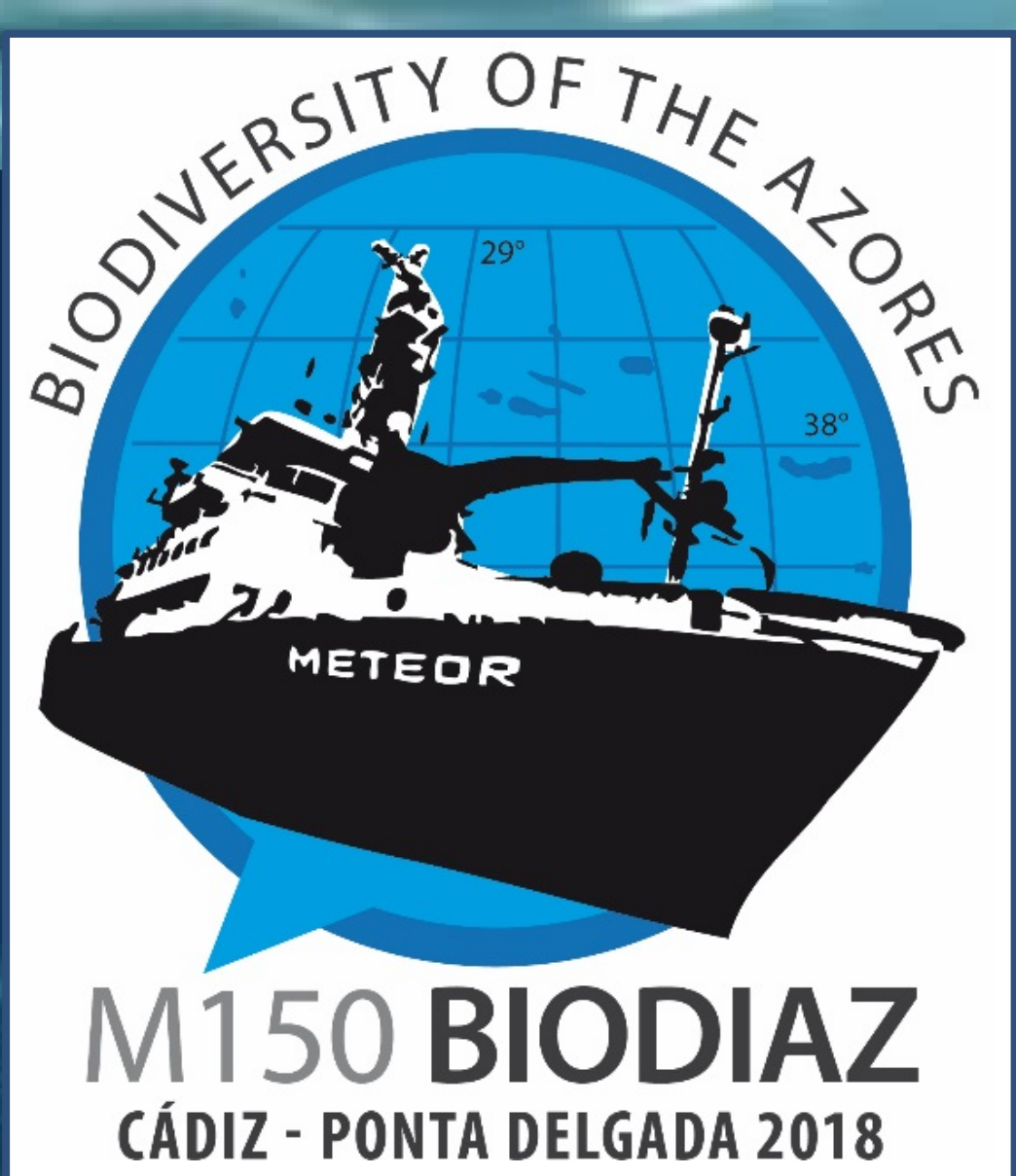


Vertical distribution of heterotrophic protists from Atlantic sublittoral to deep-sea sediments (M150 BIODIAZ)

