CH4 production via the acetoclastic pathway that results in heavier methane than the hydrogenotrophic pathway?

– Actually, we were trying to avoid speculations about acetoclastic and hydrogenotrophic pathways for depths above -35 cm, because values of α_C typically observed for the acetoclastic pathway could also arise from methanotrophic activity. Both processes would yield an enrichment in ¹³C of the residual CH₄. We believe that it is rather CH₄ oxidation, which took place because of the low water tables and the unsaturated conditions during the summer. So, for depth above -35 cm, we boiled it down to: methane production yields less enriched ¹³C - CH₄, while CH₄ oxidation would leave behind comparatively more enriched CH₄. We then interpreted more oxidation as less net production. Only for depths below -35 cm we distinguished methanogenic pathways.

Therefore, section 4.3 was completely rewritten. Please see page 17, line 6 to page 19, line 16.

* page 16, lines 20-21: Besides transporting CH4 through the aerobic peat layers without exposing it to oxygen plant-mediated transport also strongly discriminates against the heavier methane (Chanton, 2005). Because of this, plant transport can create even lighter methane that is present anywhere in the peatland. It would be important to mention this in the discussion about isotopic signatures.

– Done. Please see page 18, lines 33-34.

* page 16, lines 23-28: Yes, probably most of the methane is oxidized during the diffusion, and thus, the amount of methane reaching the atmosphere by diffusion is low. So even with low coverage of aerenchymous plants, most of the methane that is actually entering the atmosphere is emitted through them.

– That was our idea, too.

* page 16, lines 28-30: It seems to me that you have done all the necessary pre-cautions to avoid methodological biases in the data. Please specify, what actually makes you suspect some methodological problems particularly in low water table conditions.

– For gas sampling for later isotope abundance analyses we used the same chambers as used for the flux measurements. It is known that chamber measurements tend to overestimate CH₄ fluxes a bit for several reasons (spatial heterogeneity, artificial pressure fluctuations induced by the chambers...). So, artefacts cannot be fully excluded. To counteract possible concerns in terms of data quality we verified our chamber flux data with eddy covariance flux data (see the figure below). The sets of data are nicely comparable, however, CH₄ fluxes measured with chambers were slightly increased in July and August when the water tables were lowest, which we think could have something to do with deeper CH₄ pools becoming connected to the atmosphere under unsaturated conditions with dropping water tables as explained in Estop-Aragones et al. (2016). With chamber induced pressure fluctuations such CH₄ pools might have been forced out of the peat.

We would like to clarify that in the course of quality assurance/ quality checks while processing data, most likely all low-quality data was eliminated. Thus, we are sure that our data provided (fluxes and isotope data) is of very high quality. By mentioning about the issue in the first place we intended to point out the common shortcomings of the method; instead it probably downgraded our results, so we removed the related sentences from the manuscript to avoid any misunderstandings.



A comparison of our eddy covariance vs. chamber measurements for the NEE (left) and CH₄ flux (right). The gray lines indicate the 1:1 lines. A good agreement is apparent; however, the chamber measurements detected higher CH₄ emissions in July and August (circled in gray).

* page 17, line 9: Instead of just saying results, it would be better to specify which particular parameter you mean here.

- The concluding remarks section was rewritten and the confusing statement was removed.

* page 17, lines 16-19: Long and complicated sentence, please consider splitting it into two sentences.

- Done. Please see page 19, lines 28-32.

* Fig. 1: For better clarity, please mark the Luther lake reservoir in the figure.

– Done.

* Fig 6 (and page 12, lines 28-29). In this figure, you have decided to use the porewater d13C-CH4 at 5 cm. However, the methane pool at this depth does not necessarily represent very well the origin of the methane emissions, since the ebullitive and plantmediated fluxes are originating from deeper layers. Hornibrook (2009) was using the average from 0 to 50 cm. It would be interesting to see how the figure looks if the porewater methane at depth is not excluded. Although the differences between different depths were not significant, Fig. 5 shows that especially at sites 1 and 2, the porewater CH4 has different isotopic composition at 5 cm than at deeper depths.

– We redrew the panel (e) of figure 6 using also δ^{13} C-CH₄ -values from the deeper CH₄. (please see Fig. 6 in the revised manuscript.

* Fig 6. caption, line 5: A typo in the word 'circles'.

- Was corrected.

* Table 1: The differences in stoichiometry are not evident, and I do not see a clear transect here. Could you test the differences statistically and mark it in the table? Or, would the amounts instead of ratios reveal the pattern more clearly?

- Thank you for pointing it out. We removed the stochiometric ratios from the table, added more information on the vegetation and described the transect in the text while referring the reader to the SBB paper for more information on nutrient supply to the study site.

References:

Ballantyne, D. M., Hribljan, J. A., Pypker, T. G., Chimmer, R. A.: Long-term water table manipulations alter peatland gaseous carbon fluxes in Northern Michigan, Wetlands Ecology and Management, 22, 35–47, 2014.

Berger, S., Gebauer, G., Blodau, C., Knorr, K.-H.: Peatlands in a eutrophic world – Assessing the state of a poor fen-bog transition in southern Ontario, Canada, after long term nutrient input and altered hydrological conditions, Soil Biology and Biochemistry, 114, 131–144, 2017.

Estop-Aragones, C., Zajac, K., Blodau, C.: Effects of extreme experimental drought and rewetting on CO₂ and CH₄ exchange in mesocosms of 14 European peatlands with different nitrogen and sulfur deposition, Global Change Biology, 22 (6), 2285–2300, 2016.

Kim, Y., Ullah, S., Roulet, N. T., Moore, T. R.: Effect of inundation, oxygen and temperature on carbon mineralization in boreal ecosystems, Science of the Total Environment, 511, 381–392, 2015.

Robroek, B. J. M., Jassey, V. E. J., Kox, M. A. R., Berendsen, R. L., Mills, R. T.E., Cecillon, L., Puissant, J., Meima-Franke, M., Bakker, P. A. H. M., Bodelier, P. L. E.: Peatland vascular plant functional types affect methane dynamics by altering microbial community structure, Journal of Ecology, 103, 925–934, 2015.

Whiticar, M. J, Faber, E., Schoell, M.: Biogenic methane formation in marine and freshwater environments. CO_2 reduction vs. acetate fermentation—Isotope evidence, Geochimica et Cosmochimica Acta, 50 (5), 693–709, doi:10.1016/0016-7037(86)90346-7, 1986.

Dear Reviewer 2,

Thank you very much for your thoughtful review of our manuscript. We very much appreciate your time your time and your valuable ideas shared to improve our manuscript. Please find our answers to your comments below. As some of your concerns were also raised by reviewer 1, we kindly ask you to also read our response to reviewer 1.

* The manuscript by Berger and co-authors is very interesting because the authors provide a comprehensive dataset on how soil carbon cycling changes along a transect of four study sites (from undisturbed to disturbed conditions) in a peatland complex in Ontario from April 2014 until September 2015. They used a variety of methods that complement each other in space and time (e.g. chamber flux measurements of CO2 and CH4, DIC and CH4 concentration measurements at different soil depths, stable isotope measurements of CO2 and CH4, FTIR analysis of organic matter and porewater and measurements of ancillary variables such as air and water temperature, photosynthetically active radiation and water table depth below surface). The authors raise the major question, how peatland carbon fluxes respond to anthropogenically changed hydrological conditions and long-term nutrient-infiltration effects. Their major answer is that plant functional type may be a key variable to predict how soil carbon cycling in peatlands will respond to future nutrient inputs and changes in hydrology. Shrub dominated disturbed peatlands may turn into carbon sources, while

graminoid-moss dominated peatlands "may maintain the peatland's carbon storage function". However, I have few major concerns but after a thorough revision and/or modification of the manuscript it would be great to see this manuscript published in the Biogeoscience journal.

* Major comments:

The authors point out that it is not new that plant functional types may have a strong influence on ecosystem soil carbon dynamics but I completely agree with the authors that "there is a gap of knowledge in terms of interactions between peat and plants under INSITU CONDITIONS". This makes this manuscript very valuable. However, I am not an author of the paper "Peatland vascular plant functional types affect methane dynamics by altering microbial community structure. (Robroek et al. 2015, doi: 10.1111/1365-2745.12413)" but the authors of this manuscript should cite that paper and compare both results. Robroek et al. (2015) nicely demonstrate that resilience of peatland CH4 dynamics, and therefore also CO2 dynamics, to climate change may depend on interaction between microbes and plant functional types.

- Done. Please see page 3, lines 23-25; page 14, lines 24-25; page 15, lines 13-14.

* I think the manuscript would greatly benefit from a more thorough discussion about the potential role of methanogens driving soil methane dynamics at the four different sites. In the current study, the authors measured stable carbon isotope ratios of CH4 and CO2 comprehensively. Hence, apparent fractionation factors could be easily measured (Angel et al. 2011; doi:10.1371/journal.pone.0020453 or McCalley et al. 2014; doi:10.1038/nature13798), the different pathways of methanogenesis identified and discussed. Now, the authors attribute the change of isotopic signals to changes in methane oxidation. This is very speculative and not sufficient. The change in 13CH4 may result from a shift from hydrogenotrophic to aceitclastic methanogenesis, especially during drier months (see Hodgkins et al. 2014; www.pnas.org/cgi/doi/10.1073/pnas.1314641111 2014 or McCalley et al. 2014; doi:10.1038/nature13798). However, this should be discussed in the manuscript.

- We understand yours and reviewer 1's concern.

Of course, the dominant CH₄ production and transport pathway cause differences in isotopic signatures. It is not that we forgot to obtain fractionation factors and to include those pathways in the previous version of the manuscript but we had decided to exclude the issue because we thought and still think distinguishing methane production pathways with our data would be critical. Methane production strictly depends on water-saturated, anoxic conditions, but our peatland site experienced water level dropdowns down to -32 cm below peat surface. Under such conditions we assumed that methanotrophy would probably be a more likely process to leave 13 C-enriched CH₄ behind as compared with acetoclastic methanogenesis. However, since both reviewers raised that point, we decided to include fractionation factors for depths below -35 cm, assuming that here conditions would be anoxic and water-saturated.

In the revised manuscript please see: -page 2, lines 2-3 -page 3, lines 17-32 -page 10, lines 20-23 -page 13, line 27 to page 14, line 3 -page 17, lines 7-27 -page 18, lines 13-29 -table 2

* The authors state that it is clearly visible that ratios of C/N, C/Mg and C/K in peat soil are decreasing from site 1 to site 4. I do not see this pattern when I look at Table 1. C/K is higher at site 2 and 3 than at site 1. C/Mg is lowest at site 1. I guess C/N ratios do not differ between site 1, 2 and 3.

Furthermore, I guess there are no significant differences in C/P ratios between the different sites. N/P ratios are higher at site 2 than site 3 and C/Ca ratios are lowest at site 2. Please, check your data.

You are right. This table was supposed to be a short summary of the previous study, which is now published in Soil Biology & Biochemistry (114 (2017) 131-144 http://dx.doi.org/10.1016/j.soilbio.2017.07.011). We agree that this table, providing only this little information, is not convincing, as the results from the SBB paper are quite complex.

We removed the stochiometric ratios from table 1 and provided a better description of the most important results from the SBB paper, which is providing more convincing evidence that those sites in closer vicinity to the reservoir are more affected.

Instead, information on plant species abundances at each site were added to better illustrate the vegetation gradient.

* However, it would be great to have a look at the submitted publication or if the authors would incorporate more convincing information. Otherwise the authors cannot state that "it becomes evident that the peatland was exposed to nutrient infiltration form the water reservoir and thus elevated nutrient concentrations occurred in vicinity to the water reservoir." (P13, L2-L6) and should reformulate the whole discussion section!

- We would be pleased if you were to have a look at the paper (SBB 114 (2017) 131-144). Here, the observed differences in the transect are elaborated. Due to the complexity of the dataset only a short summary can be provided in the present manuscript. We revised our text to improve clarity and referred to the now published study. We apologize that the manuscript had not been available at the time of submitting this paper.

* Specific comments:

Titel: Currently, I do not see that nutrients drive carbon dynamics at your sites.

– We changed the title:

"Differential response of carbon cycling in a continental Canadian peatland to long-term nutrient input and altered hydrological conditions"

* Abstract: If you mention the other methods in the abstract, you should mention FTIR analysis as well.

– Done. Please see page 1, line 19.

* P1, L17-L19: All the sites are characterized by wet conditions. These are peat soils.

– What we were trying to say was that the site 3 hollows experienced higher water levels. This sentence was anyway removed.

* P1, L19-L20: Low 13CH4 may be caused by more hydrogenotrophically produced CH4.

* *P1, L24: or more aceticlastically produced CH4. More labile organic matter may favor aceticlastic methanogenesis.*

- Yes, we agree, but given the unsaturated conditions and strong water table drop downs during the summer and as explained further above, we think that it would be critical to distinguish methane production pathways for the shallower depths; an influence of methanotrophic conditions is much more likely. Indeed, we think that currently not many studies have presented results from around the unsaturated zone and thus most existing studies focused on discussion pathways at greater, saturated depths.

* P3, L8: I do not see a gradient in nutrient availability.

– That is probably because the provided data was in its previous presentation not convincing. The table presented only a small fraction of the data provided in the SBB paper, however, the line of argument of the SBB paper is based on a greater data set (peat ages and accumulation rates, depth profiles of element concentrations, stochiometric ratios of surface peat, δ^{15} N-values and C/N ratios of the vegetation as well as composition of the vegetation) in order to properly describe the impact of the water reservoir on the study area and its sites. When considering only single factors (e.g. only stochiometric ratios of surface peat), their explanatory power decreases; it is the entirety of factors which shapes our knowledge of the study area. We are sorry for taking the stochiometric ratios out of context. We removed the stochiometric ratios from the table and provided a more convincing description of the study sites in terms of nutrient availability in the methods section while referring the reader to the SBB manuscript. Please see page 5, lines 24-32.

After removing the stochiometric ratios from table 1 we added more information on the vegetational gradient so that differences between sites become more obvious.

* P3, L10: Please, calculate apparent fractionation factors for methanogenic pathways.

– Please see our answer above. We have calculated such fractionation factors for depths below -35 cm and added corresponding explanations. Please see page 10, lines 20-23; page 13, line 27 to page 14, line 3; and table 2.

* P3, L16: I do not see that nutrient inputs are greatest in peatland periphery (see Table 1).

– Please see our answer above.

* P3, L 18: Why should CH4 emissions be greatest at the graminoid dominated sites? There is no link to this hypothesis in the introduction.

– All 3 hypotheses were revised as also reviewer 1 requested for more specific hypotheses. Accordingly, most of the introduction needed to be rewritten.

* P3, L19-21: You should also discuss CH4 production pathways.

– Done. Please see page 2, lies 17-27.

* P4, L22-28: This paragraph is very essential for the main message of the manuscript. Unfortunately, the data do not support these statements. It would be great to see more data that support these ideas.

– We agree, the table and the statements were not convincing here. Please see our answers above on how we improved table 1 and the statements throughout paragraph 2.1.

* P5, L8: Please, write out FTIR analysis.

– Done. Please see page 6, line 13.

* P5, L8-L10: I am not familiar with FTIR analysis. "For pore-water samples 2 mg of oven-dried organic matter..." is that correct?

– Yes, it is correct, but maybe the sentence was a bit misleading. The pore water samples were dried in an oven until all the water was gone. Then the remaining solid material was scratched off the bottom of the sample containers and underwent FTIR analysis. We provided a better description. Please see page 6, lines 14-15.

* RESULTS section: The presentation of the results are too cluttered. Results that are not significant are described quite often (see P10, L7, L10) and sometimes it is not clear if results are significant or not (see P10, L16-17, L19-26). It would be better to mark significant differences in the figures and to highlight significant results or only few non-significant results in text, if they really enrich and/or support the guiding questions in the manuscript.

– As the study design was indeed very complex, we ended up with very complex results. However, we very much agree with your comment and tried to improve the data presentation. Therefore, in our revised text we focused more on significant results and provided less space for results that were not significant. To mark significant differences in the figures (in particular in figures showing time series data) was difficult because sometimes site 3 differed significantly from site 4 and sometimes site 3 and 4 differed from site 1 and 2, sometimes there were no differences, sometimes there were differences for certain dates/depths etc. but not for all sites, etc. So, we believe the figures would have gotten too complex if we added additional information. To improve clarity, we re-arranged the figures 3 and 4, following also reviewer 1's suggestion. Moreover, less space was provided for non-significant results when describing the results in the text. We also tried to organize the description of the results in a way that it now always follows the same pattern. Therefore, almost the entire results section was rewritten.

* P13, L15-L16: This is repetition of results.

- The sentence was removed.

* P13, L19-L21: This is very speculative. Did you check FTIR ratios of inflowing water? Maybe you can provide some references.

– We agree. Unfortunately, we did not sample the inflowing water. Reviewer 1 was also concerned about this statement and provided an alternative explanation (higher productivity of the graminoid vegetation \rightarrow higher input of labile compounds), which we included into the discussion. Please see page 15, lines 13-19.

* P13, L22-L23: I am not convinced by this statement. The difference between site 4 and the three other sites may be simply by chance.

– We understand your concern. You are right, the observed effects can have different causes. However, looking at the entire transect the peat quality found at the sites suggests a quite similar history before

dam construction at the site. So, based on our analysis the factor we can identify is the enrichment in nutrients and the concomitantly altered vegetation. Of course, site 4 thus had longest exposure to more minerotrophic conditions from intrusion of lake water. However, we would also consider this effect as anthropogenic then.

Our results are derived from an in-situ study; of course, experimental set-ups under controlled (laboratory) conditions provide more explicit results and such results can be more reliably related to certain factors. As compared to such studies, in-situ studies have short-comings indeed; however, in-situ studies are needed to verify concepts based on such controlled conditions and we think that we have taken all necessary pre-cautions to avoid misinterpretations and to not over-interpret our data. Hopefully you agree with us in terms of the significance of results derived from our study.

We provided statements in this regard in our methods (page 5, line 33 to page 6, line 2) and discussion (page 14, lines 18-22) sections.

* *P13, L24-L27: So, it is not the vicinity to the reservoir but the vegetation that drives carbon cycling processes?*

– We are not sure if we can provide a final answer to that question with our data. We think we are dealing with a complex interplay between vegetation, microorganisms and location factors. Site 4 and site 3 appeared to have received a similarly high amount of nutrients (well, site 4 probably received a bit more); around site 4 a dense *Myrica* belt established while at site 3 graminoids established. (The SBB paper provides information on vegetation etc.) With *Myrica* being present at the site, site 4 developed in a different way than site 3, where graminoids are established. So, we think it is both, the vicinity to the reservoir and the vegetation community that drives carbon cycling. By our study we thus also want to support that the response of a peatland to nutrient input and altered hydrological conditions may not be as simple as identified in studies with controlled variation of individual factors. Please see page 15, lines 20-24.

