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Microbial activity in deep marine sediments: does pressure make the difference?

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Abstract. We attempted to evaluate the effects of high hydrostatic pressure on microbial heterotrophic activity in deep marine sediments from the Atlantic Ocean. We investigated the potential respiration rates (acetate/glucose oxidation to CO₂) in oxic sediments recovered from up to ~4500 m water depth. Incubations were performed at ambient pressure and at near in situ pressure (~40-45 MPa) with sediments stored at ambient pressure and at in situ pressure. Potential respiration rates in sediments stored at ambient pressure were lower when measured at in situ pressure than when measured at ambient pressure, independently of the substrate used. It appears that the pressure of storage is critical since potential respiration rates of sediments stored at in situ pressure were higher than in the counterpart sediments stored at ambient pressure.

1. Introduction

Oceans cover 2/3 of the Earth's surface; marine environments including the deep-sea, the sub-seafloor sediments and the basaltic crust account for the largest high-pressure (HP) habitat for microorganisms. Microbiologists began to consider the effects of HP in sediments as early as the end of the 19th century [1]. More than half a century later, Zobell and Johnson characterized microorganisms that grow and metabolize optimally at HP as "barophilic" [2]. Yayanos reported in 1979 the isolation of the first bacterium in pure culture growing better at HP [3], and nowadays coined "piezophilic" [4]. Today, it is clear that piezosensitive and piezophilic microorganisms share the same HP habitats, however the relative proportion and activity of each group are still unknown [5]. In the last decades it was shown that microbial activity rates in deep-sea water samples are usually higher when measured at in situ pressure conditions [6 and references therein]. Some similar observations have been made in sediments [e.g. 7], however the effects of HP on microbial activity in sediments, and more generally on microbial activity important for biogeochemical cycles, have not been extensively studied.

In this communication, we present new data on the effects of the incubation and storage pressure on microbial heterotrophic activity in deep-sea oxic sub-seafloor sediments recovered in the Atlantic Ocean.

2. Material and methods

2.1. Site description and sediment collection

We visited the North Pond area in the Atlantic Ocean (23°N) during cruise MSM 11/1 on R/V Maria S. Merian in 2009 [8]. Sediment for microbial activity measurement was sampled aerobically and stored at 4°C. Samples used in this study and their incubation/storage conditions are listed in Table 1.

Table 1. List of samples used in this study and their pressure conditions of storage and incubation.

Station (water depth)	Sampling depth (cm below seafloor)	Storage pressure at 4°C (MPa)	Incubation pressure at 4°C (MPa)
GeoB 13501 (4480 m)	42.5, 105.5, 201.5, 301.5, 401.5, 501.5, 601.5, 701.5, 801.5	0.1	0.1/45
GeoB 13506 (4143 m)	57.5, 131.5, 231.5, 331.5, 431.5, 531.5	0.1	0.1/45
GeoB 13502 (4250 m)	157 204.5	40 0.1	0.1/40 0.1/40

2.2. High-pressure incubation

Sediment samples were stored and incubated at 4°C in steel pressure vessels similar to those described by Zobell and Openheimer [9]. Sediment slurries were contained in plastic syringes or in small hungate tubes without headspace and pressurized in water using a multifluid hand pump (Enerpac MP-1000).

2.3. Microbial heterotrophic activity measurements

Microbial heterotrophic activity was defined here as the oxidation of ¹⁴C-acetate or ¹⁴C-glucose to ¹⁴CO₂ in oxic conditions. Briefly, ¹⁴C-acetate or ¹⁴C-glucose was added to the sediment slurries. Incubation was performed at 4°C at ambient pressure or close to the in situ pressure (40-45 MPa). At 3 different time points, the amount of ¹⁴CO₂ was measured and potential respiration rates were calculated. The complete procedure was described elsewhere [8].