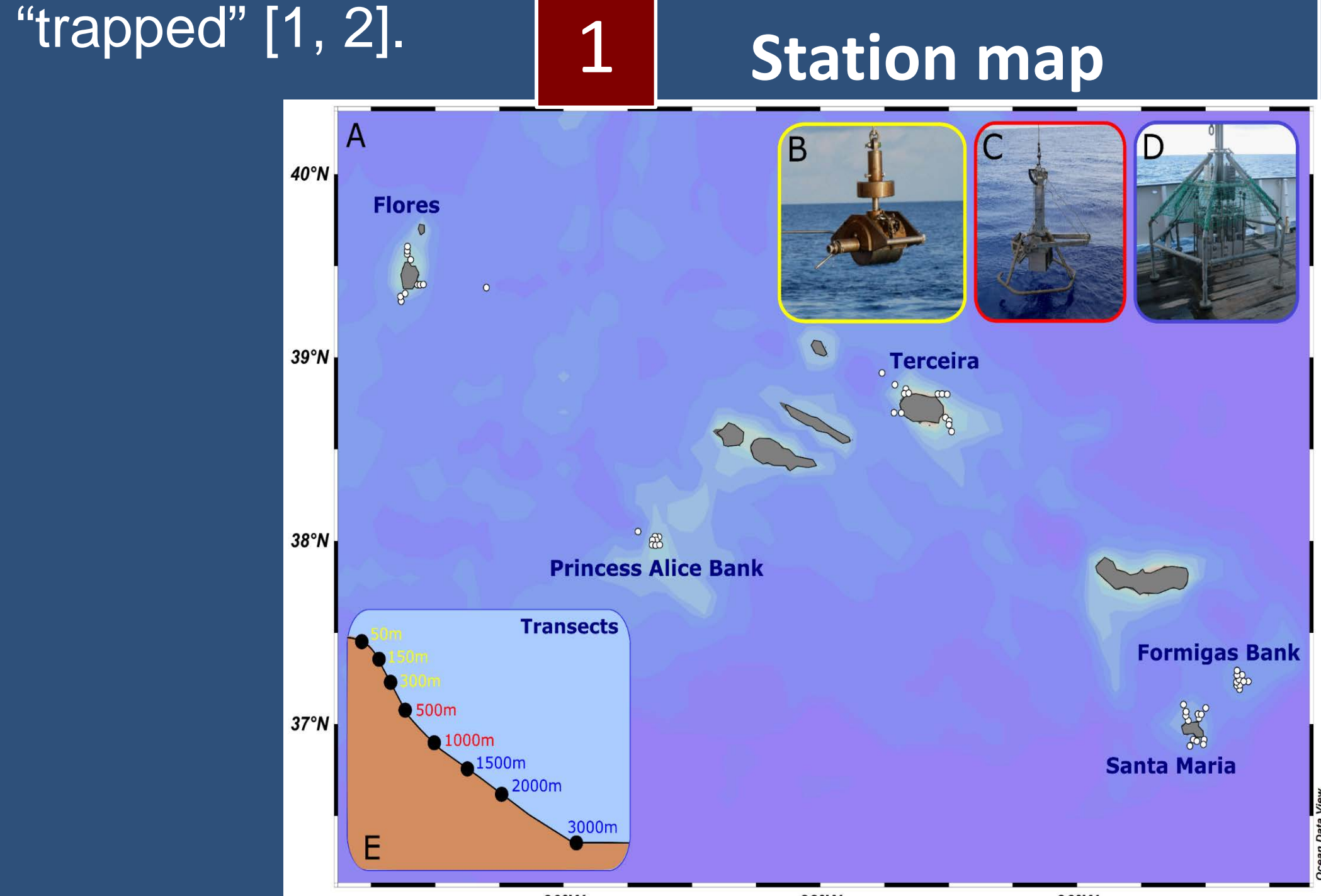


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Background

Very little is known about **vertical distribution patterns** of non-foraminiferan protists in benthic marine communities, even though they comprise key-groups to understand the role of islands/seamounts for the dispersal of species. The **Azores islands** with their isolated position in the middle of the North Atlantic are well suited to study whether they function as **stepping stones**, enabling the constant gene flow over large distances, or if they act as **trapping stones**, where the gene flow is disconnected and species become "trapped" [1, 2].

