Quantitative analysis of microbial abundance within Arctic fjord sediments assessed through direct counting

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ABSTRACT

Microbes found in the marine sediments are responsible for the production of nearly half of the carbon dioxide found in the atmosphere (Arrigo, 2005). The fjords of Svalbard (79°N) are not considered typical marine sediments because high iron content influences unique subsurface redox chemistry. Radiocarbon studies have shown that these sediments contain active bacterial sulfate-reducing communities (Finke et al., 2016). In addition to bacteria, archaeal cells within these sediments have been in aggregates encompassed by sulfate-reducing bacteria (Raveneschlag et al., 2001). These anaerobic organisms participate in mediating environmental biogeochemical cycles, including the oxidation of methane (Raveneschlag et al., 2001) and iron reduction (Vandiekene et al., 2006). These observations have led to the understanding of the important role that these microbes play in global nutrient cycling and have led to recent interest in how these communities will respond to glacial retreat associated with climate change (Arrigo, 2005). However, the abundance of microbes across the fjords in Svalbard is relatively unknown. Here, we show the abundance of is in view of two visually unstudied fjords, Van Keulenfjorden and Kongfjorden. We found a general trend of lower microbial abundance after around 10 cm in depth. For relative abundance, we saw numbers around 10^7 cells per ml in Van Keulenfjorden, which is slightly lower than counts of 165 copy numbers acquired through quantitative PCR (qPCR). Our results demonstrate that there are varying levels of microbial abundance in the fjord sediments of Svalbard. Moving forward, this research could help contribute to a better understanding of the effect that these microbes have on nutrient cycling at both a local and global level by coupling abundance data with functional analyses. Furthermore, we hope that this research will enhance the ability of predicting the effects that climate change and glacial runoff have on these microbial communities and carbon cycling in the Arctic Circle.

OBJECTIVE

The objective of this study is to determine microbial abundance in the sediments of Svalbard fjords with the intent to enhance global carbon cycling models. The trajectory of carbon dynamics in the Arctic is hard to predict with climate warming, and so we take the first steps to uncovering the effect that these microbes have on cycling of nutrients at both the local and global scale.

METHODS

Samples were collected on site (Fig. 1) and fixed with paraformaldehyde, then washed with PBS and centrifuged x 300g for 5 minutes three times. They were then stored in PBS/ETOH in a −20°C freezer. Samples were then diluted to a 1:10 dilution (to ensure the quantification limit was reached) in PBS and sonicated for 40 seconds. In preparation for staining, 20 ul of the diluted sample was combined with 5 ml of PBS and vortexed. Then, the samples were filtered using a vacuum filtration system, absorbant pads, and 0.2 um filters. The samples were then stained using SYBR Gold stain for 7 minutes. The filters were then washed with PBS for 5 seconds. Following this, the samples were mounted onto microscope slides and Vectashield was used to place the coverslips on the filters. The slides were then viewed using either an FITC or DE FGE light filter. The cells were counted using direct counting methods at 100x and then converted to cells/ml by accounting for filter size, dilution, and amount of sediment placed on filter. Two student researchers counted the same samples to test reproducibility and user bias.

PRELIMINARY RESULTS

Fig. 2 SYBR Gold Direct Counts. Counts for A:B:1 samples taken from Van Keulenfjorden. Cells per ml wet sediment around 10^7 consistently for both direct count data. The general trend of Rachel’s count went down for the deeper samples while Alex’s counts showed no general trend.

PRELIMINARY FINDINGS

Our preliminary findings are in contrast to previous studies conducted in a nearby fjord, which shows as many as 10^9 cells in sediments (Raveneschlag et al., 2001). The cause for this discrepancy could be due to a number of factors, including geography and relatively different glacial cover. However, it is more likely that washing steps introduced during sample preparation caused decreased yield. To test the hypothesis that wash and spin steps result in underreporting of cells, we designed a new protocol.

FUTURE WORK

If our hypothesis is correct, and cell loss can be traced to previously used methods, a more accurate estimate of microbial biomass in these sediments can be attained. However, should the adapted methods not alleviate cell number discrepancies between this data and the data found in previous studies, further study will be needed to determine the cause of these differences. Such a large difference in cell abundance between fjords may reveal environmental variances not before realized in these fjords responsible for microbial abundance.

This work will better our understanding of how microbial communities are shaped by their Arctic sediment environment. Ultimately, the data produced from our adapted method experiments will aid us in estimating microbial biomass living in Arctic sediment, which is the first step to understanding how microbial communities may respond to climate warming and glacial retreat.

REFERENCES