Authors' response to referee's comments (RC1) on the manuscript, "Organic matter and sediment properties determine in-lake variability of sediment CO2 and CH4 production and emissions of a small and shallow lake" by L.S.E. Praetzel et al.

Dear reviewer,

thank you very much for your comments and suggested improvements on our manuscript "Organic matter and sediment properties determine in-lake variability of sediment CO2 and CH4 production and emissions of a small and shallow lake". We are sure that consideration of your raised points and ideas will considerably improve the manuscript and therefore we will especially focus on revising the discussions part. Please find our responses to each of your comments below, they will be structured as follows:

(1) comments from referee, (2) author's response, (3) author's changes in manuscript.

We hope to be able to answer your open questions and to adequately include your suggestions.

Kind regards,
Leandra Praetzel & Co-Authors

Major points:

(1) 1.L.79-81: The Gibbs free energies given in the ms are either not found in the quoted literature (Whiticar 1999) or are different (Conrad 1999). I assume the reason is that they were calculated using energies of formation for gases in dissolved rather than in gaseous state. This would be consistent with the Nernst equations mentioned later (L.250) also probably using gas concentrations rather than partial pressures. However, the authors should clarify the procedures.

(2) Gibb's free energies were calculated using formation energies Gf0 for the elements involved in the reactions in aqueous state. Used Gf0 values are reported by Stumm & Morgan 1996 and Nordstrom & Munoz 1994. In the Nernst equation, dissolved and gaseous concentrations in incubation vials were used for calculations. This will be mentioned in the revised manuscript.

We will include the following explanation on calculation of delta Gr0 in the revised manuscript:

(3) "Values for Δ Gr0 are calculated from standard formation energies Δ Gf0 at 25°C in aqueous state listed in Stumm and Morgan (1995) and Nordstrom and Munoz (1994)."

(1) 2.L.187-200: There are no isotopic data reported, therefore the description of IRMS methodology is not necessary.

(2) delta 13C and delta 15N values are reported in Table 2 and will be discussed in more detail in the results section of the revised manuscript. IRMS was also used to determine mass contents of C and N. The authors therefore prefer to leave the methodology description in the manuscript.

(3) "C and N isotopic values did not vary much between sites and were on average -27.6 ‰ and -0.9 ‰ respectively with only one outlier for δ15N at site 3.50 (-6.3 ‰)."

"Neither CO2 nor CH4 production rates were exhibited significant correlation with C content or, C/N ratio, δ13C or δ15N,…"

(1) 3. Table 2: L.411-412 mentions strong FTIR absorption features of polysaccharides. However, this compound class is not listed in the Table.

(2) Absorption maxima for polysaccharides will be listed in Table 2 of the revised manuscript.

(1) 4.L.422-425. This is an overview of measured rates. However, the numbers seem to be slightly different from those shown in Fig.2. Although there is probably a reasonable explanation for these differences, I found it confusing. In fact I would be happy just looking at the data in the figure without reading the text. However, one could mention that the rates decreased from the shore to the centre, since this point is later relevant in the Discussion.

(2) We double-checked the accordance of production rates stated in the text and displayed in Figure 2. We confirm that values in the text are the same as in the figure. We note that production rates in the text are mean values, whereas bold lines of boxplots are median values. We suppose that this fact might have caused the perceived discrepancy.

Moreover, we will simplify the figure by removing data from 5-10 cm depth and instead include this data in the supporting information.

We will further add a sentence to emphasize that production rates increased from the center to the shore as follows:

(3) “Overall, production rates decreased from the shore to the center of the lake.”

(1) 5.L.477-487: Here applies the same as in point 4. The data in the text seem to slightly different from those seen in Fig. 7.

(2) See also answer to point 4. We double-checked the data and confirm that the values in text and figure are the same. Please note that the stated values in the text are mean values +/- standard deviations of triplicate measurements at the respective sites where minima and maxima were measured whereas boxplots in the figure represent the whole dataset at each sampling date.

(1) 6.L.507: Again the data in the text seem to slightly different from those seen in Fig. 8.

(2) We will adjust the mentioned values in the text according to the displayed flux rates in Fig. 8 as follows:

(3) “CO_2 and CH_4 fluxes measured from intact sediment core incubations ranged from 10.8 ± 4.4 to 17.9 ± 2.0 and 0.02 ± 0.01 to 1.5 ± 2.6 mmol m^{-2} d^{-1} respectively.”

(1) 7. The discussion is too wordy and should be focused to the really novel results. I also recommend a different structure for the Discussion. I think it is not ideal having individual chapters on spatial variability of OM quality, spatial variability of CO2 and CH4 production rates, and influence of OM quality on gas production, since such structure results in too much repetition and also is not very suitable for explaining gas production rates on the basis of OM quality.

(2) We will shorten the discussion and focus on novel results in the revised manuscript. The chapters 4.1.1, 4.1.2 and 4.1.3 will be summarized as one, entitled “4.1.1 Variability of CO2 and CH4

production rates and influence of OM". Like this, we will considerably shorten the paragraph and thus avoid repetitions.

(1) 9. The discussion on temperature effects can be much shorter, since it is rather well reported in the literature.

(2) In the discussion on temperature effects, we emphasize the sensitivity of small lakes to temperature changes compared to large lakes. We will shorten that section in the revised manuscript but prefer to leave that statement in the discussion, as this strong and variable temperature effect on production rates has, to our knowledge, not been shown before for small and shallow lakes.

(3) "In accordance with previous studies (den Heyer and Kalff, 1998; Sobek et al., 2009; Gudasz et al., 2015), we found that with a temperature increase of 10°C, production rates of CO2 doubled and those of CH4 were 2 to 11 times higher. Q10-values for CO2 were thus within the range of earlier reported values by Liikanen et al. (2002) and Berström et al. (2010), whereas Q10-values for CH4 production were slightly higher than values found by Duc et al. (2010). The large observed range of Q10-values, especially for CH4, implies that responses to temperature increases might not be homogeneously distributed within a lake. We point out that sediment CH4 production is more sensitive to increasing temperatures compared to CO2 production and that this leads to a stronger feedback on global warming when considering the higher global warming potential of CH4 compared to CO2 (Marotta et al., 2014). The observed negative correlation between Q10-values and FTIR peak ratios further suggests that sites with more labile OM are more susceptive to increasing temperatures in terms of CH4 production, whereas at sites with more recalcitrant OM, this recalcitrant OM may limit the degradation processes. We therefore assume that sediment greenhouse gas production in small and shallow lakes might in the course of global warming increase to a larger extent than in deeper lakes, as shallow waters, compared to deeper lakes, do not get thermally stratified in summer and therefore shallow sediments warm much faster (Jankowski et al., 2006)."

(1) 10. The discussion of methanogenic pathways (L.648-680) is not really relevant, since the data just show that both methanogenic pathways were exergonic and thus, could well operate. Everything else is speculation and not relevant. The magnitude of the Gibbs free energy does not allow to conclude whether the one pathway is more prevalent than the other. One could however discuss the correlation of the concentrations of H2 and acetate, and the respective Delta G, with sediment OM quality, since correlations were reported in the Results.

(2) We will shorten the discussion of methanogenic pathways and increase the focus on the reported relationships between Gibb's free energies and OM quality in the revised manuscript.

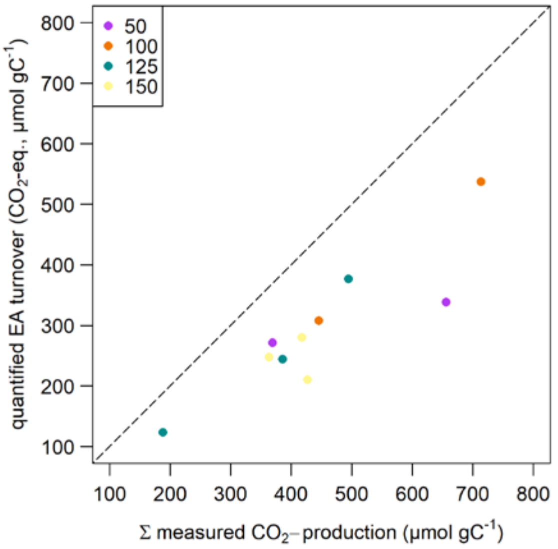
(3) "[…] suggesting that both pathways could contribute to CH4 production during the whole experiment. Still, this approach does not allow to evaluate which of the pathways predominates."

"[…] we would have expected a reverse pattern. Concomitantly, Gibb's free energy of hydrogenotrophic methanogenesis exhibited significant positive correlations with some FTIR peak ratios, although we expected that a high abundance of recalcitrant OM compounds would make hydrogenotrophic methanogenesis more feasible (Miyajima et al., 1997). Acetate and H2 concentrations on the other hand, both exhibited significant negative correlations with some FTIR peak ratios. While this seemed reasonable for acetate concentrations (less acetate available in strongly decomposed OM), this result again proved to be against our expectations in terms of H2 concentrations. One reason for these ambiguous findings might be that the system was not in a

steady state in terms of thermodynamic conditions. Similarly, long time scales of experiments still not reaching such steady state have also been observed earlier for peat columns (Bonaiuti et al., 2017).”

(1) 11. The discussion of alternative electron acceptors (L.682-698) is rather short. The authors only discuss correlations. They miss the chance to discuss stoichiometric relations of reduced EAC with the amounts of CO2 production. Although such mass comparisons apparently have recently been done by other members of the Knorr group (Gao et al. 2019), they would also be interesting for this particular lake. I have the impression that the magnitudes of reducible EACs might explain the CO2 production in the beginning of the incubations, when rates of CO2 production were larger than those of CH4 production, while methanogenic decomposition of OM should result in equal rates. I wonder why this point is not addressed.

(2) In the revised manuscript, we will, besides observed correlations, also discuss the stochiometric relationships between measured EAs and CO2 production: We find that calculated CO2 production from prevalent EAs is lower than the measured CO2 production (see. Fig.). We propose this is due to unknown consumed EAs during the incubation that we did not capture; most likely iron in the solid phase. We indeed measured solid phase iron, but are not able to make statements about its speciation. But starting from the total Fe concentration in the solid phase (2-3%), we suppose that this is high enough to explain the missing EAs to reach a 1:1 ratio of measured CO2 production and calculated EA turnover.



(1) 12. I noticed that lake sediments were anoxically preincubated for either one week (L:178) or 50 days (L.331). Please clarify! Anyway, the preincubation might have depleted most of the reducible iron and sulfur compounds. This may be the explanation for the low values of EACinorg (Fig. 7), but is not discussed.

(2) The incubations were pre-incubated for one week, data from the sediment mesocosms was only used for analysis after 50 days of deployment in the climate chamber, in order the leave the cores adapt to laboratory conditions. We verified steady state conditions via the gas concentration in the sediment profile and observed constant conditions (i.e. no increase of concentrations) after 50 days. We will give a more detailed explanation on the procedure concerning sediment mesocosms in in the revised manuscript:

(3) "For statistical analyses and discussion, we only used measurements that were made >50 days after the deployment of the intact sediment core incubations in the climate chamber. This was done in order to ensure the system had adapted to experimental conditions and had reached a steady state. Steady state conditions were indicated by quasi-constant CO_2 and CH_4 concentrations in the sediment."

(2) We will additionally change the subtitle of the sections to clarify the difference between incubations and sediment mesocosms:

(3) 2.3 Intact sediment core incubations

2.3.1 CO_2 and CH_4 fluxes

2.3.2 Sediment gas stock change

(2) Further, as suggested by the other reviewer, we will rename the sediment incubations to "slurry incubations" and the sediment mesocosms to "intact sediment core incubations".

(2) The low EACinorg values due to 1 week of preincubation will be discussed in the revised manuscript.

(3) "Nevertheless, both absolute EAC_{inorg} values as well as relative changes were very low, which might have been caused by the one-week preincubation, where most of the reducible inorganic compounds might have already been depleted."

Minor:

(1) 1.L.28: what means 'sufficiently' ? rho=0.65 is sufficient? Would rho=0.6 also be sufficient. Is there an objective criterion for sufficiency?

(2) The word sufficiently will be removed from the text.

(1) 2.L.30-32. I cannot follow the argument of this sentence. I suggest rephrasing.

(2) The sentence will be rephrased in the revised manuscript as follows:

(3) "Our results show that within a small lake, CO2 and CH4 production show significant spatial variability, which is mainly driven by spatial differences in the degradability of the sediment OM."

(1) 3.L.67: cellulose is also a polysaccharide. I suggest rephrasing.

(2) Cellulose will be replaced by natural organic matter in the revised manuscript.

(1) 4.L.83: The Delta G-zero of hydrogenotrophic methanogenesis is more negative than of aceticlastic methanogenesis. Therefore the acetoclastic pathway is less (not more) energetically favorable.

(2) This will be corrected in the revised manuscript.

(1) 5.L.91: EAC has not yet been defined. Please check also for other abbreviations.

(2) This will be corrected in the revised manuscript. We will also check for other abbreviations and make sure that they will be defined when being used the first time and that afterwards, abbreviations will be used consistently throughout the whole manuscript.

(1) 6.L.107-109: The ‘4’ in CH4 as superscript

(2) This will be corrected in the revised manuscript.

(1) 7.L.168: 12 locations; please harmonize with the 13 sampling sites mentioned in the legend of Fig.1.

(2) Samples for incubations were taken from 12 sampling site, samples for sediment mesocosms were taken from 4 sites, whereas one of the mesocosms (S.150) represents the three sites of 150 cm depth from the incubations (1.150, 2.150 and 3.150) so that there are in total 13 sampling sites. This will be explained in more detail in the caption of Fig. 1 as follows:

(3) “Location of the study area and 13 sampling sites within Lake Windsborn. Rhombus: Sampling sites for slurry incubations and intact sediment core incubations, circles: sites for slurry incubations only, asterisk: site for intact sediment core incubation only (reference for 1.150, 2.150 and 3.150).”

(1) 8.L.207: ‘relative abundance’ compared to what?

(2) FTIR absorption peaks show relative abundances of the corresponding functional moieties in a single sample compared to another moiety. We therefore calculate peak ratios normalized to the polysaccharide peak. The sentence will be changed as follows in the revised manuscript:

(3) “Distinct peaks at specific wavelengths were assigned to functional groups according to Artz et al. (2008) and normalized to the peak intensity at 1031 - 1035 cm-1 (indicative of polysaccharides) in order to obtain inter-comparable peak-ratios of functional moieties in all samples as FTIR spectra only provide information about the relative abundance of certain functional moieties in one sample.”

(1) 9.L.268: EAC/EDC: I think you mean EAC & EDC rather than the ratio between both. I found similar possible confusions at many places in the text (e.g., L.293, L.369, 370, 383 and in the labels of Fig. 7. Please check carefully.

(2) Notations will be changed in the revised manuscript as suggested by the reviewer.

(3) (EAC_{OM}, and EDC_{OM})

EAC_{OM} (EDC_{OM})

$$\Delta CO_2 = ((c(CO_2)_{end} * V_{seg}) - (c(CO_2)_{start} * V_{seg})) / \Delta t \quad (11)$$

$$\Delta CH_4 = ((c(CH_4)_{end} * V_{seg}) - (c(CH_4)_{start} * V_{seg})) / \Delta t \quad (12)$$

ΔCO_2 (ΔCH_4)

total C and N

Fig. 7 label: EAC &EDC ($\mu mol\ e^- gC^{-1}$)

(1) 10. L.299. The reference Tamura et al. (1974) only describes the analysis of Fe(II) (albeit in the presence of Fe(III)). How was Fe(III) analyzed?

(2) Fe (III) in the samples was reduced to Fe (II) with 10% ascorbic acid and determined likewise. The procedure will be explained in the revised manuscript as follows:

(3) "Because 1,10-phenanthroline can only detect Fe^{2+}*, the* Fe^{3+} *in the samples was reduced to* Fe^{2+} *with 10% ascorbic acid. Then, the determined concentration of total Fe was used to calculate the concentration of* Fe^{3+} *in the samples."*

(1) 11. L.477-479: I cannot follow this sentence. Also compare major point 6 above. Please also note, that Fig. 7 is not mentioned in the text, and that Figure number should be exchanged with that of Fig. 6, since Fig.6 is reported later in the text than Fig. 7.

(2) The order of the figures will be changed and mentioning and numbering of figures will be adjusted in the revised manuscript. The sentence in ll. 447-449 will be changes as follows:

(3) "EAC_{OM} *lay between 218.69 ± 97.15 and 545.71 ± 60.33 µmol* e^- gC^{-1} *at t0 and decreased on average by 44.85 µmol* e^- gC^{-1} *until t6. Highest values for* EAC_{OM} *were found at site 3.125 corresponding to lowest measured* CH_4 *production rates at that site."*

(1) 12.Table 3: Showing the time line as t0, t1, t2 etc. is awkward, since one has to consult the explanation in the methods section. I suggest listing the actual time points, i.e. 0, 1, 3 etc. days.

(2) The captions in Table 3 will be changed as suggested by the reviewer.

(1) 13. Table 4: The numbers in the table show too many decimal positions. Please report only those that are significant. In fact, at numerous places in the text numbers seem to show non-significant decimal positions. Please check and correct.

(2) The numbers in Table 4 as well as other number with non-significant decimal positions will be changed as suggested by the reviewer.

(3)

	CH_4 flux			**CO_2 flux**		
	rho	**p**	**n**	**rho**	**p**	**n**
Clay	0.648	< 0.05	12	0.605	< 0.05	12
Silt	0.497	*n.s.*	12	0.302	*n.s.*	12
Sand	-0.648	< 0.05	12	-0.605	< 0.05	12
Fats, waxes, lipids	-0.833	< 0.05	8	-0.333	*n.s.*	8
Phenols; humics	-0.833	< 0.05	8	-0.357	*n.s.*	8
Aromates	-0.595	*n.s.*	8	-0.524	*n.s.*	8
Lignin	-0.786	< 0.05	8	-0.381	*n.s.*	8
C/N	-0.881	< 0.01	8	-0.333	*n.s.*	8
C (%)	-0.714	*n.s.*	8	-0.190	*n.s.*	8
CH_4 sediment stock change	-0.222	*n.s.*	41	0.05	*n.s.*	35
CO_2 sediment stock change	-0.049	*n.s.*	41	-0.064	*n.s.*	35

(1) 14.L.535, 538: Should be Table 4 rather Table 5.

(2) The numbering will be changed in the revised manuscript.

(1) 15. References. Some of the references use capital letters for the titles.

(2) Capitals will be changed according to the reviewer’s suggestion.

Authors' response to referee's comments (RC2) on the manuscript, "Organic matter and sediment properties determine in-lake variability of sediment CO2 and CH4 production and emissions of a small and shallow lake" by L.S.E. Praetzel et al.

Dear reviewer,

Thank you very much for your comprehensive comments on our manuscript "Organic matter and sediment properties determine in-lake variability of sediment CO2 and CH4 production and emissions of a small and shallow lake". We appreciate your suggestions both in form and content and believe that they will substantially improve the paper. In the following, we will outline our responses to your comments one by one and thereby hope to clarify open questions and incorporate your suggestions to your satisfaction.

Each answer will be structured as follows:
(1) comments from referee, (2) author's response, (3) author's changes in manuscript.

Kind regards,

Leandra Praetzel & Co-Authors

General

(1) The manuscript needs careful line editing to take care of non-idiomatic English. An example is the frequent usage of wrong tenses (e.g. in line 60: "is mainly depending on" rather than "mainly depends on"). The authors may seek help from a native English speaker for this purpose. I have pointed out a few instances below, but these are by no means exhaustive. I must also concede that I am not a native English speaker!

(2) We asked a native speaker for help and feel certain that her corrections will substantially improve the grammatical style of the paper.

Specific

(1) Lines 14-15: Change "... to the atmosphere, following recent studies this is particularly the case for small and shallow lakes." to "... to the atmosphere; recent studies have shown that this is particularly the case for small and shallow lakes."

(2) The sentence will be rephrased in the revised manuscript as follows:

(3) “Inland waters, particularly small and shallow lakes, are significant sources of carbon dioxide (CO2) and methane (CH4) to the atmosphere.”

(1) Line 16: Delete "yet" and "thus".

(2) The terms will be deleted in the revised manuscript.

(1) Lines 21-22: Change "... were significantly negative ($p<0.05$, rho<-0.6) correlated" to "... exhibited significant negative correlation ($p<0.05$, rho<-0.6)". Please make similar changes elsewhere.

(2) The sentence will be rephrased according to the reviewer’s suggestion. Similar changes will be made elsewhere in the revised manuscript.

(1) Lines 32-34: The last sentence states the obvious. Who has suggested such a "replacement"?

(2) Some studies implicitly equal production and emission rates, e.g.

Grasset et al. 2018: doi: 10.1002/lno.10786

Sollberger et al. 2014: https://doi.org/10.1007/s00027-013-0319-2

We will adjust the statement in the revised manuscript as follows:

(3) “We highlight that studies on production rates and sediment quality need to be interpreted with care in terms of deducing emission rates and patterns as it this neglects physical sediment properties and production and oxidation processes in the water column.”

(1) Line 52: Change "has been" to "have been" (here majority is plural), and remove "is".

(2) The term will be changed in the revised manuscript and “is” will be deleted.

(1) Line 56: Remove hyphen between "in" and "lake".

(2) The hyphen will be removed in the revised manuscript.

(1) Lines 58-59: Why is it crucial? Your results show that it is not.

(2) The sentences will be rephrased in the revised manuscript as follows:

(3) "Nevertheless, anoxic sediments are important for whole lake C cycling as the CO2 and CH4 produced there can be released through the water column to the atmosphere. To understand the spatial patterns of CO2 and CH4 emissions, it is therefore of interest to also understand CO2 and CH4 production processes in the sediment as well as their major controls."

(1) Line 64: Also its origin (e.g. lignin).

(2) This will be added in the revised manuscript as follows:

(3) "…and therefore its origin and degree of decomposition…"

(1) Line 74: Remove "being".

(2) The term will be removed in the revised manuscript.

(1) Line 82: As also pointed out by the other referee a more negative deltaG change would make R2 thermodynamically more favourable.

(2) The statement will be corrected in the revised manuscript.

(1) Lines 86-87: Change "are attributed to" to "may arise from".

(2) The phrasing will be changed in the revised manuscript.

(1) Line 90: Change "remain" to "remains".

(2) The term will be corrected in the revised manuscript.

(1) Lines 92-93 and elsewhere: As also pointed out by the other referee please define each abbreviation when you use it the first time and maintain consistency.

(2) All abbreviations will be defined when being used for the first time and only abbreviations will be used afterwards in the revised manuscript.

(1) Line 96: Why "to a small extent"? In such shallow systems wind-driven turbulence could disturb the sediments. Lines 97-98: Add "penetration" after "oxygen" and remove "in our case". What do you mean by "perennial circulation".

(2) The sentence will be rephrased in the revised manuscript as follows:

(3) "…but might in the upper parts of the sediments be influenced by oxygen penetration from the water column due to a well-mixed water body."

(1) Line 99: Please use present indefinite tense, not present continuous.

(2) The tense will be corrected in the revised manuscript.

(1) Line 103: "other" sediment properties?

(2) The term "other" will be added in the revised manuscript.

(1) Line 108: Change "is accountable" by "accounts".

(2) The term will be changed in the revised manuscript.

(1) Lines 110-114: Please rephrase this sentence.

(2) The sentence will be rephrased as follows:

(3) Until now, laboratory incubations of lake sediments were mostly conducted with samples from one or few sites within one lake with a focus on comparing different lakes with each other rather than covering a high in-lake variability of production rates. Further, these studies emphasize temperature effects on production rates (Duc et al., 2010; Gudasz et al., 2010; Gudasz et al., 2015; Fuchs et al., 2016). Unlike peat soils, where a broad range of controls on CO2 and CH4 production has been investigated, to our knowledge, controls such as organic matter (OM) quality, the occurrence of alternative electron acceptors (EAs), thermodynamic processes and sediment grain size have not, or only individually, been systematically surveyed in small lakes.

(1) Line 119: Did you actually investigate "connected productions patterns to OM"?

(2) This sentence might be ambiguous. We will rewrite the sentence as follows:

(3) *"…in order to relate observed production patterns to measured OM and sediment characteristics, thermodynamics, and water-atmosphere fluxes."*

(1) Line 121 and elsewhere: I am not sure if these experiments can be termed as "mesocosm". These were incubations of cores in the lab.

(2) To clarify the experimental procedure and the differences between the two laboratory experiments, we changed the descriptions throughout the whole manuscript: *Sediment incubations* to *slurry incubations* and *Sediment mesocosms* to *Intact sediment core incubations*.

(1) Line 125: Change "hypothesize" to "hypothesized".

(2) The term will be corrected in the revised manuscript.

(1) Line 137: Change "blast" to "blasted".

(2) The term will be corrected in the revised manuscript.

(1) Line 138: Change "arose" to "formed".

(2) The term will be corrected in the revised manuscript.

(1) Figure 1 captions: Technically the depth categories are wrong. For example by <150 cm, you imply depths between 125 and 150 cm, but 20 cm is also <150 cm. This should be clarified (e.g. 125 indicates 100cm<depth_125 cm).

(2) The description of lake depths categories will be clarified in the revised manuscript as follows:

(3) "…numbers 50, 100, 125 and 150 indicate lake depth category (50: <50 cm, 100: 50-100 cm, 125: 100-125 cm, 150: 125-150 cm)."

(1) Table 1, caption: Analytical procedures do not have to be mentioned here; they should be described in methodology section.

(2) The description of analytical procedures will be deleted. These can be found in the methods section 2.4 of the revised manuscript.