* P14, L11-L19: This is repetition of results.

- The paragraph was almost entirely rewritten. Please see page 15 line 26 to page 17 line 5.

* P14, L25: It would be great to see the data.

– Please see the SBB paper.

* P15, L1-L2: Site 4 shows second highest CH4 release. Then you cannot state that graminoid sites show highest CH4 emissions. I would emphasize to reformulate the introduction and the hypothesis in such a manner that it becomes clearer to the reader why you have stated your hypothesis. Now, the discussions seems to be much too blurred.

- Introduction, hypotheses and discussion were rewritten and the misleading statement was removed.

* P15, L6: What means "healthy" Sphagnum moss community?

- The mosses at site 4 showed severe signs of desiccation and thus inactivity in 2014 and 2015 during the summer months (June ~ September) and recovered afterwards. (Reduced photosynthetic activity of *Sphagnum* mosses while facing severe drought was previously observed in several studies (e.g. Alm et al., 1999, Aurela et al., 2007)). Given the pitiful appearance of *Sphagnum* mosses during the summers at site 4, and given also that *Sphagnum* covers only 60 % of the site 4 area, we concluded

that *Sphagnum* was in retreat at site 4. In contrast, at the sites 1, 2 and 3 the *Sphagnum* mosses looked green or red (whatever their natural color was) and always moist, which we then termed a "healthy" appearance of *Sphagnum* mosses. We apologize for utilizing the colloquial expression "healthy". We worked in a more appropriate description of the vegetation of our study sites into our revised manuscript, to avoid any misunderstandings. Please see page 5, lines 8-23.

* P15, L22 – P16, L33: see major comments.

* P17, L2 – L19: see major comments.

- Please see our answers above.

* Figures: It would be great to have figures with higher resolution. In Figure 6, I can hardly identify the difference between the circles.

– Figure 6 is now provided in a higher resolution. Also, the figures 3 and 4 were redrawn.

* Table1: Please, mark significant differences.

- Stochiometric ratios from table 1 were removed as explained above.

References:

Alm, J., Schulman, L., Walden, J., Nyknen, H., Martikainen, P. J., Silvola, J.: Carbon balance of a boreal bog during a year with an exceptionally dry summer; Ecohydrology, 4, 733–743, 1999.

Aurela, M., Riutta, T., Laurila, T., Tuovinen, J.-P., Vesala, T., Tuittila, E.-S., Rinne, J., Haapanala, S., Laine, J.: CO₂ exchange of a sedge fen in southern Finland – the impact of a drought period, Tellus B, 59, 826–837, 2007.

Berger, S., Gebauer, G., Blodau, C., Knorr, K.-H.: Peatlands in a eutrophic world – Assessing the state of a poor fen-bog transition in southern Ontario, Canada, after long term nutrient input and altered hydrological conditions, Soil Biology and Biochemistry, 114, 131–144, 2017.

Differential response of carbon cycling to long-term nutrient input and altered hydrological conditions in a continental Canadian peatland

Sina Berger^{1,2,3}, Leandra Praetzel^{1,2}, Marie Goebel^{1,2}, Christian Blodau^{1,2,†}, Klaus-Holger Knorr¹

¹ University of Muenster, Institute of Landscape Ecology, Ecohydrology and Biogeochemistry Group, Heisenbergstraße 2, 48149 Muenster, Germany

² University of Guelph, School of Environmental Sciences, 50 Stone Road East, Guelph, Ontario, N1G 2W1, Canada

³ Karlsruhe Institute of Technology, Institute of Meteorology and Climate Research (IMK-IFU), Kreuzeckbahnstraße 19, 82467 Garmisch-Partenkirchen, Germany

10 Correspondence to: Sina Berger (gefleckterschierling@gmx.de), Klaus-Holger Knorr (kh.knorr@uni-muenster.de)

Abstract. Peatlands play an important role in global carbon cycling, and their responses to long-term anthropogenically changed hydrologic conditions and nutrient infiltration are not well known_x While experimental manipulation studies, e.g. fertilization or water table manipulations, exist on the plot scale, only few studies have addressed such factors under in-situ conditions along gradients within larger sites. Therefore, an ecological gradient from center to periphery of a continental

- 15 Canadian peatland bordering a eutrophic water reservoir, as reflected by increasing nutrient input, enhanced water level fluctuations, and increasing coverage of vascular plants, was used for a case study of carbon cycling along a sequence of four differently altered sites. Here we monitored carbon dioxide (CO₂) and methane (CH₄) fluxes at the soil/atmosphere interface and dissolved inorganic carbon (DIC) and CH₄ concentrations along peat profiles from April 2014 through September 2015. Moreover, we studied bulk-peat and pore-water quality and we applied δ¹³C-CH₄ and δ¹³C-CO₂ stable isotope abundance
- 20 analyses to examine dominant CH₄ production and emission pathways during the growing season of 2015. We observed differential responses of carbon cycling at the four sites, presumably driven by abundances of plant functional types (PFTs) and vicinity to the reservoir. A shrub dominated site in close vicinity to the reservoir, was a comparably weak sink for CO₂ (in 1.5 years: -1093 ±794, in 1 year: +135 ±281 g CO₂ m⁻² (=net release)) as compared to two graminoid-moss dominated sites and moss dominated site, (in 1.5 years: -1552 to -2260 g CO₂ m⁻², in 1 year: -896 to -1282 g CO₂ m⁻²). Also, the shrub-
- 25 dominated site featured notably low DIC concentrations along peat pore-gas profiles as well as comparably ¹³C enriched CH₄ (ô¹³C-CH₄: -57.81 ±7.03 ‰) and depleted CO₂ (ô¹³C-CO₂: -15.85 ±3.61 ‰) in a more decomposed peat, suggesting a higher share of CH₄ oxidation and differences in predominant methanogenic pathways. The graminoid-moss dominated site in closer vicinity to the reservoir featured a in comparison to all other sites by ~30 % increased CH₄ emission (in 1.5 years: +61.4 ±32,

1

Deleted: Plant functional types, nutrients and hydrology drive carbon cycling along a transect in an anthropogenically altered Canadian peatland complex

Deleted:

Deleted: however, the response of peatland carbon fluxes to anthropogenically changed hydrologic conditions and long-term infiltration of nutrients is still understudied. Along a transect of 4 study sites, spanning from largely pristine to strongly altered conditions within the Wylde Lake peatland complex in Ontario (Canada), Deleted: we

Deleted: in

Deleted: the

Deleted: CH_4 and CO_2 production and consumption as well as the

Deleted: with a mixture of PFTs

in 1 year: $+39.86 \pm 16.81$ g CH₄ m⁻²), and low δ^{13} C-CH₄ signatures (-62.30 \pm 5.54 ‰), indicating only low mitigation of CH₄ emission by methanotrophic activity here. Methanogenesis and methanotrophy appeared to be related to the vicinity to the water reservoir: the importance of acetoclastic CH₄ production apparently increased toward the reservoir, whereas the importance of CH₄ oxidation increased toward the peatland center. Plant mediated transport was the prevailing CH₄ emission

5 pathway at all sites even where graminoids were rare. Our study thus illustrates an accelerated carbon cycling in a strongly altered peatland with consequences for CO2 and CH4 budgets. However, our results suggest that long-term excess nutrient input does not necessarily lead to a loss of the peatland C-sink function.

1 Introduction

Since the end of the last glaciation, northern peatlands have played an important role in global carbon (C) cycling by storing atmospheric carbon dioxide (CO₂) as peat, but also emitting significant amounts of C as methane (CH₄) (Succow and Joosten, 2012). Carbon sequestration and CO₂ and CH₄ release are driven by numerous processes and the accumulation of peat results

- from only a small imbalance of photosynthetic carbon uptake over respiratory losses. CO₂ can be released through autotrophic and heterotrophic respiration under aerobic and anaerobic conditions (Limpens et al., 2008). Controls on heterotrophic respiration have been intensively studied and depend e.g. on temperature, substrate quality, energetic constraints and other
 factors (Blodau, 2002). Methanogenesis is strictly limited to anaerobic conditions (Conrad, 2005). Due to thermodynamic controls, CH₄ production is only competitive upon depletion of alternative, energetically more favorable electron acceptors for
- anaerobic respiration, such as nitrate, iron, sulfate or oxidized humics (Blodau, 2002; Klüpfel et al., 2014). <u>CH₄</u> is predominantly produced via two pathways: hydrogenotrophic and acetoclastic methanogenesis. During hydrogenotrophic methanogenesis CO_2 is reduced to CH_4 , while during acetoclastic methanogenesis acetate is split in CH_4 and CO_2 . These
- 20 pathways differ with respect to their discrimination against the heavier ¹³C-isotopes due to the kinetic isotope effect (Hoefs, 1987). Differences in the isotopic composition are thereby commonly presented as δ¹³C values, expressed as: δ¹³C = (R_{sample}/R_{standard} 1) 1000 [‰], where R is the ratio of heavy isotope to light isotope of the samples and the respective standard. Acetoclastic methanogenesis results in δ¹³C-CH₄ values of -65 to -50 ‰, while hydrogenotrophic methanogenesis, discriminates stronger against the heavier carbon isotope and results in δ¹³C-CH₄ values of -110 to -60 ‰ and considerably
- ¹³C enriched <u>CO₂</u> compared to the acetocalstic pathway (Whiticar et al., 1986). Specific patterns have been observed in terms of spatial and temporal occurrence of the major <u>CH₄</u> production pathways, with acetoclastic methanogensis typically increasing in contribution towards the surface or within the rhizosphere (Holmes et al., 2015). On the other hand, an assignment of methanogenic pathways based on ¹³C signatures of CH₄ can be biased by microbial <u>oxidation of CH₄</u>. This can in particular be the case near and in the unsaturated profile where oxygen can enter by diffusion, or in the rhizosphere where plants deliver
- 30 oxygen through aerenchyma roots (Chasar et al., 2000). Upon conversion of CH₄ into CO₂, the residual CH₄ gets enriched in 13C compared to the source CH₄ (Teh et al., 2006), a process which yields similar d13C-CH4 signatures as observed upon CH4 production by the acetoclastic pathway. CH₄ is released to the atmosphere by three different processes: i) through

2

Deleted: found that a graminoid-moss dominated site, which was exposed to wet conditions and ung-term infiltration of nutrients, was a great site of CO₂ (2260 ± 480 g CO₂ m²) but a great source of CH₄ (61 4 ± 32 g CH₄ m²). Comparably low δ^{13} C-CH₄ signatures (-62.30 ± 5.54 ‰) indicated only low mitigation of CH₄ emission by methanotrophic activity here. On the contrary, a shrub dominated site, which has been subjected to similarly high moisture conditions and loads of nutrients, was a much weaker sink of CO₂ (1093 ± 794 g CO₂ m²) as compared with all other sites. The shrub dominated site featured notably low DIC concentrations in the peat as well as comparably ¹¹C enriched CH₄ (δ^{10} C-CH₄ - 57.81 \pm 7.03 ‰) and depleted CO₂ (δ^{13} C-CO₂: -15.85 \pm 3.61 ‰) in a more decomposed and surficial aerate peat, suggesting a higher share of CH₄ oxidation. Plant mediated transport was the prevailing methane emission pathway throughout the summer of 2015 among all sites, even where graminoids covered only 10% of the area.

Deleted: supported

Deleted: earlier findings, that strongly altered, shrub dominated peatlands may turn into weak carbon sinks or even sources, while a graminoid-moss dominance may maintain the peatland's carbon storage function.

[...[1]]

Deleted: Asites is apparently less sensitive to sites . **Deleted:**

Deleted:

diffusion through the acrotelm, which is a relatively slow process, ii) through ebullition, i.e. a fast evasion of methane bubbles, and iii) through fast molecular diffusion or pressurized throughflow convection through aerenchymatous tissue of vascular plants (Morris et al., 2011; Schütz et al., 1991; Whiting and Chanton, 1996; van den Berg et al., 2016; Hornibrook et al., 2009). Due to the slow diffusion of methane in peat, up to 100 % of diffusive CH_4 is oxidized in the acrotelm before it reaches the

5 atmosphere, while the fast processes effectively bypass oxidation and thus contribute a major fraction to observed fluxes (Whalen et al., 1990; Whalen, 2005). Therefore, a change in vascular plant cover or changes in the peat structure due to altered litter inputs and stronger decomposition can be expected to affect methane emissions.

Carbon cycling and nitrogen (N) cycling in peatlands are coupled and eutrophication of peatlands is one major threat to these normally nutrient-limited ecosystems as demonstrated in several long-term fertilization experiments. For example, a decade

- 10 of fertilizer applications to bogs in Canada (Mer Bleue), in the UK (Whim Bog), in Sweden (Degerö Stormyr) and seven years of high nitrogen deposition to a bog in the Italian Alps caused a loss of mosses and an increase in vascular plant biomass (Bubier et al., 2007; Wang et al., 2016; Sheppard et al., 2013; Wiedermann et al., 2007; Bragazza et al., 2012). In the Mer Beue bog shrubs benefit most from increased nutrient availability, whereas at Whim bog, Degerö Stormyr and at an Italian mire it remained unclear whether shrubs or graminoids benefit most. Sheppard et al. (2013) further observed differential effects
- 15 on a peatland plant community when dry deposited ammonia-N and wet deposited reduced N, respectively, were applied. A number of studies supported that an increase of vascular plant cover can reduce the productivity of peat mosses and, in addition, can potentially promote the decomposition of organic matter by affecting the stoichiometry of soil enzymatic activity (Bragazza et al. 2013, Bragazza et al. 2015), ultimately leading to a decreasing ability of peatlands to sequester CO₂ from the atmosphere (Bubier et al., 2007), resulting in decomposition of peat (Rydin and Jeglum, 2013). Altered plant communities in
- 20 peatlands were repeatedly shown to alter CO₂ and CH₄ fluxes: in fact, maximum net ecosystem exchange (NEE) was found to be reduced after long-term fertilization and a concomitantly promoted vascular plant community in the Mer Bleue bog (Bubier et al., 2007), and increased CH₄ emissions were observed at Degerö Stormyr from plots with an increased vascular plant coverage after a decade of excess nutrient supply (Eriksson et al., 2010). Indeed, selective removal of plant functional types, although in this particular study combined with warming and drought experiments, demonstrated a strong impact of vegetation
- 25 changes on gas exchange (Larmola et al., 2013; Ward et al., 2013; Kuiper et al., 2014; Robroek et al., 2015). While such plot / based manipulation experiments as reported above revealed clear patterns, there is still a gap of knowledge in terms of long-term consequences of excess supply of nutrients to a peatland and the resulting interactions and feedbacks between plants and peat, especially under in-situ conditions. There is only a poor understanding of the interplay of plant functional types (PFTs), substrate quality, and anoxic-oxic conditions, and of how exchange of CO₂ and CH₄ at the soil/atmosphere-interface would
- 30 eventually be affected.

To address this research gap, we investigated C cycling of the once nutrient oligotrophic Wylde Lake peatland, which since 1954 is exposed to infiltration of nutrients and strongly pronounced water level fluctuations as induced by the nearby water reservoir. The site was in detail described by Berger et al. (2017); a particular finding was that even after decades of excess nutrient supply (currently 5.9 ± 0.1 to 4.35 ± 0.3 g m⁻² y⁻¹ of N input was determined and several more nutrients were found to

3

Deleted: s

Deleted: Since changes in distribution of plant functional types (PFT) were shown to lead to changes not only in methane en bus also in overall carbon cycling (Strack et al., 2006: Breeuwer et al., 2009, Kuiper et al., 2014), int eractions between plants, microbes and peat are increasingly being elaborated. For example, in the context of climate warming, an increasing ericaceous shrub cover was associated with increasing polyphenol content in plant litter and pore-water, as well as increasing phenol oxidase activity. Moreover, higher release of labile C from vascular plant roots was observed (Bragazza et al., 2013, Bragazza et al., 2015) and plant-derived low (Robrock et al., 2015). Also, long term nutrient inputs into peatlands are expected to change vegetation and as a consequence carbon cycling: For example, after a 10-15 years fertilization experiment in a bog in eastern Canada, both availability of substrates for microbes and activity of microbial enzymes were found to be altered (Pinsonneault et al., 2016). Thereby, the PFT of shrubs could parently buffer more effectively, used environmental nditions, as the bulk chemical composition and nutrient contents of litter seemed largely constant over a broad range of conditions with changed environmental resources (Wang et al., 2016).

Deleted: of Deleted: still

Deleted: poor

infiltrate the peatland from its periphery), the peatland still featured high peat accumulation rates of ~ 200 to ~ 300 g C m⁻² y⁻¹. However, a strong gradient in vascular plant cover was apparent. As pointed out by Berger et al. (2017) lateral nutrient influx through repeated inundation events cannot be easily compared to sites subjected to deposition from the atmosphere; nevertheless, an apparently intact peatland system, i.e. an intact mire, despite such serious anthropogenic impacts is

- 5 contradictive to findings from above mentioned studies; according to existing studies, already after one decade of N fertilization decomposition and peat degradation would be expected. Moreover, the particular scenario in our study here, the impacts of inundation on nearby ecosystems, is gaining increasing importance as there is a worldwide increase of impoundment area (Tranvik et al., 2009) and serious effects on peatland carbon cycling are likely (Ballantyne et al., 2014; Kim et al., 2015), The objective of this study was therefore to extend the existing study on nutrient impact, vegetation, and net carbon
- 10 accumulation, comparing effects on <u>C cycling</u> in more detail. To this end, we assessed current CO2 and CH4 exchange, peat quality and pore water chemistry along a transect, ranging from <u>a shrub dominated site (200 m distance to the reservoir; greatest nutrient input)</u>, over graminoid-moss dominated sites (400 and 550 m distance to the reservoir; intermediate nutrient input) to <u>a site with a mixture of mosses</u>, shrubs, graminoids and trees (800 m distance to the reservoir; smallest nutrient input) in the Wylde Lake peatland in Ontario, Canada. Moreover, to address changes in methanogenic pathways and to study predominant
- 15 pathways of emission, we assessed seasonal variation in 8¹³C of CH₄ in peat profiles and in CH₄ surface fluxes. We hypothesized (hat 1) hydrologically altered and nutrient enriched peripheral sites feature accelerated C cycling, reflected in more decomposed peat, 2) increased abundance of vascular plants can increase CO₂ uptake but also change patterns of CH₄ production and emission, in particular if graminoids dominate, and 3) long-term nutrient enrichment in combination with hydrologically altered conditions may therefore cause differential responses of carbon cycling and does not necessarily cause
- 20 a loss of the C-sink function of peatland ecosystems,

2 Methods

2.1 Description of the study area and study sites

Wylde Lake peatland has been described in detail in Berger et al., (2017). In brief, it is located in southeastern Ontario, 80 km northwest of Toronto (43.920361° N, 80.407167° W) (Fig. 1), and it is part of the Luther Lake Wildlife Management Area.
Climate is cool temperate, average July temperature is 19.1 °C, average January temperature is -8.0 °C and the mean annual temperature is about 6.7 °C. Annual precipitation amounts to 946 mm, with the major portion falling in summer (1981 to 2010, Fergus Shand Dam, National Climate Data and Information Archive, 2014). Peat formation started about 9000 years before present on calcareous limnic sediments and total peat depth today is about 5 meters.

