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## Inflow of brackish water and a preceding drought changes methanecycling microbial communities in a freshwater rewetted coastal fen

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Rewetted peatlands can be a significant source of methane, but in coastal systems, input of sulfate-rich seawater could potentially reduce these emissions. The presence of sulfate is known to suppress methanogenesis, by encouraging the growth of sulfate-reducers, which outcompete methanogens for substrate. After a drought in 2018 and a storm surge in the following winter, we investigated the effects of the drought and the brackish water inflow on the microbial communities relative to methane exchange in a rewetted fen at the southern Baltic Sea coast.

We took peat cores at four previously sampled locations along a salinity gradient to compare the soil and pore water geochemistry as well as the microbial methane and sulfate cycling communities to the common freshwater rewetting state and the drought 2018. We used high-throughput sequencing and quantitative polymerase chain reaction (qPCR) to characterize pools of DNA and cDNA targeting total and putatively active bacteria and archaea. While sequencing was done for the 16S rRNA gene, qPCR was performed on key functional genes of methane production (*mcrA*), methane oxidation (*pmoA*) and sulfate reduction (*dsrB*) in addition to 16S rRNA. Furthermore, we measured local methane (CH<sub>4</sub>) fluxes with closed chambers and retrieved soil plugs to determine the concentrations and isotopic signatures of dissolved gases in the pore water.

The sequence of the drought and the inflow of brackish water increased the absolute abundance of sulfate reducing bacteria (SRB) by two orders of magnitude. We did not observe a decrease of absolute methanogenic archaea abundance after the inflow as we expected parallel to the increase of SBRs, but saw that changes in the methanogenic communities' compositions already took place in the drought year 2018. After the inflow, absolute abundance of aerobic methanotrophic bacteria decreased back to their pre-drought level, following an increase during 2018 drought conditions. The expected establishment of methanotrophic archaea (ANME), which are capable of sulfate-mediated anaerobic methane oxidation, was not recorded though. While CH<sub>4</sub> fluxes showed a strong decline of almost 90 % to a new minimum since rewetting in 2009, dissolved CH<sub>4</sub> pore water concentrations and a strong depletion of <sup>13</sup>C-values of CH<sub>4</sub> and CO<sub>2</sub> (DIC) indicated ongoing methanogenesis and lack of methane oxidation after the brackish water inflow. The observed reduction of CH<sub>4</sub> emissions might be a result of methane oxidation and sulfate reduction in the brackish water column above the peat soil. The legacy effect of the preceding drought likely influenced the microbial communities and pore water geochemistry simultaneously suggesting a mixed effect of drought and inflow. Overall, our study revealed that the sequence of drought conditions followed by the inflow of brackish water enlarged the sulfate reducing microbial communities and substantially reduced the CH<sub>4</sub> emissions in a rewetted fen. However, unlike drought, which is associated with a rapid and irreversible peat degradation through aerobic decomposition processes, brackish water inflow encourages peat preservation by maintaining anaerobic conditions. Still, further research is needed to directly study the complex effects of brackish water rewetting on peatlands.