

# Dispersal and Distribution of Thermophilic Endospores in Deep-Sea Ecosystems

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## Abstract

The distance-decay relationship is a central concept in biogeography and spatial biodiversity, describing how two distinct entities decrease in similarity as the distance between them increases. The decay of community similarity with geographical distance is driven by multiple factors, such as gene drift, environmental selection, and the accumulation of mutations over time. While the distance-decay relationship has been recognized for several decades, there are certain circumstances where the biogeographical patterns of certain species and communities cannot be predicted by this relationship. The example addressed in this project is the case of thermophilic endospore-forming bacteria found in permanently cold deep ocean sediments.

Thermophilic endospores (thermospores) are routinely found on the deep ocean floor, a permanently cold environment that does not support their metabolic activity (Hubert et al. 2009). Thermospores are metabolically dormant states developed by some thermophilic bacteria having optimal growth temperatures between 40°C and 70°C. There is evidence that these heat-loving bacteria originate from the deep subsurface and are transported upward to the deep ocean via geological features of the oceanic crust including geofluid fluxes in high-temperature axial systems and natural hydrocarbon seeps (Gittins et al. 2022). Due to the ability of endospores to stay viable for thousands of years and resist a wide range of physicochemical stressors, they can disperse over long distances while remaining unaffected by changing factors such as selection, drift, or mutation (Gittins et al. 2022; Fig. 1). Consequently, these thermospores have the potential to challenge the distance-decay relationship and exhibit unique biogeographical patterns.

This work quantifies endospores in sediment cores at and around hydrocarbon seeps using cores from deep-sea Scotian Slope sediments. Given the challenge of quantifying specific groups of endospores (e.g., thermophiles but not mesophiles), high-temperature germination assays that allow tracking of an exponential increase in signal as spores in marine sediment samples germinate and grow can be used. Distinct exponential increases can be attributed to different populations of germinated thermospores in the post-germination growth phase and are being monitored through measurements of sulfate

reduction rates (using radiolabelled  $^{35}\text{SO}_4$ ), strain-specific quantitative PCR (qPCR), and fluorescence in-situ hybridization (FISH) (Rezende et al. 2017). These measurements revealed growth dynamics enabling an estimation of the initial cell numbers using exponential functions. These approaches are being applied to samples obtained via push coring using a remotely operated vehicle at different distances from deep sea hydrocarbon seeps, to test for the presence of abundance gradients of different thermospores. Current results already show a difference in thermospore abundance between sites at different distances from the seep.

Quantitative tracking of the dispersal of thermospores by oceanic currents in the deep sea provides an excellent opportunity to investigate their distribution and potential to colonize more remote habitats, improving the understanding of microbial biodiversity and biogeography on our planet. By investigating thermospores and their interactions with the geophysical features of the deep sea and deep subsurface, this study aims to challenge the canonical idea that speciation occurs with distance and presents new perspectives on the theme of microbial biogeography. The evidence supporting the subsurface origin of thermophilic endospores, along with the proposed model for their dispersal and distribution, will contribute toward improving an integrated understanding of ecology and geology and their intersection in the microbial realm.

## **Keywords**

extremophile, sediment, biogeography, quantification, sulfate, sulfate-reducer

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Francesco Bisiach

## **Presented at**

ISEB-ISSM 2023 poster presentation. Part 1: Natural Settings - Sediment, subseafloor, subsurface

## **Conflicts of interest**

The authors have declared that no competing interests exist.

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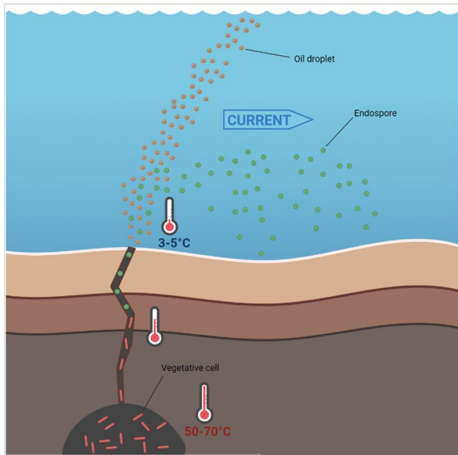


Figure 1.

The subsurface endospore dispersal theory. Thermophilic bacteria originating from deep subsurface petroleum reservoirs undergo an upward warm-to-cold translocation through the ocean crust via natural cracks in the impermeable layers of the rock. Thermospores, able to survive this transition are then expelled into the ocean along with petroleum fluids where they are further dispersed by the ocean current over long periods, before settling onto the ocean floor.