For flood control and water management, the "Luther Lake" reservoir, neighboring Wylde Lake peatland, had been created in 1954. Through flooding of the reservoir, Wylde Lake peatland has been exposed to altered hydrological conditions <u>in a way</u> that the water reservoir enhanced water level fluctuations in a large part of the site: in summer or under dry conditions, water is released from the reservoir, thereby draining <u>water out of the peatland</u>; under wet conditions, water table levels of the

4

Deleted: strong anthropogenic impacts

Deleted: we studied the interaction between cover of certain PFTs, CO₂ and CH₄ emissions and peat quality along a gradient of nutrient availability through a nearby water reservoir. Thereby, we compared a shrub-moss-dominated site vs. graminoid-moss-dominated sites vs. a site with a mixture of mosses, shrubs, graminoids and trees. Moreover, to address changes in methanogenic pathways and to study predominant pathways of emission, we assessed seasonal variation in δ^{13} C of CH₄ in peat profiles and in CH₄ surface fluxes along a transect of study sites in the Wylde Lake peatland in Ontario, Canada. Wylde Lake peatland has recently been shown to experience strong changes in vascular plant coverage and strongly increased peat accumulation as a response to long term enhanced nutrient infiltration and changed hydrological conditions (see Berger et al., (submitted for publication)), so that different areas of the peatland can serve as sentinels of future responses of peatlands to land use changes.

Deleted: We hypothesized that 1) in the peatland periphery, where nutrient input is greatest, C-cycling is accelerated as indicated by more decomposed bulk peat along peat profiles, 2) altered abundance of PFTs cause concomitant differences in C-dynamics such as high CO₂ uptake at the shrub dominated sites, and greatest CH₄ emissions at graminoid dominated sites. Moreover, 3) the dominant CH₄ emission pathway is plant-dominated transport where aerenchymatous graminoids are abundant while under predominance of shrubs diffusion is relatively more important for CH₄ emission and thus methanotrophic modification of fluxes is more likely.

Deleted: submitted for publication

Deleted:	
Deleted:	
Deleted:	
Deleted:	•
-	

Moved (insertion) [1]

Deleted: Moreover

Deleted: thicker layers of readily decomposable fibric and hemic peat were found up to a depth of approx. 48 cm at sites 1 and 2 whereas at site 3 and 4, strongly decomposed sapric peat was already found below depths of 37 cm (site 3) and 29 cm (site 4) respectively. **Deleted:** and infiltration of nutrients since the 1950s.

reservoir increase and water is pushed into the peatland. Those sites in closer vicinity to the reservoir are presumably more affected than sites further away from the reservoir (Berger et al., 2017).

Four intensively investigated measurement sites (Fig. 1) were arranged along a transect stretching from <u>nearby</u> the shoreline

of the Luther Lake reservoir about ~1 km south into the central treed bog area. Each site featured an individual mosaic of
 hummocks, hollows, and lawns, however, all measurements as considered in this study were taken in and all samples were collected from hollows.

Site 4 was located about 200 m away from the reservoir in an area overgrown by *M. gale* and where *Sphagnum* mosses were in retreat. Site 4 will hereinafter be referred to as "*shrub dominated site* 4", Site 3 and site 2 were in the open poor fen - bog transition area with site 2 being further away from "Luther Lake" reservoir (550 m) than site 3 (400 m). Site 2 and site 3 were

- 10 dominated by Sphagnum mosses and graminoids with only few shrubs and will hereinafter be referred to as "graminoid-moss dominated sites". These sites featured a variety of arenchymatous graminoid species, such as Eriophorum spp. at the sites 3 and 2, and Dulichium arundinaceum at site 3. Site 1 (~800 m away from the reservoir) accommodated equal shares of few graminoids and shrubs above dominant Sphagnum mosses, and will be referred to as "moss dominated site 1". The four sites also differed with respect to their most abundant Sphagnum species, reflecting increasingly minerotrophic conditions towards
- 15 the lake. While S. capillifolium, an ombrotrophic to slightly minerotrophic hummock species (Laine et al., 2011), was abundant at the sites 1, 2 and 3, its abundance was decreased at site 4. Moreover, site 1 featured the abundant S. magellanicum (another ombrotrophic to weakly minerotrophic hummock species (Laine et al., 2011)), site 2 featured the abundant S. angustifolium (tolerating ombrotrophic to minerotrophic conditions (Laine et al., 2011)) and site 3 featured the abundant S. girgensohnii, a minerotrophic hollow species (Laine et al., 2011). The two most abundant Sphagnum species at site 4 were S. fuscum (mostly)
- 20 on hummocks but also in hollows, an ombrotrophic species (Laine et al., 2011), with a great ability to recover from desiccation (Nijp et al., 2014)) and again the minerotrophic hollow species S. girgensohhnii. See Table 1 for a detailed overview of the vegetation at the sites and see Fig. S1 for photographs of the sites.

As presented in Berger et al. (2017) the study area is subject to nutrient infiltration most likely from the "Luther Lake" water reservoir as indicated by increasing concentrations of nitrogen, phosphorus, sulfur, potassium, calcium, magnesium, iron,

- 25 copper, and zinc as well as other_metals in peat mostly toward the peatland periphery. N input rates of 5.9 ± 0.1 g N m⁻² y⁻¹ were reported for site 4 and 4.35 ± 0.3 g N m⁻² y⁻¹ for site 1; moreover, C/P and N/P ratios of surface peat suggested fen typical P limitation, C/Ca and C/Mg ratios indicated Ca and Mg limitation, while C/K ratios indicated higher K availability as compared to bog typical values presented in Wang et al. (2015). The peatland periphery appeared to act as a buffer for nutrients in a way that site 4 received the highest loads of nutrients but also areas further away were to some extent affected.
- 30 <u>Nevertheless</u>, surface peat accumulation rates of ~200 to ~300 g C m⁻² y⁻¹ at the four sites revealed great recent carbon sequestration.

The impact of anthropogenic activities, in particular the formation of the reservoir, is evident from peat quality found at the sites: a quite similar peat quality at depths accumulated before dam construction at Wylde Lake peatland was obvious in peats

5

Deleted: Regarding Wylde Lake peatland's vegetation, its shoreline to the Luther Lake reservoir is nowadays dominated by a floating mat of cattail (*Typha latifolia* and *T. angustifolia*). Adjacent to that, the northern periphery of the peatland, which may be regarded as an open bog – poor fen transition area, is characterized by a belt of pronounced hummocks, which rise up to 0.8 m above narrow, small hollows. Those hummocks are several meters long and accommodate a dense cover of gigantically grown *Myrica gale* individuals, which suppress growth of underlying *Sphagnum* mosses. The vegetation of the adjacent more pristine open bog south is composed of dominant *Sphagnum* mosses (e.g., *S. magellanicum, S. cuspidatum, S. fuscum*), shrubs (e.g., *Myrica gale, Yaccinium spp., Chamaedaphne calyculata, Rhododendron groenlandicum, Kalmia polifolia, Antorneda polifolia, agraminois (Cyperaceae, e.g., Eriophorum spp., Carex spp.)* and few scattered herbaceous plants (e.g. *Sarracenia purpurea, Maianthemum trifolium*); the vegetation structure is rather typical of bog vegetation. The micro topography is a typical bog's mosaic of hummocks, hollows and lawn-like grass carpets. The treed bog area to the south is dominated by black spruce (*Picea mariana*), tamarack (*Larix laricina*), pine trees (*Pinus spp.*) and individuals of swamp birches (*Betula punila*).

	Deleted: in the hummocks
	Deleted: ;
	Deleted: s
Ì	Deleted: were located in
Ì	Deleted: ;
	Deleted: s
	Deleted: was located in
Ì	Deleted: the treed bog areadifferent.

Deleted: At site 4, shrubs were the dominant PFT, site 3 and 2 had a mixture of PFTs including *Sphagnum* mosses and aerenchymatous graminoids as dominant PFTs. Site 1 accommodated a mixture of

a mixture of PF1s including *Sphagnum* mosses and actenchymatous graminoids as dominant PFTs. Site 1 accommodated a mixture of PFTs, including *Sphagnum* mosses, trees, graminoid species as well as shrubs.

Deleted: summary

Deleted: site characteristics

2.2 Determination of organic matter quality of peat and pore-water

5

Peat samples were taken in July, 2014, in depths of 5, 10 and 20 cm below the living *Sphagnum* layer by manual cutting. Peat from 75 cm depth was taken with a Russian peat corer. Peat was filled in jars avoiding any headspace and closed air-tight to maintain anoxic conditions as far as possible during transport to the laboratory.

To collect in-situ pore-water, suction samplers (Macro Rhizons, Eijkelkamp, Giesbeck, The Netherlands; pore size $\sim 0.2 \,\mu$ m) were inserted into the peat at 5, 10 and 20 cm depth. Sampling was done by applying vacuum and collecting water with syringes, covering syringes with aluminum foil and peat to avoid exposure to light. Pore-water from 75 cm depth was pumped

10 from 75 cm deep piezometers that were emptied one day prior to sampling to ensure sampling of fresh pore-water. Samples from piezometers were filtered using Macro Rhizons in the laboratory to ensure similar treatment of pore-water of all depths. All samples were taken and analyzed as three replicates.

Prior to Fourier-transform infrared spectroscopic (FTIR) analysis, oven-dried (70 ° C) bulk peat samples were ground with a ball mill. Pore-water samples were oven-dried (70 ° C) until all water was evaporated; afterwards 2 mg of the remaining

- 15 organic matter were scraped off the sample bottles and ground in a mortar with 200 mg of potassium bromide (KBr) and pressed to pellets for analysis. We recorded spectra on an FT-IR Spectrometer (Varian 660, Palo Alto, USA) over a scan range of 4000 to 650 cm⁻¹ with a resolution of 2 cm⁻¹ and 32 scans per sample. A KBr background was subtracted from the spectra and spectra were baseline corrected. We identified spectral peaks (average location +/- 30 cm⁻¹) and related them to functional moieties as described in Niemeyer et al. (1992). As absorbance values do not give quantitative information on absolute values
- 20 of functional groups, we related peaks of around 1620 cm⁻¹ to 1610 cm⁻¹ (aromatic C=C compounds/aromatic moieties) to polysaccharide peaks at 1170 cm⁻¹ to 950 cm⁻¹ wavenumbers (Niemeyer et al., 1992). A relative increase in ratios thus indicates a relative decrease in the labile polysaccharide moieties and thus an increase in the degree of decomposition in regard of a residual enrichment of refractory aromatics (Broder et al., 2012).
- Pore-water samples were analyzed by absorption spectroscopy in the ultra violet and visible range (UV-VIS-spectroscopy; 25 Varian UV 1006 M005 spectrometer, Palo Alto, USA). We recorded UV-VIS spectra over a range of 200 to 800 nm with a resolution of 0.5 nm using a 1 cm quartz cuvette. Prior to measurement, a blank spectrum of ultrapure water was recorded and subtracted from each sample. We additionally recorded fluorescence properties of dissolved organic matter (DOM) on a fluorescence spectrometer (Varian Cary Eclipse, Palo Alto, USA) at a scan rate of 600 nm/min. Excitation wavelengths (ex) were 240 to 450 nm in 5 nm steps, emission wavelengths (em) 300 to 600 nm in 2 nm steps to obtain excitation-emission-
- 30 matrices (EEMs). If necessary, UV-VIS absorption at 254 nm was adjusted to a range of 0.1 to 0.3 by dilution with ultrapure water in order to be able to correct for inner filter effects in fluorescence spectroscopy. Repeated blanks were run to ensure cleanliness of the cuvette. Raman spectra of a blank were recorded each day to check analytical drift and to normalize fluorescence to Raman units (Murphy et al., 2010).

6

Moved up [1]: Moreover, thicker layers of readily decomposable fibric and hemic peat were found up to a depth of approx. 48 cm at sites 1 and 2 whereas at site 3 and 4, strongly decomposed sapric peat was already found below depths of 37 cm (site 3) and 29 cm (site 4) respectively.

Deleted: As indicated from altered stochiometric element ratios (Table 1) and as reported recently by Berger et al. (submitted for publication), those sites in closer proximity to the Luther Lake reservoir are affected by input of nutrients, such as N, P, K, S, Ca, Mg. This translated into a relative enrichment of N, Mg and K and very pronounced P and Ca limitation closer to the reservoir.

... [2]

Deleted: For
Deleted: p
Deleted: 2 mg of

Deleted: with

To evaluate DOM quality, we calculated commonly used indices, such as specific ultraviolet absorbance SUVA₂₅₄ (as a proxy for aromaticity, Weishaar et al., 2003) and the E2:E3 ratio (the ratio of UV absorbance at 250 nm divided by absorbance at 365 nm) providing information about molecular weight of organic matter (Peuravouri and Pihlaja, 1997) from UV-VIS data. From fluorescence data, we calculated a humification index HIX (Ohno, 2002). (see Table S3 for equations used).)

5 2.3 Measurements of environmental variables

Air temperature and photosynthetically active radiation (PAR) were recorded about 1 km south of site 1 in an open area by a HOBO U30 weather station (U30-NRC-SYS-B, Onset, Bourne, MA, USA) at a temporal resolution of 5 min. Water table depth below surface (wtd), water temperature (T_{water}) and air pressure were measured in 30-min intervals using one pressure transducer (Solinst Levelogger Edge) in a monitoring well at each site, corrected for barometric pressure (Barologger Gold at

10 site 2; Solinst Ltd., Georgetown, Canada). On each day of closed chamber measurements, an extra PAR sensor (Smart Sensor, Onset; Part # S-LIA-M003) and an extra temperature sensor (Temperature Smart Sensor, Onset; Part # S-TMB-M0XX) recorded PAR and air temperature at a temporal resolution of 10 secs at the site were chamber measurements were being taken.

2.4 Determination of CO₂ and CH₄ fluxes

In the hollows of sites 1-4 a set of six collars for chamber measurements were established in April 2012. The collars - installed 0.1-0.15 m into the soil- were cylindrical, had a diameter of 0.4 m and a total height of 0.2 m. Through object based image

- analysis (OBIA) based on aerial imagery obtained from UAV flights, the spatial coverages of PFTs at each site were obtained (data summarized in Table 1). Accordingly, the locations for chamber measurement collars of our study sites were defined to proportionally reflect the distribution of PFTs.
- Closed chamber measurements were performed following Burger et al., (2016). Measurements were taken every 10 to 30 days
 at each site from <u>April 20th</u>, 2014 through September 22nd, 2015. In total 19 to 23 daily courses per site could be accomplished.
 Cylindrical Plexiglas chambers were used for the flux measurements: a transparent chamber to measure net ecosystem exchange (NEE), a chamber covered with reflective insolation foil for ecosystem respiration (R_{eco}). Chamber closure time was 180 secs.
- Air was circulated between the chamber and a trace gas analyzer (Ultraportable Greenhouse Gas Analyzer 915-001, Los Gatos Research Inc., Mountain View, USA) through 2 mm inner diameter polyethylene tubing, recording trace gas concentrations of CO₂ and CH₄ at a temporal resolution of 1 sec. According to the manufacturer, the reproducibility of CH₄ and CO₂ is < 2 ppb and < 300 ppb, respectively. The analyzer was factory-calibrated immediately before the campaign. Stability of the calibration was checked repeatedly during summer of 2014. In January and July 2015, the analyzer was again re-calibrated. If CH₄ concentrations increased sharply within the first 60 secs of the measurement due to CH₄ bubble release caused by the

30 positioning of the chamber, the measurement was discarded and repeated.

7

Deleted: To preserve the soil and plant structure, and to avoid any bias in gas fluxes during the chamber placement, boardwalks were installed.

Deleted: April,

During each measurement day, each collar was monitored several times with the transparent and dark chamber at different times (typically between 5 am and 8 pm) and different PAR levels (typically 5 to 2000 μ mol m⁻² s⁻¹) throughout the day. Unfortunately, due to the remoteness of our study site, measurements at night were not possible.

5 Gas fluxes were determined by Eq. 1:

$$F_{chamber} = \frac{\Delta c}{\Delta t \ A} \cdot \frac{P \ V}{R \ T}$$

based on the changes of concentration over time inside the chamber, applying the ideal gas law with the ideal gas constant R,

10 and correcting for atmospheric pressure *P* and temperature inside the chamber *T*. The chamber volume *V* and basal area *A* were calculated from the chamber's physical dimensions, taking into account each collar's vegetation volume, determined in May, July and October 2014 as well as in April and August 2015 and extrapolated for the other campaigns. The concentration change over time was derived from the slope of a linear regression of concentration vs. time. The first 40 secs after chamber deployment were discarded to account for the analyzer's response time. If the slope was not significantly different from 0 15 (tested with an F-test, $\alpha = 0.05$), the flux was set to zero.

An empirical description of the measured NEE fluxes of each site was accomplished with the help of a hyperbolic light response model (Owen et al., 2007). The non-linear least squares fit of the data to the model was done according to Eq. 2:

$$NEE = \frac{\alpha\beta Q}{\alpha\beta} + y$$

20

where *NEE* is in mol m⁻² s⁻¹, α is the initial slope of the light response curve (in mol CO₂ m⁻² s⁻¹ per mol photon m⁻² s⁻¹), β is the maximum NEE in mol CO₂ m⁻² s⁻¹, Q is the photosynthetic active radiation in mol photon m⁻² s⁻¹, and γ is an estimate of the average R_{eco}. Integration of NEE over the course of one day gave net daily ecosystem production (NEP). Gross primary productivity (GPP) was retrieved by subtracting R_{eco} from NEP.