(1) Line 167: Change "at three occasions" to "on three occasions".

(2) The term will be changed in the revised manuscript.

(1) Line 169: Why randomly? It should be selectively based on a reason.

(2) We randomly chose the sites in order to avoid detecting differences between sites, water depths or transects due to the sampling date. E.g. if we took all four samples from one transect on the same sampling date, we could not have been sure that potentially observed differences in production rates were because of different site characteristics or because of the sampling date. The same would be true for water depths, so we decided to perform a random sampling at each date.

(1) Line 172: Add "respectively" at the end of the sentence.

(2) The term will be added in the revised manuscript.

(1) Line 176: Change "added with" to "containing".

(2) The term will be changed in the revised manuscript.

(1) Line 180: Change "stored" to "maintained".

(2) The term will be changed in the revised manuscript.

(1) Line 192: Referring to a comment by the other reviewer, I note that some isotope data are presented in Table 2 (not for sulphur though), although not at all discussed in the text. It is not clear whether the sample was decalcified. Also what was the reproducibility of measurements? In fact the precision of analysis is not given for any parameter.

(2) Isotopic values of C and N will be mentioned in the revised manuscript in the results sections 3.1.1 and 3.1.2.

Sulphur isotope data was not measured an will therefore be deleted from the methods section.

The samples were not decalcified before analyses. But we analyzed samples for carbonate content which confirmed that carbonate contents were very low (< 0.9 mg/g). We therefore assume that carbonates in the samples only have a minor influence on the results of isotopic data.

During every run of samples, multiple working standards were measured to assure reproducibility of measurements. Precision of standards are: < 1% for C, < 0.1 % for N, < 0.05 ‰ for delta13C, < 0.5 ‰ for delta15N. Information on precision will be added to the methods section 2.2.2 of the revised manuscript.

(1) Line 201: Change "Therefore" to "For this purpose".

(2) The term will be changed in the revised manuscript.

(1) Line 253: Something missing in the sentence.

(2) The term will be corrected in the revised manuscript.

(1) Line 267: Change "analyzed for" to "measured".

(2) The term will be corrected in the revised manuscript.

(1) Lines 274, 291: Change "measured" to "analyzed". (Note samples are analyzed, parameters are measured).

(2) The term will be corrected in the revised manuscript.

(1) Line 301: Change "Therefore" to "For this purpose".

(2) The term will be corrected in the revised manuscript.

(1) Line 313: These are lab experiments, NOT mesocosms!

(2) See comment on line 121.

(1) Line 331: Change "conducted" to "made".

(2) Will be changed in the revised manuscript. Please note that the whole sentence should be rephrased in order to the other reviewer's suggestion as follows:

(3) "For statistical analyses and discussion, only measurements that were made > 50 days after the deployment of the sediment mesocosms in the climate chamber were used. This was done in order to ensure the system had adapted to experimental conditions and had reached a steady state. Steady state conditions were indicated by quasi-constant CO2 and CH4 concentrations in the sediment."

(1) Line 361: There is no other way to quantify inputs is ebullition?

(2) The only way to directly quantify ebullition is via inverse funnels that could trap the emitted methane bubbles. We tested this method in our sediment cores, but without success so that we decided to only measure total methane fluxes and separate diffusive and ebullitive fluxes mathematically as this has been suggested by Bastviken et al. 2004 and adapted by many others when measuring in-situ methane fluxes with a floating chamber approach.

(1) Line 408: Change "nor" to "or".

(2) The term will be changed in the revised manuscript.

(1) Fig. 2: Figure difficult to digest. I could not follow "Different letters indicate significant differences between these sites." What do letters "a"-"d" mean? Am I missing anything?

(2) For more clarity, the figure will be split into two and production rates in 5-10 cm depth will be displayed in the supporting information.

a-d denote if there are significant differences between sites. Same letters mean no significant differences. The description will be rephrased in the revised manuscript as follows:

(3) "Identical lowercase letters indicate production rates that were not significantly different (i.e. $p > 0.05$) from each other."

(1) Line 483: Change "by averagely" to "on a average by".

(2) The term will be changed in the revised manuscript.

(1) Lines 490, 493, 499: See earlier comments on Lines 21-22.

(2) The sentences will be rephrased according to the above-noted suggestion.

(1) Line 508 and elsewhere: I believe sediment ebbulition in inferred from k>o. I am not sure. Was there any bioturbation that could increase the emission?

(2) Ebullition was inferred from piston velocity $k > 2$ as described in the methods section 2.3 (see lines 359-361 of the submitted manuscript).

We did not observe any bioturbation during the experiment.

(1) Table 4: "n.s." presumably means not significant ($p<0.05$). Is is mentioned somewhere? Significance also depends on the number of values that are not given.

(2) n.s. means not significant. An explanation will be added to the table’s description as follows:

(3) “n.s. means that correlations were not significant ($p > 0.05$).”

We will revise the table including number of values as follows:

	CH_4 flux			**CO_2 flux**		
	rho	**p**	**n**	**rho**	**p**	**n**
Clay	0.648	< 0.05	12	0.605	< 0.05	12
Silt	0.497	*n.s.*	12	0.302	*n.s.*	12
Sand	-0.648	< 0.05	12	-0.605	< 0.05	12
Fats, waxes, lipids	-0.833	< 0.05	8	-0.333	*n.s.*	8
Phenols; humics	-0.833	< 0.05	8	-0.357	*n.s.*	8
Aromates	-0.595	*n.s.*	8	-0.524	*n.s.*	8
Lignin	-0.786	< 0.05	8	-0.381	*n.s.*	8
C/N	-0.881	< 0.01	8	-0.333	*n.s.*	8
C (%)	-0.714	*n.s.*	8	-0.190	*n.s.*	8
CH_4 sediment stock change	-0.222	*n.s.*	41	0.05	*n.s.*	35
CO_2 sediment stock change	-0.049	*n.s.*	41	-0.064	*n.s.*	35

(2) Numbers of values for other calculated correlations will be given in the Supporting information Tab. S1.

(1) Lines 528: Change "concentration" to "concentrations" and "was" to "were".

(2) The term will be changed in the revised manuscript.

(1) Line 536: Change "were significantly negative correlated" to "showed significant negative correlation"

(2) The term will be changed in the revised manuscript.

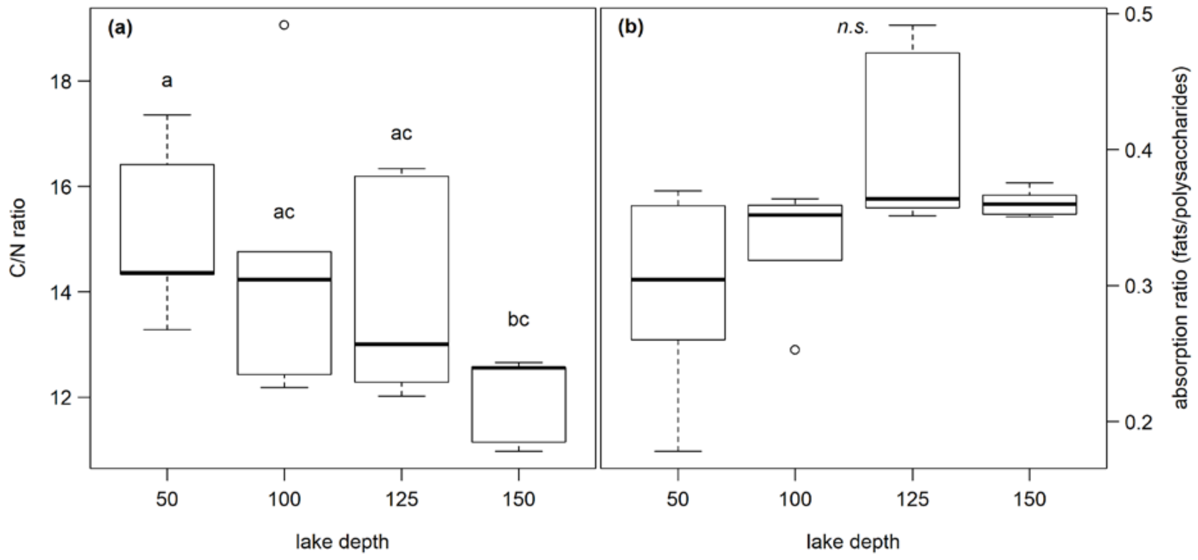
(1) Line 542: What do you mean by "narrower"? lower?

(2) Narrower will be changed to *lower* in the revised manuscript.

(1) Lines 545-556: Authors have emphasized on C/N ratio. They have observed increase in C/N with depth in the inner part of the lake. C/N ratio may not be a very efficient parameter to characterize organic source owing to rapid remobilization of nitrogen as well as reabsorption of ammonium on particulates. The paragraph 405 “The C content in the samples was between 2.15 and 33.16% with lowest values at site 3.50 and highest at site 1.50. C/N ratios ranged from 10.97 at site 1.150 to 19.06 at site 3.100. Neither C content nor C/N ratio showed significant changes with sediment nor lake depth, but C/N ratio was significantly higher in samples taken close to the shore (50) than in samples from the lake center (150) ($p < 0.01$).” is very confusing. A graph showing distribution of C/N ratio across the horizontal length of the lake would suitable to comprehend the results better.

(2) We will include a figure showing C/N ratios and absorption ratios for fats/polysaccharides to better illustrate our results.

(3)



(1) Line 551-552: I do not believe in shallow depths it matters.

(2) We additionally propose the mechanism of resuspension and focusing of small particles, that could alter the degree of decomposition of OM.

(3) *"…As this process might not be of the same importance in shallow lakes compared to deeper lakes, we additionally suggest that the more decomposed OM in the lake center might have undergone degradation processes during resuspension and focusing of small particles as a result of wind-induced bed-shearing (Mackay et al., 2011)."*

(1) Line 559: Change "buried" to "getting buried"

(2) The term will be changed in the revised manuscript.

(1) Line 571: Change "role for" to "role in"

(2) The term will be changed in the revised manuscript.

(1) Line 578: Change "e.g." to "among other things"

(2) The term will be changed in the revised manuscript.

(1) Line 587: Change "in the following" to "below"

(2) The term will be changed in the revised manuscript.

(1) Line 591: Remove "the" before "in other studies"

(2) The term will be removed in the revised manuscript.

(1) Lines 612-614: Laborious sentence. All you are saying is that such shallow depths do not get thermally stratified in summer.

(2) We will rewrite the sentence as follows:

(3) "…especially regarding the fact that shallow waters, as against deeper lakes, do not get thermally stratified in summer and therefore shallow sediment warm much faster (Jankowski et al., 2006)."

(1) Lines 619-620: Change the tense to present indefinite.

(2) The term will be changed in the revised manuscript.

(1) Line 627-630: What do you mean by "wider" and "narrower"? I do not follow this sentence.

(2) "Wider" and "narrower" will be changed to "higher" and "lower". We will restructure the whole discussions part about OM quality and CO2 and CH4 production rates according to the other reviewer's suggestions. To explain what we mean by this statement: C/N ratios can be interpreted in two ways: a) high C/N ratios = low decomposition state, low C/N ratios = high decomposition state; b) but C/N ratio can also be used to differentiate between OM of terrestrial and aquatic origin (see Meyers 1994) whereas high ratios = terrestrial and low ratios = aquatic origin. It is known that OM of a low decomposition state is easier degradable for microorganisms and therefore leads to higher production rates of CO2 and CH4, but on the other hand, aquatic OM is usually easier degradable for aquatic microorganisms and would therefore lead to higher production rates compared to OM of terrestrial origin (see e.g. Grasset et al. 2018). We therefore conclude that, although there exist two contradicting effects (low vs. high decomposition or aquatic vs. terrestrial origin), the fact that OM closer to the shore is in a lower decomposition state (although it is probably of terrestrial origin) fuels CO2 and CH4 production. The paragraph on C/N ratio and production rates will be revised as follows:

(3) "C/N ratios are frequently used to characterize the degradation state of OM, but we did not find correlations between C/N ratios and CO2 and CH4 production rates in the slurry incubations. Although OM of autochthonous origin was found to fuel higher degradation rates than allochthonous OM (West et al., 2012; Grasset et al., 2018) we found evidence of predominant inputs of allochthonous (terrestrial) material at sites with higher production rates close to the shore (higher C/N ratios), whereas sites with lower production rates in the lake center received mainly autochthonous (aquatic) OM as indicated by lower C/N ratios (Meyers, 1994). On the other hand, high C/N ratios also indicate a lower degradation state and therefore higher degradation potential whereas low C/N ratios are usually typical of highly decomposed OM having a lower CO2 and CH4 production potential (Malmer and Holm, 1984; Kuhry and Vitt, 1996). These two possibilities of interpreting C/N ratios might be the reason for apparently contradicting findings and the missing relationship between C/N ratios and CO2 and CH4 production rates."

(1) Line 637: Change the tense to present indefinite.

(2) The term will be changed in the revised manuscript.

(1) Line 654: But the acetate concentration increased!

(2) Please not the discussion in section 4.1.4 of this observation in lines 660-665 of the submitted manuscript.

(1) Line 655: Remove "of" before "importance"

(2) The term will be changed in the revised manuscript.

(1) Line 671: OM quality is not quantified so instead of low you should perhaps use poor.

(2) The term will be changed in the revised manuscript.

(1) Line 673: Change "of energy" to "in energy"

(2) The term will be changed in the revised manuscript.

(1) Line 675: Change "... acetate, but rather is fermentation" to " ... acetate. Instead fermentation may be rate limiting"

(2) The term will be changed in the revised manuscript.

(1) Line 677: Bring "Further" before "it".

(2) The term will be changed in the revised manuscript.

(1) Line 678: Change "finding emphasizes" to "supports"

(2) The term will be changed in the revised manuscript.

(1) Line 692: If the relationship was insignificant the trend cannot be "clear".

(2) The term will be removed in the revised manuscript.

(1) Line 693: Not at all clear, and so is the following conclusion. I find this whole paragraph speculative.

(2) We will rewrite the whole paragraph 4.1.5 - also following the other reviewer's suggestions – where we will elaborate the relationships between CO2 and CH4 production and alternative EAs more precisely. Instead of discussing relationships between EAC and CH4 production, we emphasize that measured inorganic and organic EAs can explain 40-80% of measured CO2 production. The missing capacity can probably be explained by solid-phase iron, which we found ranging from 2 to 3 %, but whereof we do not have information on its speciation.
We further emphasize, that missing correlations between EAC and CH4 production are due to our experimental set-up: the one-week pre-incubation might have already depleted a large amount of reducible organic and inorganic EAs so that subsequent changes and therefore correlations were low.

(1) Line 702: Change "something" to "somewhat"

(2) The term will be changed in the revised manuscript.

(1) Line 706: Change "approaches" to "factors"

(2) The term will be changed in the revised manuscript.

(1) Line 730: Authors attempt to correlate ebullition with grain size. They believe that higher sand content leads to lesser ebullition. Which is highly unlikely since ebullition depends on permeability of sediments and not porosity. Sand always has higher permeability than silt and clay although lesser porosity. You need to elaborate your concept with more clarity

(2) When explaining CH4 ebullition with the concept of grain size distribution/porosity, it is not primarily of importance how permeable the material is, but how effective bubbles can actually accumulate in the sediment (so that they can subsequently be released). Lui et al. 2018 found that the dominant pathway of bubble formation is by displacing the surrounding sediment, and that this is easier in soft, silty sediment compared to sandy sediments. This sediment displacement would lead to more macropores and therefore a higher connectivity creating conduits for bubble release.

We will change the paragraph explaining these mechanisms more precisely as follows:

(3) "We found ebullition supporting significantly to total CH_4 fluxes in two of our four intact sediment core incubations, whereas sites with higher shares of sand exhibited less ebullitive fluxes confirming the findings of Liu et al. (2016) and (2018). The authors explain their findings with the dominant pathway of bubble formation in the sediment, which is by displacing surrounding sediment particles. As this mechanism is more efficient in soft silty sediments compared to sandy material, CH_4 bubbles likely accumulate more easily in silt, creating a network of macropores and therefore conduits for subsequent bubble release. We further found OM quality partly exhibiting significant negative correlations with CH_4 fluxes, but to a lesser extent than with CH_4 production. When preparing slurry incubations, the physical sediment structure is destroyed, so that OM quality becomes the major controlling factor for gas production. These findings suggest that grain size distribution is besides OM quality a main driver of spatial CH_4 flux patterns in intact sediment core incubations and that only a combination of physical characteristics and sediment OM quality could sufficiently explain CH_4 emission patterns from lakes."

(1) Line 747: Change "experiment" to "results"

(2) The term will be changed in the revised manuscript.

(1) Line 749: Remove "especially"

(2) The term will be changed in the revised manuscript.

(1) Line 753: Change "vulnerable" to "sensitive"

(2) The term will be changed in the revised manuscript.

(1) Line 754: Change "unroll" to "expect"

(2) The term will be changed in the revised manuscript.

(1) Line 755: Change "lower water columns" to "shallow depths"

(2) The term will be changed in the revised manuscript.

(1) Line 761: Change "refer" to "attribute"

(2) The term will be changed in the revised manuscript.

(1) Lines 764-765: Then why do you not find strong relationship between methane production and (EACorg)?

(2) Please see also comment on Line 693. The statement will be discussed in more detail in the revised manuscript.

(1) Line 770: Measuring "production rate" does not neglect water column processes, interpretation of these data alone would.

(2) The sentence will be rephrased in the revised manuscript as follows:

(3) "Further, measuring production rates only would neglect the importance of the water column as a sink of sediment generated CH4…"

List of relevant changes made in the manuscript

Overall

1. Editing of English language and grammar
2. „...were significantly correlated to...“ was changed to „...exhibited significant negative correlation...“
3. rephrasing of „incubation“ and „mesocosms“ to „slurry incubations“ and intact sediment core incubations“
4. correction of non-significant decimal places
5. revision of use of abbreviations

Introduction

L. 105 ff. Correction of standard formation energies

L. 144 ff. Revision of the whole paragraph describing knowledge gaps

Materials and Methods

Fig. 1 caption: Renaming of lake depth categories

L. 466 ff.: Adding of description of methods for determining lake water parameters

Results

L. 501: Adding of Fig. 2, which displays C/N ratio and fats/polysaccharide ratio at different lake depth categories

Fig. 3: Deletion of production rates from 5-10 cm depth for better clarity

L. 567 ff.: Adding of description of correlations of acetate and hydrogen concentrations with OM quality

L. 610 ff.: Adding of calculation of potential CO2-production from prevalent electron acceptors + Figure

Discussion

L. 669 ff.: The chapters „Spatial variability of OM“, „Spatial variability and temperature dependency of CO2 and CH4 production“, and „Influence of OM quality on CO2 and CH4 prodcution rates“ have been merged and therefore shortened

L. 871 ff.: The chapter „Methanogenic pathways“ has been shortened and refocussed on correlations between methanogenic pathways and OM quality

L. 919 ff.: The chapter „Alternative Eas“ has been refocussed; the speculative part on CH4 production has been removed and the discussion on CO2 production has been added

L. 1009 ff.: The discussion on correlation between CH4 bubble formation and grain size distribution has been described in more detail

Organic matter and sediment properties determine in-lake variability of sediment CO_2 and CH_4 production and emissions of a small and shallow lake

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Abstract

Inland waters, particularly small and shallow lakes, are significant sources of carbon dioxide (CO_2) and methane (CH_4) to the atmosphere. ~~, following recent studies this is particularly the case for small and shallow lakes. The~~ However, the spatial in-lake heterogeneity of CO_2 and CH_4 production processes and their drivers in the sediment ~~yet~~ remain poorly studied. We ~~thus~~ measured potential CO_2 and CH_4 production in ~~sediment~~ slurry incubations from 12 sites within the small and shallow crater lake Windsborn in Germany as well as fluxes at the water-atmosphere interface of intact sediment core incubations ~~at~~ from four sites. Production rates were highly variable and ranged from 7.2 ~~and~~ to 38.5 µmol CO_2 gC^{-1} d^{-1} and from 5.4 to 33.5 µmol CH_4 gC^{-1} d^{-1}. Fluxes ~~lay~~ ranged ~~between~~ from 4.5 ~~and~~ to 26.9 mmol CO_2 m^{-2} d^{-1} and ~~between~~ from 0 ~~and~~ to 9.8 mmol CH_4 m^{-2} d^{-1}. Both CO_2 and CH_4 production rates and the CH_4 fluxes exhibited a significant and negative correlation ($p < 0.05$, rho < -0.6) ~~were significantly negative (p<0.05, rho<-0.6) correlated~~ with the prevalence of recalcitrant organic matter (OM) compounds in the sediment as identified by Fourier-transformed infrared ~~FTIR~~ spectroscopy. The ~~C/N~~carbon/nitrogen ratio exhibited a significant negative correlation ($p<0.01$, rho=-0.88)~~was significantly (p<0.01, rho=-0.88) correlated~~ with CH_4 fluxes, but ~~neither~~ not with production rates nor CO_2 fluxes. Availability of i~~I~~norganic (nitrate, sulfate, ferric iron) and organic (humic acids) electron acceptors failed to~~gether could~~ explain differences in CH_4 production rates, assuming a competitive suppression, ~~(R^2=0.22) whereas~~ but observed non-methanogenic CO_2 production could be explained by up to 91% by prevalent electron acceptors. ~~we~~ We did not find clear relationships between ~~organic matter~~OM quality, thermodynamics of methanogenic pathways (acetoclastic vs. hydrogenotrophic) and electron accepting capacity of the ~~organic matter~~OM. Differences in CH_4 fluxes were ~~also able~~interestingly to a large part ~~be~~ explained by ~~Grain~~ grain size distribution ~~could sufficiently~~ ($p < 0.05$, rho = ±0.65)~~ explain differences in CH_4 fluxes~~. Surprisingly though, sediment gas storage, potential production rates and water-atmosphere fluxes were decoupled from each other and did not show any correlations. Our results show that within a small lake, ~~there exists a significant spatial variability of~~ sediment ~~gas~~ CO_2 and CH_4 production show significant spatial variability,~~even within a small lakes~~ which is mainly driven by spatial differences in the degradability of the sediment ~~organic matter~~OM~~can be explained by the origin and pre-processing, and therefore the degradability of the organic matter~~. We highlight that studies on production rates and sediment quality need to be interpreted with care, though, in terms of deducing emission rates and patterns, ~~measuring production rates is not a suitable way to replace in-situ flux measurements~~ as ~~it that approach~~approaches based on production rates only neglect~~s~~ physical sediment properties and production and oxidation processes in the water column as major controls on actual emissions.

1 Introduction

Inland waters play an important role in the global carbon (C) cycle and contribute significantly to ~~the~~ natural emissions of the greenhouse gases carbon dioxide (CO_2) and methane (CH_4) (Cole et al., 2007; Battin et al., 2009; Bastviken et al., 2011; Raymond et al., 2013; Regnier et al., 2013). Lakes and reservoirs are estimated to emit in total 0.32 – 0.39 Pg C of CO_2 and 0.58 Pg C (CO_2 -eq.) of CH_4 year^{-1} (Cole et al., 2007; Bastviken et al., 2011; Raymond et al., 2013~~).~~), but ~~Especially~~ especially small lakes (< 0.1 km²) have previously been underestimated ~~in the past in regard to~~regarding their spatial expansion and, therefore, their contribution to global greenhouse gas emissions (Downing, 2010). Although small lakes account for ~~approx.~~ ~1/3 of the total lake area and cover less than 1% of the global land surface, they contribute 35% of the CO_2 and 72% of the CH_4 emissions from lakes worldwide (Downing et al., 2006; Holgerson and Raymond, 2016). Further, ~~Even~~ even ~~if the highest number of lakes can be~~though most lakes are found in boreal zones, the largest areas occur in lower latitudes around 50° (Verpoorter et al., 2014). In small lakes, ~~Due~~ due to their shallowness, shorter water residence times and ~~a~~ smaller ~~perimeter~~ perimeter-~~to~~ to-volume ratios, metabolic processes and ~~carbon (~~C~~)~~ turnover ~~in small lakes is~~ are much faster ~~compared to~~than in larger lakes, making small lakes potentially ~~and they can therefore be expected to be~~ more ~~vulnerable~~ susceptible to environmental and climatic changes ~~than the latter~~ (Wetzel, 1992; Downing, 2010).

Recently, many studies have shown that both CO_2 and CH_4 fluxes are highly variable both on a spatial and temporal scales, but the majority of these measurements ~~has~~ have been ~~taken out~~performed on larger lakes and ~~is~~ concentrated in boreal regions (Schilder et al., 2013; Wik et al., 2013; Bastviken et al., 2015; Natchimuthu et al., 2016; Natchimuthu et al., 2017; Spafford and Risk, 2018). Beyond that, few studies have examined ~~on~~ greenhouse gas (GHG) production processes in the sediments~~,~~ or have attempted to link ~~and studies linking~~ sediment ~~gas~~ GHG production to emissions~~ both remain scarce~~. ~~Nevertheless, anoxic sediments play a crucial role in lake C cycling as they are the main producer of CO_2 and CH_4 which is subsequently released through the water column to the atmosphere.~~ Nevertheless, anoxic sediments are important for whole-lake C cycling, as the CO_2 and CH_4 produced there can be released through the water column to the atmosphere. To understand the spatial patterns of CO_2 and CH_4 emissions, it is therefore ~~crucial~~ of interest to also understand CO_2 and CH_4 production processes in the sediment as well as their major controls.