3. Results

3.1. Effects of high pressure on microbial heterotrophic activity profiles in oxic sediments

Potential respiration rates were evaluated in two sediment cores, GeoB 13501 and GeoB13506, recovered at water depths of 4480 and 4143 m, respectively. Sediment samples from GeoB 13501 were amended with ¹⁴C-glucose and ¹⁴C-acetate and incubated at 4°C at ambient pressure and at near in situ pressure (45 MPa). Potential respiration rates, shown in Fig. 1, are comparable with both substrates (<0.1 nmol organic compound respired cm⁻³ d⁻¹), although a bit lower using glucose. With glucose, rates measured at high pressure are all lower than rates measured at 0.1 MPa (Fig. 1A and 1B). Using acetate, rates measured at high pressure are in the same range as those measured at atmospheric pressure, although rates appeared to be slightly enhanced during the 8-day incubation under pressure (Fig. 1C and 1D).

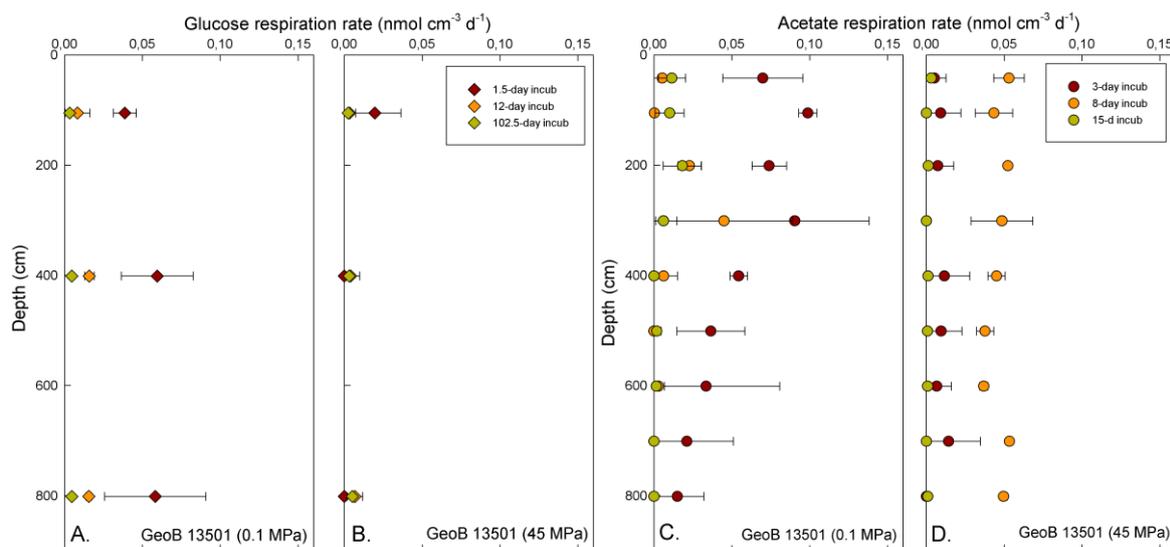


Figure 1. Microbial activity profiles at Station GeoB 13501. Glucose respiration rates measured at 0.1 MPa (A) and 45 MPa (B). Acetate respiration rates measured at 0.1 MPa (C) and 45 MPa (D).

At Station GeoB13506, rates are clearly lower at high pressure at all incubation times (Fig. 2).

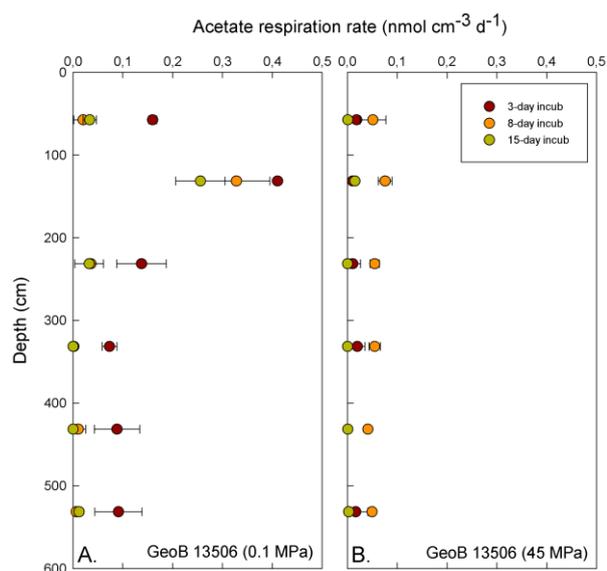


Figure 2. Microbial activity profiles measured at 0.1 MPa (A) and 45 MPa (B) at Station GeoB 13506.