25 Average CH₄ fluxes of measurement days of each site were obtained and lastly, cumulative emissions of CO₂ and CH₄ were calculated likewise according to Tilsner et al. (2003).

To determine isotopic signatures of CH_4 fluxes, we carried out additional chamber flux measurements once a month from May to September 2015 using a shrouded chamber and the Los Gatos UGGA. The chamber was closed until CH_4 concentrations reached > 10 ppm for analysis of isotopic composition, but not more than 30 min. Samples for isotopic analysis were extracted

30 from the chamber with 60 ml syringes through a polyethylene tube with a three-way stopcock on one end and filled into 40 ml crimp vials that had before been flushed with nitrogen (N₂) and sealed with rubber stoppers. To correct isotopic values of CH₄ for background isotopic signature in the chamber, we collected six air samples at each site on every sampling day. Analysis was carried out as outlined for dissolved gases (see below).

2.5 Sampling of gases and dissolved gases in the peat

Concentrations of CH₄ and dissolved inorganic carbon (DIC/ Σ CO₂) along peat profiles were analyzed in 5, 15, 25, 35, 45 and 55 cm depth with three replicates at each site using diffusive equilibration samplers made of permeable silicon₂ tubes (Kammann et al. 2001). Using three-way valves, samples were taken with 10 ml syringes every two to three weeks from June 2014, to September 2015. Samples were stored overnight at 5 ° C and analyzed the next day.

- To determine temporal dynamics of isotopic signatures of CH₄ and CO₂ in the peat, we installed a separate set of silicon<u>e</u> tubes in 5, 15, 25 and 35 cm depth with three replicates each per site. <u>Silicone</u> tubes for isotope sampling had an inner diameter of <u>1</u> or <u>0.5 cm</u>, corresponding to a volume of 20 or <u>5 mL</u>. The samplers with a volume of <u>20 mL</u> were installed in <u>5 cm</u> depth and the smaller samplers below, as close to surface larger volumes of samples were necessary in order to obtain sufficiently high concentrations (2.5 < x < 2000 ppm) for isotope analysis.
- Tightness of all samplers was confirmed prior to installation. Equilibrium of gases such as N₂O, CH₄, and CO₂ at the silicon membrane has been shown to adjust with hours to days and isotopic fractionation can be expected to be negligible <u>(Nielsen et al., 1997; Panikov et al., 2007; Pack et al., 2015)</u>. All samplers were installed one month prior to the first sampling and samples were taken once a month from May 2015 to September 2015 with 10 and 60 ml syringes and filled in 10 respectively.
- 15 <u>40 ml crimp vials that were before flushed with N₂ and sealed with rubber stoppers. Silicone samplers were refilled with N₂ to avoid oxygen entering the system.</u>

To obtain high resolution depth profiles of concentration and isotopic signatures of CH_4 and DIC, pore-water peepers of 60 cm length and a 1 cm resolution (Hesslein 1976) were inserted on three occasions in June, July and September 2015 and allowed to equilibrate for four weeks prior to sampling. As results of pore-water peepers generally confirmed the data of the silicon<u>c</u> samplers, results are not presented here but described in the supporting information (see Fig. S6).

2.6 Analyses of CO2 and CH4 concentrations and δ¹³C-CO2 and δ¹³C-CH4-values

Gaseous CO_2 and CH_4 concentrations were analyzed with a gas chromatograph (SRI 8610 C, SRI Instruments, Torrance, US) equipped with a Flame Ionization Detector (FID) and a Methanizer. Samples from pore-water peepers were analyzed by measuring the headspace concentration in the vials.

- 25 Rations of δ¹³C of CO₂ and CH₄ were determined by Cavity Ringdown Spectroscopy (CRDS; Picarro G2201-*i*, Picarro Inc., Santa Clara, US), simultaneously determining ¹³C isotopic composition of CO₂ and CH₄ with a precision of <0.16 ‰ for δ¹³C-CO₂ and <1.15 ‰ for δ¹³C-CH₄. The analyzer was calibrated before each measurement with two working standards of CO₂ (1000 ppm, -31.07 ‰) and CH₄ (1000 ppm, -42.48 ‰). Standard deviation for δ¹³C-CO₂ was below 2 ‰ and below 4 ‰ for δ¹³C-CH₄. Isotopic signatures were expressed in the δ-notation in ‰ versus VPDB-Standard according to Eq. 3:
- 30

5

 $\delta^{13}C = (R_{sample}/R_{standard} - 1) \cdot 1000 \ [\%]$

9

Deleted: These latter silicon
Deleted: 0.5 or
Deleted: and
Deleted: ml in 5
Deleted: depth
Deleted: and 5
Deleted: order to
Deleted: Samples were taken once a month from May 2015 to September 2015 with 10 and 60 ml syringes and filled in 10 respectively 40 ml crimp vials that were before flushed with N_2 and sealed with rubber stoppers. Silicon samplers were refilled with N_2 to avoid oxygen entering the system.
Deleted: All samplers were installed one month prior to the first

Deleted: All samplers were installed one month prior to the first sampling to assure equilibration, and Deleted: t

Deleted: 5.2

where R_{Sample} is the ¹³C/¹²C ratio of the sample and $R_{Standard}$ is the ¹³C/¹²C ratio of the standard.

As the accuracy of δ^{13} C-CO₂ values was affected by high CH₄ concentrations present in the samples, we established a correction formula to revise δ^{13} C-CO₂ values. This formula was applied for molar concentration ratios of CO₂:CH₄ between 0.3 and 1.5. Samples with CO₂:CH₄ ratios < 0.3 could not be corrected and were discarded; samples with higher ratios did not

- 5 need correction. To cross-check values of δ¹³C-CO₂, two additional standards, carbonic acid from fermentation (-26.61 ‰), natural carbonic acid (-0.19 ‰) and a mixture of both (-15.16 ‰) were measured both with CRDS and an isotope-ratio mass spectrometer (EA/TC-IRMS Nu Horizon, Hekatech/Nu Instruments, Wrexham, UK). Additionally, δ¹³C-CO₂ values had to be corrected for a storage effect. As samples were stored for several weeks, CO₂ was lost from the vials and isotopic signatures increased by 0.056 ‰ per day. There was no such effect detectable for CH₄.
- 10 Dissolved concentration of CO₂ and CH₄ were recalculated from partial pressures inside the silicon samplers applying Henry's Law according to Eq. 4:

 $c = K_H * p$

15 where *c* is the concentration in μ mol/L, *p* is the pressure in atm and *K*_H is the in-situ temperature corrected Henry-constant in mol L⁻¹ atm⁻¹ (Sander, 1999). CO₂ dissociation was considered using equilibrium constants from Stumm and Morgan (1996) to calculate the total amount of DIC.

DIC and CH₄ concentrations in samples from pore-water peepers were recalculated from gas concentrations in the headspace, applying the ideal gas law and temperature corrected Henry-constants for laboratory conditions.

20 To gain information about the dominant CH₄ production pathway, the isotope fractionation factor α_C (for 35 cm depth) was calculated according Eq. 5 after Whiticar et al. (1986):

$\underline{\alpha_{\rm C}} = (\delta^{13}\text{C-CO}_2 + 1000) / (\delta^{13}\text{C-CH}_4 + 1000).$

2.7 Statistical analysis

25 Statistics software R i386 version 3.1.0 was used to verify if observed differences in organic matter quality varied between depths and sites were statistically relevant for each indicator separately. Data was tested for normal distribution (Shapiro-Wilk-Test, $\alpha = 0.05$) and homogeneity of variance (Levene-Test, $\alpha = 0.05$). In case both requirements were met, we carried out a one-way ANOVA (Analysis of Variance) ($\alpha = 0.05$) with a post-hoc Tukey's Honest Significant Difference (HSD) test ($\alpha = 0.05$) to identify which depths or which sites differed significantly. If either normal distribution or homogeneity of variance

30 were not given, a Kruskal-Wallis test ($\alpha = 0.05$) with a multiple comparison test after Kruskal-Wallis ($\alpha = 0.05$) as post-hoc test was executed.

Using *RStudio* Version 0.99.902 as well as R i386 3.2.3 we examined whether there were significant differences in δ^{13} C values of CO₂ and CH₄, CO₂ and CH₄ concentrations and cumulative emissions between the sites. Means were compared with t-Tests

Deleted: 6

(if <u>data was</u> normally distributed) respectively Kruskal-Wallis and post hoc Wilcoxon-Mann-Whitney-Test (if <u>data was</u> not normally distributed) <u>The confidence level for the statistical tests was $\alpha = 0.05$. Normality was tested with Shapiro-Wilk-Test</u> ($\alpha = 0.05$) and homogeneity of variance was confirmed with Levene-Test ($\alpha = 0.05$). Correlations between environmental variables and fluxes, concentrations and isotopic signatures were determined with Pearson's product-moment correlation for

5 normally distributed data or with Spearman's rank correlation if data was not normally distributed. With ANOVA ($\alpha = 0.05$), the effect of categorical variables on CH₄ fluxes and δ^{13} C values was computed.

3 Results

3.1 Organic matter quality of peat and pore-water

- 10 The highest degree of bulk peat decomposition, as indicated by the highest 1618.5/1033.5 absorption ratios, was found at site 4 between 5 and 20 cm depth (p < 0.05 in 10 and 20 cm depth), indicating the highest degree of bulk peat decomposition at this site and these depths (Fig 2 (a)), whereas the 1618.5/1033.5 ratios of the sites 1-3 were not significantly different. Porewater samples' 1618.5/1033.5 ratios of site 3 were smallest between 5 and 20 cm depth as compared to all other sites (p < 0.05), indicating the lowest degree of decomposition of DOM here (Fig 2 (b)). Aromaticity as determined with *SUVA*254 (Fig
- 15 2 (c)) did not show significant differences between sites in pore-water samples (exception: site 1 and site 3 in 20 cm depth (p = 0.033), where site 1 SUVA₂₅₄ was significantly higher than site 3 SUVA₂₅₄). The degree of humification, as depicted by *HLX* (Fig 2 (d)), was significantly lowest in site 3 pore-water (5 cm site 3 and 4: p = 0.026; 10 cm site 1 and 3: p = 0.014; 20 cm site 3 and 4: p = 0.020). The slope ratio *E2:E3* (Fig 2 (e)), indicative of molecular size and aromaticity, was not significantly different at the sites 1-4.

20 3.2 Development of wtd and T_{water} during the study period

During our study period, hollow wtd showed strong seasonal fluctuations; maximum wtd (i.e. highest water table levels) throughout the study period were reached during snowmelt in spring 2014 (site 1: 6.94 cm, site 2: 4.99 cm, site 3: 16.26 cm, site 4: 23.18 cm above hollow surface), minimum wtd (i.e. lowest water table levels) were reached during the summer of 2015 (site 1: 32.5 cm, site 2: 31.75 cm, site 3: 13.34, site 4: 19.11 cm below hollow surface). The sites 1 to 4 all showed similar
courses of wtd, however, at site 3 and site 4 water levels were generally higher as compared to site 1 and 2 (p < 0.05). The range between maximum and minimum wtd at all sites was overall similar (site 1: ~39.5 cm, site 2: ~36.7 cm, site 3: ~30 cm (logger failure when water levels were lowest), site 4: ~42.3 cm). T_{water} varied between ~2 °C in winter and ~16 °C in summer. Detailed courses of wtd and T_{water} are presented in panels (a) and (b) of Fig. 3.

Dele	eted: data
Dele	eted: at a
Dele	eted: confidence level
Dele	eted: of

Deleted:
Deleted: The
Deleted:

... [4]

Deleted: Deleted: Deleted: the top panels of the figures 3 and

Deleted:



3.3 Fluxes of CO₂ and CH₄ at the soil/atmosphere interface, concentrations of CH₄ and DIC along soil profiles during the study period

<u>Fluxes of CH₄ and CO₂ (Fig. 3, panels (c) - (f)) showed strong annual variability. CH₄ fluxes (Fig. 3 (c)) were positive (fluxes from soil to atmosphere) throughout the entire study period; greatest fluxes occurred during the growing season, minute fluxes fluxes occurred during the growing season, minute fluxes fluxes occurred during the growing season for the fluxes fluxes occurred during the growing season fluxes fluxes fluxes fluxes fluxes occurred during the growing season fluxes fl</u>

- 5 were detected during the dormant season. In general, site 3 CH_4 fluxes plotted above the other sites' fluxes, whereas site 4 fluxes plotted below the other sites' fluxes (except for August 16th, 2015 when a mean flux of 0.76 ± 0.58 g CH_4 m⁻² d⁻¹ was detected, exceeding the fluxes measured at all other sites. During the entire study period (April 2014 through September 2015), hollows of site 3 released significantly (p < 0.001) more methane (61.4 ± 32 g CH_4 m⁻²) than the sites 1 (41.8 ± 25.4 g CH_4 m⁻²), 2 (44.6 ± 13.7 g CH_4 m⁻²), and 4 (46.1 ± 35.2 g CH_4 m⁻²); see also Fig. S5. Annual CH_4 emissions from May 2014 to May
- 10 2015 were 22.18 \pm 8.96 at site 1, 30.66 \pm 7.63 at site 2, 39.86 \pm 16.81 at site 3, and 12.53 \pm 11.38 g CH₄ m⁻² at site 4; thus emissions at site 3 were significantly (p<0.05) higher than at site 4, but CH₄ emission at sites 3 and 4 did not differ significantly from sites 1 and 2 emissions.

Fluxes of CO₂ (Fig. 3 (d), (e), (f)) showed a strong seasonal variability, with highest photosynthetic uptake and highest ecosystem respiration in summer and lowest fluxes in the dormant season. Site 3 NEP plotted below all other sites' fluxes,

- 15 indicating most CO_2 net uptake, whereas site 4 NEP apparently plotted above all other sites' fluxes, indicating less net uptake of CO_2 if not a net emission of CO_2 (Fig. 3 (d)). Regarding R_{geo} (Fig. 3 (e)), paztterns were similar at all sites. Regarding GPP (Fig. 3 (f)), site 3 plotted below all other sites, indicating highest photosynthetic uptake here, whereas site 4 mostly plotted above the other sites, indicating smallest productivity. During the study period, the cumulative NEP of hollows of site 1 was 1552 ± 652 g CO_2 m⁻², site 2 accumulated 1637 ± 184 g CO_2 m⁻², site 3 accumulated 2260 ± 480 g CO_2 m⁻² and site 4
- 20 accumulated 1093 ± 794 g CO₂ m⁻² (see Fig. S4). Thus, between May 19th, 2014 and September 23rd, 2015 site 4 accumulated significantly less CO₂ as compared with the other three sites (p < 0.001), while there were no statistically significant differences in terms of CO₂ uptake for the sites 1, 2 and 3. Annual cumulative NEP from May 2014 to May 2015 was -896 ± 151 g CO₂ m⁻² at site 1, -1023 ± 615 g CO₂ m⁻² at site 2, -1282 ± 361 g CO₂ m⁻² at site 3, while site 4 released 135 ± 281 g CO₂ m⁻². Annual cumulative NEP of the sites 1, 2 and 3 was significantly lower than the site 4 NEP (p < 0.05).</p>
- 25 Interestingly, site 4 CH₄, NEP and GPP fluxes differed notably between the growing seasons of 2014 and 2015. This was particularly caused by two plots, which in 2015 dramatically increased productivity and CH₄ emissions as compared to the previous year (data not shown).

Concentration of CH_4 along depth profiles (Fig. 4, top panels) of all sites varied strongly throughout the year; they generally increased during the growing season, reached maximum values in the winter season 2014/2015 and comparably decreased

30 during snowmelt in spring. A similar pattern was observed for DIC concentrations along depth profiles (Fig. 4, lower panels). Maximum DIC concentrations were observed below 20 cm depth in autumn 2014 and winter 2014/2015 and minimum concentrations were observed during snowmelt in March and April 2015. DIC concentrations at site 4 at all depths were overall lower and significantly decreased (p < 0.05) in comparison to all other sites from February 23rd through April 4th, 2015;</p>

12

Moved (insertion)	[2]
Deleted: CO ₂ and	
Deleted:	
Deleted: and 4	
Deleted: a	

Deleted: Overall, NEP, GPP, and R_{eco} at all sites were highest during the growing seasons. Also, CH₄ fluxes were highest during the growing season, despite comparably low concentrations of dissolved CH₄ in the investigated upper 50 cm of the peat profile, suggesting that exchange of gases at the peat/atmosphere interface and concentrations in the uppermost peat were decoupled.

Deleted: Accordingly, NEP, GPP, R_{eco} and CH_4 fluxes at almost all sites were positively correlated with T_{water} (p < 0.05), however, negatively correlated with wid (lowest water tables co-occurred with highest fluxes). As suggested from visual inspection, R_{eco} , GPP and CH_4 fluxes of the sites 1, 2 and 3 were in most cases negatively correlated with DIC and CH_4 concentrations along soil profiles, i.e. increasing CO_3 and CH_4 fluxes were correlated with decreasing DIC and CH_4 concentrations. Only at site 4 greater DIC and CO_3 concentrations along peat profiles coincided with higher effluxes of CO_3 and CH_4 . Statistical results are summarized in the Table $\{\dots, [5], Deleted: 3$ and

moreover, site 4 DIC concentrations were significantly (p < 0.05) lower than site 3 DIC concentrations on August 6th, 2014 and between April 19th through July 18th, 2015. <u>Concentrations in the uppermost depths of both CO₂ and CH₄ were strongly affected by fluctuations of wtd, with strong decreases upon water table decline and vice versa (see table S4 for statistical results).</u>

5

20

3.4 Temporal and spatial variability of δ^{13} C-CO₂ and δ^{13} C-CH₄ -values in peat pore-gas profiles during the growing season in 2015

Values of δ¹³C of the sampled CH₄ in the peat ranged from -78.74 to -26.77 ‰, δ¹³C signatures of CO₂ ranged from -25.81 to
 +4.03 ‰ (see Fig. 5). Highest δ¹³C-CH₄ and CO₂ values were measured at site 1 in 5 respectively 35 cm depth in September.
 Lowest δ¹³C-CH₄ and CO₂ values were detected at site 1 in 15 cm depth in June and at site 2 in 15 cm depth in August respectively.