The~~ degradability of OM, and therefore the~~ amount of ~~produced~~ CO_2 and CH_4~~,~~ produced in lake sediments relates to the degradability of the organic matter (OM) present, which ~~is mainly depending~~ depends primarily on its ~~components~~quality, the microbial biomass and enzyme activities (Updegraff et al., 1995; McLatchey and Reddy, 1998; Fenchel et al., 2012). While ~~C/N ratios can be used to determine~~ the origin and the degradation state of OM can be determined using carbon/nitrogen (C/N) ratios, ~~Fourier-transformed infrared (FTIR) spectroscopy can provide~~ qualitative information about OM components can be determined via Fourier-transformed infrared (FTIR) spectroscopy. The latter technique ~~, which~~ can, therefore, ~~also~~further provide information about~~and therefore~~ its origin and ~~grade~~ degree of decomposition (Meyers, 1994; Broder et al., 2012; Biester et al., 2014; Li et al., 2016). The prevalence of different OM compounds leads to specific features in FTIR spectra, as functional moieties have wavelength-specific absorption maxima (Niemeyer et al., 1992; Cocozza et al., 2003). Artz et al. (2008) compiled a range of functional moieties that is used to characterize OM in peat soils, ~~but is they are~~ also applicable to OM in general. While polysaccharides and proteins are preferentially degraded by microorganisms, ~~cellulose natural organic matter~~aliphatic (e.g. waxes) ~~or~~ and aromatic compounds (e.g. lignin) are ~~due to their molecular structure~~ more recalcitrant (due to their molecular structure) and, therefore, residually accumulate in the anoxic sediment (Fenchel et al., 2012; Tfaily et al., 2014). ~~The prevalence of different OM compounds leads to specific FTIR spectra, as functional moieties have wavelength-specific absorption maxima (Niemeyer et al., 1992; Cocozza et al., 2003).~~

In anoxic sediments, CO_2 and CH_4 are produced during the breakdown of OM over a cascade of microbially ~~induced~~ mediated processes. After ~~the fermentation of~~ cleavage of complex organic polymers, resulting monomers are fermented, which mainly produces hydrogen (H_2) and ~~OM~~ low molecular weight organic compounds ~~of low molecular weigh~~t (e.g. acetate).~~,~~ ~~the~~ The ~~latter~~ low molecular weight ~~products~~compounds are ~~being~~ further oxidized to CO_2, and H_2 to H_2O, ~~together with a set~~upon consumption of electron acceptors (EAs: nitrate, sulfate, ferric iron or humic substances). ~~before Then~~Only subsequently, CH_4 production ~~initiates asis~~as the final step of terminal ~~electron~~ electron-accepting processes, ~~which happens~~initiates after ~~depletion of~~ all other~~,~~ thermodynamically more favorable ~~electron acceptor~~EAs are depleted (Achtnich et al., 1995; Blodau, 2011). CH_4 ~~in turn~~ is mainly produced via two different pathways, the acetoclastic pathway and the hydrogenotrophic pathway~~ with~~, ~~where~~for which either acetate is the substrate (acetoclastic, R1) or H_2 ~~as~~ is the substrate and CO_2 ~~as~~ is the ~~electron acceptor~~EA (hydrogenotrophic, R2) (Conrad, 1999; Whiticar, 1999). Values for Δ G_r^0 for these terminal electron accepting processes and methanogenesis can be ~~are~~ calculated from standard formation energies Δ G_f^0 in aqueous state listed in Stumm and Morgan (1995) and Nordstrom and Munoz (1994).

$$CH_3COOH \rightarrow CO_2 + CH_4 \qquad \Delta G_r^0\ (25°C) = -50.99\ kJ\ mol^{-1} \qquad R1$$

$$CO_2 + 4\ H_2 \rightarrow 2\ H_2O + CH_4 \qquad \Delta G_r^0\ (25°C) = -193.03\ kJ\ mol^{-1} \qquad R2$$

Several studies have shown that in anoxic wetland and marine sediments rich in labile organic compounds, the acetoclastic pathway dominates~~,~~. ~~which is energetically more less favorable for microorganisms, dominates,. whereas Thus, w~~With increasing recalcitrance, the hydrogenotrophic pathway becomes more important as acetate as direct precursor of CH_4 ~~gets~~ is depleted (Schoell, 1988; Hornibrook et al., 1997; Miyajima et al., 1997). As suggested by Lojen et al. (1999), the OM quality in lake sediments might thus be responsible for~~suggested that~~ seasonal changes in the dominant ~~methane~~ CH_4 pathway~~ in a lake sediment are attributed to OM quality~~.

Whereas many studies have shown that~~the potential of~~ inorganic ~~electron acceptor~~EAs ~~to ~~can suppress methanogenic activity ~~is well studied ~~(e.g. Yao et al., 1999; Fenchel et al., 2012), information on the role of humic substances as organic ~~electron acceptor~~EAs remains scarce. In one study, Klüpfel et al. (2014) revealed ~~the potential of~~that humic substances ~~to ~~can be reduced and re-oxidized at oxic-anoxic interfaces in peatlands, sediments or soils underlying water table fluctuations and another study showed ~~it has recently been shown ~~that in peat soils poor in inorganic EAs, the electron accepting capacity (EAC) of ~~availability of EAC in ~~OM ~~controls ~~represents the major control on CO_2 and CH_4 production ~~in peat soils poor in inorganic electron acceptors (EA)s~~ (Gao et al., 2019). As sediments from the lake ~~under study~~we investigated ~~here ~~are also rich in OM, we wanted to verify ~~if ~~whether the ~~electron accepting (~~EAC~~)~~ and electron-~~-~~donating capacities (EDC) of humic substances also play a vital role in explaining the spatial variabilities of CO_2 and CH_4 production in lake sediments. Although ~~that ~~they are not subject~~ed~~ to water table fluctuations, ~~but ~~they might, ~~to a small extent ~~in the upper parts of the sediments, be influenced by oxygen penetration from the water column due to ~~the ~~a ~~in our case prevalent perennial circulation~~well-mixed water body particularly in shallow lakes (Lau et al., 2016).

Overall, ~~The ~~the amount of CO_2 and CH_4 produced ~~is ~~therefore either ~~depending ~~depends on the availability and degradability of the OM itself, the presence of EAs, and the ~~concentration ~~levels of H_2 and acetate as substrates for methanogenesis (Segers, 1998; Conrad, 1999; Megonigal et al., 2003; Blodau, 2011; Fenchel et al., 2012).

Within lakes, ~~T~~the spatial distribution of OM and other sediment properties ~~within lakes has been found to be highly variable~~vary considerably in terms of their origin (terrestrial vs. aquatic), degradability, elemental geochemistry and grain size (Muri and Wakeham, 2006; Ostrovsky and Tęgowski, 2010; Tolu et al., 2017). ~~It has also been shown that~~Further, sediment grain size is an important factor for the evolution of CH_4 bubbles in sediments (Ostrovsky and Tęgowski, 2010; Liu et al., 2018~~). ~~); for example, Liu et al. (2016) ~~for example ~~revealed ~~a decrease in~~that CH_4 ebullition decreases with ~~increased~~ increasing shares of sand in lake sediments. Overall though, CH_4 ebullition ~~in turn is accountable~~is thought to account for a large proportion (75%) of CH_4 emissions from lakes (Bastviken et al., 2011).

Until now, laboratory incubations of lake sediments were mostly conducted with samples from one or few sites within one lake with a focus on comparing different lakes with each other rather than covering a high in-lake variability of production rates. Further, these studies emphasize temperature effects on production rates (Duc et al., 2010; Gudasz et al., 2010; Gudasz et al., 2015; Fuchs et al., 2016). Unlike in peat soils, where a broad range of process based controls on CO_2 and CH_4 production has been studie, in small lakes, controls such as OM quality, the occurrence of alternative EAs, thermodynamic processes and sediment grain size have not, or have only individually, been systematically surveyed. ~~So far, experimental incubations of lake sediments were only conducted with samples from one or few sites within one lake, rather comparing different lakes with each other than to study within-lake variations of~~ ~~CO_2~~ ~~and~~ ~~CH_4~~ ~~production and with a focus on temperature effects on production rates (Duc et al., 2010;~~

~~Gudasz et al., 2010; Gudasz et al., 2015; Fuchs et al., 2016). Although a broad range of controls on CO_2 and CH_4 production has been widely investigated for anoxic peatland soils, studies on lake sediment are rare. To our knowledge, controls such as organic matter (OM) quality, the occurrence of alternative electron acceptors (EAs), thermodynamic processes and sediment grain size have not, or only individually, been systematically surveyed.~~

To close this knowledge gap, we determined the magnitude and spatial variability of sediment CO_2 and CH_4 production in a small and shallow temperate lake~~,~~ in order to relate observed production patterns to~~and connected productions patterns to~~measured OM and sediment characteristics, thermodynamics of methanogenesis, and water-atmosphere fluxes. To this end, we conducted slurry and intact sediment ~~mesocosm~~ core incubations with sediment from the crater lake Windsborn in Germany. This site was chosen as a model system ~~having~~ because it has a high sediment OM content (~30%), a very small catchment area and no surficial in- or outflows, meaning minimal ~~in order to keep~~ influence~~s~~ from the surrounding ~~area~~catchment ~~as small as possible~~.

We hypothesized ~~that~~ (I) that ~~there exists a noticeable spatial variability of~~ CO_2 and CH_4 production vary spatially in the sediment, (II) that the variability ~~of~~ in the production rates is reflected in the flux patterns~~,~~; and ~~that~~ (III) that the variation in production rates, methanogenic pathways and flux patterns can be explained by factors of OM degradability, ~~the occurrence of~~ availability of organic and inorganic EAs, and grain size distribution, or a combination of these factors.

2 Materials and Methods

2.1 Study site

The studied Lake Windsborn is a polymictic, small~~ and~~, shallow crater lake in the Volcanic Eifel, Rhineland Palatinate, south-west Germany and is part of the ~~“~~Mosenberg~~”~~ volcano group (see Fig. 1) (LfU, 2013). The climate is temperate with a mean annual temperature of 8.3°C and 931 mm precipitation (multi-annual mean 1981-2010; DWD, 2019). Lake Windsborn is the only genuine crater lake north of the Alp~~e~~s. It emerged approx. 29,000 years ago after a volcanic eruption when the top of the volcano ~~was ~~had been blasted away and the newly formed crater was subsequently filled with water (LfU, 2013). The present lake ~~arose ~~formed around 1850, after drainage of the lake and the partial removal of the peat from the lake bottom (Kappes and Sinsch, 2005). The lake is part of a conservation area that was established in 1927 (Kappes and Sinsch, 2005). From 1950 until the 1990s, the lake was used a fishing ground, and was, therefore, stocked~~ up~~ with fish and limed (LfU, 2013). The lake is nearly circular and surrounded by a 20- to ~~30 ~~30-~~meter ~~meter-high rampart which consists of an alternation of red-brown ashes, slag and lapilli from the eruption. Therefore, it has a very small catchment of only about 8 ha compared to the lake surface of 1.41~~ ha without any~~, and it has no surficial in- and outflows and is only fed by groundwater and precipitation (LfU, 2013; Meyer, 2013). The maximum lake depth varies between 1.3 and 1.7 meters (Kappes and Sinsch, 2005). The area is underlain by ~~devonic~~ Devonian quartzite (Kampf, 2005).

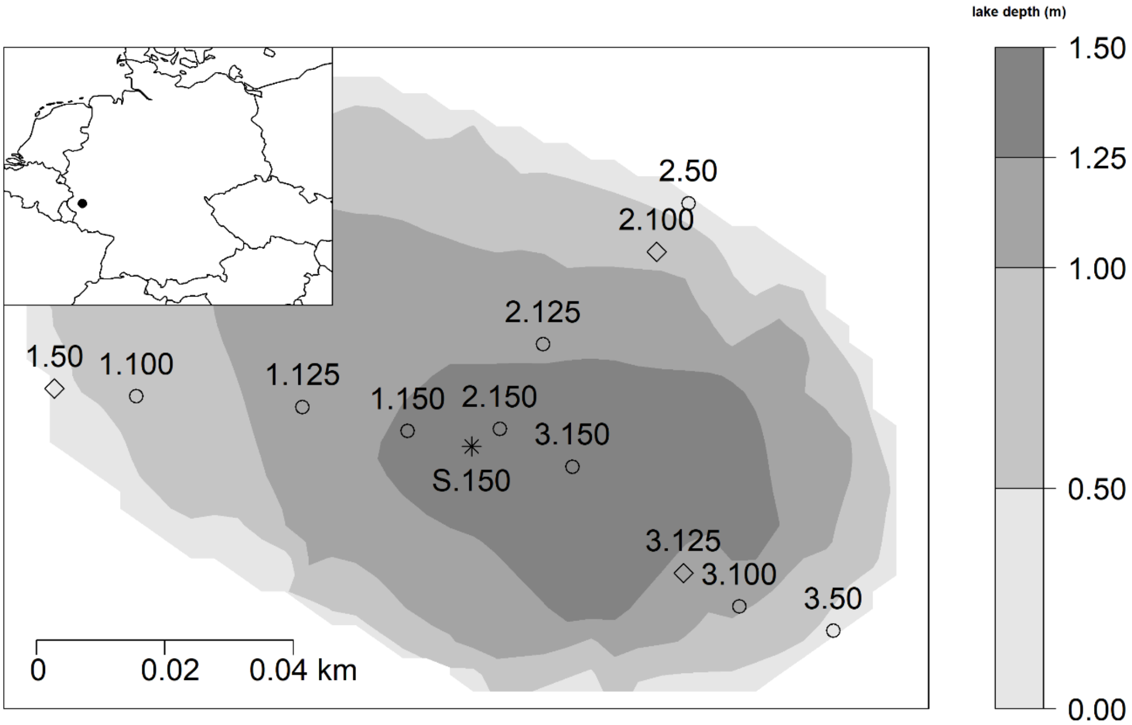


***Figure 1:** Location of the study area in Germany (black spot on the map top left) and 13 sampling sites within Lake Windsborn. ~~Rhombus~~Diamonds: Sampling sites for slurry incubations and intact sediment ~~mesocosms~~core*

incubations, circles: sites for slurry incubations only, asterisk: site for intact sediment ~~mesocosm~~ core incubation only (reference for 1.150, 2.150 and 3.150). Depths were interpolated by bivariate linear interpolation. Numbers 1, 2 and 3 refer to the transect number~~ of transect~~ from the lake shore to center, numbers 50, 100, 125 and 150 indicate lake depth category (50: <50 cm, 100: ~~<~~50-100 cm, 125: ~~<~~100-125 cm, 150~~<~~: 125-150 cm).

The lake’s shoreline is vegetated with *Carex rostrata*, *Comarum palustre* and *Menynathes trifoliata*, all indicating poor nitrogen (N) supply (Ellenberg et al., 2001). At the north-western riparian zone, there is ~~emerges~~ a quaking bog ~~of~~ mainly dominated by *Sphagnum spec.* whose expansion will slowly lead to the silting up of the lake. Lake Windsborn was previously considered ~~as~~ humic-oligotrophic, but in the early 1990’s it ~~has~~ transitioned to a~~n~~ eutrophic lake, and ~~in the early 1990’s~~ now it is slowly recovering from human impacts and nutrient input (Kappes et al., 2000). During our measurement campaigns in 2017 and 2018, the lake exhibited partly meso- and eutrophic features as shown in Table 1.

Table 1: *Selected lake water characteristics measured in Lake Windsborn in 2017 and 2018~~.~~: pH, conductivity (cond.), dissolved oxygen (O_2), chlorophyll alpha (Chl. α), dissolved organic carbon (DOC), total dissolved nitrogen (TN), chloride (Cl), calcium (Ca), iron (Fe), potassium (K), magnesium (Mg), sodium (Na), phosphorous (P) and sulfur (S).~~pH, conductivity and O2 concentration were determined in-situ, DOC and TN were determined by catalytic oxidation, chloride was determined by ion chromatography, and other elements by inductively coupled plasma optical emission spectroscopy (ICP-OES).~~*

parameter	**pH**	**cond.**	**O_2**	**Chl.~~orophyll~~ α**	**DOC**	**TN**	**Cl^-**
unit		µS cm^{-1}	mg L^{-1}	µg L^{-1}		mg L^{-1}	
n	*398*	*387*	*397*	*163*	*419*	*419*	*361*
average	6.8	19.~~35~~4	9. ~~6~~7	27.~~6~~7	13.7~~4~~	~~0.96~~1.0	2.~~86~~9
± SD	0.8~~4~~	1.7~~2~~	0.9~~1~~	18.~~66~~7	2.3~~0~~	1.~~8~~9	3.4~~4~~
parameter	**Ca**	**Fe**	**K**	**Mg**	**Na**	**P**	**S**
unit				mg L^{-1}			
n	*379*	*378*	*379*	*379*	*379*	*329*	*379*
average	1.2~~17~~	0.1~~1~~	0.8~~77~~	0.7~~2~~	4.0~~1~~	0.1~~06~~	0.4~~1~~
± SD	0.~~27~~3	0.1~~06~~	0.4~~2~~	0.1~~3~~	5.0~~1~~	0.1~~06~~	0.1~~3~~

2.2 ~~Sediment~~ Slurry incubations

2.2.1 Sampling and preparation of slurry incubations

~~Samples f~~For the slurry incubation experiment, samples were taken ~~at~~ on three occasions (in March, April and May 2018) from ~~in total~~ 12 of the~~ in total~~ 13 ~~locations~~ sampling sites within the lake from three transects covering multiple water depths (<50, <100, <125, and <150 cm) (see. Fig. 1). On each sampling date, four of the 12 sampling sites were chosen randomly as it was not possible to set up the experiment with all samples at once. At the sampling dates, ~~Measured~~ measured air, water and sediment temperatures at the sampling dates were 7.7°C, 5.6°C and 5.2-°C (March), 13.8°C, 15.1°C and 10.1-°C (April) and 23.9°C, 23.6°C and 13.8 °C (May), respectively. Sediment samples were taken in duplicates with a gravity corer (UWITEC, Mondsee, Austria) from a boat ~~with a gravity corer (UWITEC, Mondsee,~~

~~Austria)~~ in 60 cm long PVC tubes and transported in an insulated box at ~5°C. The next day, ~~Sediment~~ sediment cores were cut with a core cutter in the laboratory ~~the next day~~ in segments of 5 cm thickness (0-5 and 5-10 cm sediment depth). Duplicate samples were homogenized, and then 20 g of each sediment was filled into 120 mL crimp vials ~~added with~~containing 20 mL lake~~-~~ water, after which the vials were ~~and~~ closed with a butyl~~-~~ rubber stopper and aluminum crimp cap. Samples were flushed with ~~nitrogen~~ N_2 for 30 minutes in order to remove any remaining oxygen from the water and headspace, and then they were pre-incubated for one week ~~to have them~~so they were fully anoxic. Then, they were ~~and then~~ again flushed with ~~nitrogen~~ N_2 prior to the actual incubation. Slurry ~~Incubations~~ incubations were set up in triplicates and ~~stored~~ maintained at 25°C (corresponding to maximum measured in-situ sediment temperatures in summer 2018) in the dark. ~~At~~ During each run, one set of parallel samples was incubated at 10°C in order to determine a Q_{10}-value for CO_2 and CH_4 production rates.

The remaining sample material was freeze-dried (Alpha 1-4 ~~LPplus~~LDplus, Christ, Osterode, Germany), ground with a ball mill (Mixer Mill MM 400, Retsch, Haan, Germany) and used for solid phase analyses, as outlined below.

2.2.2 Sediment ~~organic matter~~OM quality

Freeze-dried and ground sediment samples were analyzed for total ~~carbon (~~C~~),~~ and ~~nitrogen (~~N~~) and sulfur (S)~~ concentrations and ~~C and N~~ stable isotopes using isotope-ratio mass spectrometry (IRMS; Eurovector EA3000 coupled with Nu Instruments Nu Horizon, Hekatech, Wegberg, Germany) and for ~~organic matter (~~OM~~)~~ components using FTIR-~~Spectroscopy~~ spectroscopy (Cary 670 FTIR Spectrometer, Agilent, Santa Clara, USA).

For IRMS, 5 mg of sample ~~were~~ was weighed out into a tin cup together with 4 mg of ~~Vanadium~~vanadium-~~Pentoxide~~ pentoxide (V2O5). The combustion and reduction furnace were set to 1000°C and 650 °C, respectively, and the resultant gaseous compounds were quantified by IRMS. Results are provided in % by mass for C~~,~~ and N~~ and S~~ contents (precision < 1% for C and < 0.1% for N) and in ‰ vs. VPDB/AIR~~/VCDT~~ for C~~,~~ and N~~ and S~~ isotopic signatures (precision < 0.05‰ for ^{13}C and < 0.5‰ for ^{15}N). For isotope analyses, appropriate certified reference materials were used: IAEA 600 ($\delta^{13}C$ = -27.77~~1~~ ‰; $\delta^{15}N$ = 1.0 ‰)~~ and S-1 ($\delta^{34}S$ = -0.30 ‰) for calibration~~, and BBOT (2.5-Bis-(5-tert.-butyl-2-benzo-oxazol-2-yl)thiophen; Hekatech, Wegberg, Germany), birch leaf, wheat flour and sorghum flour standards (IVA Analysetechnik e. K., Meerbusch, Germany) as working standards covering a range of -27.5‰ to -13.68~~-~~‰ for ^{13}C, and -0.6‰ to 2.12~~-~~‰ for ^{15}N~~, and -9.3 to -1.42 ‰ for ^{34}S~~.

Functional groups of OM compounds were identified by FTIR spectroscopy. ~~Therefore~~For this purpose, 2 mg of freeze-dried sample ~~were~~ was ground together in a mortar with 200 mg KBr (potassium bromide)~~ in a mortar~~, pressed into 13 mm pellets, and analyzed. Each sample was scanned from 599 to 4000 cm^{-1} with a resolution of 2 cm^{-1} and baseline corrected. Distinct peaks at specific wavelengths were assigned to functional groups according to Artz et al. (2008) and normalized to the peak intensity at

1031 - 1035 cm^{-1} (indicative of polysaccharides) in order to obtain inter-comparable peak~~-~~ ratios of functional moieties in all samples, as FTIR spectra only provide information about the relative abundance of certain functional moieties in one sample.

2.2.3 CO_2 and CH_4 production rates from slurry incubations

Potential CO_2 and CH_4 productions rates were determined by measuring the increase in concentration of CO_2 and CH_4 in the incubation vials over time. Concentrations were obtained from ~~analysis of~~analyzing the headspace at the beginning of the experiment (t0), and after 1, 3, 8, 11, 14 and 18 days (t1-t6). Samples were taken from the vial with a 10 mL ~~PP~~ polypropylene syringe equipped with a three~~3~~-way~~-~~ stopcock and a 0.6 mm needle. Before each sampling, the pressure inside the vial was determined with a pressure sensor (GMH 3110, Greisinger, Regenstauf, Germany), and the syringe was ~~three times~~ flushed three-times with ~~nitrogen~~N_2~~,~~. ~~then~~ Then 2 mL of N_2~~nitrogen~~ ~~were~~ was left in the syringe before ~~stabbing~~ the needle was stabbed through the vial's stopper, whereby the N_2~~nitrogen~~ was added to the headspace, mixed, and, subsequently 2 mL of the sample ~~were~~ was taken from the vial so that the volume inside the vial remained constant. The gas samples were analyzed for CO_2 and CH_4 concentrations with a gas chromatograph (8610 GC-TCD/FID, SRI Instruments, Torrance, USA) equipped with a f~~F~~lame i~~I~~onisation ~~D~~detector (FID) and ~~M~~methanizer to simultaneously measure CO_2 and CH_4. Before every sampling day, ~~T~~the gas chromatograph was calibrated with standard gas mixtures of known concentrations (CO_2: 385, 5,000 and 50,000 ppmV; CH_4: 5, 1,000 and 50,000 ppmV)~~ before every sampling day~~.

First, measured concentrations in ppmV were pressure corrected and converted using the ideal gas law:

$$n = (p * V) / (R * T) \tag{1}$$

where n is the amount of substance in mol, p is the gas partial pressure in atm, V is the headspace volume in L, R is the ideal gas constant (0.082 L atm mol^{-1} K^{-1}) and T is the laboratory temperature in K.

Total CO_2 and CH_4 concentrations in the gas and water phase in the incubation vials were calculated from headspace concentrations ~~with~~ using Henry's ~~L~~law:

$$c = K_h * p \tag{2}$$

where c is the concentration in the water phase in mol L^{-1}, K_h is the temperature-dependent Henry-constant (CO_2, 25°C = 0.0339 mol L^{-1} atm $^{-1}$~~,~~; CH_4, 25°C = 0.00129 mol L^{-1} atm $^{-1}$ (Sander, 2015)) and p is the gas partial pressure in atm.

Moreover, CO_2 concentrations were pH-corrected in order to obtain pH-independent values for total CO_2 concentrations using the Henderson-Hasselbalch-equation and equilibrium constants according to Stumm and Morgan (~~1996~~1995):

$$n_{\Sigma CO2} = n_{water} * 10^{pH-6.4} + (n_{water} * 10^{pH-6.4}) * 10^{pH-10.25} \tag{3}$$

where $n_{\Sigma CO2}$ is the pH-corrected CO_2 amount in mol, and n_{water} is the calculated amount of CO_2 in the water phase in mol.

Finally, production rates were calculated ~~as~~ by linear regression ($R^2 > 0.8$ for CO_2 and > 0.9 for CH_4) from concentration change in gas and solute phase over time.

To evaluate the effect of temperature on CO_2 and CH_4 production, we calculated Q_{10}-values, describing the relative increase of production rates with an increase in temperature of 10 K~~elvin~~ (Fenchel et al., 2012).

$$Q_{10} = (R2/R1)^{[10/(T2-T1)]} \qquad (4)$$

where R2 is the production rate at T2 (25°C), and R1 is the production rate at T1 (10°C).