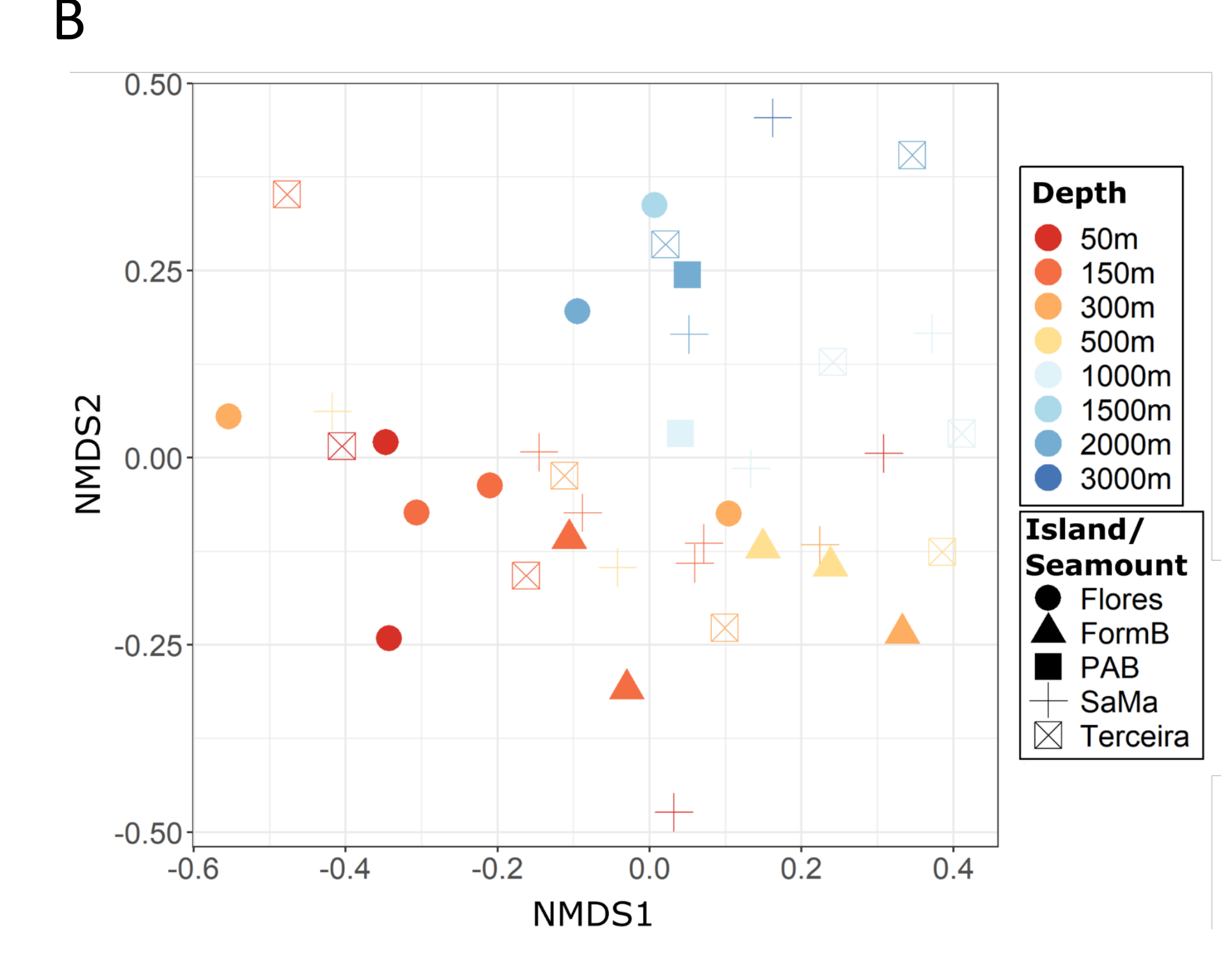