Overall, δ^{13} C-CH₄ values showed an increasing trend with time from June to August in all depths. Average signatures in 5 to 35 cm depth differed significantly between sampling dates at all sites except between August and September (p < 0.05).

15 Concomitant to a decline in water table levels in August and September, δ^{13} C-CH₄ signatures shifted to less negative values in the upper 5 cm at sites 1 to 3; this shift was most distinctive at site 1 and least distinctive at site 3. At site 4, such shift occurred at 15 cm depth.

For δ^{13} C-CO₂ signatures, significant differences between some sampling dates were found at sites 1, 2 and 4 for average values in 5 to 35 cm depth. At sites 1 and 2, signatures in August and September were higher than in June and July, paralleling the trend in δ^{13} C-CH₄. At site 3 and 4, such significant shifts could not be observed.

- No significant differences between depths could be found for either δ^{13} C-CH₄ or δ^{13} C-CO₂ signatures at any site. Anyway, at sites 1 and 2, δ^{13} C-CH₄ signatures apparently increased with depth in June and July, no trend was observable at sites 3 and 4. In August and September, δ^{13} C-CH₄ signatures seemed to decrease with depth except for site 4. Values of δ^{13} C of CO₂ increased with depth except at site 1 in July and at site 2 in July and August.
- 25 Mean signatures of δ^{13} C-CH₄ at site 4 (-57.81 ±7.03 ‰) differed significantly from those at the other sites (site 1: -61.48 ±10.71 ‰, site 2: -60.28 ±5.57 ‰, site 3: -62.30 ±5.54 ‰) for the whole sampling period (p < 0.01, p < 0.05, p < 0.001). δ^{13} C-CO₂ signatures at site 3 were significantly higher than at the other sites in July (p < 0.05, p < 0.01, p < 0.01). Overall, highest mean values were found at site 1 (-12.05 ± 8.23 ‰) whereas site 4 revealed lowest δ^{13} C-CO₂ signatures (-15.85 ± 3.61 ‰).
- 30 Isotopic composition of CH₄ and CO₂ as determined from porewater peepers confirmed results obtained from the silicon gas samplers. Data is presented in the Fig. S5 in the supplemental information.

13

Deleted:

Moved up [2]: Fluxes of CO₂ and CH₄ (Fig. 3 and 4) showed a strong annual variability. Overall, NEP, GPP, and R_{eco} at all sites were highest during the growing seasons. Also, CH₄ fluxes were highest during the growing season, despite comparably low concentrations of dissolved CH₄ in the investigated upper 50 cm of the peat profile, suggesting that exchange of gases at the peat/atmosphere interface and concentrations in the uppermost peat were decoupled. -

[... [6]

<u>Fractionation factors $\alpha_{\rm C}$ to characterize methanogenic pathways (according to Whiticar et al. (1986))</u> were calculated for water saturated, presumably anoxic conditions at <u>-35 cm depth</u> only (Table 2), as frequent or prevailing unsaturated conditions above this depth would rather favor methanotrophy. <u>Given that $\alpha_{\rm C}$ values between 1.04 and 1.055 indicate the prevalence of the</u> acetoclastic methane production pathway, whereas $\alpha_{\rm C}$ values higher than 1.065 support a shift towards the hydrogenotrophic

5 pathway, the acetoclastic pathway was apparently favored in July and August at the sites 1 and 2, in August at site 3 and in July, August and September at site 4, indication shift towards a higher contribution of the hydrogenotrophic pathway was only observed in June and September at site 1, and in June at site 2

$3.5 \, \delta^{13}$ C signatures of emitted CH₄ during summer 2015

Values of δ¹³C of emitted CH₄ ranged from -81.87 ± 3.81 and -55.61 ± 1.20 ‰ (see Fig. 6, panel (a) to (d)). Thereby, δ¹³CCH₄ signatures increased from July to August and slightly decreased again in September. This pattern was thus related to the course of the wtd. Significant differences were only found at sites 3 and 4 between July and August (p < 0.01, p < 0.05), however, from visual inspection of the panels (a) – (d) of Fig. 6 δ¹³C- CH₄ values seemed to increase between May and September at site 3, while they appeared to decrease at site 4, with very distinct values in August. There was no such pattern observable at sites 1 and 2. Summing up δ¹³C- CH₄ signatures from all sites, isotopic signatures in July differed significantly from those in August and September (p < 0.05). In September, isotopic signatures of the CH₄ flux at site 2 differed significantly from those at the other sites (p < 0.05).

Comparing isotopic signatures of dissolved CH4 in the peat and emitted CH4, plant mediated transport was the dominant CH_4 emission pathway during the summer 2015 at all sites and all sampling dates according to Hornibrook (2009) (Fig. 6 (e)).

20 4 Discussion

As expected from our studied transect ranging from a strongly altered (site 4) to an only slighty altered site (site 1) in terms of nutrient supply, hydrological conditions, and coverage of PFTs, we observed pronounced differences in gas fluxes and peat guality. On the other hand, the dominant CH_4 emission pathway was plant-mediated transport at any site. We are aware that observed effects of anthropogenic impact are much more difficult to constrain in an in-situ study as ours, compared to well

25 defined ecosystem manipulation studies. Nevertheless, our results support a clear and a_obvious<u>interplay of processes</u>, fluxes, and vegetation that can be related to the observed impacts of nutrient enrichment and altered hydrology, as <u>discussed in the</u> following paragraphs.

4.1 Long-term insights into carbon cycling at the sites

Long-term plant community changes were recently shown to affect peatland organic matter composition (Hodgkins et al.,
 2014), while such an effect was not identified in a short-term study (Robroek et al., 2015). Along our transect of study sites, affected by consequences of a construction of a dam in 1954, we observed the highest degree of bulk peat decomposition in

14

Deleted: Lowest δ^{13} C values were measured in May at site 3 (Fig. 6 (c)). Highest values were observed in August at site 2 (Fig. 6 (b)).

Deleted: The distinct patterns of August may have to be interpreted with caution due to limitations in our sampling technique, in particular when water tables were lowest (see discussion Sect. 4.3).

Deleted: Along our studied gradient of four sites it became evident that the peatland was exposed to nutrient infiltration from the water reservoir and thus elevated nutrient concentrations occurred in vicinity to the water reservoir, as reported elsewhere (Berger et al., submitted for publication). This was clearly visible in C/N, C/Mg and CK ratios, decreasing in the order of site 4 < 3 < 2 < 1, while C/P, N/P and C/Ca ratios increased in vicinity to the reservoir, indicating a limitation of P and Ca, which is typical of eutrophic peatlands (see Berger et al., submitted for publication and Table 1). Moreove pronounced differences in terms of vegetation were evident: While at site 4 the predominant PFT were shrubs and *Sphagnum* mosses were in retreat, sites 2 and 3 were dominated by a mixture of Sphagnum mosses and graminoids, and site 1 accommodated a mixture of PFTs. such as Sphagnum mosses, graminoids, dwarf shrubs and few trees. In addition to those characteristics, further differences in terms of water table regimes were observed, i.e. sites 3 and 4 were apparently wheter that sites 1 and 2. Moreover, there were pronounced differences in gas fluxes and peat quality, while the dominant CH_4 emission pathway was plant-mediated transport at any site. An interpretation of the interplay of processes, fluxes, and vegetation d on our results will be discussed in the following.

Deleted: Peat quality

Deleted:, affected by consequences of a construction of a dam in 19...,

the upper peat layers of shrub dominated site 4, which was located in closest vicinity to the water reservoir and which was the most altered one among our four sites (Fig 2 (a)). Our initial hypothesis 1 that peripheral sites feature accelerated C cycling, reflected in more decomposed peat, could thus only partly be verified: the fact that we did not find a gradual decrease in terms of degree of bulk peat decomposition with increasing distance from the reservoir, but observed significant differences only for

- 5 site 4, suggests that the observed differences could also be primarily induced by the shift to a predominance of shrubs. Shrubs contain more woody parts and thus have higher lignin contents and more phenolic groups than graminoids or mosses and they are also more productive than mosses and graminoids (Bragazza et al., 2007). In recent studies, an increasing ericaceous shrub cover was associated with increasing polyphenol content in plant litter and pore-water, as well as increasing phenol oxidase in litter of ericaceous shrubs. Also, a higher release of labile C from vascular plant roots was observed. These changes, however
- 10 along an altitudinal gradient, were accompanied by a decreasing *Sphagnum* productivity (Bragazza et al., 2013; Bragazza et al., 2015). Even though at site 4 we primarily dealt with eutrophication, rather than warming, there might be similar processes explaining our observations: Shrubs outcompete *Sphagnum* mosses after long term-nutrient infiltration and a reduced recalcitrance of the peat arising from shrub litter may result in a reduced C storage, i.e. peat accumulation (Turetsky et al., 2012; Larmola et al., 2013; Ward et al., 2013). This was further suggested by the lowest observed CO₂ uptake and lowest DIC concentrations along peat profiles throughout the study period at that particular site.
- At the graminoid-moss dominated site 3, pore-water DOM quality indices revealed a significantly lower share of aromatic compounds, and thus suggested a lower degree of humification and comparably increased molecular weight at that site (Fig 2 (c)-(e)). This more labile nature of dissolved organic matter compared to otherwise similar bulk peat quality suggested either an input from the vegetation (Robroek et al., 2015) or some inflow of water and solutes from the nearby reservoir. Given that
- 20 the site 4 pore-water DOM characteristics differed strongly from those at site 3, as did predominant PFTs, the distinctive features of the site 3 pore-water were probably also induced by the vegetation. However, the fact that the vegetational composition of site 3 and site 2 were rather similar, whereas the pore-water DOM quality was again significantly different, suggested that DOM properties were likely affect by vegetation, i.e. photosynthetic productivity and concomitantly higher input of labile compounds, as well as inflow of DOM from the reservoir.
- 25 The nature of our results does of course not allow for an unambiguous conclusion in terms of whether it is the vicinity to the reservoir or the plant community composition, which drives carbon cycling and peat accumulation at the sites. However, since peatland plant community compositions are known to be remarkably stable over time but experience changes in relative abundances (Rydin and Barber, 2001, Bragazza et al., 2006), we suggest that it was probably the vicinity to the reservoir that shaped the plant community composition at the sites over time, whereas the plant community actually drives carbon cycling.

30 4.2 Seasonal development of carbon fluxes

Different PFTs were recently shown to have a strong impact on peatland ecosystem CO₂ fluxes (Ward et al., 2013; Kuiper et al., 2014). This could be confirmed by our results: Our shrub dominated, strongly altered site 4 showed the lowest cumulative CO₂ uptake, whereas, at our graminoid-moss dominated sites 3 and 2, and at the least altered moss dominated site 1, very high

15

Deleted: The FTIR ratio derived peat quality index revealed the highest degree of bulk peat decomposition in the upper peat layers of shrub dominated site 4 (Fig 2 (a)), while there were no significant differences in bulk peat decomposition degree as far as the other sites were concerned. On the other hand, in the upper depths of graminoid-moss dominated site 3, pore-water DOM quality indicators mostly revealed a significantly lower share of aromatic comparably increased molecular weight at that site (Fig 2 (c)-(e)). This more labile nature of dissolved organic matter compared to otherwise similar bulk peat quality suggested some inflow of water and solutes, presumably from the reservoir, delivering labile matter to fuel decomposition.

FTIR ratios of our study compare well with other studies (e.g. Blodau and Siems, 2010; Biester et al., 2014), as well as ranges of UV-vis and fluorescence spectroscopy based indices (see Table S7). The degree of humification (HIX) was high at Wylde Lake peatland complex but not as high as at poorly drained thermokarst wetland sites, moderately well drained and well drained sites in central Alaska, USA (Wickland et al., 2007).

Deleted: FTIR ratios of our study compare well with other studies (e.g. Blodau and Siems, 2010; Biester et al., 2014), as well as ranges of UV-vis and fluorescence spectroscopy based indices (see Table S7). The degree of humification (HIX) was high at Wylde Lake peatland complex but not as high as at poorly drained thermokarst wetland sites, moderately well drained and well drained sites in central Alaska, USA (Wickland et al., 2007).

Deleted: water table regimes and

Deleted: Water tables and water temperatures followed a clear seasonal pattern. During the growing seasons, water tables notably dropped and temperatures increased as expected, while during the winter season and snowmelt period water levels were highest and temperatures were lowest. Among the four sites, water levels tended to be on average higher towards the shoreline of the water reservoir (sites 3 and 4). The hollows of these sites thus do not only reflect conditions. - CO_2 uptake rates were observed. The CO_2 uptake rates of our sites 1, 2 and 3 exceeded reported CO_2 uptake rates of bogs by far; for instance Teklemariam et al. (2010) reported that net ecosystem exchange of the ombrotrophic, continental Mer Bleue bog ranged from -140 to -20 g C m⁻² while hollow CO_2 uptake rates of our study were notably higher, and were rather comparable to uptake rates reported for fens (Lund et al., 2010). These latter values also compare well with the surface peat

- 5 accumulation rates of ~200 to ~300 g C m² observed at our site (Berger et al. (2017)). Net CO₂ exchange of shrub dominated site 4 was significantly lower compared to the other sites fluxes, however, a strong inter-annual variability was apparent. In the light of the strong alterations in terms of vegetation cover and the most decomposed surface peat at shrub dominated site 4, our findings from an in-situ transect support earlier findings of a reduced net CO₂ exchange and a concomitantly promoted vascular plant community in a controlled long-term fertilization experiment at the Mer Bleue bog (Bubier et al., 2007).
- Partitioning of NEP into R_{eco} and GPP further illustrated the observed differences in CO₂ fluxes between sites. While R_{eco} of all sites was in a comparable range, differences were predominantly driven by GPP. At shrub dominated site 4 during the growing season of 2014 photosynthetic uptake was clearly decreased (maximum GPP of only -18.41 ± 1.54 g CO₂ m⁻² d⁻¹) as compared to the other sites but during the growing season of 2015, maximum GPP increased up to -26.34 ± 5.1 g CO₂ m⁻² d⁻¹, which was well comparable to other sites' GPP, indicating a strong inter-annual variability in terms of photosynthetic activity
 at our shrub dominated site 4.
- With regard to CH_4 emissions, the graminoid-moss dominated site 3 exceeded the other three sites by on average 30 %. In existing studies, greatest emissions were **similarly** found in wetter habitats dominated by graminoids (Levy et al., 2012, Gray et al. 2013). Given that CH_4 emissions of site 2 were significantly smaller than those from site 3, even though the two sites feature a very similar graminoid-moss dominated vegetation cover, the differences in CH_4 fluxes could either be attributed to
- i) the wetter conditions at site 3 or ii) a greater nutrient supply to site 3, stimulating greater CH₄ production and emissions as observed earlier (Eriksson et al., 2010) or iii) a mixture of both effects. Interestingly, shrub dominated site 4, which experienced similar water table fluctuations like site 3, but featured a notably different vegetation cover, emitted CH₄ in a similar range as the graminoid-moss dominated site 2 and mixture of vegetation site 1. High methane production due to input of labile organic matter nearby the reservoir was probably outweighed by lower methane transport and therefore emission due to a lower
 graminoid cover.

In our study NEP, GPP and CH_4 emissions were negatively correlated with CH_4 and DIC concentrations in the uppermost 50 cm of the profiles at the <u>mixture-of-PFTs site L</u> and at the <u>graminoid-moss dominated sites</u> 2 and 3. Such a decoupling of CO_2 and CH_4 fluxes from pools in the peat was already observed in previous studies: Graminoids are known to be important facilitators of CH_4 emissions because they can transport CH_4 from deeper, water-saturated layers of the peat into the

30 atmosphere via aerenchymatous tissue and bypass the zone of CH₄ oxidation (Shannon and White, 1994; Marushchak et al., 2016). Moreover, they supply exudates via their roots, stimulating microbial activity and accordingly methanogenesis (Bubier et al., 1995). Through their deeper rooting system, graminoids may thus have connected the CH₄ pools of deeper layers below our studied profile, i.e. below 50 cm depth, to fuel the observed surface fluxes. The decreasing concentrations of CH₄ in near surface layers due to a decrease in water table levels and partial aeration did thus not translate in lower fluxes, a similar effect

16

Deleted: As observed for wtd and T_{water} , NEP, R_{weo} , GPP and CH_4 fluxes followed seasonal patterns as well (see Fig. 3 and 4). Over the entire study period (April 2014 through September 2015) cumulative NEP increased in the order of site 4 < 1 < 2 < 3, meaning that the two graminoid dominated sites accumulated the highest amount of CO₂, in the case of site 3 his was also statistically significant. In contradiction to the first part of our hypothesis 2 (greatest CO₂ uptake at the shrub dominated and mostly fertilized site) our wet and shrub dominated site 4 showed the lowest cumulative CO₂ uptake. In previous studies, a greatest decrease in the CO₂ sink-strength was observed when graminoids were dominant (Larmola et al., 2013; Ward et al., 2013; Kuiper et al., 2014), while in our study site 3, where Sphagnum mosses and graminoids were the dominant PTT and where wid was comparable to shrub dominated site, the highest CO₂ uptake was observed. Moreover, the CO₂ uptake rates of all our sites exceeded reported CO₂ uptake rates of bogs by far, irrespective of predominant PTTs, and were comparable to uptake rates reported for fens (Lund et al., 2010), which can be well related to increased nutrient availability as discussed in Berger et al., (submitted for publication).

Deleted: further were predominantly driven bythis was particularly obvious at , asPartitioning of NEP into R_{so} and GPP illustrated the observed differences in CO₂ fluxes between sites. Our wet and graminoid-moss dominated site 3 reached great rates in GPP of up to -26.65 g CO₂ m² d⁻¹, while R_{co} never exceed rates of 22.07 g CO₂ m² d⁻¹. The maximum GPP of the comparably drier, graminoid-moss dominated site 2 (-19.79 g CO₂ m² d⁻¹) also remained well above maximum R_{eco} here (15.49 g CO₂ m² d⁻¹.) In contrast, R_{eco} of site 1 with a mixture of PFTs but comparably dry, as well as of wet and shrub dominated site 2 and 3 (maximum CO₂ uptake rates of -24.22 g CO₂ m² d⁻¹). Thus, rather an increase in R_{eco} than a decrease in GPP, accounted for the differences in NEP between our study sites.

Deleted: Cumulative CH₄ release increased in the order of site 1 < 2 < 4 < 3, partly confirming that graminoid sites show highest CH₄ emission (hypothesis 2). In existing studies, greatest emissions were typically found in wetter habitats dominated by graminoids (Levy et al., 2012, Gray et al. 2013).

