Elevated bacterial endospores associated with thermogenic hydrocarbon seeps in deep sea sediments.

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Abstract

Introduction and approach

Bacterial endospore distributions in marine sediments are influenced by geological conduits providing routes for subsurface to surface microbial dispersal. To examine this phenomenon in more detail, endospore abundance was determined by quantifying the biomarker 2,6-pyridine dicarboxylic acid (dipicolinic acid or DPA) in 16 deep sea sediment cores from hydrocarbon prospective areas in the NW Atlantic Ocean. DPA is specific to endospore-forming bacteria from the phylum Firmicutes and constitutes a significant percentage of endospore dry weight. DPA is therefore a potential biomarker for sediment dwelling endospores and geological conduits.

Piston cores (10), gravity cores (3) and box cores (3) were collected during two expeditions to the Scotian Slope in the NW Atlantic Ocean off the east coast of Canada aboard the CCGS Hudson in 2016 and 2018 (Campbell (2016), Campbell and Normandeau (2018), Campbell and MacDonald. (2016)). Sampling sites were 1970 to 2791 m water depth, with piston cores (n=3) ranging from 344 to 953 cmbsf and gravity cores (n=10) ranging from 43 to 739 cmbsf, box coring captured the top 25 cmbsf. To address the efficacy of DPA biomarker analysis as a tool for hydrocarbon seep location we established a modified Tb^{3+} chelation method (Lomstein and Jørgensen (2012), Rattray (2021)). Sediment samples were extracted using acid hydrolysis, chelated with Tb3+ and analysed using HPLC fluorescence, measuring at 270 nm emission and 545 nm excitation. DPA concentrations were converted to Endospore numbers were calculated using 2.24 fmol DPA per endospore (Fichtel 2007), a conversion factor routinely used in other studies (Braun 2017, Gittins 2022, Heuer 2020, Lomstein 2012, Rattray 2022, Wörmer 2019, Lomstein and Jørgensen 2012). DPA concentrations were compared with measurements of over 250

different gaseous and liquid hydrocarbon compounds used to assess for the presence of thermogenic hydrocarbons.

Results and discussion

Samples and locations were assessed as being thermogenic hydrocarbon gas positive (stations 16-41, 18-07) or thermogenic hydrocarbon negative based on the abundance of C1-C5 hydrocarbons in sediments sampled from the same cores. Station 18-14 contained hydrocarbons from biogenic origin. Station 18-06 is the only site with higher endospore abundance but that was determined to be hydrocarbon negative.

Deep water Scotian Slope sediment cores show high endospore abundance correlates with thermogenic hydrocarbon seeps (Fig. 1). Cores from locations lacking evidence for thermogenic hydrocarbons generally contained significantly lower endospore abundances, with the notable exception of site 18-06. This potential paleoenvironmental hydrocarbon seep site highlights the utility of a DPA proxy for potentially identifying ancient hydrocarbon seeps and investigating past geological systems. The association of high endospore abundances with thermogenic hydrocarbons and the quantity of gas expulsion points to an interesting new biological tool for understanding hydrocarbon seepage in the deep biosphere, based on DPA assays in marine sediments.

Keywords

Endospores, Dipicolinic acid, hydrocarbon seeps, HPLC metal chelation chromatography

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Conflicts of interest

The authors have declared that no competing interests exist.

References

- Braun S, et al. (2017) Microbial turnover times in the deep seabed studied by amino acid racemization modelling. Scientific Reports 7 (1).
- Campbell D, et al. (2016) CGS Hudson Expedition 2016-011, phase 2. Cold seep investigations on the Scotian Slope, offshore Nova Scotia, June 15-July 6, 2016. Geological Survey of Canada Open File 8525. https://doi.org/10.4095/313603
- Campbell D, Normandeau A (2018) CCGS Hudson Expedition 2018-041: high-resolution investigation of deep-water seabed seeps and landslides along the Scotian Slope, offshore Nova Scotia, May 26 - June 15, 2018. https://doi.org/10.4095/314695
- Campbell DC, MacDonald. AW (2016) CCGS Hudson Expedition 2015-018. Geological Survey of Canada, Open File 8116.
- Fichtel J, et al. (2007) Spore dipicolinic acid contents used for estimating the number of endospores in sediments. Fems Microbiology Ecology 61 (3): 522-532. https://doi.org/10.1111/j.1574-6941.2007.00354.x
- Gittins DA, et al. (2022) Geological processes mediate a microbial dispersal loop in the deep biosphere. Science Advances 8 (34).
- Heuer VB, et al. (2020) Temperature limits to deep subseafloor life in the Nankai Trough subduction zone. Science 370 (6521).
- Lomstein BA, et al. (2012) Endospore abundance, microbial growth and necromass turnover in deep sub-seafloor sediment. Nature 484 (7392).
- Lomstein BA, Jørgensen BB (2012) Pre-column liquid chromatographic determination of dipicolinic acid from bacterial endospores. Limnology and Oceanography-Methods 10: 227-233. https://doi.org/10.4319/lom.2012.10.227
- Rattray JE, et al. (2021) Sensitive quantification of dipicolinic acid from bacterial endospores in soils and sediments. Environmental Microbiology 23 (3): 1397-1406. https://doi.org/10.1111/1462-2920.15343

- Rattray JE, et al. (2022) Endospores associated with deep seabed geofluid features in the eastern Gulf of Mexico. Geobiology 20 (6).
- Wörmer L, et al. (2019) Microbial dormancy in the marine subsurface: Global endospore abundance and response to burial. Science advances 5 (2).

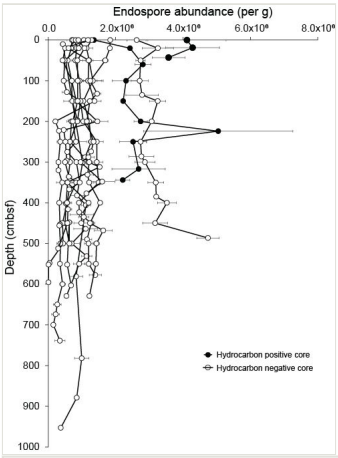

Figure 1.

Endospores g^{-1} calculated from concentrations of the biomarker dipicolinic acid measured along the full length of piston cores (3) and gravity cores (10). Mean endospore abundances per depth are plotted with error bars showing the standard deviation of triplicate measurements.