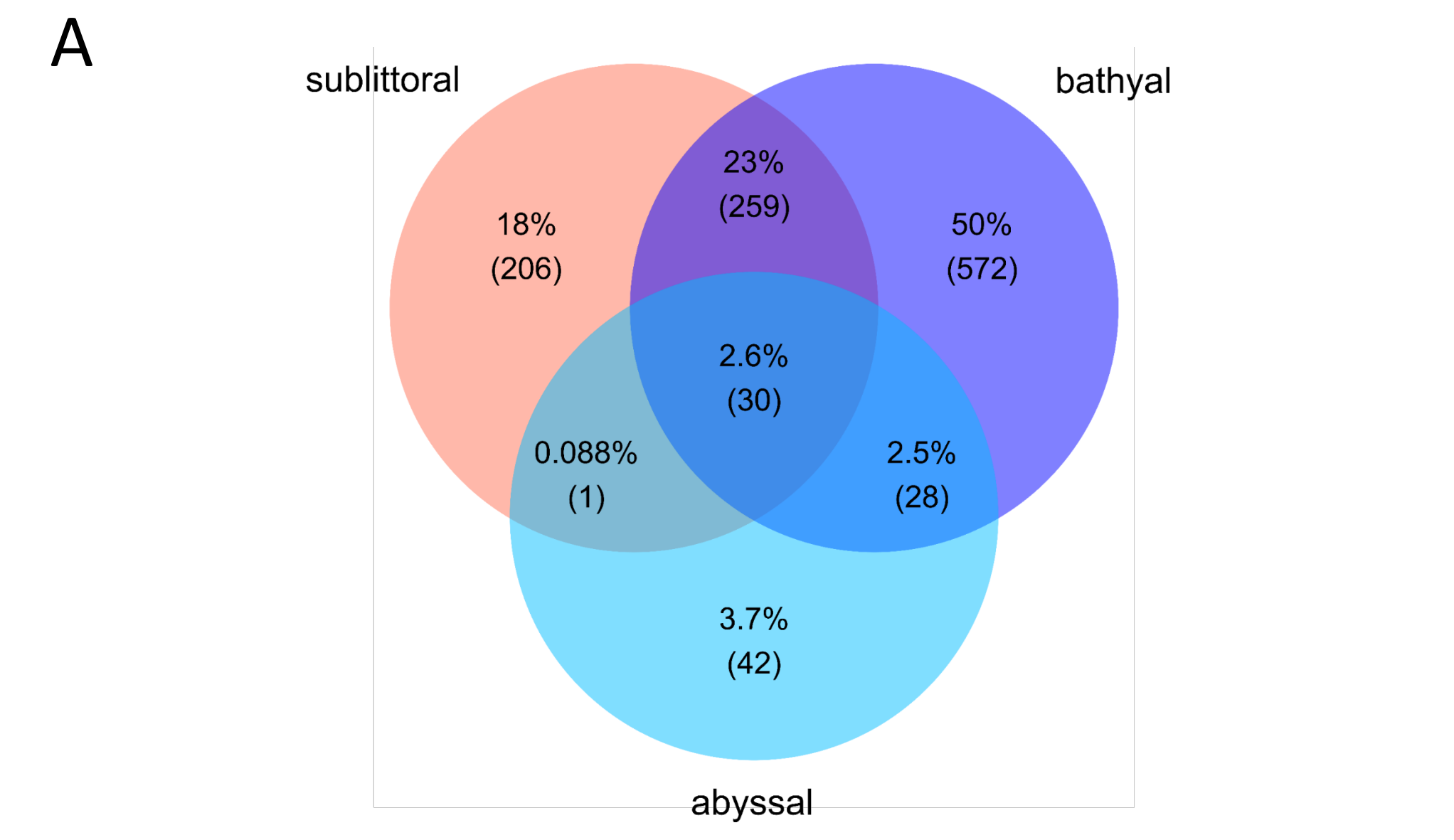