Deleted: i Deleted: sites

Deleted: 1 Deleted:

Deleted: The sites 2 and 3 were predominated by a greater share of graminoids, while all the sites 1, 2 and 3 appeared to accommodate a healthy *Sphagnum* moss community and a bog typical vegetation, even though our mixture of PFTs site 1 featured comparably less graminoids.

Deleted: surface

as suggested by Strack et al. (2006). Also, at low water tables and unsaturated conditions higher diffusivity for CO_2 can occur, leading to notably higher diffusive fluxes despite low concentrations (Knorr et al., 2008). It is striking that DIC concentrations at the shrub dominated site 4 were notably lower as compared with other sites. A reasonable explanation is a lower peat quality resulting from repeated peat oxygenation upon water table fluctuations of the reservoir, stimulating microbial decomposition

5 in the presence of deciduous shrubs (Bragazza et al., 2016), which are apparently promoted in closer vicinity to the eutrophic water reservoir. Such effect of aeration might appear contradictory, as wetter conditions would be expected near the water reservoir. However, repeated water table fluctuations driven by management of the reservoir could effectively recharge electron acceptor pools to support ongoing decomposition, as e.g. shown in water table manipulation experiments (Blodau et al., 2004; Knorr et al., 2009). Moreover, near the reservoir, also an advective redistribution and removal of CO₂ and CH₄
 10 through advective flow cannot be excluded.

4.3 Methane production, methanotrophy and pathways of CH4 emissions as inferred from stable isotopes

Distinguishing CH_4 production pathways in peatlands using $\delta^{13}C$ -signatures along depth profiles is a common approach (e.g. Holmes et al., 2015; McCalley et al., 2014; Hodgkins et al., 2014; Kotsyurbenko et al., 2004; Chasar et al., 2000). However, methanogenesis is a strictly anaerobic process and thus saturated, anoxic conditions are a prerequisite for an unbiased

- 15 differentiation of pathways using ¹³C only (Conrad, 1996). Methanotrophy would otherwise bias the interpretation of ¹³C isotopic signatures of methane, as residual methane gets enriched in ¹³C, mimicking values as observed under methanogenic conditions predominated by the acetoclastic pathway (Whiticar, 1999). Indeed, summer water table levels at all sites at Wylde Lake peatland dropped down to 32.5 cm (site 1), 31.75 cm (site 2), 13.34 (site 3) and 19.11 cm (site 4) below surface and we could thus only assume saturated, anoxic conditions below that depth. Therefore, we will limit the discussion of CH₄ production
- 20 pathways to depths below -35 cm. For shallower depths (-5 to -25 cm), effects of under such conditions much more favorable methanotrophic activity can be expected to predominate: If the proportion of methanogenesis vs. methanotrophy is comparatively shifted toward methanogenesis, a relative ¹³C-CH₄ depletion would be detected, and if the proportion of methanogenesis vs. methanotrophy is comparatively shifted toward methanotrophy, a relatively ¹³C enrichment in CH₄ would be detected). Methane oxidation is known to cause ¹³C-enriched CH₄ that is even more ¹³C enriched compared to the ambient
- 25 CH₄, as was observed in the top -5 to -15 cm along our study transect during the summer months and α_C values typically observed for the acetoclastic pathway would in this case clearly arise from methanotrophic activity (Alstad and Whiticar, 2011). Our least negative values (-26.77 ‰), observed at 5 cm depth of moss-dominated site 1, markedly exceeded those found in other studies (e.g. Bellisario et al., 1999; Corbett et al., 2013). This is not surprising, though, as the latter studies had been conducted in peats under anoxic conditions, in which oxidation of CH₄ did not play an important role. Moreover, δ¹³C-CH₄
- 30 signatures at 5 cm depth of different sampling dates appeared to be most variable at the sites 1 and 2, which were also found to be drier than the sites 3 and 4, where less pronounced shifts of δ¹³C-CH₄ signatures occurred throughout the sampling period. However, also at the latter sites, variations in δ¹³C-CH₄ were apparently driven by fluctuations of the water table levels. Isotopic composition of CO₂ and CH₄ thus indicated clear differences between sites and depth₂ (see Fig. 5), corresponding to

17	н.	-
1/	н	1
		1

-	Deleted:	wet and	

Deleted:

Deleted: However, repeated water table fluctuations driven by management of the reservoir could effectively recharge electron acceptor pools to support or ongoing decomposition, as e.g. shown in water table manipulation experiments. HoweverMoreover, in close proximity to the reservoir, also an advective redistribution and removal of CO₂ and CH₄ through advective flow cannot be excluded **Deleted:**

Deleted: D using δ^{13} C-signatures along depth profiles ogenesis is a strictly anaerobic process and thus prerequisiten unbiaseddifferentiation using 13C onlyMethanotrophy would otherwise bias the interpretation of 13C isotopic signatures of methane, as residual methane gets enriched in 13C, mimicking values as observed under methanogenic conditions predominated by the acetoclastic pathway Indeed s levelsall sites at could thus only assume, anoxicTherefore, limit the ion oftodepths belowshallower depthsa effects of under such conditions much more favorable methanotrophic activity can be expected to predominate: If the, and if the ament in Methane oxidation is known to cause more ¹³C-enriched CH₄ that is even more ¹³C enriched compared to the ambient CH₄ (Alstad and Whiticar, 2011), as was observed here(...) would in this case clearly. Our least negative values (-26.77 ‰), observed at 5 cm depth of mixture-of-PFTs site 1, markedly exceeded those found in other studies (e.g. Bellisario et al., 1999; Corbett et al., 2013). This is not surprising, though, as the latter studies had, which had been conducted in peats subjected tounder anoxic conditions, wherein which oxidation of CH_4 did not play an important role. Moreover, $\delta^{13}C$ - CH_4 signatures at 5 cm depth of different sampling dates appeared to be most variable at the sites 1 and 2, which were also ind to be drier than the sites 3 and 4, where less pronounced shifts ³C-CH₄ signatures occurred throughout the sampling period. However, also at the latter sites, variations in d13C-CH4 were apparently driven by fluctuations of the water table levels.

Moved (insertion) [3]

observed patterns in concentrations and inferred production, consumption, or emissions. In general, δ^{13} C-CH₄ signatures along soil profiles of all sites increased between June and August, i.e. CH₄ got more enriched in ¹³C, suggesting that CH₄ oxidation must have been an important factor throughout the dry season in summer when the water table notably dropped. <u>Another</u> interesting finding was the strong d13C-CH4 signal pointing to notable CH4 oxidation at 15 cm depth of shrub dominated site

- 5 4 in August 2015 (-39.10 ‰) as compared to more 13C depleted CH4 (-57.73 ‰) in 5 cm depth. This was probably due to the particularly low CH₃ concentrations and more negative δ¹³C-CH₄, suggesting an input of atmospheric methane into the surface peat the shrub dominated site 4 also featured the most enriched δ¹³C-CH₄ signatures in general, suggesting either least CH₄ production or most CH₄ oxidation here. The graminoid-moss dominated site 3, showed the smallest variations in δ¹³C-CH₄ signatures throughout the sampling period, suggesting least modification of δ¹³C-CH₄ from oxidation here, which corresponds well with greatest CH₄ emissions measured at that site.
- Our δ^{13} C-CO₂ values ranging from -25.81 to + 4.03 ‰ also agreed well with other studies (e.g. Landsdown et al., 1992; Hornibrook et al., 2000), although comparably less data on ¹³C in CO₂ from other studies is available. Overall lowest values were found at shrub dominated site 4, where least negative values of d13C-CH4 were observed, suggesting a higher share of CO₂ from increased CH₄ oxidation.,Our δ^{13} C-CO₂ values generally got more enriched in ¹³C with depth at all sites and sampling
- 15 dates, as expected from ongoing fractionation by methanogenesis. Great shifts in δ^{13} C-CO₂ values of the drier sites 1 and 2 during the entire sampling period could again be explained by increased exchange of peat derived CO₂ with atmospheric CO₂ under unsaturated conditions with dropping water tables in August.
- Regarding observed ranges of a_C values at -35 cm depth at the sites, also a gradient in terms of methane production along the transect of study sites became apparent: moss dominated site 1 and graminoid-moss dominated site 2, which experienced the lowest water tables during the summer, and which were located in farthest distance from the water reservoir, feature a distinct shift from mostly hydrogenotrophic CH₄ production in June to acetoclastic CH₄ production in July and August and another shift back to hydrogenotrophic CH₄ production in September, with these shifts being more pronounced at site 1. This could be related to increased vascular plant activity in the growing season and concomitant substrate supply to methanogens, e.g. though
- exudation; an increased share of acetoclastic methanogenesis within the rhizosphere has previously been reported (Chasar et al., 2000; Hornibrooket al., 1997). At graminoid-moss dominated site 3 and at shrub dominated site 4, such obvious shifts of methane production pathways could not be observed, though; α_C values indicated either acetoclastic CH₃ production or a co-occurrence of acetoclastic and hydrogenotrophic CH₄ production. As acetoclastic methanogenesis is in particular supported in minerotrophic peatlands in presence of vascular plants (Alstad and Whiticar, 2011; Chasar et al., 2000), predominance of that
- pathway -in particular in closer vicinity to the reservoir- is not a surprising finding for Wylde Lake peatland. Indeed, under
 predominance of sedges, which supply labile organic matter through roots, aceotelastic CH₄ production prevailed (Bellisario et al., 1999; Popp et al., 1999; Strom et al., 2003). However, the fact that CH₄ production pathways at site 3 and 4, featuring a very different vegetation, were similar, whereas CH₄ production pathways at the site 2 and 3 were different, even though the sites featured a similar vegetation, suggested that variation of α_C would rather reflect the impact of the reservoir, either a) by sustaining higher water tables, or b) by increased nutrient input, than the presence of sedges at the sites,

18

Moved up [3]: Methane oxidation is known to cause more ¹³Cenriched CH4 that is even ¹³C enriched compared to the ambient CH4 (Alstad and Whiticar, 2011), as was observed here. Our least negative values (-26.77 ‰), observed at 5 cm depth of mixture-of-PFTs site 1, markedly exceeded those found in other studies (e.g. Bellisario et al., 1999; Corbet et al., 2013), which had been conducted in peats subjected to anoxic conditions, where oxidation of CH4 did not play an important role. Moreover, δ^{13} C- CH4, signatures at 5 cm depth of different sampling dates appeared to be most variable at the sites 1 and 2, which were also found to be drier than the sites 3 and 4, where less pronounced shifts of δ^{16} C-CH4 signatures occurred throughout the sampling period.

Deleted: , which could have to do with more aerated peat due to rooting of shrubs and the deepening of soil oxygenation probably promoting a highly active methanotrophic bacteria community, which drew CH₄ from the atmosphere down to that depth, resulting in a more pronounced CH₄ oxidation signal in 15 cm than in 5 cm depth.

Deleted: T

Deleted: wet and

Deleted: High CO₂ emission despite strikingly low DIC concentrations, which we observed along the site 4 peat profiles during the entire study period, suggest that the CO₂ stemmed from increased respiration through stronger aeration and parallel CH₄ oxidation in the site 4 peat.

Deleted: Moreover, according to Hornibrook et al. (2007), methanogenesis in semi-terrestrial environments typically leads to δ^{13} C-CO₂ values between -20 and -5 ‰ if dominated by an input of C3 vegetation, which is the case in the Wylde Lake peatland complex.

Deleted: Regarding observed ranges of at the sites, also became apparentsuch obvious of, thoughorin particular in presence of vascular plants predominance of, ing.,variation of α_c would rather reflect the impact, by sustaining by Regarding isotopic signatures of the emitted CH_4 , in general, the emitted CH_4 (see Fig. 6 (a) – (d)) was lighter than the CH_4 in the pore-gas (see Fig. 5) of <u>all sampled</u> peat layers. This suggests that the emitted CH_4 must have been produced in the deeper peat layers (Marushchak et al., 2016), where $\delta^{13}C$ -CH₄ signatures were probably more depleted and during transport through plant aerenchyma, the lighter CH₄ could bypass oxidation. Moreover, plant-mediated transport also discriminates against the

- 5 $\frac{13}{\text{C-CH}_4 \text{(Chanton, 2005)}}$. Interestingly, plant mediated transport was the dominant CH₄ emission pathway even at the shrub dominated site 4 and mixture-of-PFTs site 1, where graminoid cover accounted only for about 10 % We suggest that this is due to the great CH₄ oxidation in the upper peat layers and rather high concentrations at greater depth, facilitating plant mediated transport and ebullition. From visual inspection of the panels (a) (d) of Fig. 6 and Fig. 5 we suggest that the emitted CH₄ originated from at least -35 cm depth or below.
- 10 Regarding hypothesis 2 stating that increased abundance of vascular plants can increase CO_2 uptake but also change patterns of CH_4 production and emission, in particular if graminoids dominate, this hypothesis can only partly be accepted. If increased vascular plant cover translated into increased CO_2 uptake, we should have observed increasing uptake in the order of site $1 \le 2 = 3 \le 4$, but in fact we observed only significantly decreased uptake at site 4. The CO_2 uptake at mixture-of PFTs site 1 (the site with the least coverage of vascular plants) was not statistically different from the cumulative NEP observed at the
- 15 graminoid-moss dominated sites 2 and 3. Moreover, we cannot directly state that CH₄ production and emission was increased where graminoids dominated. Although greatest CH₄ emission was observed at graminoid-moss dominated site 3, cumulative CH₄ emission at site 2 was significantly lower, despite relatively similar vegetation. Moreover, besides by PFTs, CH₄ production appeared to be affected by the vicinity of the water reservoir, whereas plant-dominated CH₄ emission was the dominant CH₄ emission pathway at all study sites, even where graminoid coverage accounted for only 10%. So, we conclude
- 20 that an interplay of nutrient input, water table depth and composition of vegetation shaped CO₂ uptake, CH₄ production and emissions and there is likely no unique driver in our in-situ study, compared to well defined manipulation experiments.

5 Concluding remarks

Our study and earlier work at this particular site confirm <u>that despite long-term increased nutrient supply</u>, <u>peatland ecosystem</u> functioning in terms of C sequestration was largely maintained. However, along a sequence of study sites it became apparent

- 25 that the affected sites responded differently to the altered conditions after dam construction in 1954. A shrub dominated site, which was in closest vicinity to the reservoir and accordingly faced greatest nutrient input and most pronounced water level fluctuations, indeed showed indications of degradation, such as most decomposed bulk peat, least atmospheric CO₂ uptake, and reduced coverage of <u>Sphagnum mosses</u>. However, even here, overall net CO₂ uptake still exceeded net CO₂ release. Two graminoid-moss dominated sites and a mixture-of-PFTs site featured very high CO₂ uptake rates despite apparent impact of
- 30 nutrients and altered hydrology_Therefore, as hypothesized, our case study supports that long-term nutrient enrichment in combination with hydrologically altered conditions may cause differential responses of C cycling and does not necessarily cause a loss of the C-sink function of peatland ecosystems.

19

|--|

Deleted: . Here,

Deleted:

Deleted: So, our assumption that the dominant CH_4 emission pathway is plant-dominated transport where aerenchymatous graminoids are abundant while the shrub dominated site features mostly CH_4 diffusion (hypothesis 3) is to be rejected.

Deleted: Plant-mediated transport was the dominant CH_4 emission pathway at all sites and at all sampling dates.

Deleted: As already mentioned in the results Sect. (3.5) we would prefer to not overinterpret the variation in δ^{13} C-CH₄ signatures throughout the growing season, because we think that by placing our chambers for isotope sampling when wid was lowest (in August 2015), we might have pushed additional CH₄, which was subject to oxidation, out of the upper peat layers. However, given the limitations in our sampling technique, the δ^{13} C-CH₄ values of emitted CH₄ were still much lower than the pore-gas δ^{13} C-CH₄ values of the upper peat layers, further underlining that plant-mediated transport was the dominant CH₄ emission pathway. Deleted: 2 Moreover, methanogenesis and methanotrophy featured a pattern which appeared to be related not predominantly to vegetation, but primarily to the vicinity to the reservoir and thus nutritional status and hydrologic regime. On the other hand plant-mediated transport was determined to be the dominant methane emission pathway along our transect at all sites, even if graminoid cover was only 10 %. All surface peat layers indicated high methanotrophic activity, mitigating CH₄ emission

5 through diffusion.

Lastly, our results suggest that a graminoid-moss dominated peatland site can obviously withstand eutrophication in combination with frequent inundation better than a shrub dominated peatland site, when regarding the overall carbon budget. Straightforward results from manipulation experiments of individual factors (e.g. fertilization or water table changes) may therefore not be easily transferred to complex in-situ conditions. We suggest that there could be a tipping point, when a peatland system shifts from still being a net C sink -even though experiencing eutrophic conditions- to decreasing productivity, which

might be related to an expansion of shrub dominated vegetation, decreasing overall carbon uptake,

6 Data availability

The data can be accessed by email request to the corresponding authors.

15

Author contributions. Christian Blodau, Klaus-Holger Knorr and Sina Berger designed the experiments; Sina Berger, Leandra Praetzel and Marie Goebel conducted field work and analyses with help of Klaus-Holger Knorr. Sina Berger prepared the manuscript with contributions from Klaus-Holger Knorr, Leandra Praetzel and Marie Goebel.

20 The authors declare that they have no conflict of interest.

Acknowledgements. This study was funded by the Deutsche Forschungsgemeinschaft (German Research Foundation - DFG) (BL563/21-1) We thank Martin Neumann from the Grand River Conservation Authority for the permission to carry out research in the Luther Marsh Wildlife Management Area and we thank Claudia Wagner-Riddle, Peter Smith and Linda Wing

- 25 for their help on organizational issues. We also thank Inge-Beatrice Biro, Magdalena Burger, Ines Spangenberg, Niclas Kolbe, Eike Esders, Michael Rammo, Nils Vickus, Fabian Benninghoff, Leonie Fröhlich, Jörg Rostek and Cornelia Mesmer for their support in the field. Analyses of CO₂ and CH₄ concentrations, δ^{13} C abundance and spectral analyses of various samples were carried out at the institutional lab or the Institute of Landscape Ecology, University of Münster. We thank Stefanie Holm, Ronya Wallis and Madelaine Supper for assistance during analysis of numerous samples in the laboratory.
- 30 This paper is dedicated to the memory of Christian Blodau, who led the Wylde Lake peatland project until he tragically passed away in July 2016.

We thank two anonymous reviewers for their thoughtful comments that helped to improve this manuscript.