2.2.4 Thermodynamics and methanogenic pathways

In order to calculate the thermodynamic energy yield for hydrogenotrophic and acetoclastic methanogenesis we measured ~~hydrogen (~~H_2~~)~~ concentrations at the beginning ~~(t0)~~, after 8 days (t3) and at the end ~~(t6)~~ of the experiment, and we measured acetate ~~(H_3COO^-)~~ concentrations at the beginning and at the end of the incubation. The thermodynamic energy yield, expressed as Gibb's free energy, was calculated using the Nernst equation and total dissolved and gaseous concentrations of educts and products in the incubation vials as described in Beer and Blodau (2007):

$$\Delta G_r = \Delta G_r^0 + R * T \ln (\Pi_i(\text{products})^{vi} / \Pi_i(\text{educts})^{vi}) \qquad (5)$$

By calculating the Gibb's free energy (ΔG_r), it is possible to evaluate whether these processes are feasible under given conditions. In order for each reaction to occur, ~~Therefore~~ a theoretical threshold of $\Delta G_r =$ -20 to -25 kJ mol^{-1} ~~for~~ ha~~s~~d to be exceeded (Schink, 1997; Conrad, 1999; Blodau, 2011).

For H_2 concentration measurements, 2 mL of sample were taken from the incubation headspace with a syringe and needle and replaced with the same amount of N2. Samples were analyzed with a r~~R~~eduction g~~G~~as d~~D~~etector (RGD) ~~H~~hydrogen and c~~C~~arbon m~~M~~onoxide ~~A~~analyzer (ta3000R Gas Analyzer, Ametek, Pittsburgh, USA) that was calibrated with gas standards of 5, 25, and 50 ppmV H_2. Measured H_2 concentrations were corrected for pressure and converted into dissolved concentrations using Henry's ~~L~~law ($K_h(H_2$, 25°C) = 0.00078 mol L^{-1} atm^{-1} (Sander, 2015)) analogous to CO_2 and CH_4.

Acetate concentrations were determined by ion chromatography ~~IC~~ with chemical suppression (883 Basic IC plus, Metrohm, Filderstadt, Germany; A-supp 5 column, Metrohm, Filderstadt, Germany). Aqueous samples were filtered with 0.45 µm ~~N~~nylon + ~~G~~glass ~~M~~micro~~-F~~fibre syringe filters (Simplepure, BGB Analytik, Rheinfelden, Germany) and kept frozen at -21°C until analysis.

2.2.5 Alternative ~~Electron Acceptor~~EAs

To quantify alternative ~~Electron Acceptors (~~EAs~~)~~ that could support anaerobic respiration and potentially suppress methanogenesis in anoxic incubations, we ~~analyzed for~~measured nitrate (NO_3^-), sulfate (SO_4^{2-}), ferr~~ous~~ic iron ~~(III)~~ (Fe^{3+}), and the ~~electron accepting~~EAC and ~~donating capacity~~EDC of the ~~organic matter~~OM (EAC_{OM}, and EDC_{OM}) at the beginning ~~(t0)~~ and the end ~~(t6)~~ of the slurry incubation.

As the analysis of EAC_{OM} and EDC_{OM} is ~~so far~~ only possible on finely ground materials, thus providing potential capacities rather than true in-situ capacities, we set up a second set of slurry incubations for a sediment depth of 0-5 cm with samples from 10 sites (all except 3.50 and 3.100) ~~was set up with~~using 0.4 g of the freeze-dried sediment material and 100 mL of ~~Millipore~~ Milli-Q water. ~~Incubations~~ Slurry incubations were set up in six replicates, flushed with N_2, pre-incubated and stored analogous to the first set of slurry incubations. Samples were ~~measured~~ analyzed at the beginning and at the end of the experiment, ~~whereas~~ since at every sampling occasion, three of the replicates had to be sacrificed (destructive sampling). ~~To this end~~Prior to analysis, samples were transferred into a glovebox (O_2 < 1 ppm, Innovative Technology, Amesbury, USA) ~~prior to analysis~~ to avoid alteration of the samples' redox state.

EAC_{OM} and EDC_{OM} were measured ~~chronoamperometrically~~ using chronoamperometry (CHI1000C, CH Instruments, Austin, USA) by mediated electrochemical reduction (MER) and oxidation (MEO) (Aeschbacher et al., 2010; Klüpfel et al., 2014). The cell consisted of a cylindrical glassy ~~carbon~~ C working electrode, a p~~P~~latinum wire counter electrode in a glass-ceramic frit, and an Ag/AgCl reference electrode. Cells were filled with 10 mL of 0.01 M/0.1 M MOPS/KCl-b~~B~~uffer to stabilize the pH at 7 and were continuously stirred during measurement. To facilitate electron transfer, organic mediators were added to the buffer~~;~~: 180 µL DQ (diaquat-dibromide monohydrate, Sigma-Aldrich) for MER~~,~~ and 180 µL ABTS (2,2'-azino-bis(3-ethylbenzthiazoline-sulfonic acid), Sigma-Aldrich) for MEO at a potential of E_h = -0.69 V and E_h = +0.41 V, respectively (reported vs. the standard ~~hydrogen~~ H_2 electrode but experimentally measured vs. the Ag/AgCl reference electrode). To determine the electron transfer, 100 µL of suspended samples were added to the buffer solution (Lau et al., 2015), which resulted in an increase of the current, recorded as a peak in the analysis software. After ~~approx.~~ ~30 minutes, when the baseline was reached again, the next sample was added to the cells. Samples were ~~measured~~ analyzed in duplicate~~s~~. The electron transfer was calculated with the Nernst~~-~~ equation and normalized to the C content in the samples (Lau et al., 2015).

EAC_{OM} ~~/~~ (EDC_{OM}) = peak area / (V * F * C) (6)

with EAC_{OM} ~~/~~and EDC_{OM} in µmol e^- gC^{-1}, peak area in µA sec, V = sample volume in µL, F = Faraday constant 96,485 A sec / mol e^-, and C = ~~carbon~~ C content in mg L^{-1}.

EAC_{OM} and EDC_{OM} had to be corrected for either ferric iron (Fe^{3+}), or ~~iron (II)~~ferrous iron (Fe^{2+}) and sulfide (S^{2-}) concentrations, respectively, ~~as~~ since with the applied potential ~~also~~ Fe^{3+} would be reduced and Fe^{2+} and S^{2-} would be oxidized (Lau et al., 2015; Agethen et al., 2018).

Fe^{2+}, Fe^{3+} and S^{2-} were determined colorimetrically (Gilboa-Garber, 1971; Tamura et al., 1974) with a spectrophotometer (Cary 100 UV-Vis, Agilent, Santa Clara, USA). ~~As the determination with~~Because 1,10-phenanthroline can only detect Fe^{2+}, the Fe^{3+} in the samples was reduced to Fe^{2+} with 10% ascorbic acid. ~~The likewise~~~~Then, the~~Ferric iron was thus determined as the difference of ~~concentration of~~ total Fe and ~~could then be used to calculate the concentration of Fe^{3+}~~ferrous iron in the samples.

NO_3^- and SO_4^{2-} concentrations were determined with IC, as described above. ~~Therefore~~For this purpose, samples were filtered with a 0.45 µm syringe filter, ~~filled~~ added to in micro-centrifuge tubes, retrieved from the glovebox and stored frozen at -21°C until analysis.

Total ~~electron accepting capacity~~EAC (EAC_{tot}) was calculated as the sum of EAC_{OM} and EAC_{inorg} (EAC from nitrate, sulfate, ferric iron ~~(III)~~) considering the respective amounts of electrons transferred during the main pathways of dissimilatory reduction, i.e., assuming a reduction of NO_3^- to N_2, of SO_4^{2-} to S^{-2}, and of Fe^{3+} to Fe^{2+} (Konhauser, 2009):

$$EAC_{tot} = EAC_{OM} + NO_3^- * 5e^- + SO_4^{2-} * 8e^- + Fe^{3+} * 1e^- \qquad (7)$$

~~In theory~~Assuming reversibility of electron transfer to and from OM, EAC_{OM} and EDC_{OM} correlate with each other.~~:~~ ~~if~~ If EAC_{OM} decreases, EDC_{OM} increases equivalently as quinones are reduced to hydroquinones. But in practice, values of EDC_{OM} are potentially biased as MEO does not only capture EDC_{OM}, but may also irreversibly oxidize phenolic moieties, which are sensitive to slightest changes in pH and potentials (Aeschbacher et al., 2011; Walpen et al., 2016; Walpen et al., 2018). The discussion will therefore focus on EAC_{OM} data.

2.3 ~~Sediment mesocosms~~Intact sediment core incubations

2.3.1 ~~:~~ CO_2 and CH_4 fluxes ~~and sediment gas stock change~~

To obtain ex-situ CO_2 and CH_4 gas fluxes and estimate changes in sediment CO_2 and CH_4 stocks, intact sediment cores (PVC tubes, 60 cm length, 5.8 cm diameter) were taken in triplicate~~s~~ from four ~~sites~~ out of the ~~twelve~~ 12 sites ~~above~~ in November 2017 (1.50, 2.100, 3.125, S.150; see Fig. 1). S.150 was chosen as one site ~~representing~~ to represent the sites 1.150, 2.150 and 3.150 from the same lake depth category. Sediment cores were transported cool~~ed~~ and deployed in a climate chamber (CLF Plant Master, CLF Plant Climatics GmbH, Wertingen, Germany) at constant conditions (temperature 20°C, humidity 60 %). Cores were taken to ensure that each tube contained a sediment layer ~~of on~~with average thickness of 35 cm~~ thickness,~~ and was covered with a lake water column of 20 cm; in the lab, we created a headspace of ~~approx.~~ ~150 mL. The cores were equipped with eight sampling ports~~;~~: one in the headspace, one in the water phase and six in the sediment. Of the ~~latter~~ports in the sediment, three were used to sample dissolved gases in the sediment~~,~~ and three were used for sediment pore water extraction. The gas samplers consisted of ~~a gas~~ gas-permeable silicon tubes of 5 cm in length and 0.8 cm in diameter equipped with a ~~3~~three-way~~-~~ stopcock, modified after Kammann et al. (2001). This technique allows for sampling of dissolved gases in the sediment by diffusive equilibration through the silicon membrane.

For pore water sampling, a vacuum was applied with a syringe to suction samplers (Rhizon, Eijkelkamp Agrisearch, Giesbeek, Netherlands) of 5 cm in length, 0.25 cm in diameter, and about 0.1 µm pore size (Seeberg-Elverfeldt et al., 2005). Gas and pore water samplers were deployed in average depths of 5.0 ± 2.8, 15.3 ± 2.9, and 23.6 ± 2.1 cm below the sediment surface.

For ~~statistical~~data analyses~~ and discussion~~, we only used measurements that were ~~conducted~~made >-50 days after the deployment of the intact sediment core incubations in the climate chamber~~ were used~~. This was done in order to ensure the system had adapted to experimental conditions and had reached a steady state. Steady state conditions were~~All measurements were conducted after 50 days of incubation of the cores in the climate chamber when the system had reached a steady state as~~ indicated by quasi-constant CO_2 and CH_4 concentrations in the sediment, as identified by repeated monitoring of dissolved gases.

~~For the determination of~~To determine CO_2 fluxes, the cores were closed gas~~-~~ tight with a stopper and connected to a laser-based, portable greenhouse~~-~~ gas analyzer (Los Gatos Research, San Jose, USA), which allowed ~~to ~~for ~~measure ~~measuring real-time increase of CO_2, CH_4 and H_2O concentrations in the headspace of the cores with a resolution of 1 Hz. As the headspace was too small for the instrument's flow rate, a gas bag with a volume of 150 mL was interposed between the headspace and the analyzer. The headspace was closed for 10 minutes, and the diffusive CO_2 flux was calculated by linear regression ($R^2 > 0.8$) ~~of ~~using the increase in concentration over time and by the ideal gas law, corrected for air pressure and temperature and related to the water surface area:

$$F = \Delta c/\Delta t * (p * V) / (A * R * T) \qquad (8)$$

where F is the CO_2 flux in µmol m^{-2} d^{-1}, $\Delta c/\Delta t$ is the slope of the linear regression in ppm d^{-1}, p is the air pressure in atm, V is the sum of headspace and gas bag volume in m^3, A is the water surface area in m^2, R is the ideal gas constant $8.2*10^{-5}$ m^3 atm mol^{-1} K^{-1}, and T is the temperature in K.

CH_4 fluxes were determined by closing the cores with the stopper for 24 hours~~,~~ and then taking a gas sample right after closing~~,~~ and then again after 24 hours with a syringe from the headspace. CH_4 fluxes were calculated according to Bastviken et al. (2004):

$$F(CH_4) = k * (C_w – C_{fc}) \qquad (9)$$

where F is the CH_4 flux in mmol m^{-2} d^{-1}, k is the piston velocity in m d^{-1}, C_w is the measured CH_4 concentration in the water phase in mmol m^{-3} and C_{fc} is the CH_4 equilibrium concentration in the headspace at the given CH_4 water concentration.

The piston velocity k was determined as:

$$k = (-\ln((c_{sat} - c_{end})/(c_{sat} - c_{start}))/ \Delta t * V) / (A * K_h * R * T) \qquad (10)$$

where c_{sat} is the saturation concentration in the chamber headspace at the measured CH_4 water concentration, c_{end} is the measured CH_4 concentration in the chamber headspace at the end of the flux measurement, and c_{start} is the measured CH_4 concentration in the chamber headspace at the beginning of the flux measurement (all in µatm).

The flux was corrected for the non-linear increase of CH_4 concentration in the headspace over time due to saturation and divided into diffusive and ebullitive proportions based on the piston velocity ($k < 2$ = diffusion, $k > 2$ = ebullition).

2.3.2 Sediment gas stock change

CH_4 and CO_2 concentrations in the sediment were obtained from gas-permeable silicon tubes, determined by gas chromatography as described above (2.2.3) and calculated by Henry's ~~L~~law using ~~temperature~~ temperature-corrected Henry's constants (see ~~formula~~ equation 4). Measured CO_2 concentrations were corrected for pH (~~formula~~ equation 5).

The storage change of CO_2 and CH_4 in the sediment was calculated for each depth segment between two sampling ports as the difference ~~between~~ of CO_2 and CH_4 concentrations obtained from silicon gas samples at the beginning and at the end of gas flux measurements:

$$\Delta CO_2\cancel{/CH_4} = ((c(CO_2\cancel{/CH_4})_{end} * V_{seg}) - (c(CO_2\cancel{/CH_4})_{start} * V_{seg})) / \Delta t \qquad (11)$$

$$\Delta CH_4 = ((c(CH_4)_{end} * V_{seg}) - (c(CH_4)_{start} * V_{seg})) / \Delta t \qquad (12)$$

where Δ~~-~~CO_2 (Δ~~/~~CH_4~~-~~) is the storage change in mmol d^{-1}, $c(CO_2)_{end/start}$~~/~~ ($c(CH_4)_{end/start}$) is the CO_2 or~~/~~ CH_4 sediment pore gas concentration at the end/beginning of the flux measurement in mmol m^{-3}, and V_{seg} is the volume of the sediment core segment between two samplers in m^3.

After completion of flux measurements, intact sediment ~~mesocosms~~ core incubations were eventually cut into 10 cm slices, freeze-dried and ground for solid phase analyses, as described above.

2.4 Other chemical and physical parameters in ~~water and~~ sediment and lake water samples

Total phosphorus (P), ~~sulphur~~ sulfur (S), manganese (Mn) and iron (Fe) in the sediment were determined by wavelength dispersive X-ray fluorescence (WD-XRF; ZSX Primus II, Rigaku, Tokyo, Japan). To this end, 500 mg of freeze-dried and ground sample were pressed into pellets together with 50 mg of wax (Hoechst Wax C, Merck, Darmstadt, Germany) as a pelleting agent. Calibration of the instrument was done using a set of 22 certified reference materials.

~~Dissolved organic carbon (DOC) and total nitrogen (TN) concentrations in watery samples and sediment pore water were determined by catalytic oxidation and subsequent NDIR detection with a total~~

~~carbon/nitrogen analyzer (TOC-L/TNM-L, Shimadzu, Kyoto, Japan). Calibration was verified at each measurement day with potassium hydrogen phthalate (5 and 25 mg L^{-1}) and potassium nitrate (1 and 10 mg L^{-1}) standard solutions.~~

Grain size distribution was determined after Austrian standards (OENorm B 4412; OENorm L 1050; OENorm L 1061) by the Physio-geographic Lab of the Institute of Geography and Regional Research of the University of Vienna. To this end, the organic ~~substance~~ matter was removed from the samples with hydrogen peroxide (H_2O_2) prior to analyses and mineral fine ~~soil~~ sediment was divided into clay (< 2 µm), silt (fine (2-6 µm), medium (6-20 µm), coarse (20-63 µm)) and sand (fine (63-200 µm), medium (200-630 µm), coarse (630-2000 µm)).

Values of pH, conductivity and dissolved O_2 concentration were determined in-situ with a multi-probe (WTW Multi 3420 + IDS sensor, WTW GmbH, Weilheim, Germany). DOC and TN in lake water samples were determined by catalytic oxidation and subsequent NDIR detection with a total C and N analyzer (TOC-L/TNM-L, Shimadzu, Kyoto, Japan). Total Cl, Ca, Fe, K, Mg, Na, P and S concentrations in lake water samples were determined by inductive-coupled plasma optical emission spectroscopy (ICP-OES, Spectroblue, SPECTRO Analytical Instruments GmbH, Kleve, Germany).

2.5 Statistics

All statistical analyses were conducted with R Studio, Version 3.5.~~-~~2 (R Core Team, 2018). Data was tested for normal distribution and homoscedasticity with the Shapiro-Wilk and the Levene~~-~~ ~~T~~test (Fox and Weisberg, 2011), respectively. For non-normally distributed data, significant differences between groups were identified using the Kruskal-Wallis test and the ~~P~~post-hoc Dunn~~-~~ t~~T~~est (Dinno, 2017) for more than two groups and using the Mann-Whitney~~-T~~ test for comparing two groups. If the condition of homoscedasticity was not fulfilled, groups were compared with Mood's ~~M~~median ~~T~~test. Correlations and regressions between production rates and sediment parameters were calculated by Spearman's ~~R~~rank ~~C~~correlation and by linear regression models respectively. All data was tested on a 95% confidence interval and a significance level of α = 0.05.

3 Results

3.1 ~~Sediment~~ Slurry incubations

3.1.1 Sediment ~~Organic Matter~~OM Quality

The C content in the samples was between 2.~~15~~ 2% and 33.~~16~~2%, with the lowest values at site 3.50 and the highest at site 1.50. C/N ratios ranged from ~~10.97~~11.0 at site 1.150 to 19.~~06~~ 1 at site 3.100. Neither the C content nor the C/N ratio showed significant changes with sediment ~~n~~or lake depth, but the C/N ratio was significantly higher in samples taken close to the shore (50) than in samples from the lake center (150) ($p < 0.01$) (see Fig. 2a). C and N isotopic values did not vary much between sites and were on average -27.6 ‰ and -0.6 ‰ respectively.

~~Organic matter~~OM quality as identified by FTIR~~-~~ analysis was predominated by strong absorption features of polysaccharides, lignin, humic acids, phenolic and aliphatic structures, aromatic compounds, and fats, waxes and lipids (see Fig. S1). Except for lignin, which was not identified at sites 2.50, 2.100, 2.125, 3.50, and 3.100, all components were ~~found~~ abundant at all sites. Ranges of peak ratios for ~~every~~ each component class can be found in Tab~~.~~le 2. Overall, the lowest FTIR peak ratios were present at site 3.50 and the highest at site 3.125, corresponding to highest and lowest CH_4 production rates (see below). All peak ratios correlated with each other. They tended to increase (i.e., indicate more decomposed material) with sediment depth and toward~~s~~ the lake center~~, respectively,~~ but this change was not significant (see Fig. 2b).

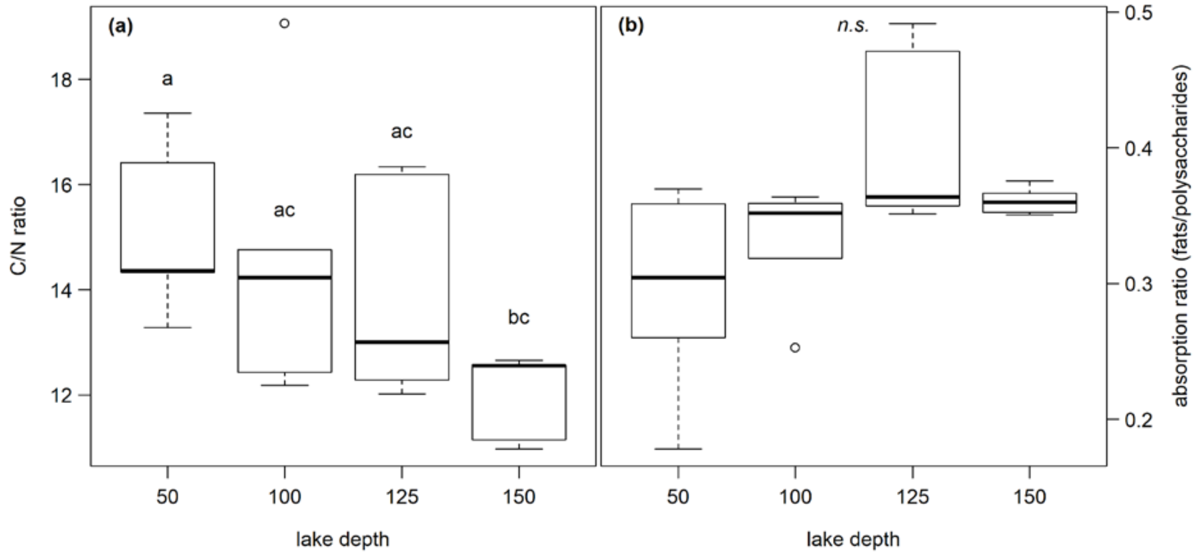


***Figure** 2: (a) C/N ratio and (b) absorption ratio of fats and polysaccharides at different lake depth categories. Identical lowercase letters indicate C/N ratios that were not significantly different (i.e. $p > 0.05$) from each other. N.s. means that absorption ratios did not exhibit significant differences between lake depths. n(50, 100) = 5, n(125,150) = 6*

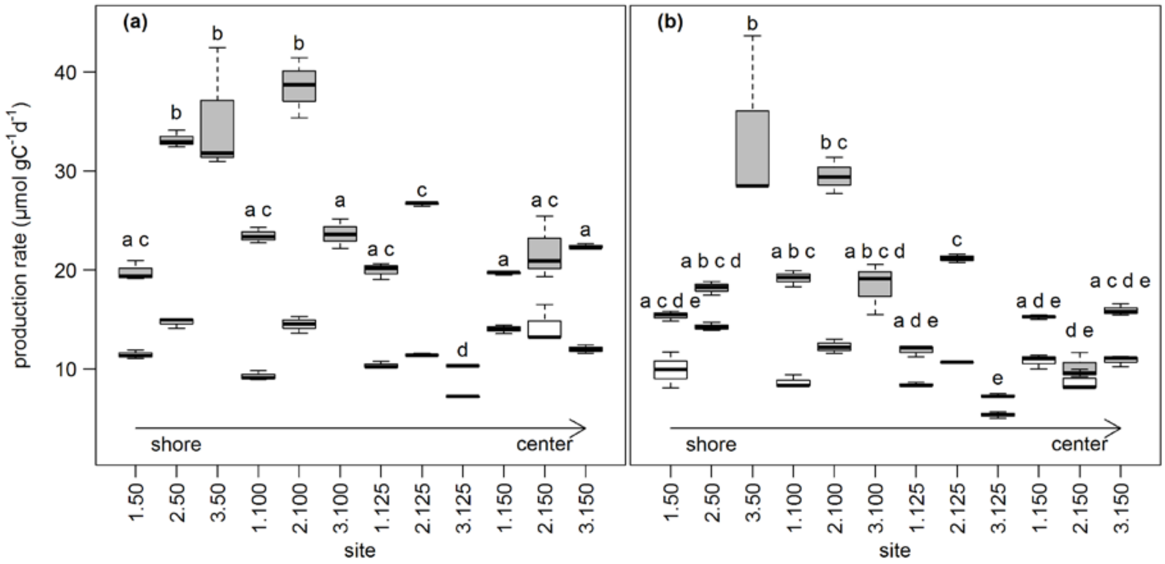
Table 2: *C, N, P, S, Mn, and Fe contents, C/N and FTIR peak ratios (peak maximum for polysaccharides) of identified OM compounds and their minimum and maximum values. n = 16-22.*

	Min	**Max**	**Average**
C (%)	2.~~15~~2	33.~~16~~2	26. ~~4~~5
N (%)	0.~~15~~2	2.~~4~~5	~~1~~2.~~97~~0
C/N ratio	1~~0.97~~1.0	19.~~06~~1	13.8~~1~~
δ^{13}C (‰)	-27.8~~4~~	-27.3	-27.6
δ^{15}N (‰)	-6.~~27~~3	1.3~~3~~	-0.9~~3~~
P (%)	0.3	0.5~~1~~	0.~~37~~4
S (%)	0.1~~3~~	1.2~~2~~	0.9~~3~~
Mn ~~(%)~~(ppm)	~~0.037~~370	~~0.12~~1160	~~0.049~~490
Fe (%)	2.1~~3~~	7.0~~1~~	~~2.95~~3.0
Polysaccharides (1033-1035 cm^{-1})	0.58	1.24	0.81
Lignin (1220-1234 cm^{-1})	0	0.~~437~~44	0.~~235~~24
Humic acids (1417-1419 cm^{-1})	0.~~039~~04	0.49~~1~~	0.~~328~~33
Phenols & aliphatics (1456 cm^{-1})	0.~~018~~02	0.48~~0~~	0.32~~4~~
Other Aromatics (1623-1646 cm^{-1})	0.21~~0~~	0.~~825~~83	0.~~629~~63
Fats, waxes, lipids (2850-2856 cm^{-1})	0.1~~7~~8	0.49~~2~~	0.~~349~~35

3.1.2 CO_2 and CH_4 production rates

Overall, production rates decreased from the shore to the center of the lake. Rates for potential CO_2 production ~~in~~ at 0-5 cm depth ranged from 10.3~~09~~ ± 0.0~~03~~ µmol gC^{-1} d^{-1} at site 3.125 to 38.5~~15~~ ± 2.~~48~~ 5 µmol gC^{-1} d^{-1} at site 2.100. Potential CH_4 production ~~lay~~ was between 7.3 ± 0.1~~4~~ µmol gC^{-1} d^{-1} at site 3.125 and 33.5~~3~~ ± 7.~~151~~ 2 µmol gC^{-1} d^{-1} at site 3.50. Production rates ~~in~~ at 5-10 cm depth were always lower compared to the upper sediment layer and were between 7.~~189~~ 2 ± 0.~~072~~ 1 and 14.7~~14~~ ± 0.4~~35~~ µmol gC^{-1} d^{-1} for CO_2 and between 5.~~361~~ 4 ± 0.~~259~~ 6 and 14.~~264~~ 3 ± 0.3~~41~~ gC^{-1} d^{-1} for CH_4. ~~Overall, production rates decreased from the shore to the center of the lake.~~ Both CO_2 and CH_4 production rates showed significant differences between sites (Kruskal-Wallis, $p < 0.001$) and were significantly (Dunn's, $p < 0.05$) higher at the shore (50+100) than in the center of the lake (125+150) (see Fig. ~~2~~ 3 + Fig. S2 & S3).