3.2. Effects of storage pressure conditions on microbial activity in sediments

One sediment sample was selected in Core GeoB 13502 (157 cm below seafloor) and was pressurized at 40 MPa shortly after sampling. The pressure vessel was stored at 4°C and pressure was checked regularly for ~8 months until the experiment was performed. Since it was not possible to obtain two samples from the same depth, we used the sample taken at 204.5 cm in a similar lithological horizon as a reference stored at 0.1 MPa. The activity rates were evaluated with acetate in the two samples and incubations were performed both at 0.1 and 40 MPa (Fig. 3). Activity was apparently increased in the sample that has been stored under pressure, independently of the pressure of incubation.

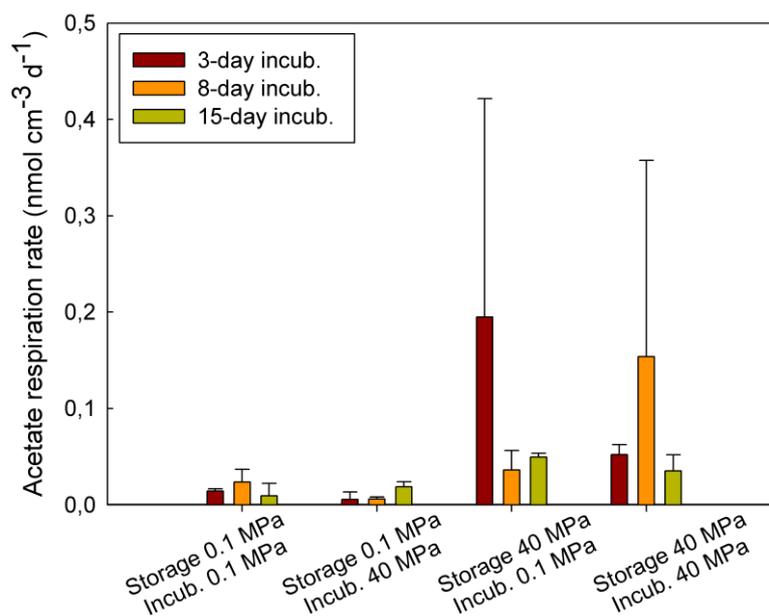


Figure 3. Acetate respiration rates measured in samples from GeoB 13502. The sample stored at 0.1 MPa was taken at 204.5 cm depth below seafloor, while the sample stored at 40 MPa was taken at 157 cm below seafloor. Both samples are of similar lithology.

4. Discussion

We show here that pressure inhibits microbial activity in deep-sea sediments recovered and stored at ambient pressure. Our results suggest also that microbial activity is preserved in sediments that have been stored at in situ pressure. How pressure affects microbial activity in sediments is not clearly established yet. Ideally, microbial activity has to be measured as soon as possible after the recovery of the sediment samples. However, in practice, an important amount of sediment samples are stored for several weeks to several months (especially when the samples have to be shipped to the laboratory via container) until performing the experiments. We suggest here that quickly restoring the in situ pressure after the recovery of deep-sea sediments could maintain the initial metabolic potential of the microbial community. Further technical developments would be necessary to sample, store and incubate deep-sea sub-seafloor sediment samples without any decompression. In conclusion, more studies on deep environmental samples are needed to draw a general conclusion. Moreover studies on the effects of high pressure on “simple” systems, such as microbial isolates from deep environments, would also greatly improve the understanding on the effects of high pressure on more complex systems, such as sub-seafloor sediments. The field of “high-pressure geomicrobiology” is obviously still in its infancy.

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