20

Deleted:

References

Alstad, K. P., Whiticar, M. J.: Carbon and hydrogen isotope ratio characterization of methane dynamics for Fluxnet peatland ecosystems, Org Geochem, 42(5), 548–558, 2011.

5 Ballantyne, D. M., Hribljan, J. A., Pypker, T. G., Chimner, R. A.: Long- term water table manipulations alter peatland gaseous carbon fluxes in Northern Michigan, Wetl Ecol Manag, 22, 35–47, 2014.

Berger, S., Gebauer, G., Blodau, C., Knorr, K.-H.: Peatlands in a eutrophic world – assessing the state of a poor fen-bog transition in southern Ontario, Canada, after long term nutrient Input and altered hydrological conditions, Soil Biol Biochem,
 10 114, 131–144, 2017.

Bellisario, L. M., Bubier, J. L., Moore, T. R.: Controls on CH₄ emissions from a northern peatland, Global Biogeochem Cycles, 13(1), 81–91, 1999.

15 Biester, H., Knorr, K.-H., Schellekens, J., Basler, A., and Hermanns, Y.-M.: Comparison of different methods to determine the degree of peat decomposition in peat bogs, Biogeosciences, 11, 2691–2707, 2014.

Blodau, C.: Carbon cycling in peatlands - A review of processes and controls, Environ Rev, 10, 111-134, 2002.

20 Bragazza, L.: A decade of plant species changes on a mire in the Italian Alps: vegetation-controlled or climate-driven mechanisms?, Clim Change, 77, 415–429, 2006.

Bragazza, L., Siffi, C., Lacumin, P., Gerdol, R.: Mass loss and nutrient release during litter decay in peatland: The role of microbial adaptability to litter chemistry, Soil Biol Biochem, 39(1), 257–267, 2007.

25

Bragazza, L., Buttler, A., Habermacher, J., Brancaleoni, L., Gerdol, R., Fritze, H., Hanjík, P., Laiho, R., Johnson, D.: High nitrogen deposition alters the decomposition of bog plant litter and reduces carbon accumulation, Glob Chang Biol, 18, 1163– 1172, 2012.

30 Bragazza, L., Parisod, J., Buttler, A., Bardgett, R. D.: Biogeochemical plant-soil microbe feedback in response to climate warming in peatlands, Nat Clim Change, 3, 273–277, 2013.

Bragazza, L., Bardgett, R. D., Mitchell, E. A. D., Buttler, A.: Linking soil microbial communities to vascular plant abundance along a climate gradient, New Phytol, 205, 1175–1182, 2015.

Bragazza, L., Buttler, A., Robroek, B., Albrecht, R., Zaccone, C., Jassey, V., Signarbieux, C.: Persistent high temperature and
low precipitation reduce peat carbon accumulation, Glob Change Biol, 22, 4114–4123, 2016.

Broder, T., Blodau, C., Biester, H., Knorr, K.-H.: Peat decomposition records in three pristine ombrotrophic bogs in southern Patagonia, Biogeosciences, 9, 1479–1491, 2012.

10 Bubier, J. L., Moore, T. R., Bellisario, L., Comer, N. T., Crill, P. M.: Ecological controls on methane emissions from a Northern Peatland Complex in the zone of discontinuous permafrost, Manitoba, Canada, Glob Biogeochem Cycles, 9(4), 455–470, 1995.

Bubier, J. L., Moore, T. R., Bledzki, L. A.: Effects of nutrient addition on vegetation and carbon cycling in an ombrotrophic bog, Glob Chang Biol, 13, 1168–1186, 2007.

15

Burger, M., Berger, S., Spangenberg, I., Blodau, C.: Summer fluxes of methane and carbon dioxide from a pond and floating mat in a continental Canadian peatland, Biogeosciences, 13, 3777–3791, 2016.

<u>Chanton, J. P.: The effect of gas transport on the isotope signature of methane in wetlands, Org Geochem, 36 (5), 753–768,</u>
2005.

Chasar, L. S., Chanton, J. P., Glaser, P. H., Siegel, D. I.: Methane Concentration and Stable Isotope Distribution as Evidence of Rhizospheric Processes: Comparison of a Fen and Bog in the Glacial Lake Agassiz Peatland Complex, Ann Bot, 86, 655– 663, 2000.

25

Conrad, R.: Quantification of methanogenic pathways using stable carbon isotopic signatures. A review and a proposal, Org Geochem, 36(5), 739–752, 2005.

<u>Corbett, J. E., Tfaily, M. M., Burdige, D. J., Cooper, W. T., Glaser, P. H., Chanton, J. P.: Partitioning pathways of CO₂
 production in peatlands with stable carbon isotopes, Biogeochemistry, 114(1-3), 327–340, 2013.
</u>

Eriksson, T., Öquist, M. G., Nilsson, M. B.: Production and oxidation of methane in a boreal mire after a decade of increased temperature and nitrogen and sulfur deposition, Glob Chang Biol, 16, 2130–2144, 2010.

Gray, A., Levy, P. E., Cooper, M. D. A., Jones, T., Gaiawyn, J., Leeson, S. R., Ward, S. E., Dinsmore, K. J., Sheppard, L. J., Ostle, N. J., Evans, C. D., Burden, A., Zielinski, P.: Methane indicator values for peatlands: a comparison of species and functional groups, Glob Change Biol, 19, 1141–1150, 2013.

5 Heidel, D.: Isotopenfraktionierungseffekte bei der Beprobung von Bodenluft und gelösten Gasen, unpublished Bacherlor's Thesis, 2009.

Hesslein, R. H.: Insitu Sampler for close interval pore water studies, Limnol Oceanogr, 21(6), 912-914, 1976.

10 Hodgkins, S. B., Tfaily, M. M., McCalley, C. K., Logan, T. A., Crill, P. M., Saleska, S. R., Rich, V. I., Chanton, J. P.: Can changes in peat chemistry associated with permafrost thaw increase greenhouse gas production, PNAS, 111(16), 5819–5824, 2014.

Hoefs, J.: Stable Isotope Geochemistry, 3rd ed., Springer, Berlin, 1987.

15

Holmes, M. E., Chanton, J. P., Tjaily, M. M., Ogram, A.: CO₂ and CH₄ isotope compositions and production pathways in a tropical peatland, Glob Biochem Cycles, 29, 1–18, 2015.

Hornibrook, E. R. C., Longstaffe, F. J., William, F. S.: Evolution of stable carbon isotope compositions for methane and carbon
 dioxide in freshwater wetlands and other anaerobic environments, Geochim Cosmochim Acta, 64(6), 1013–1027, 2000.

Hornibrook, E. R. C., Bowes, H. L.: Trophic status impacts both the magnitude and stable carbon isotope composition of methane flux from peatlands, Geophys Res Lett, 34(21), 2007.

25 Hornibrook, E. R. C.: The stable carbon isotope composition of methane produced and emitted from northern peatlands. Andrew J. Baird, Lisa R. Belyea, Xavier Comas, A. S. Reeve und Lee D. Slater (Ed.): Carbon Cycling in Northern Peatlands, Bd.184. Washington, D. C.: American Geophysical Union (Geophysical Monograph Series), pp. 187–203, 2009.

Kammann, C., Grünhage, L., Jäger, H.-J.: A new sampling technique to monitor concentrations of CH₄, N₂O and CO₂ in air at
 well-defined depths in soils with varied water potential, Eur J Soil Sci, 52, 297–303, 2001.

Kim, Y., Ullah, S., Roulet, N. T., Moore, T. R.: Effect of inundation, oxygen and temperature on carbon mineralization in boreal ecosystems, Sci Total Environ, 511, 381–392, 2015.

Klüpfel, L., Piepenbrock, A., Kappler, A., Sander, M.: Humic substances as fully regenerable electron acceptors in recurrently anoxic environments, Nat Geosci, 7, 195–200, 2014.

Knorr, K.-H., Oosterwoud, M. R., Blodau, C.: Experimental drought alters rates of soil respiration and methanogenesis but not
carbon exchange in soil of a temperate fen, Soil Biol Biochem, 40(7), 1781–1791, 2008.

Kotsyurbenko, O. R., Chin, K.-J., Glagolev, M. V., Stubner, S., Simankova, M. V., Nozhevnikova, A. N., Conrad, R.: Acetoclastic and hydrogenotrophic methane production and methanogenic populations in an acidic West-Siberian peat bog, Environ Microbiol, 6 (11), 1159–1173, 2004.

10

Kuiper, J. J., Mooij, W. M., Bragazza, L., Robroek, B. J. M.: Plant functional types define magnitude of drought response in peatland CO₂ exchange, Ecology, 95(1), 123–131, 2014.

Laine, J., Harju, P., Timonen, T., Laine, A., Tuittila, E.-S., Minkkinen, K., Vasander, H.: The Intricate Beauty of *Sphagnum* mosses – a Finnish Guide to Identification, 2nd amended edition, 2011.

Lansdown, J. M, Quay, P. D., King, S. L.: CH₄ production via CO₂ reduction in a temperate bog: A source of ¹³C-depleted CH₄, Geochim Cosmochim Acta, 56(9), 3493–3503, 1992.

20 Larmola, T., Bubier, J. L., Kobyljanec, C., Basiliko, N., Juutinen, S., Humphreys, E., Preston, M., Moore, T. R.: Vegetation feedbacks of nutrient addition lead to a weaker carbon sink in an ombrotrophic bog, Glob Change Biol, 19, 3729–3739, 2013.

Levy, P. E., Burden, A., Cooper, M. D. A., Dinsmore, K. J., Drewer, J., Evans, C., Fowler, D., Gaiawyn, J., Gray, A., Jones, S. K., Jones, T., Mcnamara, N. P., Mills, R., Ostle, N., Sheppard, L. J., Skiba, U., Sowerby, A., Ward, S. E., Zielinski, P.:
 Methane emissions from soils: synthesis and analysis of a large UK data set, Glob Change Biol, 18, 1657–1669, 2012.

Limpens, J., Berendse, F., Blodau, C., Canadell, J. G., Freeman, C., Holden, J., Roulet, N., Rydin, H., Schaepman-Strub, G.: Peatlands and the carbon cycle: from local processes to global implications – a synthesis, Biogeosciences, 5, 1475–1491, 2008.

30 Lund, M., Lafleur, P. M., Roulet, N. T., Lindroth, A., Christensen, T. R., Aurela, M., Chojnicki, B. H., Flanagan, L. B., Humphreys, E. R., Laurila, T., Oechel, W., Olejnik, J., Rinne, J., Schubert, P., Nilsson, M. B.: Variability in exchange of CO₂ across 12 northern peatland and tundra sites, Glob Change Biol, 16, 2436–2448, 2010.

Marushchak, M. E., Friborg, T., Biasi, C., Herbst, M., Johannson, T., Kiepe, I., Liimatainen, M., Lind, S. E., Martikainwn, P. J., Virtanen, T., Soegaard, H., Shurpali, N. J.: Methane dynamics in the subarctic tundra: combining stable isotope analyses, plot- and ecosystem-scale flux measurements, Biogeosciences, 13, 597–608, 2016.

5 McCalley, C., Woodcroft, B., Hodgkins, S., Wehr, R., Kim, E.-H., Mondav, R., Crill, P., Chanton, J., Rich, V., Tyson, G., Saleska, S.: Methane dynamics regulated by microbial community response to permafrost thaw, Nature, Vol. 514, no 7523, 478–481, 2014.

Morris, P. J., Waddington, J. M., Benscoter, B. W., Turetsky, M. R.: Conceptual frameworks in peatland ecohydrology: looking
 beyond the two-layered (acrotelm–catotelm) model, Ecohydrology, 4, 1–11, 2011.

Murphy, K. R., Butler, K. D., Spencer, R. G., Stedmon, C. A., Boehme, J. R., Aiken, G. R.: Measurement of dissolved organic matter fluorescence in aquatic environments: an interlaboratory comparison, Environ Sci Technol, 44, 9405–9412, 2010.

15 National Climate Data and Information Archive, 2014. Canadian Climate Normals. Dataset for the climate station Fergus, Shand Dam, 1981 to 2010. URL http://climate.weather.gc.ca/climate normals/index e.html (accessed November 18th, 2015).

Niemeyer, J., Chen, Y., Bollag, J. M.: Characterization of humic acids, composts, and peat by diffuse reflectance Fourier-Transform Infrared Spectroscopy, Soil Sci Soc Am J, 56, 135–140, 1992.

20

Nijp, J. J., Limpens, J., Metselaar, K., van der Zee, S. E. A. T. M., Berendse, F., Robroek, B. J. M.: Can frequent precipitation moderate the impact of drought on peatmoss carbon uptake in northern peatlands?, New Phytol, 203, 70–80, 2014.

Ohno, T.: Fluorescence inner-filtering correction for determining the humification index of dissolved organic matter, Environ 25 Sci Technol, 36, 742–746, 2002.

Owen, K. E., Tenhunen, J., Reichstein, M., Wang, Q., Falge, E., Geyer, R., Xiao, X. M., Stoy, P., Amman, C., Arain, A., Aubinet, M., Aurela, M., Bernhofer, C., Chojnicki, B. H., Grainer, A., Gruenwald, T., Hadley, J., Heinsch, B., Hollinger, D., Knohl, A., Kutsch, W., Lohila, A., Meyers, T., Moors, E., Moureaux, C., Pilegaard, K., Saigusa, N., Verma, S., Vesala, T.,
 Vogel, C.: Linking flux network measurements to continental scale simulations: ecosystem carbon dioxide exchange capacity

under non-water-stressed conditions, Glob Change Biol, 13(4), 734–760, 2007.

Popp, T. J., Chanton, J. P., Whiting, G. J., Grant, N.: Evaluation of methane in the rhizosphere of a Carex dominated fen in north central Alberta, Canada, Biogeochemistry, 51, 259–281, 2000.

R Core Team: R: A language and environment for statistical computing, R Foundation for Statistical Computing, Vienna, 2015.

5 Robroek, B., Jassey, V. E. J., Kox, M., A. R., Berendsen, R. L., Mills, R. T. E., Cécillon, L., Puissant, J., Meima-Franke, M., Bakker, P. A. H. M., Bodelier, P. L. E.: Peatland vascular plant functional types affect methane dynamics by altering microbial community structure, J Ecol, 103, 925–934, 2015.

Rydin, H., Barber, K. E.: Long-term and fine-scale coexistence of closely related species, Folia Geobot, 36, 53-61, 2001.

Rydin, H., Jeglum, J. K.: The Biology of Peatlands, 2nd ed., Oxford University Press, 2013.

10

 Sander, R.: Compilation of Henry's Law Constants for Inorganic and Organic Species of Potential Importance in Environmental Chemistry, Max Planck Institute of Chemistry, Mainz, 1999. Online at http://www.henrys-law.org/henry 3.0.pdf.

Schütz, H., Schröder, P., Rennenberg, H.: Role of plants in regulating the methane flux to the atmosphere, Sharkey, T.D., Holland, E.A., Mooney, H.A. (Eds.), Trace Gas Emission by Plants, Academic Press, NY, pp. 29–69, 1991.

20 Shannon, R. D., White, J. R.: A Three-Year Study of Controls on Methane Emissions from Two Michigan Peatlands, Biogeochemistry, 27(1), 35–60, 1994.

Sheppard, L. J., Leith, I. D., Leeson, S. R., vam Dijk, N., Field, C., Levy, P.: Fate of N in a peatland, Whim bog: immobilisation in the vegetation and peat, leakage into pore water and losses as N2O depend on the form of N, Biogeosciences, 10, 149–160,
 25 2013.

Strack, M., Waller, M. F., Waddington, J. M.: Sedge succession and peatland methane dynamics: A potential feedback to climate change, Ecosystems, 9(2), 278–287, 2006.

30 Strom, L., Ekberg, M., Mastepanov, M., Christensen, T. R.: The effect of vascular plants on carbon turnover and methane emissions from a tundra wetland, Glob Change Biol, 9, 1185–1193, 2003.

Stumm, W., Morgan, J. J.: Aquatic chemistry. Chemical Equilibria and Rates in Natural Waters. 3rd ed. Hoboken: Wiley (Environmental Science and Technology), 1996.

Succow, M., Joosten, H.: Landschaftsökologische Moorkunde. Zweite, völlig neu bearbeitete Auflage. 2. Auflage. E. Schweizerbart`sche Verlagsbuchhandlung, Stuttgart. 2012.

- Teh, Y. A., Silver, W. L., Conrad, M. E., Borglin, S. E., Carlson, C. M.: Carbon isotope fractionation by methane-oxidizing bacteria, J Gephys Res.: Biogeosci, 111, G01003.
 Tilsner, J., Wrage, N., Lauf, J., Gebauer, G.: Emission of gaseous nitrogen oxides from an extensively managed grassland in NE Bavaria, Germany. I. Annual budgets of N₂O and NO_x emissions, Biogeochemistry, 63, 229–247, 2003.
- 10 Teklemariam, T. A., Lafleur, P. M., Moore, T. R., Roulet, N. T., Humphreys, E. R.: The direct and indirect effects of interannualmeteorological variability on ecosystem carbon dioxide exchange at a temperate ombrotrophic bog, Agric For Meteorol, 150, 1402–1411.

 Tranvik, L. J., Downing, J. A., Cotner, J. B., Loiselle, S. A., Striegl, R. G., Ballatore, T. J., Dillon, P., Finlay, K., Fortino, K.,
 Knoll, L. B., Kortelainen, P. L., Kutser, T., Larsen, S., Laurion, I., Leech, D. M., McCallister, L., McKnight, D. M., Melack, J. M., Overholt, E., Porter, J. A., Prairie, Y., Renwick, W. H., Roland, F., Sherman, B. S., Schindler, D. W., Sobek, S., Tremblay, A., Vanni, M. J., Verschoor, A. M., von Wachenfeldt, E., Weyhenmeyer, G. A.: Lakes and reservoirs as regulators of carbon cycling and climate, Limnol Oceanogr, 54(6, part 2), 2298–2314, 2009.

20 Turetsky, M. R., Bond-Lamberty, B., Euskirchen, E., Talbot, J., Frolking, S., McGuire, M. R., Tuittila, E.-S.: The resilience and functional role of moss in boreal and arctic ecosystems, New Phytol, 196(1), 49–67, 2012.

van den Berg, M., Ingwersen, J., Lamers, M., Streck, T.: The role of *Phragmites* in the CH₄ and CO₂ fluxes in a minerotrophic peatland in southwest Germany, Biogeosciences, 13, 6107–6119, 2016.