Methods

Sediment was sampled along vertical depth gradients from **50-3,000m** depth around **three islands** and **two seamounts** of the Azores archipelago (Fig. 1). We analysed for example:

- Metabarcoding of the V9-region on the SSU rDNA of total DNA aided by enlargement of databases (Fig. 2)
- Abundance and biovolume estimates of heterotrophic protists by live-counting (Fig. 3)
- Cultivation combined with molecular, morphological and ecological characterization of protist species (Fig. 4)

2 Metabarcoding Approach



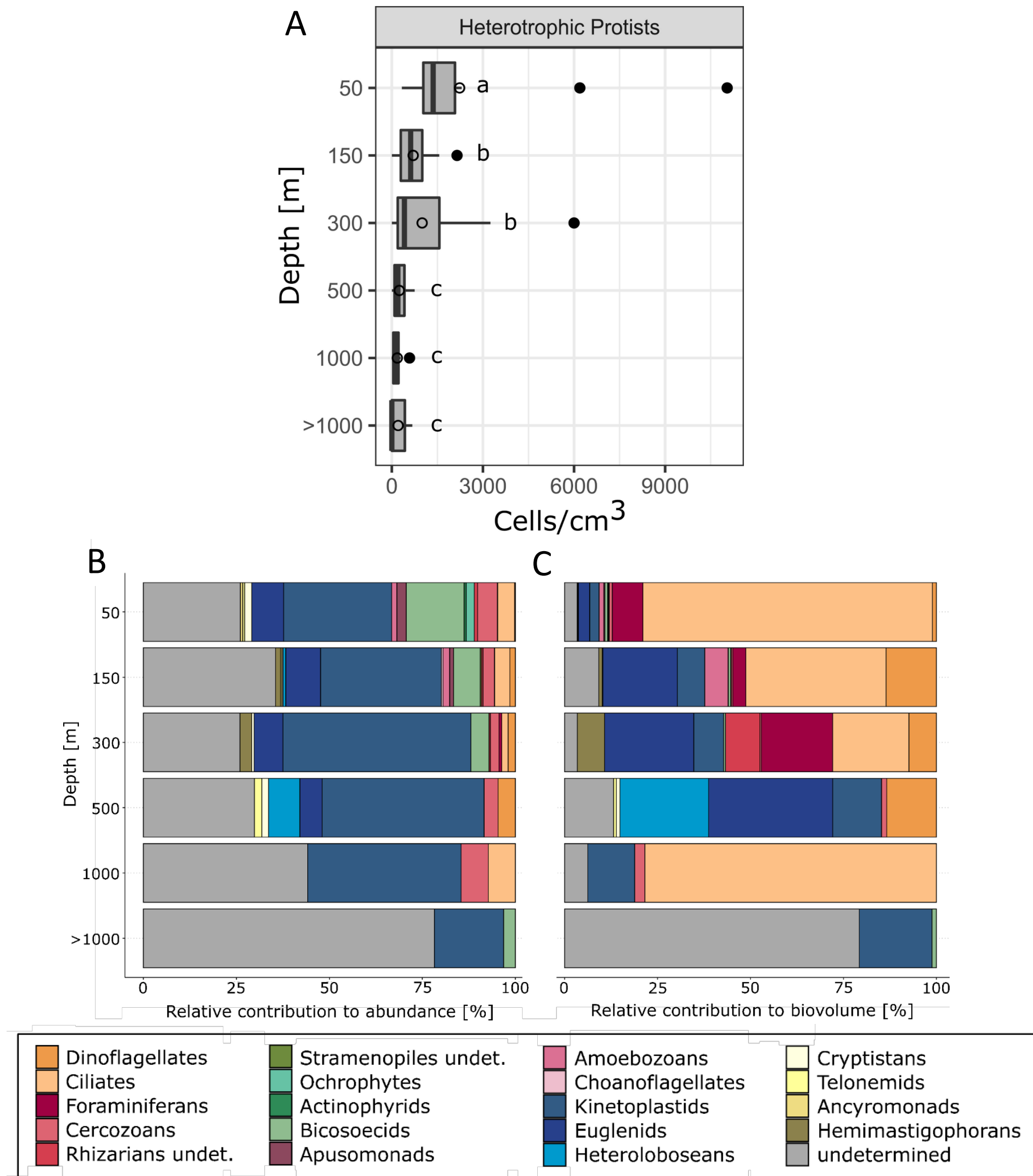
Metabarcoding

- Sequencing of samples from up to now 30 stations resulted in a total of about 50,000,000 **filtered reads (incl. mock community)** clustering into **1,138 OTUs**
- The **sublittoral** and **bathyal** shared only low amounts of OTUs with the **abyssal** (Fig. 2 A)
- Samples from **50-500m** and samples from **1,000-3,000m** clustered closely together in the NMDS analysis (Fig. 2 B)