CO_2 and CH_4 production rates decreased with time. CO_2 production was highest at the beginning, while CH_4 production ~~had its peak~~was retarded and hig~~e~~hest only after three to eight days of incubation (s. Fig. 4~~3~~, Tab~~.~~le 3). The CO_2/ CH_4 amount ratio constantly decreased during the incubation, with a maximum value of 62.1~~3~~ ± 58.4~~4~~ at the beginning of the incubation at site 1.125 and minimum values of 1.~~17~~ 2 ± 0.0~~04~~ at the end of the incubation at site 2.50, approaching ratios of 1 as expected under strictly methanogenic conditions (s. also Tab~~.~~ le 3).



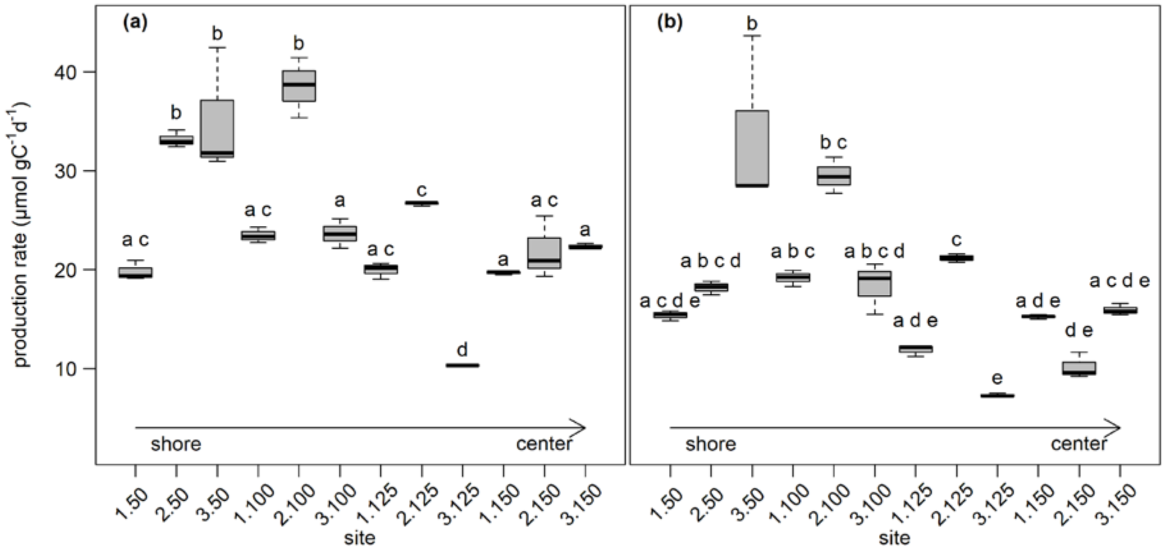


Figure 3: *CO$_2$ (a) and CH$_4$ (b) production rates ~~in~~ at 0-5 ~~(grey) and 5-10 (white)~~ cm sediment depth. n = 3. Production rates ~~are~~ were calculated from the linear regression of concentration increase over time. Bold lines are the median, boxes show the 25 and 75 percentile, and whiskers indicate minima and maxima within 1.5 times the interquartile range~~. Different letters indicate significant differences between these sites~~ Identical lowercase letters indicate production rates that were not significantly different (i.e. $p > 0.05$) from each other.*

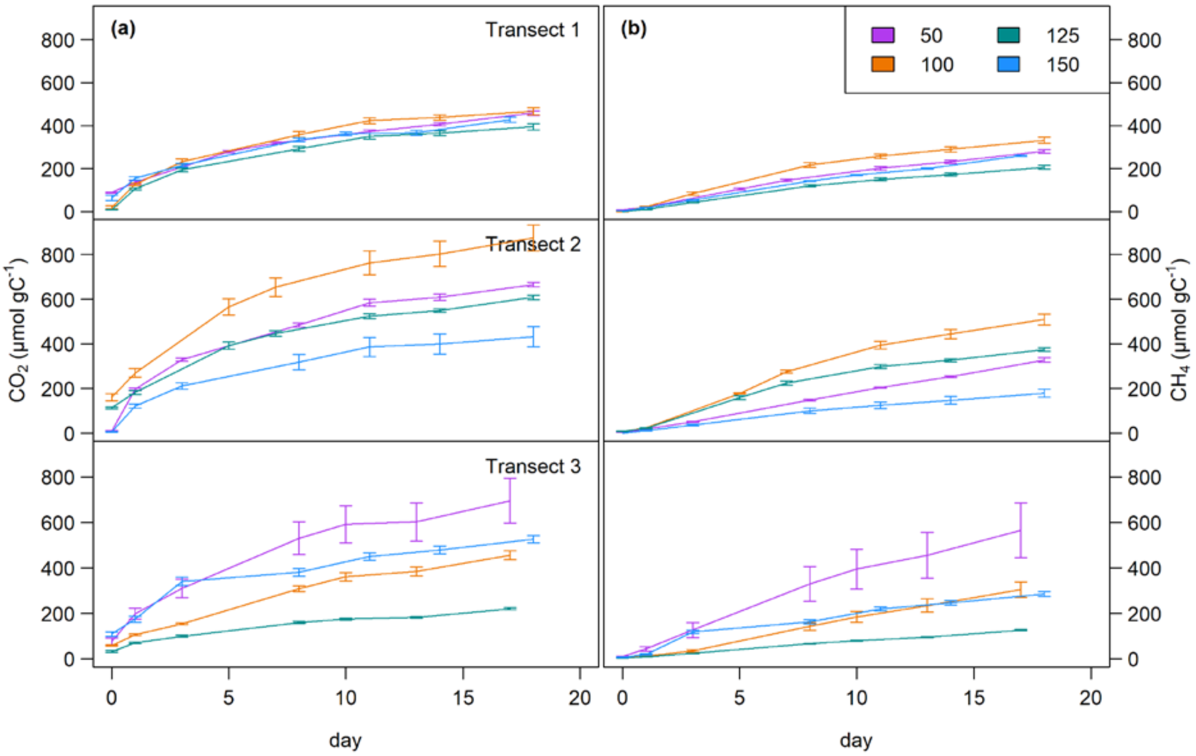


Figure 4: *CO$_2$ (a) and CH$_4$ (b) production over time at all sites ~~in~~ at 0-5 cm sediment depth. Top: transect 1~~,~~; middle: transect 2~~,~~; bottom: transect 3. Lines are average values of triplicate measurements ± SD.*

Table 3: *CO$_2$ and CH$_4$ production rates and the CO$_2$/ CH$_4$ amount ratio over time. Rates are in µmol gC^{-1} d^{-1}~~,~~; ratio is in µmol. Values in brackets are SDs.*

	sediment depth	**day of incubation**						
		~~t~~0	**~~t~~1**	**~~t2~~3**	**~~t3~~8**	**~~t4~~11**	**~~t5~~14**	**~~t6~~18**
CO$_2$ production rate	0-5 cm		91.~~49~~5 (40.~~29~~3)	44.~~6~~7 (16.~~77~~8)	26.~~49~~5 (9.~~69~~7)	21.1~~4~~ (7.6~~59~~)	6.~~79~~8 (3.6~~59~~)	13.3~~2~~ (5.0~~0~~)
	5-10 cm		39.~~86~~9 (14.~~18~~2)	19.6~~3~~ (~~5~~6.~~95~~0)	11.0~~1~~ (2.~~69~~7)	11.4~~0~~ (3.~~06~~1)	6.0~~0~~ (2.2~~0~~)	8.9~~1~~ (2.7~~4~~)
CH$_4$ production rate	0-5 cm		14.~~37~~4 (7.4~~4~~)	22.8~~0~~ (11.~~26~~3)	23.~~88~~9 (11.~~47~~5)	16.7~~4~~ (7.9~~86~~)	11.6~~0~~ (4.~~69~~7)	13.~~58~~6 (5.~~7~~8)
	5-10 cm		9.~~36~~4 (2.~~67~~7)	11.~~68~~7 (2.~~85~~9)	11.~~56~~6 (3.~~13~~1)	9.~~72~~7 (2.~~34~~3)	7.~~89~~9 (2.~~10~~1)	9.~~32~~3 (2.~~72~~7)
CO$_2$/CH$_4$ ratio	0-5 cm	14.~~55~~6 (6.~~71~~7)	8.~~70~~7 (2.~~26~~3)	3.~~84~~8 (1.~~32~~3)	2.~~33~~3 (0.~~48~~5)	2.~~11~~1 (0.~~45~~5)	1.~~89~~9 (0.~~37~~4)	1.~~73~~7 (0.~~30~~3)
	5-10 cm	16.~~19~~2 (24.~~90~~9)	5.~~61~~6 (1.~~60~~6)	2.~~92~~9 (0.~~88~~9)	1.~~93~~9 (0.~~35~~4)	1.~~74~~7 (0.~~34~~3)	1.~~57~~6 (0.~~25~~3)	1.~~44~~4 (0.~~20~~2)

Q$_{10}$-values were between 1.~~56~~ 6 ± 0.~~13~~ 1 and 2.~~19~~ 2 ± 0.~~14~~ 1 for CO$_2$ production rates and between 2.~~65~~ 7 ± 0.~~48~~ 5 and 11.~~37~~ 4 ± ~~0.96~~1.0 for CH$_4$ production rates.

Sampling dates did not have any impact on production rates as confirmed by a Kruskal-Wallis-test (CH$_4$: p = 0.17~~3~~, CO$_2$: p = 0.~~755~~76)

Neither CO_2 nor CH_4 production rates ~~were correlated~~exhibited significant correlations with C content ~~or~~, the C/N ratio, $\delta^{13}C$ or $\delta^{15}N$, but we found significant negative correlations ($p < 0.01$, rho < -0.6) between all FTIR peak ratios and CO_2 and CH_4 production rates as well as of FTIR peak ratios ~~with~~ and Q_{10}-values of CH_4 production ($p < 0.05$, rho = -0.82) (see Fig. ~~4~~5, test statistics see Table S1, SI).

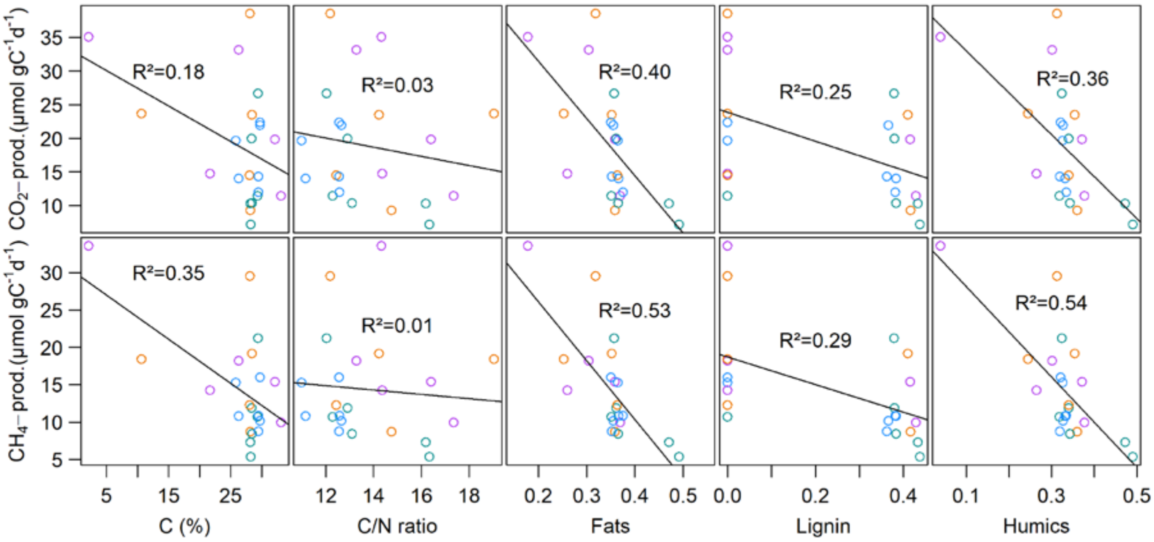


Figure 5: *Correlations between CO_2 and CH_4 production rates and OM quality parameters as determined by elemental analysis or FTIR spectroscopy. Different colors denote different depth categories (see Fig. 4).*

3.1.3 Methanogenic pathways

Both H_2 and acetate concentrations increased during the incubation. H_2 concentrations were between 0 and 1587.~~42~~4 ± 170.~~12~~1 nmol L^{-1} and acetate concentrations ranged from 219.~~74~~7 ± 104.~~25~~3 to 1212.~~71~~7 ± 11.~~35~~4 µmol L^{-1} (see Fig. ~~5~~6). Gibb's free energy for acetoclastic methanogenesis was between - 96.~~32~~3 ± 0.~~84~~8 and -66.~~37~~4 ± 0.~~28~~3 kJ mol^{-1} and for hydrogenotrophic methanogenesis it was between -62.~~41~~4 ± 0.~~85~~9 and -7.~~41~~4 ± 0.0 kJ mol^{-1}. Energy yields decreased for the acetoclastic pathway and significantly increased (Mood's median, t0-t6: $p < 0.05$, t3-t6: $p < 0.001$) ~~increased~~ for the hydrogenotrophic pathway throughout the incubation (see Fig. ~~5~~6). Energy yields did not differ significantly between lake depths except for the acetoclastic pathway at the end of the incubation (~~t6,~~ $p < 0.001$), when the energy yield was highest in samples from the center of the lake. Significant differences between all sites were found for the acetoclastic and hydrogenotrophic pathway at the beginning ~~of the incubation~~ (Kruskal-Wallis, $p < 0.01$) and at ~~t6~~ the end of the incubation for acetoclastic methanogenesis only (Kruskal-Wallis, $p < 0.01$). H_2 concentrations at the end of the experiment exhibited significant positive correlation ~~were significantly correlated~~ with average CO_2 ($p < 0.001$, rho = +0.51) and CH_4 ($p < 0.001$, rho = +0.45) production rates (test statistics see Table S2, SI). Further, Gibb's free energy of acetoclastic methanogenesis exhibited a significant positive correlation ~~was positively correlated~~ with C/N ratio ($p < 0.05$, rho = +0.45) at the end of the experiment ~~(t6)~~. Acetate concentration at the end of the incubation exhibited significant negative correlations with FTIR peak ratios and C/N ratio ($p < 0.05$, rho < -0.44). Gibb's free energy of hydrogenotrophic methanogenesis exhibited significant positive correlations with FTIR peak ratios of fats, humic acids, phenolics and C content ($p < 0.05$, rho > 0.17), i.e. a high energy yield was associated with low FTIR peak ratios and C content. Correspondingly, H_2 concentrations exhibited significant negative correlations with all FTIR peak ratios and C content ($p < 0.05$, rho < -0.15) (test statistics see Table S2, SI).

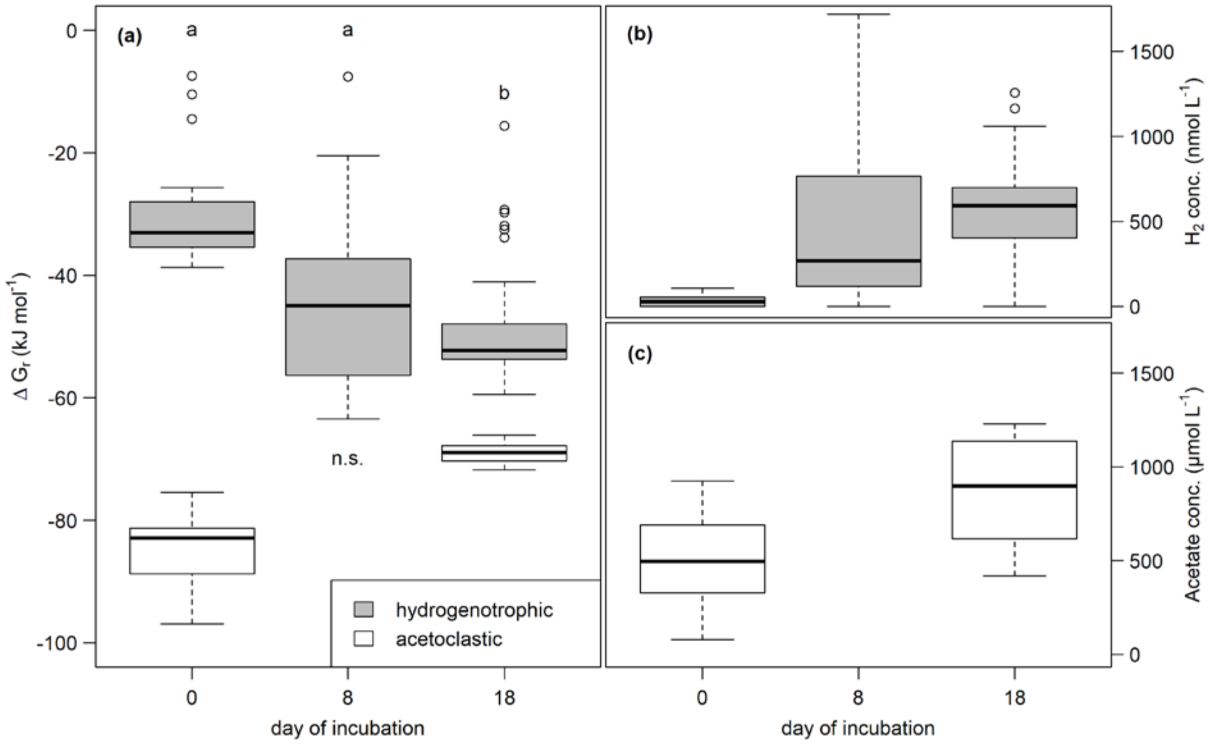


Figure 6: *(a) Change of Gibb's free energy of hydrogenotrophic (grey) and acetoclastic (white) methanogenesis and of H_2 (b) and acetate (c) concentrations over time. More negative Δ G_r values correspond to a higher energy yield. Different lowercase letters denote significant differences between sampling dates.*

3.1.4 Alternative Organic and Inorganic ~~Electron Acceptor~~EAs

EAC_{OM} ~~lay~~ was between 218.~~69~~ 7 ± 97.~~15~~ 2 and 545.~~71~~ 7 ± 60.~~33~~ 3 μmol e^- gC^{-1} at ~~t0~~ the beginning and decreased ~~by an~~on average ~~of~~ by 44.~~85~~ 9 μmol e^- gC^{-1} ~~to 170.76 ± 32.70 to 460.79 ± 51.47 μmol e⁻ gC⁻¹ at~~ until ~~t6~~the end of the slurry incubation. ~~with h~~The highest values for EAC_{OM} were found at site 3.125 corresponding to lowest measured CH_4 production rates at that site. EAC_{inorg} was, with values between ~~19.82~~ 2.9 ± ~~10.52~~ 1.0 and ~~218.01~~ 39.2 ± ~~24.79~~ 4.4 μmol e^- gC^{-1}, much lower than EAC_{OM} and also decreased during the incubation by a mean of ~~42.40~~ 6.4 μmol e^- gC^{-1}. EAC_{tot} ranged from ~~226.95~~ 250.8 ± ~~83.87~~ 93.9 μmol e^- gC^{-1} at site 3.150 to ~~614.58~~ 551.4 ± ~~80.72~~ 60.3 μmol e^- gC^{-1} at site 3.125 and ~~significantly~~ decreased from the beginning to the end of the incubation~~(t-test, p < 0.001) decreased by averagely~~ by on average ~~87.25~~ 51.2 μmol e^- gC^{-1} ~~from t0 to t6~~. EDC_{OM} was between 149.~~54~~ 5 ± 26.~~72~~ 7 and 462.~~69~~ 7 ± 18.~~57~~ 6 µmol e$^-$ gC^{-1} at the beginning and between 152.~~94~~ 9 ± 53.~~78~~ 8 and 370.~~65~~ 7 ± 196.~~22~~ 2 µmol e$^-$ gC^{-1} at the end of the incubation ~~with~~ showing a slight ~~increase~~ decrease by, on average, 31.~~38~~ 4 µmol e$^-$ gC^{-1}. The l~~L~~owest EDC$_{OM}$ at both ~~t0~~ the beginning and ~~t6~~ the end of the incubation was found at site 2.100, corresponding to the highest CO$_2$ production rates there (see Fig. ~~6~~7).

We further found significant differences for EAC$_{OM}$ and EAC$_{tot}$ (ANOVA, $p < 0.001$) ~~and EAC$_{tot}$ (ANOVA, $p < 0.01$)~~ between all sites, with the highest average values at site 3.125 and the lowest at sites 1.125 (t0) and 3.150 (t6) (see Fig. ~~S3~~S4). Average CO$_2$ and CH$_4$ production rates exhibited a significant negative correlation ~~were significantly ($p < 0.05$, rho = 0.7) negative correlated~~ with initial EDC$_{OM}$ ($p < 0.05$, rho = -0.7), whereas we did not find any significant correlation between the production rates and~~with~~ EAC, electron exchange capacity (EEC, sum of EAC and EDC) ~~nor~~ or the EAC/EDC ratio, although low CH$_4$ production rates were associated with high EAC. ~~CO$_2$, CH$_4$ and acetate concentration were significantly ($p < 0.05$, < 0.05, < 0.001) negative correlated with EAC$_{tot}$. Gibb’s free energy of hydrogenotrophic methanogenesis was significantly ($p < 0.05$) positive and acetate concentration significantly ($p < 0.01$) negative correlated to EAC$_{OM}$~~. Acetate concentration exhibited significant negative correlations with EAC$_{OM}$ and EAC$_{tot}$ ($p < 0.01$, <0.001, rho < -0.38). Gibb’s free energy of hydrogenotrophic methanogenesis exhibited significant positive correlations with EAC$_{OM}$ and EAC$_{tot}$ ($p < 0.05$, rho = 0.43). We did not find any significant correlations between EAC or EDC and OM quality parameters except for EDC$_{OM}$ and the FTIR peak ratio indicative of fats, waxes and lipids ~~and EDC$_{OM}$~~ ($p < 0.05$, rho = -0.76). Of the inorganic EAs, only total ~~sulfur~~ S content in the sediment exhibited a significant negative correlation ~~was significantly negative correlated~~ with ~~CH$_4$~~ CO$_2$ production rates ($p < 0.05$, rho = -0.45) (test statistics see Tables S1, SI).

Calculated potential CO$_2$-production from prevalent organic and inorganic EAs was always lower than the measured CO$_2$-production in slurry incubations and was between 110.5 and 586.4 µmol CO$_2$-eq. gC^{-1} (see. Fig. 8) and could explain 38 to 91% of measured CO$_2$-production, whereof 4 to 51% where explained by EAC$_{OM}$ alone.