25

Ward, S. E., Ostle, N. J., Oakley, S., Quirk, H., Henrys, P. A., Baedgett, R. D.: Warming effects on greenhouse gas fluxes in peatlands are modulated by vegetation composition, Ecol Lett, 16, 1285–1293, 2013.

Wang, M., Moore, T. R., Talbot, J., Riley, J. L.: The stochiometry of carbon and nutrients in peat formation, Glob Biochem
 30 Cycles, 29, 113–121, 2015.

Wang, M., Larmola, T., Murphy, M. T., Moore, T. R., Bubier, J.: Stoichiometric response of shrubs and mosses to long-term nutrient (N, P and K) addition in an ombrotrophic peatland, Plant Soil, 400, 403–416, 2016.



Weishaar, J. L., Aiken, G. R., Bergamischi, B. A., Fram, M. S., Fujii, R., Mopper, K.: Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon, Environ Sci Technol, 37, 4702–4708, 2003.

5 Whalen, S. C., Reeburgh, W. S., Sandbeck, K. A.: Rapid methane oxidation in a landfill cover soil, Appl Environ Microbiol, 56, 3405–3411, 1990.

Whalen, S. C.: Biogeochemistry of methane exchange between natural wetlands and the atmosphere, Environ Eng Sci, 22, 73– 94, 2005.

10

τ.

Whiticar, M. J., Faber, E., Schoell, M.: Biogenic methane formation in marine and freshwater environments. CO₂ reduction vs. acetate fermentation—Isotope evidence, Geochim Cosmochim Acta, 50 (5), 693–709, 1986.

Whiting, G. J., Chanton, J. P.: Control of the diurnal pattern of the methane emission from emergent aquatic macrophytes by
 gas transport mechanisms, Aquat Bot, 54, 237–253, 1996.

Wiedermann, M. M., Nordin, A., Gunnarsson, U., Nilsson, M. B., Ericson, L.: Global Change Shifts vegetation and plantparasite interactions in a boreal mire, Ecology, 88(2), 454–464, 2007.

Deleted: Alstad, K. P., Whiticar, M. J.: Carbon and hydrogen isotope ratio characterization of methane dynamics for Fluxnet peatland ecosystems, Org Geochem, 42(5), 548–558, 2011.







Figure 1: Location of Wylde Lake peatland complex in North America (left), and sampling sites (black dots) within Wylde Lake peatland complex (right). Source: ArcGIS.



Figure 2: FTIR ratios 1618.5/1033.5 in bulk peat (a) and pore-water (b) as well as SUVA₂₅₄, indicating aromaticity, (c), HIX, humification index, (d) and E2:E3, indicative of molecular size and aromaticity, (e) for pore-water samples of the sites 1 to 4. n = 3. Error bars indicate +/- standard deviation.







mean CH₄ concentrations [µmoi L], CO₂ mixes (NEF partitioned into Reco and GPP) [g CO₂ m² d⁻¹], CH₄ fluxes [g CH₄ m² d⁻¹] \pm 1 SD (n=6) in hollows of the sites 1 and 2 from April 1st, 2014 through September 22nd, 2015. Negative CO₂ and CH₄ fluxes indicate uptake, positive fluxes indicate a release to the atmosphere. Dashed gray lines in the flux graphs indicate a 0flux.













Table 1: Overview of the four sites' distances to the water reservoir and of the composition of the vegetation in terms of coverage [%] of plant functional types (PFT) in hollows and abundances of plant species (vascular plants and mosses, excluding liverworts and hornworts). Abundances are abbreviated as follows: "d" means "dominant" (> 75 %), "a" means
5 "abundant" (51-75 %), "f" means "frequent" (26-50 %), "o" means "occasional" (11-25 %), and "r" means "rare" (1-10 %).

Because Sphagnum mosses were very hard to distinguish in the field, we only determined the abundance of the most abundant

5

Sphagnum species of each site.

Deleted: Table 1: Overview of study sites' hollow properti	es within
Wylde Lake peatland, including coverages of dominant plant	
functional types (PFT), peat age at 60 cm depth ± 1 SD (n=3)), pH,
stochiometric ratios (C/N, C/P, N/P, C/Ca, C/Mg, C/K) at the	peat
surface ± 1 SD (n=3). Stoicniometric ratios have been presen	ted in
Berger et al., (submitted for publication).	
Deleted: Site	[0]

Site 1		Site 2		Site 3		Site 4	
Distance to reservoir [m]							
800		550		400		200	1
_		_					
Coverage of PFT [%]							
Sphaanum spp.: 100		100		100		60	1
Graminoids: 10		30		30		- <u></u> 10	1
Shruhe: 0		<u>50</u>		<u>- 50</u>		- 10	1
511005. 6		<u>2</u>		- 4			1
- Diant an asian		-					1
Plant species		-					-
<u>Spriugriu</u>	1.1	-		- C	-		-
<u>S. magenanicum</u>	<u>a</u> .	<u>S. magellanicum</u>		<u>S. mageilanicum</u>		<u>S. mageilanicum</u>	-
<u>S. cupinijonum</u>	<u>a</u> _	<u>S. cupinijonum</u>	₫ -	<u>S. cupinijonum</u>	<u>d</u>	<u>S. cupinijonum</u>	÷
<u>S. juscum</u>		<u>5. juscum</u>		<u>S. juscum</u>	-	<u>S. juscum</u>	-
<u>S. squarrosum</u>		<u>S. wuijiunum</u>		<u>S. girgensonni</u>	<u>d</u>	<u>S. yirgensonni</u>	1
<u>s. ungustijonum</u>		<u>s. ungustijonum</u>	₫ -	S. squarrosum		<u>s. wujjuliulii</u>	-
-		-		<u>S. wujjunum</u>			-
-		-		<u>S. ungustijonum</u>			1
-		-		<u>5. cuspidutum</u>			7
Other mosses		-		-	-		1
Polytrichum spp.	0	- Polvtrichum spp.	0	Polytrichum spp.	0	Polvtrichum spp.	r
Rhytidiadelphus triquestrus	r	Polytrichum spp.	0	Polytrichum spp.	0		-
			_				÷.
Graminoids					_		_
Carex disperma	f	Scheuchzeria palustris	0	C. disperma	0	C. disperma	0
Carex oligosperma	r	C. disperma	0	C. magellanica	<u>o</u>	E. angustifolium	r
Eleocharis palustris	r	C. oligosperma	<u>r</u>	Dulichium arundinaceum	f	E. vaginatum	<u>0</u>
Eriophorum angustifolium	<u>r</u>	Carex limosa	<u>r</u>	E. palustris	<u>o</u>	E. virginicum	<u>o</u>
Eriophorum vaginatum	0	Carex magellanica	<u>r</u>	E. angustifolium	0	J. effusus	<u>r</u>
Eriophorum virginicum	<u>o</u>	E. palustris	<u>o</u>	E. vaginatum	<u>f</u> .		2
Juncus effusus	<u>r</u> .	E. angustifolium	<u>o</u>	E. virginicum	<u>f</u>		1
-		E. vaginatum	<u>f</u>	J. effusus	<u>o</u>		1
-		E. virginicum	f				2
-		<u>J. effusus</u>	<u>r</u> .		÷.,		1
-		-					-

<u>Shrubs</u>		_		_			_
Aronia melanocarpa	<u>r</u> .	Myrica gale	<u>r</u>	M. gale L.	0	M. gale	a
Andromeda glaucophylla	<u>r</u> .	A. glaucophylla	<u>r</u>	A. glaucophylla	r	A. glaucophylla	r
Chamaedaphne calyculata	0	C. calyculata	0	C. calyculata	0	C. calyculata	0
Kalmia polifolia	<u>o</u>	K. polifolia	<u>r</u>	<u>K. polifolia</u>	<u>r</u> .	K. polifolia	r
Rhododendron groenlandicum	0	R. groenlandicum	r	R. groenlandicum	r	R. groenlandicum	0
Vaccinium myrtilloides	<u>r</u> .	_		_		V. oxycoccos	0
Vaccinium oxycoccos	<u>r</u> .	_		-	÷		_
-		=		-			-
Trees					÷		2
Larix laricina	<u>r</u> .	<u>L. laricina</u>	<u>r</u> .	P. strobus	<u>r</u> .	<u>B. pumila</u>	<u>r</u>
Picea mariana	<u>r</u>	P. strobus	<u>r</u>		÷		2
Pinus strobus	<u>r</u>	B. pumila	<u>r</u>		÷		2
<u>Betula pumila</u>	<u>r</u> .	-		. <u> </u>	÷		_
-		-			÷		-
Herbs					÷		-
Sarracenia purpurea	<u>r</u> .	<u>S. purpurea</u>	<u>r</u> .	Maianthemum trifolium	<u>r</u> .	S. purpurea	<u>r</u>
Drosera rotundifolia	<u>r</u>	D. rotundifolia	<u>r</u>	S. purpurea	<u>r</u> .	D. rotundifolia	<u>r</u>
		_		<u>D. rotundifolia</u>	<u>r</u>		_

<u>Table 2: α_C values and standard deviation obtained from silicone samplers in 35 cm depth at sites 1 to 4 from June to September.</u>
 <u>α_C values between 1.04 and 1.055 indicate the prevalence of the acetoclastic methane production pathway, α_C values higher
 <u>than 1.065 indicate the hydrogenotrophic pathway.</u>
</u>

Site	<u>1</u>		2		3	3		
	<u>α</u>	<u>std.dev</u>	<u>α</u> _c	<u>std.dev</u>	<u>α</u> _c	<u>std.dev</u>	<u>α</u> _c	<u>std.dev</u>
June	1.068		1.064	0.004	1.061	0.004	1.056	0.004
July	1.042		1.044		1.058	0.001	1.048	
August	1.043		1.046	0.001	1.052	0.004	1.045	0.002
<u>September</u>	1.066	0.007	<u>1.057</u>	0.002	<u>1.058</u>	0.003	<u>1.051</u>	0.002

Page 2: [1] Deleted	Sina Berger	9/13/17 10:22:00 AM
	-	

Ample of abiotic and biotic factors have been identified to further influence production and emission of CH_4 on the field scale, such as water level, temperature of air, soil and water as well as provision of fresh substrates or inhibitors through root exudates, which is coupled to photosynthetic activity (e.g. Shannon and White, 1994; Moore and Basiliko, 2006; Dorodnikov et al., 2011).

Page 6: [2] Deleted	Sina Berger	9/18/17 8:49:00 PM
Page 6: [2] Deleted	Sina Berger	9/18/17 8:49:00

All measurements were taken and all samples were collected in hollows.

Page 9: [3] Deleted	Sina Berger	10/14/17 6:14:00 PM

Temporal dynamics of CO₂ and CH₄ concentrations and δ^{13} C-CO₂ and δ^{13} C-CH₄-values along peat pore-water profiles

2.5.1

Page 11: [4] Deleted	Sina Berger	9/14/17 8:15:00 AM
	-	

Aromaticity as determined with $SUVA_{254}$ (Fig 2 (c)) did not show significant differences between sites in porewater samples (exception: site 1 and site 3 in 20 cm depth (p = 0.033), where site 1 SUVA₂₅₄ was significantly higher than site 3 SUVA₂₅₄). Site 3 had the lowest aromaticity at the upper layers (5 cm to 20 cm depth), however, the difference was not statistically significant.

The degree of humification, as depicted by *HIX* (Fig 2 (d)), was significantly lowest in site 3 pore-water (5 cm site 3 and 4: p = 0.026; 10 cm site 1 and 3: p = 0.014; 20 cm site 3 and 4: p = 0.020).

The slope ratio E2:E3 (Fig 2 (e)), indicative of molecular size and aromaticity, indicated decreasing aromaticity and decreasing molecular weight in the order of site 4 > site 1 > site 2 > site 3.

Page 12: [5] Deleted	Sina Berger	9/14/17 11:04:00 AM
Accordingly, NEP, GPP, R_{eco} and C	H ₄ fluxes at almost all sites were positively	v correlated with T_{water} (p < 0.05),
however, negatively correlated with	wtd (lowest water tables co-occurred with h	ighest fluxes). As suggested from
visual inspection, R_{eco},GPP and CH	4 fluxes of the sites 1, 2 and 3 were in mos	t cases negatively correlated with
DIC and CH ₄ concentrations along so	il profiles, i.e. increasing CO_2 and CH_4 fluxe	es were correlated with decreasing
DIC and CH ₄ concentrations. Only at	site 4 greater DIC and CO ₂ concentrations	along peat profiles coincided with
higher effluxes of CO ₂ and CH ₄ . Stat	istical results are summarized in the Table	S4.
During the study period, hollows of	site 1 accumulated 1552 ± 652 g CO ₂ m ⁻² a	nd released 41.8 ± 25.4 g CH ₄ m ⁻

², site 2 accumulated 1637 ± 184 g CO₂ m⁻² and released 44.6 ± 13.7 g CH₄ m⁻², site 3 accumulated 2260 ± 480 g CO₂ m⁻² and released 61.4 ± 32 g CH₄ m⁻² and site 4 accumulated 1093 ± 794 g CO₂ m⁻² and released 46.1 ± 35.2 g CH₄ m⁻² (see Fig. S5). Between May 19th, 2014 and September 23rd, 2015 site 4 accumulated significantly less CO₂ as compared with the other three sites (p < 0.001), while there were no statistically significant differences in terms of CO₂ uptake for the sites 1, 2 and 3. Moreover, site 3 emitted significantly more CH₄ as compared with the sites 1, 2 and 4 (p < 0.001).

A similar pattern was observed for CH_4 concentrations (Fig. 3 and 4), as they were increasing during the growing season, reached maximum values in the winter season 2014/2015 and comparably decreased during snowmelt in spring. Concentrations in the uppermost depths of both CO_2 and CH_4 were strongly affected by wtd fluctuations, with strong decreases upon water table decline and vice versa.

Page 13: [7] Moved to page 0 (Move #2)	Sina Berger	9/14/17 8:33:00 AM

Fluxes of CO₂ and CH₄ (Fig. 3 and 4) showed a strong annual variability. Overall, NEP, GPP, and R_{eco} at all sites were highest during the growing seasons. Also, CH₄ fluxes were highest during the growing season, despite comparably low concentrations of dissolved CH₄ in the investigated upper 50 cm of the peat profile, suggesting that exchange of gases at the peat/atmosphere interface and concentrations in the uppermost peat were decoupled. Accordingly, NEP, GPP, R_{eco} and CH₄ fluxes at almost all sites were positively correlated with T_{water} (p < 0.05), however, negatively correlated with wtd (lowest water tables co-occurred with highest fluxes). As suggested from visual inspection, R_{eco}, GPP and CH₄ fluxes of the sites 1, 2 and 3 were in most cases negatively correlated with decreasing DIC and CH₄ concentrations. Only at site 4 greater DIC and CO₂ concentrations along peat profiles coincided with higher effluxes of CO₂ and CH₄. Statistical results are summarized in the Table S4.

During the study period, hollows of site 1 accumulated 1552 ± 652 g CO₂ m⁻² and released 41.8 ± 25.4 g CH₄ m⁻², site 2 accumulated 1637 ± 184 g CO₂ m⁻² and released 44.6 ± 13.7 g CH₄ m⁻², site 3 accumulated 2260 ± 480 g CO₂ m⁻² and released 61.4 ± 32 g CH₄ m⁻² and site 4 accumulated 1093 ± 794 g CO₂ m⁻² and released 46.1 ± 35.2 g CH₄ m⁻² (see Fig. S5). Between May 19th, 2014 and September 23rd, 2015 site 4 accumulated significantly less CO₂ as compared with the other three sites (p < 0.001), while there were no statistically significant differences in terms of CO₂ uptake for the sites 1, 2 and 3. Moreover, site 3 emitted significantly more CH₄ as compared with the sites 1, 2 and 4 (p < 0.001).

Page 20: [8] Deleted	Sina Berger	9/20/17 10:20:00 AM
----------------------	-------------	---------------------

Our case study shows that certain PFTs co-occurred with pronounced differences in carbon cycling and associated peat quality. A large uptake of CO_2 and a remarkable release of CH_4 in 1.5 years at the graminoid-moss dominated site 3 was accompanied by more degradable and less aromatic pore-water quality. On the contrary, the shrub dominated site 4, which experienced the same water table fluctuations and similar nutrient supply like site 3, featured lowest productivity in terms of CO_2 uptake during the study period, highest degree of bulk peat decomposition and strikingly low DIC concentrations. Two more study sites, of which one had a very similar vegetation cover like site 3 and the other one inhabited a mixture of PFTs, however, being less affected by the nutrient supplying and wtd altering water reservoir, had similar to each other, but different results as compared to the sites 3 and 4.

Our results suggest that a graminoid-moss dominated peatland site can withstand eutrophication better than a shrub dominated site (which was also suggested by Wu et al., (2015)). We suggest that there could be a tipping point, when a peatland system shifts from still being productive even though experiencing eutrophic conditions to decreasing productivity, which might co-occur with a spreading of shrub dominated vegetation. If such systems shift to shrub dominated systems, which are exposed to high nutrient deposition even at high water tables, there would be a negative feedback in terms of carbon sequestration.

Our results further suggest that in a eutrophic peatland, which experiences strong water table dropdowns during the growing season, plant mediated transport is the dominant CH_4 emission pathway, even in a shrub dominated site and in mixture of vegetation site, where graminoid cover was only 10 %, because oxidation of CH_4 strongly counteracted CH_4 emission through diffusion.

Pa	ge 35: [9]	5: [9] Deleted Sina Berger							9/14/17 2:36:00 PM			
Site	Cover			Peat age	<u>pH</u>		stoch. Ratios					
	Sphagn.	Gram.	Shrubs	at 60 cm	at surface	at 75 cm	C/N	C/P	N/P	C/Ca	C	
	[%]	[%]	[%]	[years]								
1	100	10	8	70 (± 21)	3.6	3.7	52.6 ± 4.7	1207.2 ± 85.9	35.1 ± 0.9	129.1 ± 3.4	926.0	
2	100	30	5	50 cm: 135	4.3	3.7	47.4 ± 6.4	1256.4 ± 50.2	41.8 ± 2.5	99.1 ±11.0	1228	
3	100	35	4	148 (± 45)	3.8	3.8	51.7 ± 2.7	1403.0 ± 123.0	37.6 ± 2.8	146.5 ± 18.1	1063	
4	60	10	30	184 (± 17)	4.0	4.1	41.5 ±4.2	1342.6 ± 244.7	45.7 ±3.4	187.8 ± 11.0	962.	