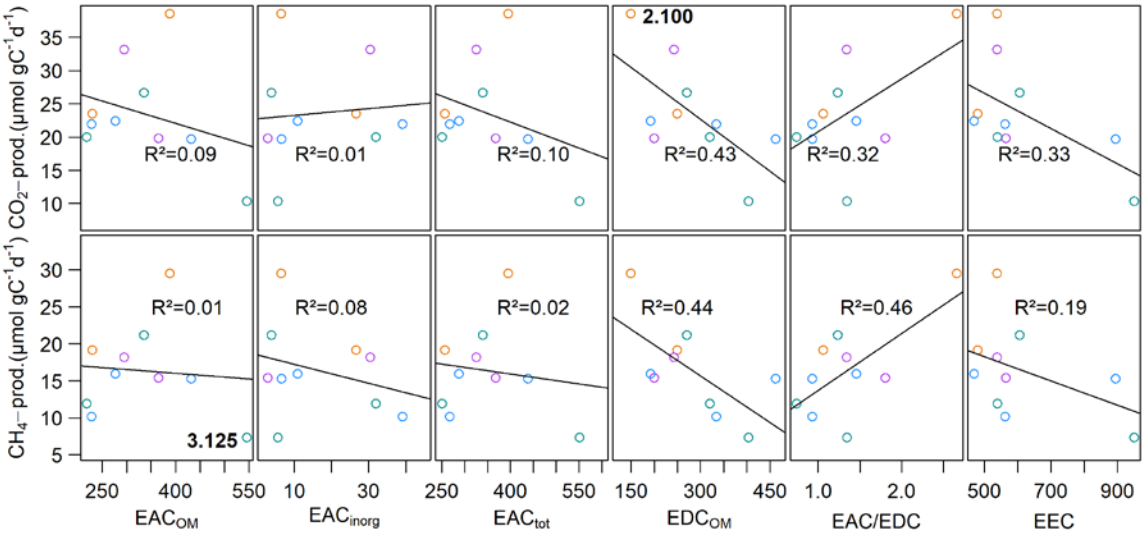


Figure ~~6~~7: *Linear regression of CH_4 and CO_2 production rates with the initial ~~electron accepting~~EAC and ~~donating~~ EDC~~capacities~~. Points are mean values of triplicate measurements at each site. At site 2.100, highest CO_2-production rate concurred with lowest EDC_{OM}, at site 3.125 lowest CH_4-production rate concurred with highest EAC_{OM}. Different colors denote different depth categories (see Fig. 4).*

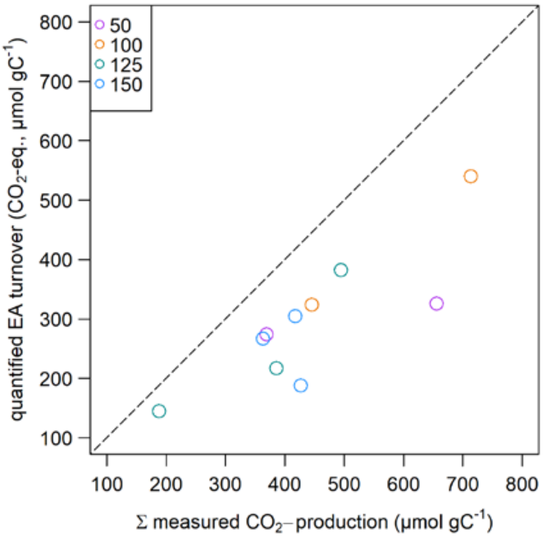
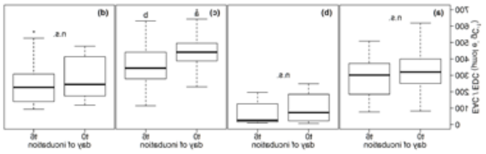
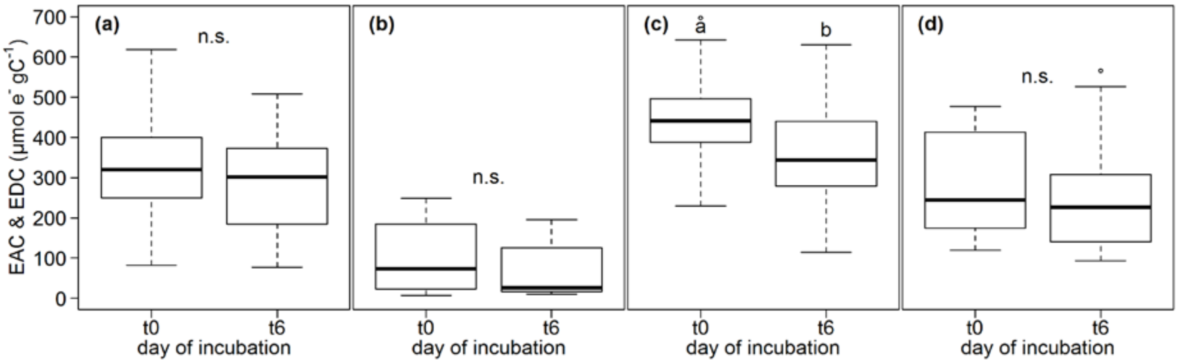


Figure 8: Calculated expected CO_2-production rate from prevalent EAs vs. measured potential CO_2-production rate in slurry incubations. Dashed line shows the closed budget of expected and measured CO_2-production.

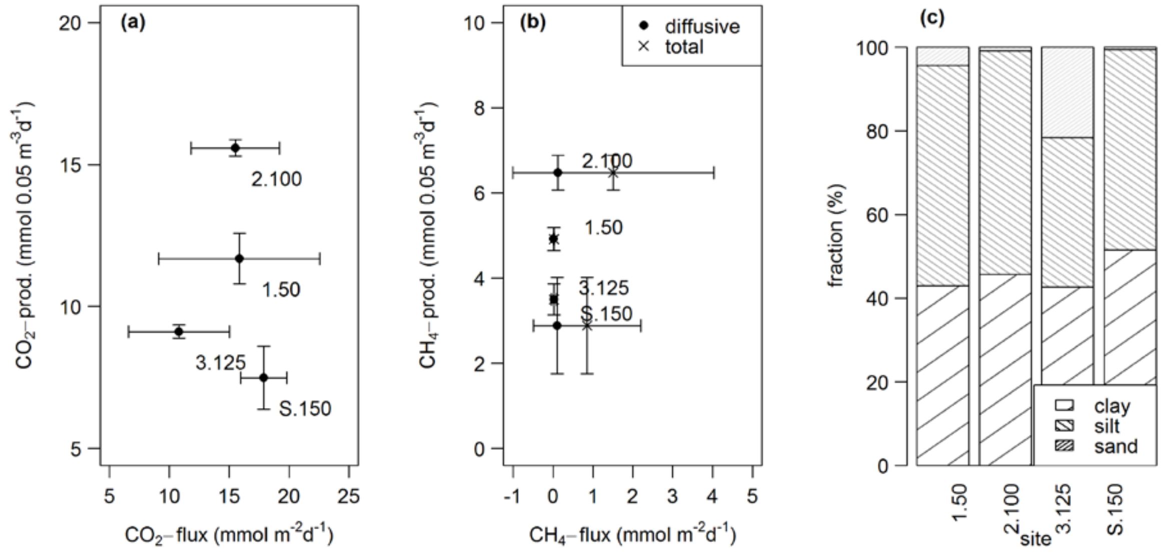




~~**Figure 7:** EAC$_{OM}$ (a), EAC$_{inorg}$ (b), EAC$_{tot}$ (c) and EDC$_{OM}$ (d) at the beginning and the end of the incubation at all sites. n = 60-88. Different letters denote significant differences.~~

3.2 ~~Sediment mesocosms~~Intact sediment core incubations: Fluxes, sediment storage change and grain size

CO$_2$ and CH$_4$ fluxes measured from intact sediment core ~~mesocosms~~ incubations ranged from ~~4.46~~10.8 ± 4.4 to ~~26.9~~17. 9 ± 2.0 and ~~0~~ 0.02 ± 0.01 to ~~9.8~~1.5 ± 2.6 mmol m^{-2} d^{-1}, respectively. CH$_4$ ebullition played a major role in cores from sites 2.100 and S.150 and accounted for up to 100% of the fluxes there. CO$_2$ fluxes at site 3.125 were significantly lower (t-test, p < 0.05) ~~lower~~ than at the other sites while total CH$_4$ fluxes at site 2.100 were significantly higher (Kruskal-Wallis, p < 0.01) ~~higher~~ compared to all other sites. Fluxes were within the same range ~~like~~ as potential production rates in slurry incubations but ~~did~~ only partly followed the pattern observed in slurry incubations (see Fig. ~~8~~9a+b).



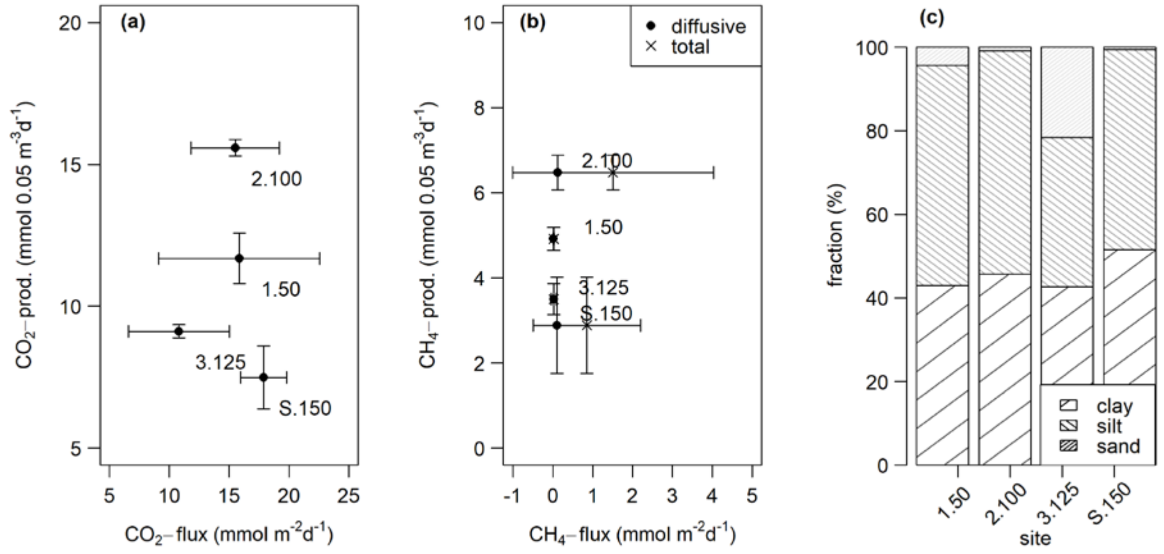


Figure 7: *Comparison of fluxes obtained from intact sediment ~~mesocosms~~ core incubations and potential production rates from the slurry incubations (~~in~~ at 0-5 cm sediment depth) for CO_2 (a) and diffusive and total CH_4 (b). Points are averages (n(slurry incubations) = 3-9, n(CO_2 fluxes) = 9-14, n(CH_4 fluxes) = 11-13), horizontal and vertical lines are standard deviations. We decided to display production rates from the 0-5 cm sediment depth, as this depth turned out to be the most important for sediment ~~gas~~ GHG production (see sections 3.1.2 & 4.1.~~2~~1). (c) Relative grain size distribution in intact sediment ~~mesocosms~~core incubations.*

Table 4: *Spearman's rank correlation coefficients for CH_4 and CO_2 fluxes (means of each intact sediment ~~mesocosm~~core incubation except for correlations with sediment stock changes where values from each sampling date were used) and different sediment characteristics. n.s. means that correlations were not significant ($p > 0.05$).*

	~~CH_4 flux~~		~~CO_2 flux~~	
	~~rho~~	~~p~~	~~rho~~	~~p~~
~~**Clay**~~	~~0.6477503648~~	~~0.02275< 0.05~~	~~0.6045669605~~	~~< 0.050.03731~~
~~**Silt**~~	~~0.49665085~~	~~*n.s.*~~	~~0.3022835~~	~~*n.s.*~~
~~**Sand**~~	~~-0.6477503648~~	~~< 0.050.02275~~	~~-0.6045669605~~	~~< 0.050.03731~~
~~**Fats, waxes, lipids**~~	~~-0.8333333~~	~~< 0.050.01538~~	~~-0.3333333~~	~~*n.s.*~~
~~**Phenols; humics**~~	~~-0.8333333~~	~~< 0.050.01538~~	~~-0.3571429~~	~~*n.s.*~~
~~**Aromates**~~	~~-0.5952381~~	~~*n.s.*~~	~~-0.5238095524~~	~~*n.s.*~~
~~**Lignin**~~	~~-0.7863867~~	~~< 0.050.02063~~	~~-0.3805097381~~	~~*n.s.*~~
~~**C/N**~~	~~-0.8809524881~~	~~0.007242< 0.01~~	~~-0.3333333~~	~~*n.s.*~~
~~**C (%)**~~	~~-0.7142857~~	~~*n.s.*~~	~~-0.1904762~~	~~*n.s.*~~
~~**CH_4 sediment stock change**~~	~~-0.2219706222~~	~~*n.s.*~~	~~0.0498599405~~	~~*n.s.*~~
~~**CO_2 sediment stock change**~~	~~-0.04872526049~~	~~*n.s.*~~	~~-0.06414566~~	~~*n.s.*~~

	CH_4 flux			**CO_2 flux**		
	rho	**p**	**n**	**rho**	**p**	**n**
Clay	0.648	< 0.05	12	0.605	< 0.05	12
Silt	0.497	*n.s.*	12	0.302	*n.s.*	12
Sand	-0.648	< 0.05	12	-0.605	< 0.05	12
Fats, waxes, lipids	-0.833	< 0.05	8	-0.333	*n.s.*	8
Phenols; humics	-0.833	< 0.05	8	-0.357	*n.s.*	8
Aromatics	-0.595	*n.s.*	8	-0.524	*n.s.*	8
Lignin	-0.786	< 0.05	8	-0.381	*n.s.*	8
C/N	-0.881	< 0.01	8	-0.333	*n.s.*	8
C (%)	-0.714	*n.s.*	8	-0.190	*n.s.*	8
CH_4 sediment stock change	-0.222	*n.s.*	41	0.05	*n.s.*	35
CO_2 sediment stock change	-0.049	*n.s.*	41	-0.064	*n.s.*	35

Concentrations of dissolved CH_4 and CO_2 ($\Sigma(CO_2, HCO_3^-, CO_3^{2-})$) in the sediment ranged from ~~3.97~~4.0 to 129.~~4~~5 mmol m^{-3} (CH_4) and from 322.9~~2~~ to 3811.~~36 ~~4 mmol m^{-3} (CO_2), ~~respectively ~~with the lowest values found at site 3.125 and the highest values at S.150. We did not see significant changes in CO_2 and CH_4 concentrations in the depth profile (see Fig. ~~S4~~S5).

Observed changes of CH_4 and CO_2 concentrations in the sediment of intact cores ~~was ~~were overall very low and ~~lay ~~were between 0.~~06 ~~1 and 2.~~4~~5 (CH_4) and 0.~~55 ~~6 and 57.~~1~~1 mmol d^{-1} (CO_2) in the upper 5 cm of the sediment. ~~It was within~~These changes were on the same order of magnitude as measured fluxes but did not correlate with the~~ latter~~m.

Grain sizes distribution differed between the four sites, with site 3.125 having the highest share of sand (21.6%) and the lowest shares of silt and clay (35.~~68 ~~7% and 42.7~~3~~%) whereas the other sites were dominated by finer material and ~~consisted of~~contained less than 5% of sandy components (see Fig. ~~8~~9c). CH_4 and CO_2 fluxes exhibited significant correlation ~~were significantly correlated ~~with clay and sand content, but not with silt (see Tab~~.~~le 4~~5~~).

Similar to slurry incubations, CH_4 fluxes exhibited significant negative correlations ~~were significantly negative correlated ~~with some FTIR peak ratios indicative of recalcitrant OM compounds ~~as well as~~and with the C/N ratio but not with C content. CO_2 fluxes did not show any significant correlation with OM quality parameters at all (see Tab~~.~~le ~~5~~4).

4 Discussion

4.1 ~~Sediment ~~Slurry incubations

4.1.1 Variability of CO_2 and CH_4 production rates and OM quality~~Spatial variability of OM quality~~

Results from the slurry incubation experiment showed that both CO_2 and CH_4 production rates were – despite its small size - highly variable within the examined lake. CH_4 production rates were within the range of previously reported values from lakes in central Sweden, a reservoir in Brazil, and in sediments from Lakes Stechlin and Geneva in Germany and Switzerland, respectively (Duc et al., 2010; Fuchs et al., 2016; Grasset et al., 2018). CO_2 production rates were high compared to rates found in Lake Kinneret in Israel and in the Pantanal and Amazon regions in Brazil, exceeding reported values by a factor of 7 to 100 (Schwarz et al., 2008; Conrad et al., 2011), supporting the assumption that small lakes generally have higher C turnover rates compared to larger ones.

In all samples, production rates were higher at the beginning of the experiment than toward the end. While CO_2 production rates were highest right after the start and then constantly decreased until reaching a plateau around day 8, CH_4 production rates peaked after 3 to 8 days and then slowly decreased afterwards approaching a 1:1 CO_2:CH_4 production ratio. This typically observed three-phase pattern (Yao et al., 1999) might among other things be due to thermodynamic constraints, such as end product accumulation in the sample vials and subsequent inhibition of respiration and methanogenesis (Beer and Blodau, 2007; Blodau et al., 2011; Bonaiuti et al., 2017). The observed delay in notable CH_4 production was expected as prevalent alternative EAs (NO_3^-, Fe^{3+}, SO_4^{2-}, humic substances) likely suppressed methanogenic activity (Lovley et al., 1996; Yao et al., 1999; Fenchel et al., 2012).

Both CO_2 and CH_4 production rates were higher in the topmost sediment layer compared to the 5-10 cm sediment depth, which suggests that the first centimeters of the sediment play a major role in GHG production as a consequence not only of temperature but also of microbial community distribution and changes in OM quality (Falz et al., 1999; Sobek et al., 2009; Wilkinson et al., 2015). This finding corresponded with decreasing C/N ratios and increasing FTIR peak ratios with sediment depth. While sediment age typically increases with depth, the OM quality and, thus, reactivity usually decreases with sediment depth, as recalcitrant compounds are not being completely mineralized but instead residually enrich get buried and preserved in the sediment (Avnimelech et al., 1984; Burdige, 2007; Sobek et al., 2009).

Similar to den Heyer and Kalff (1998), we found higher production rates in samples close to the lake shore in contrast to lower production rates in the center, which suggests either that the input of OM (quantity) in the sediments is higher at the shore sites and/or the OM quality is more labile and, therefore, more easily degradable. As we did not find any correlations between CO_2 or CH_4 production rates and sediment C content as proposed e.g. by Conrad et al. (2011) or Romeijn et al. (2019), we instead suggest that the OM quality rather than quantity might be the determining factor for CO_2 and CH_4 production.

C/N ratios are frequently used to characterize the degradation state of OM, but we did not find correlations between C/N ratios and CO_2 and CH_4 production rates in the slurry incubations. Although OM of autochthonous origin was found to fuel higher degradation rates than allochthonous OM (West et al., 2012; Grasset et al., 2018) we found evidence of predominant inputs of allochthonous (terrestrial) material at sites with higher production rates close to the shore (higher C/N ratios), whereas sites with lower production rates in the lake center received mainly autochthonous (aquatic) OM as indicated by lower C/N ratios (Meyers, 1994). On the other hand, high C/N ratios also indicate a lower degradation state and therefore higher degradation potential whereas low C/N ratios are usually typical of highly decomposed OM having a lower CO_2 and CH_4 production potential (Malmer and Holm, 1984; Kuhry and Vitt, 1996). These two possibilities of interpreting C/N ratios might be the reason for apparently contradicting findings and the missing relationship between C/N ratios and CO_2 and CH_4 production rates.

The abundance of recalcitrant OM compounds (lignin, aromatics, humics, phenols, and lipids) as indicated by FTIR peak ratios, proved to be a more suitable explanation for observed CO_2 and CH_4 production patterns, as indicated by strong significant negative relationships between peak ratios and production rates. All FTIR peak ratios indicative of more refractory components increased toward the lake center, which indicated an increasing predominance of more recalcitrant OM with greater distance from the lake shore and corresponded with decreasing production rates from lake shore to center. At first sight this was unexpected as allochthonous material is known to be richer in recalcitrant compounds like lignin or aromatics compared to autochthonous biomass, therefore, is effectively buried in lake sediments (Sobek et al., 2009). However before reaching the sediment, the internally produced material in the lake center must pass through a deeper water column, meanwhile undergoing degradation processes, such that more decomposed OM reaches the sediment (Meyers, 1994; Torres et al., 2011). As this process might not be of the same importance in shallow lakes compared to deeper lakes, we additionally suggest that the more decomposed OM in the lake center might have undergone degradation processes during resuspension and focusing of small particles as a result of wind-induced bed-shearing (Mackay et al., 2011). The less decomposed OM close to the shore might further originate from labile aquatic plant substrates growing at the shoreline (Wetzel, 1992; Cole et al., 2007).

To summarize, the OM degradability does not necessarily depend on its origin, and the OM of terrestrial origin is not necessarily more recalcitrant to degradation by aquatic microorganisms in this study. Sites closer to the shore probably received higher rates of terrestrial OM matter that was less decomposed, whereas autochthonous production in the lake center was overall low, due to the low nutrient status of the lake, and was already undergoing degradation during sedimentation; this likely lead to more recalcitrant OM in the sediment. Earlier studies by West et al. (2012) and Grasset et al. (2018), that found allochthonous material to be less decomposable compared to autochthonous matter were conducted in larger lakes, where the influence of the watershed was overall much lower than in our case, because larger lakes have a lower perimeter to volume ratio. We hypothesize that in small and nutrient-poor lakes, the pattern might be reversed as the system adapts to overall low productivity and a simultaneously high input of terrestrially produced OM.

In accordance with previous studies (den Heyer and Kalff, 1998; Sobek et al., 2009; Gudasz et al., 2015), we found that with a temperature increase of 10°C, production rates of CO_2 doubled and those of CH_4 were even 2 to 11 times higher. Q_{10}-values for CO_2 were within the range of earlier reported values by Liikanen et al. (2002) and Bergström et al. (2010), whereas Q_{10}-values for CH_4 production were slightly higher than values found by Duc et al. (2010). The large observed range of Q_{10}-values, especially for CH_4, implies that responses to temperature increases might not be homogeneously distributed within a lake. The observed negative correlation between Q_{10}-values and FTIR peak ratios further suggests that sites with more labile OM are more susceptive to increasing temperatures in terms of CH_4 production, whereas at sites with more recalcitrant OM, the inherent quality of this recalcitrant OM may limit the degradation processes. We therefore assume that sediment GHG production – particularly CH_4 production - in small and shallow lakes might in the course of global warming increase to a larger extent than in deeper lakes regarding the fact that shallow waters, as against deeper lakes, do not develop a stable stratification in summer and therefore shallow sediments warm much faster (Jankowski et al., 2006).

The high measured variability in production rates proves (1) that it is necessary to sample lake sediments from different locations in one lake in order to obtain potentials of CO_2 and CH_4 production representative of the whole lake and (2) that upscaling production rates based on single point measurements may be highly biased. Considering only sediment production rates, based on findings from other studies it seems that the observed spatial variability of CO_2 and CH_4 emissions from lakes might to a large extent be controlled by variability in sediment GHG production (Schilder et al., 2013; Bastviken et al., 2015; Natchimuthu et al., 2016; Natchimuthu et al., 2017; Spafford and Risk, 2018). But still, as sediment production and emission patterns have so far only been considered separately, other factors driving actual emissions might have been neglected. Therefore, in section 4.2 we further discuss the relationships between CO_2 and CH_4 production and actual emissions, as assessed in the intact sediment core incubation, and relate them to respective sediment properties.

~~In our study, we found higher C/N ratios in samples close to the lake shore and narrower ratios in the lake center indicating a predominant input of allochthonous (terrestrial) material at the shore and mainly sedimentation of autochthonous (aquatic) OM in the lake center (Meyers, 1994).~~

~~Besides C/N ratios, FTIR peak ratios revealed information about the OM composition and quality in Lake Windsborn. We found FTIR peak ratios for refractory components (lignin, aromatics, humics, phenols, and lipids) increasing towards the lake center, indicating increasing predominance of more recalcitrant OM with higher distance from the lake shore. At first sight, this was unexpected as allochthonous material is known to be richer in recalcitrant compounds like lignin or aromates compared to autochthonous biomass and therefore being effectively buried in lake sediments (Sobek et al., 2009). On the other hand, the internally produced material in the lake center has to bypass a deeper water column before reaching the sediment, meanwhile undergoing degradation processes and leading to more decomposed OM reaching the sediment (Meyers, 1994; Torres et al., 2011). The less decomposed OM close to the shore might further originate from labile aquatic plant substrates growing at the shoreline (Wetzel, 1992; Cole et al., 2007).~~

~~Not only on a spatial scale, but also with sediment depth, C/N ratios decreased and FTIR peak ratios increased, corresponding with a decrease in CO_2 and CH_4 production rates. While sediment age typically increases, OM quality and thus reactivity usually decreases with sediment depth, as recalcitrant compounds are not being mineralized completely but instead buried and preserved in the sediment (Avnimelech et al., 1984; Burdige, 2007; Sobek et al., 2009).~~

~~4.1.2 Spatial variability and temperature dependency of CO_2 and CH_4 production rates~~

~~Our incubation experiment showed that both CO_2 and CH_4 production rates were highly variable within one lake of only small spatial extent. CH_4 production rates were within the range of formerly reported values from lakes in central Sweden, a reservoir in Brasil, and in sediments from Lakes Stechlin and Geneva in Germany and Switzerland (Duc et al., 2010; Fuchs et al., 2016; Grasset et al., 2018). CO_2 production rates were high compared to rates found in Lake Kinneret in Israel and in the Pantanal and~~

~~Amazon regions in Brazil and exceeded reported values by a factor of 7 to 100 (Schwarz et al., 2008; Conrad et al., 2011). Compared to production rates after addition of fresh OM to lake sediment, our CO_2 and CH_4 production rates remained low (Grasset et al., 2018).~~

~~Both CO_2 and CH_4 production rates were higher in the topmost sediment layer compared to 5-10 cm sediment depth, suggesting that the first centimeters of the sediment play a major role for gas production as a consequence of temperature and microbial community distribution and changes in OM quality (as discussed below) (Falz et al., 1999; Sobek et al., 2009; Wilkinson et al., 2015).~~

~~In all samples, production rates were higher at the beginning of the experiment than towards the end. While CO_2 production rates were highest right after the start and then constantly decreased until reaching a plateau around day 8, CH_4 production rates peaked after 3 to 8 days and then slowly decreased afterwards approaching a 1:1 CO_2:CH_4 production ratio. This typically observed 3-phase pattern (Yao et al., 1999) might e.g. be due to thermodynamic constraints, such as end product accumulation in the sample vials and subsequent inhibition of methanogenesis (Beer and Blodau, 2007; Blodau et al., 2011; Bonaiuti et al., 2017). The observed delay in notable CH_4 production can be explained by the prevalent alternative EAs (NO_3^-, Fe^{3+}, SO_4^{2-}, humic substances) suppressing methanogenic activity (Lovley et al., 1996; Yao et al., 1999; Fenchel et al., 2012).~~

~~Similar to den Heyer and Kalff (1998), we found higher production rates in samples close to the lake shore in contrast to lower production rates in the center, suggesting that either the input rate of OM to the sediment is higher and therefore fueling higher degradation rates or/and that the organic material at these sites is more labile and therefore more easily degradable for microorganisms. More details on that hypothesis will be given in the following.~~

~~The high measured variability in production rates showed that it is necessary to sample lake sediments from different locations in one lake, in order to understand potentials of CO_2 and CH_4 production representative of the whole lake and that upscaling production rates based on single point measurements may be highly biased. Considering only sediment production rates, it seems that the in other studies observed spatial variability of CO_2 and CH_4 emissions from lakes (Schilder et al., 2013; Bastviken et al., 2015; Natchimuthu et al., 2016; Natchimuthu et al., 2017; Spafford and Risk, 2018) might to a large extent be controlled by sediment gas production. But still, as sediment production and emission patterns have so far only been considered separately, important relations might have been neglected. In section 4.2, we therefore further discuss the relationships between CO_2 and CH_4 production and actual emissions and relate them to sediment properties.~~

~~Many studies revealed that higher temperatures lead to enhanced mineralization of OM due to higher microbial activity and thus increased production of CH_4 and CO_2 in sediments (den Heyer and Kalff, 1998; Sobek et al., 2009; Gudasz et al., 2015). Accordingly, we found that production rates of CO_2 were 2-times and of CH_4 2 to 11-times higher with a temperature increase of 10°C. Q_{10} values for CO_2 were~~

~~within the range of earlier reported values by Liikanen et al. (2002) and Berström et al. (2010) whereas Q_{10}-values for CH_4 production were slightly higher than values found by Duc et al. (2010). The high observed range of Q_{10}-values, especially for CH_4, implies that responses of temperature increase might not be homogeneously distributed within one single lake. The observed negative correlation between Q_{10}-values and OM quality indicators suggests that sites with more labile OM are more vulnerable to increasing temperatures in terms of CH_4 production and that at sites with more recalcitrant OM, the latter may limit the degradation processes. Aben et al. (2017) and Kiuru et al. (2018) showed that increasing temperatures caused higher CO_2 and CH_4 emissions from lakes. Assuming that gas production in the sediment and efflux to the atmosphere are coupled, global warming may thus drastically enhance emissions from small lakes like Lake Windsborn, especially regarding the fact that these sediments warm much faster (max. bottom-water temperature in summer 2018: 26.65°C, surface water temperature at the same time: 27.14 °C) compared to deeper lakes which have a deeper water column and stable thermal summer stratification as buffer (Jankowski et al., 2006).~~

~~4.1.3 Influence of OM quality on CO_2 and CH_4 production rates~~

~~The amount and the quality of OM usually determines the degradability and therefore the production of end-products of anaerobic mineralization processes, CO_2 and CH_4.~~

~~Against our expectations and previous findings e.g. by Conrad et al. (2011) and Romeijn et al. (2019), C content was not correlated with CO_2 nor CH_4 production rates. As the production potential is obviously not depending on the amount of C, we suggest that instead the OM quality might be the determining factor for CO_2 and CH_4 production.~~

~~Typically, OM of autochthonous origin fuels higher degradation rates than allochthonous OM (West et al., 2012; Grasset et al., 2018) as the latter is rich in compounds like lignin, cellulose and humic acids, being recalcitrant to decomposition and therefore being effectively buried in lake sediments (Sobek et al., 2009). We however measured higher CO_2 and CH_4 production rates at sites close to the shore receiving higher inputs of allochthonous material compared to the center of the lake receiving mainly autochthonous OM as indicated by the C/N ratio. As a wider C/N ratio does not only indicate OM of terrestrial origin, but also implies that the OM is still less decomposed and therefore has a higher decomposition potential whereas a narrower C/N ratio is not only typical of aquatic OM but also indicates a higher degradation state, higher production rates at sites closer to the shore seem to be reasonable (Malmer and Holm, 1984; Meyers, 1994; Kuhry and Vitt, 1996; Gudasz et al., 2015; Grasset et al., 2018). Anyway, the correlation between production rates and C/N ratio was not significant, implying that the C/N ratio might, due to before explained contradictions, not be a suitable proxy for the mineralization potential in our case. Instead, all FTIR peak ratios were significantly negative correlated with CO_2 and CH_4 production rates, supporting the hypothesis that OM mineralization is highly dependent on the quality of the OM.~~

~~To summarize, the quality of OM is not necessarily depending on its origin and that OM of terrestrial origin is not necessarily more recalcitrant to degradation by aquatic microorganisms. Sites closer to the shore probably receive higher rates of terrestrial OM matter being less decomposed, whereas autochthonous production in the lake center is overall low, due to the low nutrient status of the lake, and already undergoing degradation processes during sedimentation. This leads to more recalcitrant OM in the sediment. Earlier studies which found that allochthonous material is less decomposable compared to autochthonous matter were conducted in larger lakes, where the influence of the watershed is overall lower due to a lower perimeter to volume ratio. We hypothesize that in small and nutrient-poor lakes the pattern might be reversed as the system adapts to overall low productivity and simultaneous high input of terrestrially produced OM.~~

4.1.~~4~~ 2 Methanogenic pathways

To make methanogenesis a profitable reaction for microorganisms, theoretical thresholds of -10.6 kJ mol^{-1} for the hydrogenotrophic pathway and -12.8 kJ mol^{-1} for the acetoclastic pathway ~~have to~~must be exceeded (Hoehler et al., 2001). Except for a few samples at the beginning of the incubation for the hydrogenotrophic pathway, the theoretical thresholds were always exceeded, suggesting that both pathways could potentially contribute~~d~~ to CH_4 production during the whole experiment. Still, this approach does not allow to evaluate which of the pathways actually predominated.

~~During the incubation, Gibb's free energy increased for the hydrogenotrophic pathway, implying an increasing contribution of that pathway to total~~ ~~CH_4~~ ~~production, whereas it was opposite for the acetoclastic pathway. This suggests that labile organic polymers as precursors of acetate got depleted during the incubation so that~~ ~~H_2~~ ~~gained of importance as substrate for methanogenesis. But when considering~~ ~~H_2~~ ~~and acetate concentrations, the picture gets more complicated. An overall increase of~~ ~~H_2~~ ~~concentrations was observed during incubation, but average~~ ~~H_2~~ ~~concentrations at t3 had a high standard deviation (see Fig. 5) because some samples showed a peak in~~ ~~H_2~~ ~~concentrations at t3 instead of a constant increase. These patterns could hint at an imbalance of the system, where fermenting, syntrophic and methanogenic bacteria were not yet equilibrated (Fey and Conrad, 2003). It is also remarkable, that the energy yield for acetoclastic methanogenesis decreased although acetate concentrations increased. We propose two different explanations for this finding: First, acetate is also used to reduce other EAs, which were present in higher concentrations at the beginning of the incubation, and therefore might have kept acetate concentrations low. Second, higher acetate concentrations do not necessarily mean more favorable thermodynamic conditions due to end product accumulation. During degradation of carbohydrates,~~ ~~H_2~~ ~~and acetate are being produced in a 2:1 ratio. We found acetate concentration exceeding~~ ~~H_2~~ ~~concentrations by a factor of~~ ~~10^3~~~~, suggesting that homoacetogenesis contributed significantly to acetate production in our incubations (Conrad, 1999).~~

Against our expectations, Gibb's free energy of acetoclastic methanogenesis at the end of the incubation was significantly ~~lowest~~ lower (i.e. higher energy yield) in the center of the lake ~~at t6~~ and ~~was~~

~~additionally~~ ΔG_r exhibited significant positive correlation ~~positively correlated~~ with C/N ratio. Considering that we found the ~~lowest~~ most decomposed material (following peak ratios from FTIR analyses and low C/N ratios) ~~OM quality~~ in the lake center and that low C/N ratios in this case indicate OM of high recalcitrance whereas the acetoclastic pathway is ~~attributed~~ thought to predominate with high quality substrates, we would have expected a reverse pattern (Liu et al., 2017; Berberich et al., 2019). Concomitantly, Gibb's free energy of hydrogenotrophic methanogenesis exhibited significant positive correlations with some FTIR peak ratios indicative of higher contributions of recalcitrant OM compounds, although we expected that a high abundance of recalcitrant OM compounds would make hydrogenotrophic methanogenesis more feasible (Liu et al., 2017). Acetate and H_2 concentrations on the other hand, both exhibited significant negative correlations with some FTIR peak ratios. While this seemed reasonable for acetate concentrations (less acetate available in strongly decomposed OM), this result again proved to be against our expectations in terms of H_2 concentrations. This suggests that more recalcitrant material in general slows fermentative processes, delivering both acetate and H_2, as rate determining step, and all fermentation products are thus kept at low levels.

~~Also, we found significant differences of energy yields between the sites, but could not relate them to OM quality. This implies that acetate as intermediate in the anaerobic OM mineralization cascade is not the rate determining step, but rather is fermentation and that OM quality only explains a part of the variance in~~ ~~CO_2 and CH_4 production patterns.~~

~~It further has to be mentioned, that correlations with~~ ~~CO_2 and CH_4~~ ~~production rates were only observed for~~ ~~H_2~~ ~~concentrations at the end of the incubation, but not with acetate. This finding emphasizes the hypothesis that the system was not in balance from the beginning of the incubation, and that fermentation seems to be the rate determining step for OM mineralization.~~

4.1.~~5~~ 3 Alternative ~~electron acceptor~~EAs

~~A~~The average EAC_{OM} measured in our experiment (302.8 ± 124.6 µmol e^- gC^{-1}) was slightly lower but in the same order of magnitude compared to values found by Lau et al. (2015) and Gao et al. (2019) in other lake sediments or peat soils ~~and other lake sediments,~~ respectively. Given that C contents in the study by Gao et al. (2019) were much higher than in our study (46.2 – 49.4~~ ~~%, Agethen and Knorr (2018)) and that Lau et al. (2015) fully oxidized their samples prior to analyses (C content 11.0 – 27.4 %), our measured capacities seem reasonable.

We found that both organic and inorganic EAs ~~decreasing~~ decreased during the experiment, indicating that all EAs ~~are being~~become similarly depleted with time as they are being reduced by microorganisms. Nevertheless, both absolute EAC_{OM} and EAC_{inorg} values as well as relative changes were rather low, which might have been caused by the one-week preincubation, where a majority of the reducible organic and inorganic compounds might have already been depleted. Klüpfel et al. (2014) showed that the EAC of oxidized humic acids strongly declined already after one day of anoxic incubation and did not change much afterwards. Inorganic EAs were also shown to decrease exponentially during anoxic incubation

(Yao et al., 1999). The high CO_2 production rates (91.5 µmol gC^{-1} d^{-1}) at the beginning of the slurry incubation are an indication of these processes.

Sites with higher EAC_{tot} had lower CH_4 production rates and vice versa, as an increased concentration of oxidized humic substances would suppress methanogenic activity on longer time scales, supporting the findings of Agethen et al. (2018) and Gao et al. (2019). Although the relationship between EAC and CH_4 production was not significant – probably due to the low number of samples and the sensitivity of the measuring method - ~~not significant,~~ a ~~clear~~ trend was observable.

Non-methanogenic CO_2-production could by 38 to 91% be explained by prevalent organic and inorganic EAs, whereas organic EAs alone were able to explain up to 52% of measured CO_2-production in slurry incubations. Similar contributions were observed in previous studies from freshwater systems and peatlands, where up to 70 and 95% of non-methanogenic CO_2-production was explained by total EAC and 25 to 55% by decreases in EAC_{OM} alone (Lau et al., 2015; Agethen et al., 2018; Gao et al., 2019). The authors suggest several possible reasons for the gap between calculated and experimentally measured CO_2-production: Fermentation processes could explain excess CO_2, but in this case reduced equivalents of fermentation products, such as H_2 would be expected to accumulate. Moreover, decarboxylation processes that occur in oxygen-rich, labile OM might occur. Further, the assumption of a nominal oxidation state of C of zero – as assumed in our approach – might be wrong, so that when assuming a higher oxidation state, measured changes in EAC_{OM} might explain a higher share of CO_2-production (Gao et al., 2019). As we neither observed accumulation of fermentation products nor was the sediment under study strongly oxidized, we instead assume these processes are of minor importance and instead suggest that the proportion of unexplained CO_2-production can be explained by unknown consumed inorganic EAs during the slurry incubation. In this case, this was most likely solid phase iron which we found to occur 2-3% (≙ 53-137 mg Fe gC^{-1}) in our samples, but whereof we were not able to make statements about its speciation. We suppose this concentration is high enough to reach a closed budget of measured CO_2-production and calculated EA turnover; if on average 7.8% (4-18.7%) of solid phase iron were present as ferric iron, this would suffice to close the CO_2 budget.

The large range (4 to 52%) of the contribution of organic EAs to CO_2-production shows that there exists a distinct small-scale in-lake variability of terminal electron accepting processes and their contribution to CO_2 formation in the sediment of Lake Windsborn. Interestingly, the contribution of EAC_{OM} was highest at site 3.125, where we observed lowest CO_2-production rates and highest OM recalcitrance. Given that both the absolute C content in the incubation vials and the C:Fe ratio were highest at this site, this support findings of Lau et al. (2015), who stated that the contributions of EEC_{OM} to total EEC increase with the OC:Fe ratio, indicating more peat-like material.

~~Additionally, relationships were stronger with EAC_{tot} that with EAC_{inorg} or EAC_{org} only, suggesting that humic substances play a crucial, but variable role as terminal electron acceptors and should not be neglected when estimating the contribution of EAs to methanogenic activity in lake sediments rich in~~

~~OM (~30%) (Lau et al., 2016). The spatial variability of measured EAC and C content in the samples shows that substrate composition is critical when evaluating the contribution of different EAs and CH_4 production patterns.~~

4.2 Comparison of production rates, fluxes, and sediment parameters

Comparing the order of magnitude of potential production rates from slurry incubations and fluxes from intact sediment cores, results were in good accordance with each other. While CO_2 production rates were overall ~~something~~ somewhat below measured fluxes, CH_4 production rates were considerably higher than actual CH_4 fluxes. The differences in CO_2 production rates and fluxes can be explained by the influence of the water column: CO_2 is not only produced in the sediment, but also in the water column, e.g. by the degradation of OM or zooplankton respiration (Kling et al., 1992) and can therefore contribute to water-atmosphere fluxes. The differences concerning CH_4 could be explained by several ~~approaches~~factors. First, preparing slurry incubations leads to the homogenization of the sediment and higher surface area, and therefore, an even distribution of substrates and microorganisms, ~~which is then better~~such that the substrates are more readily available ~~for the latter~~to the microorganisms (Hoehler et al., 2001). Second~~ly~~, sediment fluxes from intact sediment cores may be low compared to homogenized slurry incubations, as maximum sediment concentrations there were already reached in shallow depths~~we did not observe significant increases in the depth profile of sediment cores,~~ ~~and suggest~~suggesting that strong changes in sediment gas concentrations ~~which are driving~~that drive fluxes are likely to occur only in the upper ~~mm~~ millimeters to ~~cm~~centimeters. Additionally, the preparation of ~~incubations~~ slurries reduces the thermodynamic constraints ~~which~~ that usually exist in intact profiles due to end product accumulation (Blodau et al., 2011; Bonaiuti et al., 2017). Third~~ly~~, not all CH_4 produced in and emitted by the sediment will reach the water-atmosphere interface, because some of the CH_4 will be consumed in the oxygenated water column on the way to the surface: ~~of consumption processes in the oxygenated water column that has to be passed. Through~~ through ~~CH_4~~ its oxidation in the water column, the amount~~s~~ of CH_4 emitted from lakes to the atmosphere can be substantially reduced by 51 to 100~~-~~% (Bastviken et al., 2002; Bastviken et al., 2008).

Interestingly, spatial patterns of CO_2 and CH_4 emissions could not be well reproduced from ~~sediment~~ slurry incubations. Although highest CO_2 emissions were observed at site S.150, we found lowest CO_2 production rates there. Additionally, we did not see any correlations between sediment OM quality and CO_2 fluxes, suggesting that CO_2 production processes in the sediment overlying water column play a much more important role than in the sediment itself. Moreover, CH_4 production rates at sites 1.50 and 3.125 differed significantly from each other, while this was not the case when comparing fluxes. We assume that the different patterns of CH_4 fluxes and production can be explained by grain size distribution and, thus, physical sediment properties. Sediment grain size determines sediment pore size which is essential for the evolution of ~~methane~~ CH_4 bubbles in the sediment (Boudreau et al., 2005). Ebullition is the major component of CH_4 fluxes in shallow waters, as it can efficiently bypass oxidation in the water column (Bastviken et al., 2004; Wik et al., 2013; Natchimuthu et al., 2016). We found that ebullition ~~supporting significantly to total~~was responsible for most of the CH_4 fluxes in two of our four intact sediment ~~mesocosms~~core incubations, whereas sites with higher shares of sand exhibited less ebullitive fluxes, confirming the findings of Liu et al. (2016) and (2018). The authors justify their findings with the dominant pathway of bubble formation in the sediment, which is by displacing surrounding sediment particles. As this mechanism is more efficient in soft silty sediments compared to sandy material, CH_4 bubbles likely accumulate more easily in silt, creating a network of macropores and therefore conduits for subsequent bubble release. We further found that OM quality ~~being~~ partly exhibited significant negative correlations with ~~significantly correlated to~~ actual CH_4 fluxes, but to a lesser extent than ~~to~~ with sediment CH_4 production. As said, when preparing slurry incubations, the physical sediment structure is destroyed, so that OM quality likely becomes the major controlling factor for GHG production. These findings suggest that ~~grain size distribution is~~ besides OM quality, grain size distribution is a main driver of spatial CH_4 flux patterns in intact sediment core incubations~~mesocosms~~ and that only a combination of the sediment's physical characteristics and its OM quality could sufficiently explain CH_4 emission patterns from lakes.

~~On the other hand, when preparing incubations, the physical sediment structure is destroyed, so that OM quality becomes the major controlling factor for gas production. Although we also found OM quality being significantly correlated to mesocosm fluxes, we suppose that the combination of physical characteristics and sediment OM quality can sufficiently explain~~ ~~CH_4~~ ~~emission patterns from lakes.~~

The missing link between sediment gas storage and both diffusive and ebullitive emissions ~~could~~ can potentially be explained by our sampling procedure. While fluxes were partly dominated by bubbles, these bubbles cannot be captured in the sediment by silicon samplers. Silicon samplers rely on the slow diffusion of gas between the pore gas and the sampler~~.~~, whereas ~~B~~bubbles ~~however~~ may form on shorter time-scales and ~~could~~ can quickly be~~ing~~ released to the water~~-~~ column. Thus, concentrations in the bulk sediment stay ~~similar~~ consistent while the released bubbles cause high fluxes. Moreover, the resolution of our gas samplers was probably not high enough, as they could not cover~~not covering~~ the uppermost centimeters of the sediment, which we assume to be crucial for diffusive fluxes.

5 Conclusion

Our ~~experiment~~ results show~~ed~~ that there exists a significant spatial variability of CO_2 and CH_4 production in the sediment of a small and shallow lake and, therefore, that it is~~ therefore~~ not possible to upscale sediment production rates from single point measurements. We further proved that ~~especially~~ CH_4 production ~~is~~ strongly ~~depending~~ depends on temperature, and that the extent of temperature dependence might differ within ~~one~~ a lake, especially between littoral and central parts due to ~~different~~ differences in OM quality. Small lakes seem to be very ~~vulnerable~~ sensitive to temperature increases in terms of accelerated C cycling, which implies they might become larger sources of CO_2 and particularly of CH_4 emissions under climate change scenarios. ~~Still, they~~Although such lakes might not react homogeneously, but we ~~could~~ would ~~unroll~~ expect that sediments in shallower parts might react more sensitively due to ~~lower water columns~~shallow water depths and input of reactive allochthonous OM, which is particularly important for oligotrophic lakes without significant contribution of autochthonous primary production. The strong negative correlations we found between recalcitrant OM compounds and CO_2 and CH_4 production rates ~~proved~~ provide evidence that OM quality plays a major role in controlling the mineralization of anoxic lake sediment and can be used to explain and predict within-lake patterns of production rates. Both hydrogenotrophic and acetoclastic methanogenesis were feasible during slurry incubations, but we could ~~neither~~ not clearly ~~refer~~ attribute observed patterns to OM quality ~~n~~or ~~electron exchange capacities~~EEC~~,~~; ~~and~~ therefore, we suggest that ~~both H_2 and acetate were not limited as substrates but that~~ fermentation was the rate-determining step of OM mineralization and subsequent consumption of acetate and H_2 by electron accepting processes or methanogenesis kept the levels of these fermentation products low as expected. We did not find clear patterns between EAC and CH_4 production, but organic and inorganic EAs could explain up to 91% of the observed non-methanogenic CO_2-production in slurry incubations. ~~More than organic or inorganic EAs alone, EAC~~~~$_{tot}$~~ ~~could explain observed variabilities in CH_4 production, implying that also organic EAs play a major role in constantly anoxic lake sediments with high C contents.~~

~~However~~In sum, measured production rates were only partly reflected in CO_2 and CH_4 flux patterns obtained from ~~whole~~ intact sediment core incubations. We ~~suggest~~ suppose that this ~~is~~ was because the physical sediment structure (e.g. pore size) were destroyed in the slurry incubations~~due to the destruction of physical sediment parameters in the incubations (e.g. pore size)~~ , but these parameters are crucial for the evolution of CH_4 bubbles in the sediment.~~ which are crucial for the evolution of CH_4 bubbles in the sediment~~ We also suppose this was because in the slurry incubations, ~~and due to the lack of~~ thermodynamic and gas transport constraints that exist in intact anoxic sediment profiles were removed. Further, ~~measuring~~ the interpretation of production rates only would neglect~~s~~ the importance of the water column as a sink of sediment generated CH_4 through oxidation and source of CO_2 through degradation and respiration processes which might then cause discrepancies between observed production and flux rates.

So far, direct flux measurements of CO_2 and CH_4 between water and the atmosphere seem to be the most accurate way ~~for determining~~to determine the magnitude and variability of emissions from lakes, but still our study contributes substantially to the understanding of controls ~~on~~ underlying potential CO_2 and CH_4 production ~~processes~~ in lake sediments and sediment-inherent factors determining emission.

6 Data availability

The underlying research data of this study can be found in the Supplement.

7 Author contribution

LP, MS, SS, NP and KHK designed the study. NP and SS carried out the experiments and sample analyses with the help of LP and MS. LP performed data analyses and wrote the manuscript with the help of NP, SS, KHK and GB.

8 Competing interests

The authors declare that they have no conflict of interest.

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Supporting Information to: Organic matter and sediment properties determine in-lake variability of sediment CO_2 and CH_4 production and emissions of a small and shallow lake

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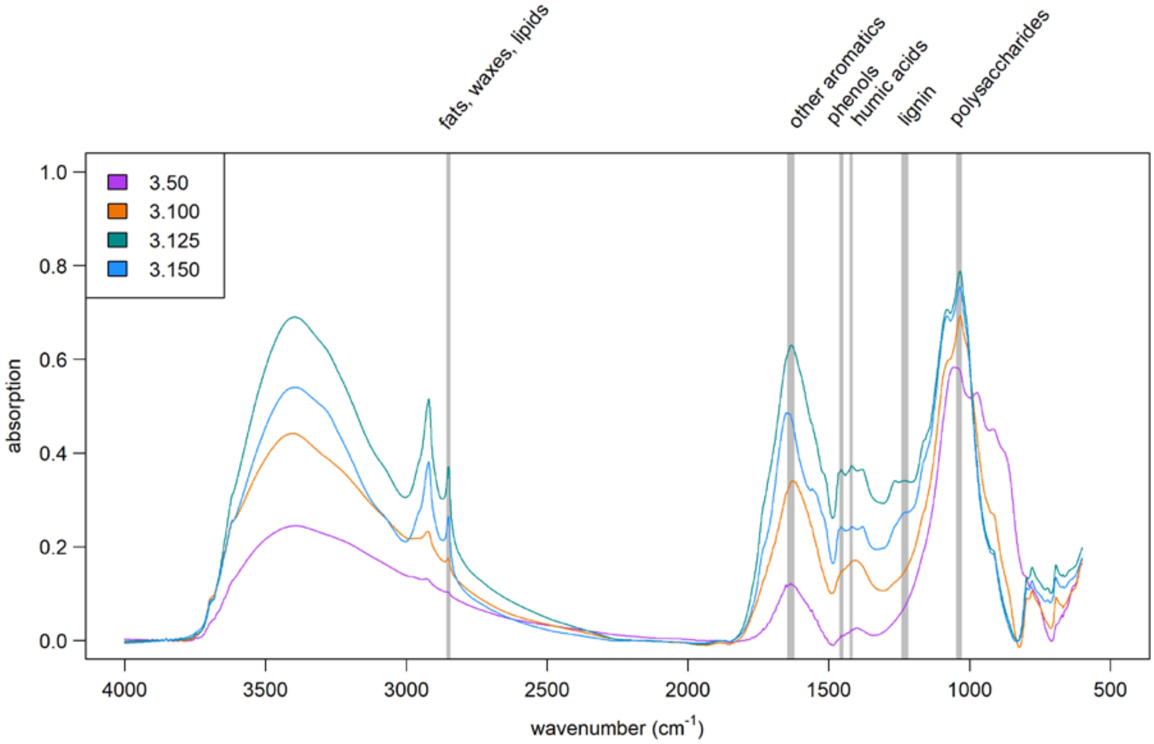


Figure S1: *FTIR spectra of samples from transect 3.*

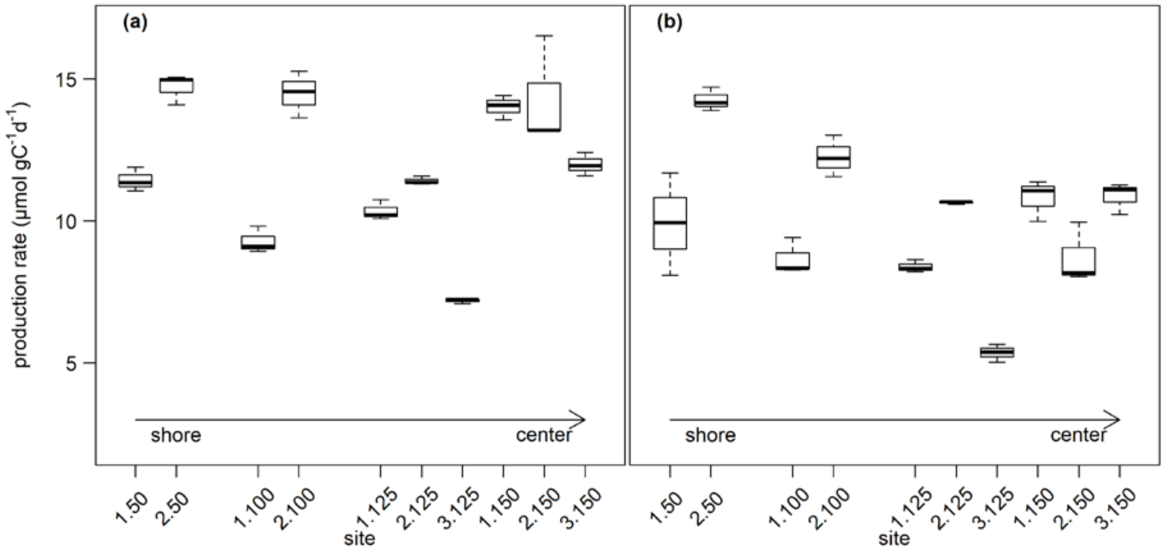


***Figure S2:** CO$_2$ (a) and CH$_4$ (b) production rates in 5-10 cm sediment depth. n = 3.*

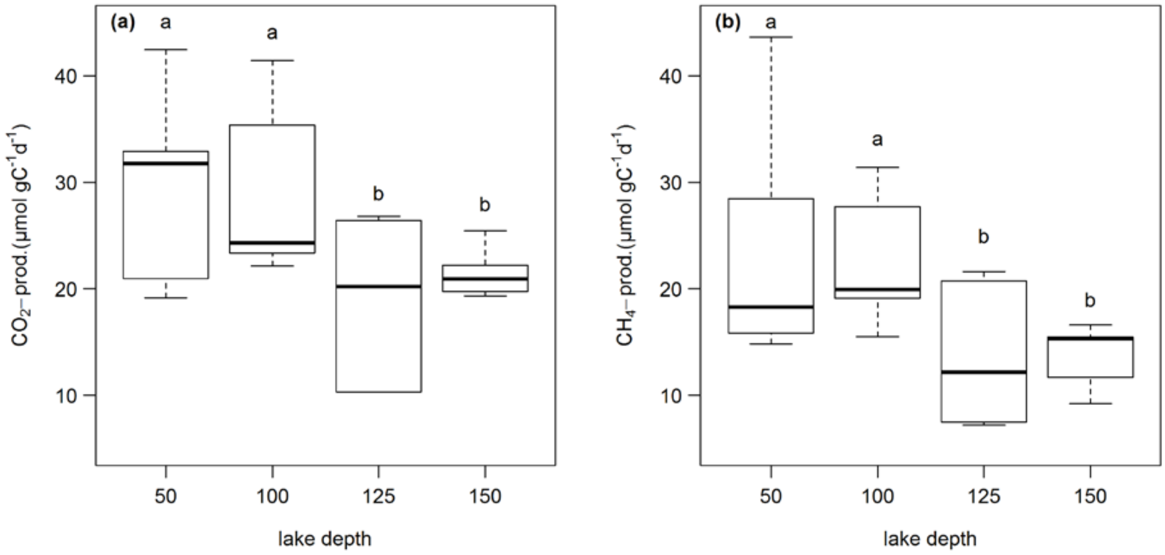
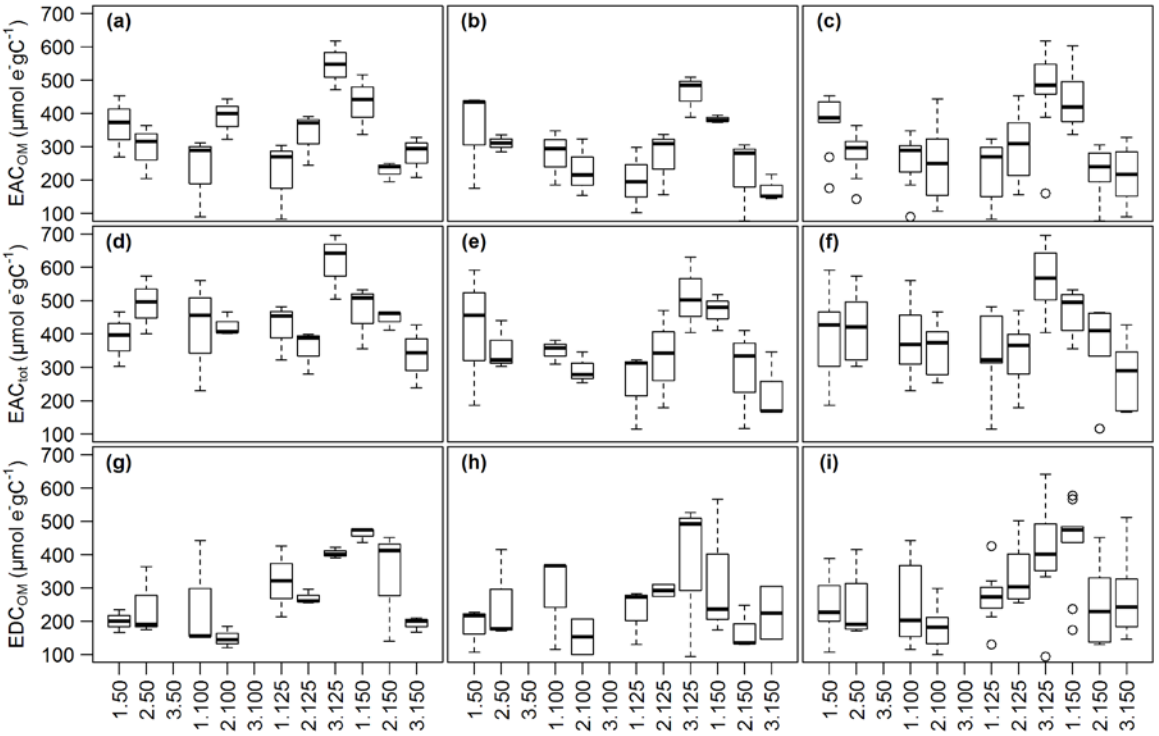


***Figure S3:** CO$_2$ (a) and CH$_4$ (b) production rates in the upper 5 cm of the sediment vs. lake depth. n=12. Different letters denote significant differences between groups.*



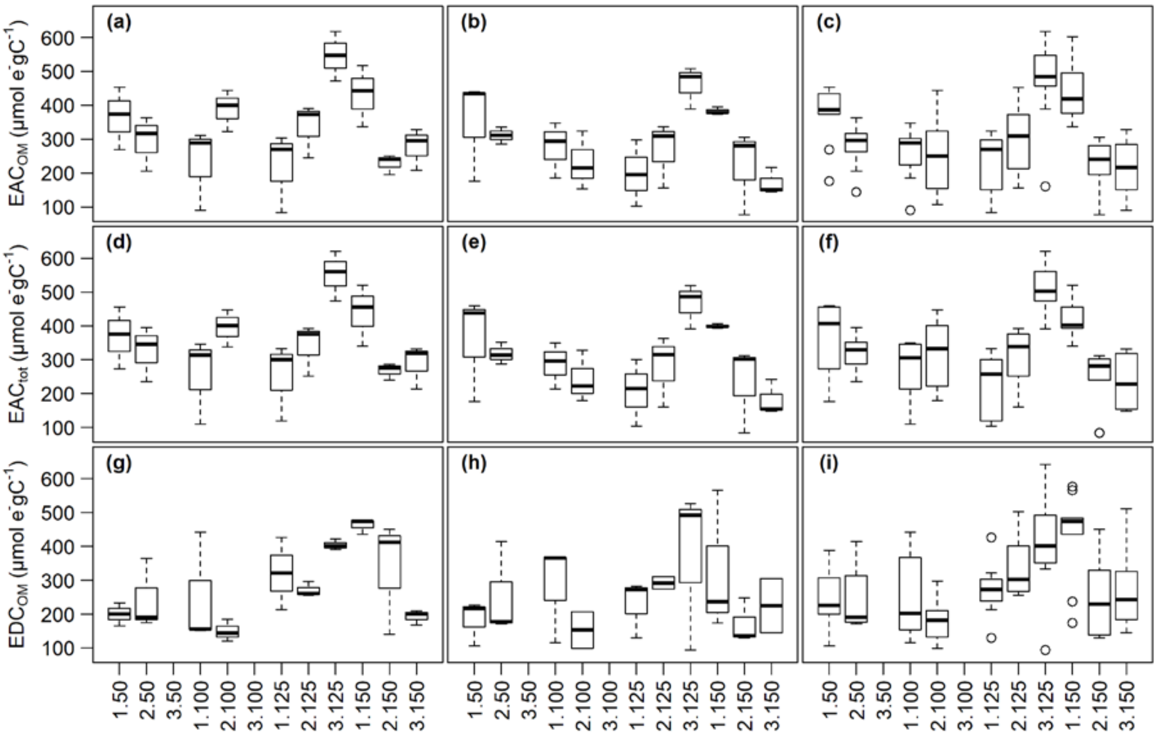


Figure ~~S3~~S4: *Spatio-temporal variability of* EAC_{OM} *(a, b, c),* EAC_{tot} *(d, e, f) and* EDC_{OM} *(g, h, i) in the incubation experiment at the beginning (a, d, g, n = 3), and the end (b, e, h, n = 3) of the experiment as well as average values for the whole experiment (c, f, i, n = 6).*

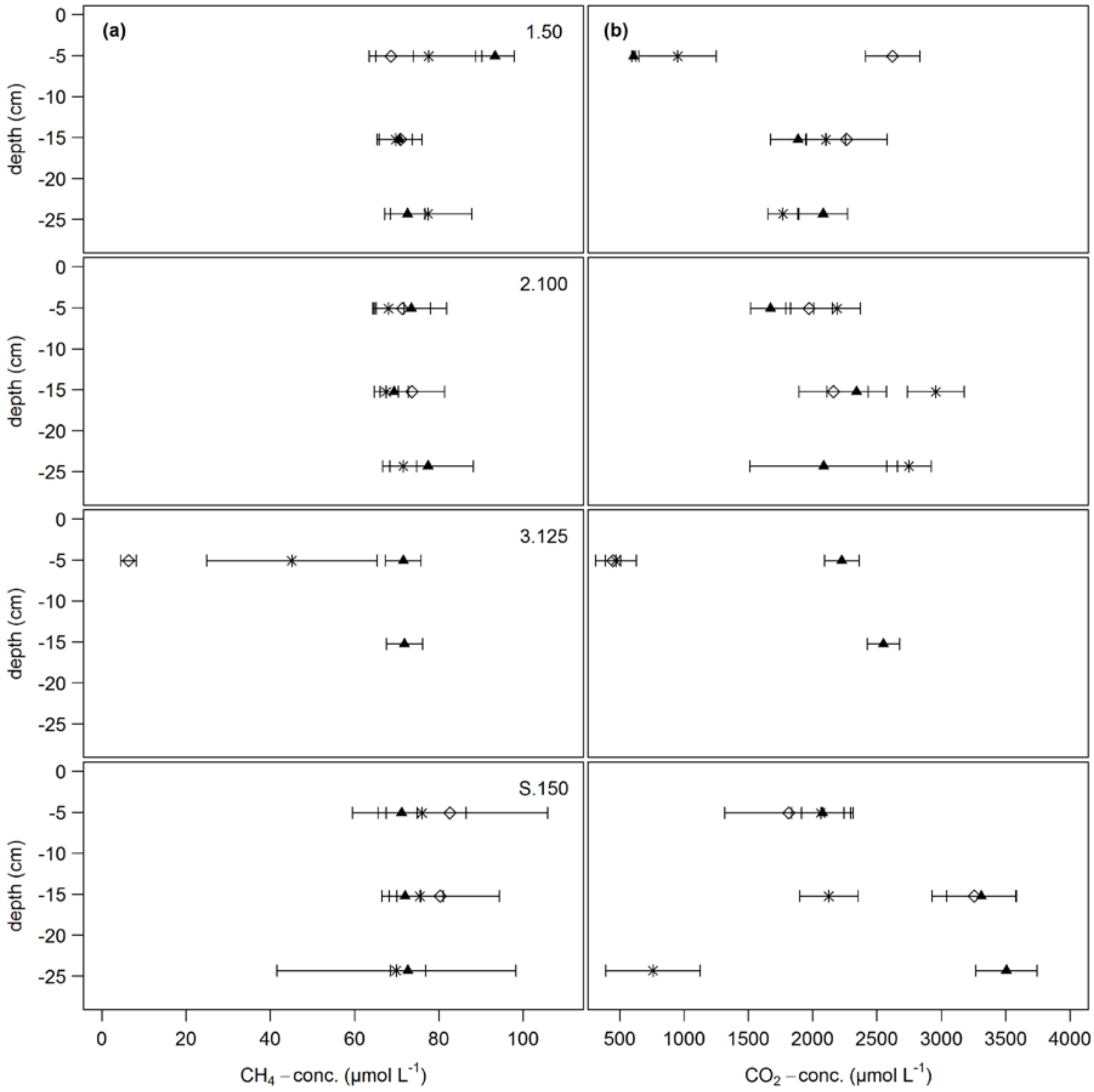


***Figure ~~S4~~S5:** Depth profile of CH_4 (a) and CO_2 (b) concentration in the sediment of the mesocosms. Different symbols denote three replicates at each site. Values are means over sampling period ± SD. n = 2-10*

Table S1: *Spearman's rank correlation coefficients and significance levels for CO_2 and CH_4 production, Q_{10}-values and EAC and EDC with all other ~~all~~ measured parameters.* ~~*Italic numbers denote no significant correlations*~~*n.s. means that correlations were not significant ($p > 0.05$). ACM = acetoclastic methanogenesis, HTM = hydrogenotrophic methanogenesis*.

	~~**CH_4 production**~~		~~**CO_2 production**~~		~~**Q_{10} (CH_4)**~~		~~**acetoclastic methanog.**~~		~~**EDC**~~		~~**EAC_{OM}**~~		~~**CH_4 flux**~~		~~**CO_2 flux**~~	
	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~	~~***p***~~	~~***rho***~~
~~**aromatics**~~	~~0.001~~	~~-0.669~~	~~0.002~~	~~-0.641~~	~~0.034~~	~~-0.821~~							~~*0.132*~~	~~*-0.595*~~	~~*0.197*~~	~~*-0.524*~~
~~**fats, waxes, lipids**~~	~~0.000~~	~~-0.700~~	~~0.000~~	~~-0.736~~	~~0.034~~	~~-0.821~~			~~0.001~~	~~0.565~~			~~0.015~~	~~-0.833~~	~~*0.428*~~	~~*-0.333*~~
~~**humic acids**~~	~~0.003~~	~~-0.618~~	~~0.001~~	~~-0.653~~	~~0.034~~	~~-0.821~~							~~0.015~~	~~-0.833~~	~~*0.389*~~	~~*-0.357*~~
~~**lignin**~~	~~0.003~~	~~-0.606~~	~~0.003~~	~~-0.610~~	~~0.034~~	~~-0.821~~							~~0.021~~	~~-0.786~~	~~*0.352*~~	~~*-0.381*~~
~~**phenols**~~	~~0.003~~	~~-0.606~~	~~0.001~~	~~-0.667~~	~~0.034~~	~~-0.821~~							~~0.015~~	~~-0.833~~	~~*0.389*~~	~~*-0.357*~~
~~**C**~~	~~*0.194*~~	~~*-0.287*~~	~~*0.327*~~	~~*-0.219*~~									~~*0.058*~~	~~*-0.714*~~	~~*0.665*~~	~~*-0.190*~~
~~**C/N**~~	~~*0.356*~~	~~*-0.206*~~	~~*0.378*~~	~~*-0.197*~~			~~0.013~~	~~0.447~~					~~0.007~~	~~-0.881~~	~~*0.428*~~	~~*-0.333*~~
~~**H_2 concentration**~~	~~0.000~~	~~0.450~~	~~0.000~~	~~0.515~~							~~*0.157*~~	~~*-0.285*~~				
~~**acetate concentration**~~	~~*0.492*~~	~~*0.248*~~	~~*0.191*~~	~~*0.455*~~							~~0.002~~	~~-0.387~~				
~~**hydrogenotrophic**~~											~~0.031~~	~~0.426~~				
~~**acetoclastic**~~											~~*0.748*~~	~~*0.042*~~				
~~**EAC_{OM}**~~	~~*0.946*~~	~~*0.030*~~	~~*0.470*~~	~~*-0.261*~~												
~~**EAC_{inorg}**~~	~~*0.204*~~	~~*-0.442*~~	~~*1.000*~~	~~*0.006*~~												
~~**EAC_{tot}**~~	~~*0.191*~~	~~*-0.455*~~	~~*0.470*~~	~~*-0.261*~~												
~~**EDC**~~	~~0.031~~	~~-0.697~~	~~0.039~~	~~-0.673~~												
~~**EAC/EDC**~~	~~*0.247*~~	~~*0.406*~~	~~*0.407*~~	~~*0.297*~~												
~~**EEC**~~	~~*0.166*~~	~~*-0.479*~~	~~*0.054*~~	~~*-0.636*~~												
~~**S (%)**~~	~~0.019~~	~~-0.577~~	~~*0.087*~~	~~*-0.441*~~												
~~**clay**~~													~~0.023~~	~~0.648~~	~~0.037~~	~~0.605~~
~~**silt**~~													~~*0.101*~~	~~*0.497*~~	~~*0.340*~~	~~*0.302*~~
~~**sand**~~													~~0.023~~	~~-0.648~~	~~0.037~~	~~-0.605~~
~~**CH_4 stock change**~~													~~*0.163*~~	~~*-0.222*~~	~~*0.714*~~	~~*-0.064*~~
~~**CO_2 stock change**~~													~~*0.762*~~	~~*-0.049*~~	~~*0.776*~~	~~*0.050*~~

	CH_4 production			**CO_2 production**			**Q_{10} (CH_4)**			**EAC_{OM}**			**EAC_{tot}**			**EDC**		
	p	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***
aromatics	< 0.001	-0.669	22	< 0.01	-0.641	22	< 0.05	-0.821	7	*n.s.*	*0.030*	10	*n.s.*	*0.042*	10	*n.s.*	*0.515*	10
fats, waxes, lipids	< 0.001	-0.700	22	< 0.001	-0.736	22	< 0.05	-0.821	7	*n.s.*	*0.321*	10	*n.s.*	*0.333*	10	< 0.05	0.758	10
humic acids	< 0.01	-0.618	22	< 0.01	-0.653	22	< 0.05	-0.821	7	*n.s.*	*0.067*	10	*n.s.*	*0.042*	10	*n.s.*	*0.455*	10
lignin	< 0.01	-0.606	22	< 0.01	-0.610	22	< 0.05	-0.821	7	*n.s.*	*0.031*	10	*n.s.*	*-0.006*	10	*n.s.*	*0.275*	10
phenols	< 0.01	-0.606	22	< 0.001	-0.667	22	< 0.05	-0.821	7	*n.s.*	*0.152*	10	*n.s.*	*0.115*	10	*n.s.*	*0.479*	10
C	*n.s.*	*-0.287*	22	*n.s.*	*-0.219*	22	*n.s.*	*-0.393*	7	*n.s.*	*-0.394*	10	*n.s.*	*-0.370*	10	*n.s.*	*-0.321*	10
C/N	*n.s.*	*-0.206*	22	*n.s.*	*-0.197*	22	*n.s.*	*-0.074*	7	*n.s.*	*-0.079*	10	*n.s.*	*-0.115*	10	*n.s.*	*-0.103*	10
$\delta^{13}C$	*n.s.*	*0.134*	22	*n.s.*	*0.091*	22	*n.s.*	*-0.321*	7	*n.s.*	*0.309*	10	*n.s.*	*0.224*	10	*n.s.*	*0.164*	10
$\delta^{15}N$	*n.s.*	*-0.267*	22	*n.s.*	*-0.281*	22	*n.s.*	*-0.071*	7	*n.s.*	*0.055*	10	*n.s.*	*-0.006*	10	*n.s.*	*-0.164*	10
H_2 conc.	< 0.001	0.450	22	< 0.001	0.515	22				*n.s.*	*-0.285*	26	*n.s.*	*-0.335*	26	*n.s.*	*-0.139*	26
acetate conc.	*n.s.*	*0.248*	10	*n.s.*	*0.455*	10				< 0.01	-0.387	60	< 0.001	-0.418	60	*n.s.*	*-0.035*	60
HTM	< 0.01	-0.394	22	< 0.001	-0.516	22				< 0.05	0.426	26	< 0.05	0.426	26	*n.s.*	*0.162*	26
ACM	*n.s.*	0.491	10	*n.s.*	0.297	10				*n.s.*	*0.042*	60	*n.s.*	*0.042*	60	*n.s.*	*-0.208*	60
EAC_{OM}	*n.s.*	*0.030*	10	*n.s.*	*-0.261*	10												
EAC_{inorg}	*n.s.*	*-0.261*	10	*n.s.*	*0.152*	10												
EAC_{tot}	*n.s.*	*-0.042*	10	*n.s.*	*-0.285*	10												
EDC	< 0.05	-0.697	10	< 0.05	-0.673	10												
EAC/EDC	*n.s.*	*0.406*	10	*n.s.*	*0.297*	10												
EEC	*n.s.*	*-0.479*	10	*n.s.*	*-0.636*	10												
S	*n.s.*	*-0.411*	16	< 0.05	-0.446	16												
P	*n.s.*	*0.282*	22	*n.s.*	*0.305*	22												
Fe	*n.s.*	*0.338*	22	< 0.05	0.453	22												
Mn	*n.s.*	*0.020*	22	*n.s.*	*0.121*	22												
CO_2 conc.										*n.s.*	*-0.161*	20	*n.s.*	*-0.146*	20	*n.s.*	*-0.252*	20
CH_4 conc.										*n.s.*	*-0.083*	20	*n.s.*	*-0.080*	20	*n.s.*	*-0.219*	20

Table S2: Spearman's rank correlation coefficients and significance levels for Gibb’s free energy of acetoclastic (ACM) and hydrogenotrophic (HTM) methanogenesis, acetate and H_2 *concentrations with OM quality parameters. n.s. means that correlations were not significant (*$p > 0.05$*).*

	ACM (t6)			**acetate conc. (t6)**			**HTM**			**H_2 conc.**		
	p	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***	***p***	***rho***	***n***
aromatics	*n.s.*	*-0.052*	30	< 0.001	-0.585	30	*n.s.*	*0.136*	154	< 0.05	-0.187	174
fats, waxes, lipids	*n.s.*	*-0.237*	30	< 0.05	-0.440	30	< 0.01	0.255	154	< 0.001	-0.352	174
humic acids	*n.s.*	*0.082*	30	< 0.001	-0.634	30	< 0.05	0.183	154	< 0.01	-0.229	174
lignin	*n.s.*	*0.265*	30	< 0.01	-0.533	30	*n.s.*	*0.140*	154	< 0.05	-0.190	174
phenols	*n.s.*	*0.104*	30	< 0.001	-0.668	30	< 0.05	0.196	154	< 0.01	-0.246	174
C	*n.s.*	*-0.062*	30	*n.s.*	*0.074*	30	< 0.05	0.174	154	< 0.05	-0.149	174
C/N	< 0.05	0.447	30	< 0.01	-0.538	30	*n.s.*	*-0.057*	154	*n.s.*	*0.102*	174