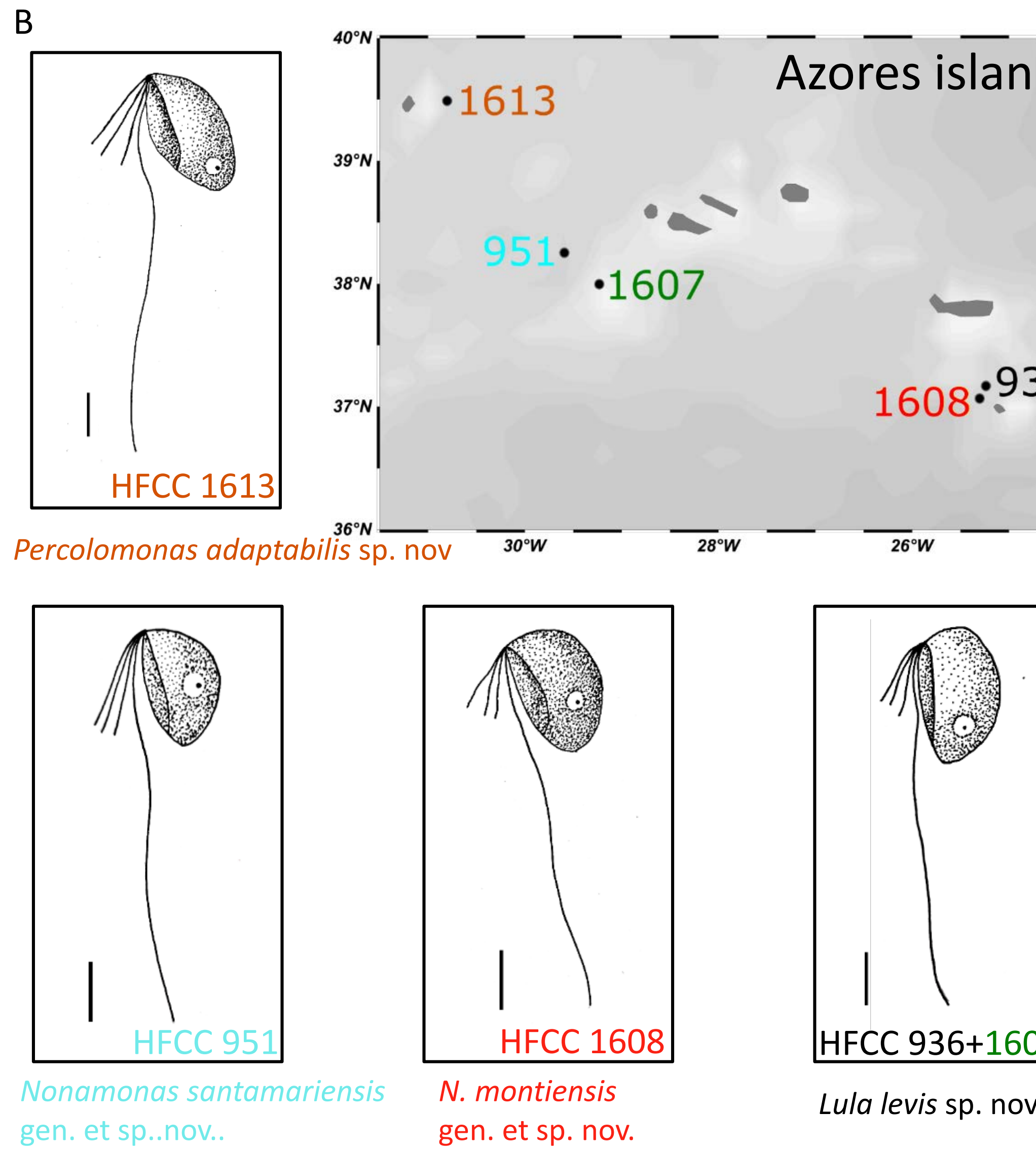
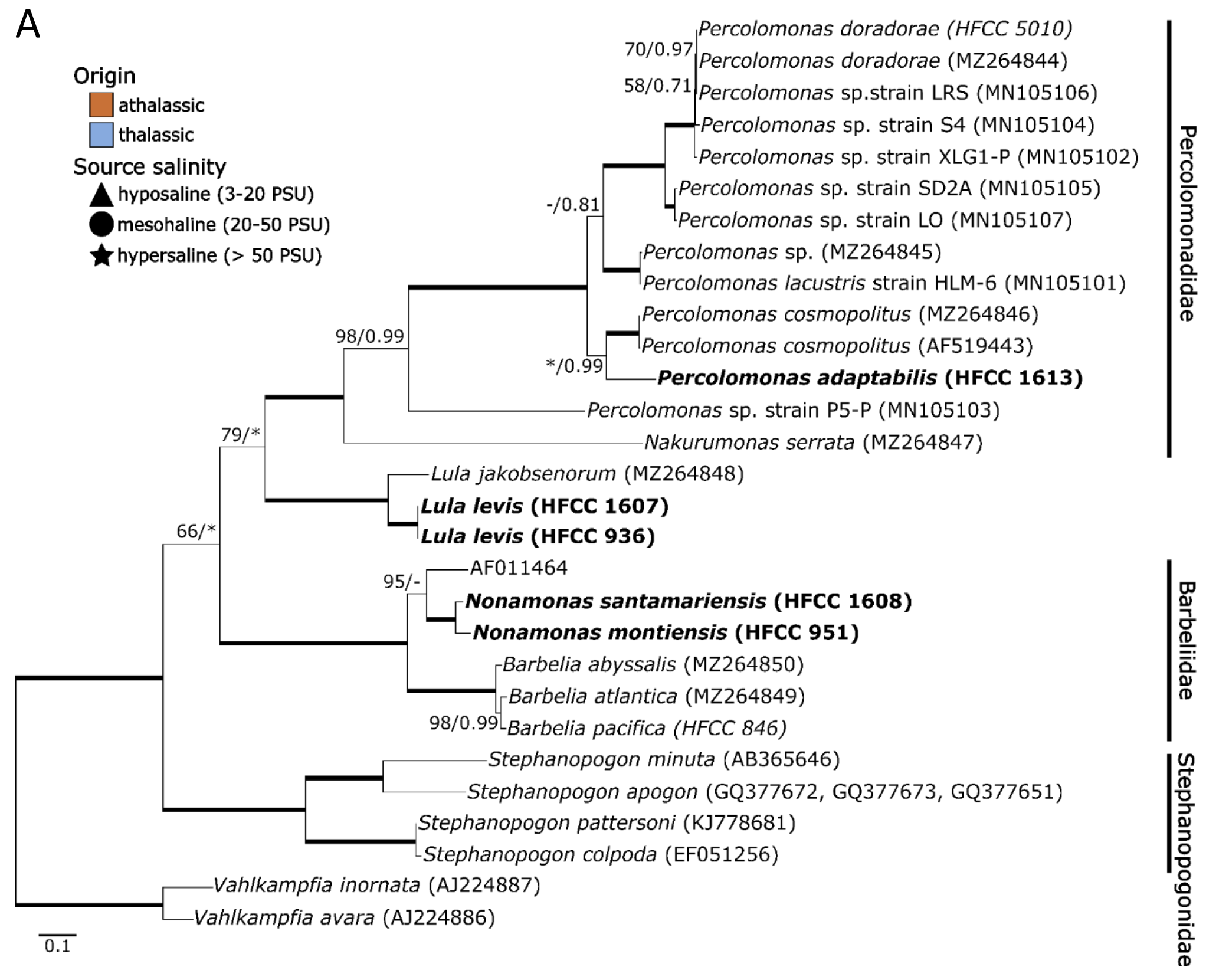
Abundance and Biovolume Estimates

- In total **202** different morphotypes were identified by live-counting
- **Abundance and biovolume** of heterotrophic protists was significantly **higher at 50m** than compared to all other depths (Fig. 3 A)
- At **50-1000m**, **kinetoplastids** were most abundant (Fig. 3 B)
- At **>1000m**, organisms which could **not be identified** contributed with up to 78% of abundance (Fig. 3 B)
- **Ciliates** made up the largest part of the **biovolume** in most depths (Fig. 3 C)

3 Community Structure of Benthic Protists



4 Morphology and Phylogeny of Selected Groups: Example Percolomonadida



Cultivation Approach

- Protist species of **various taxonomic groups** were isolated and cultivated (esp. Cafeteriaceae, Goniomonada, Percolomonadida)
- Example: **Percolomonadida** (Heterolobosea), several species seem to show a very restricted distribution based on our worldwide comparisons (Genbank data)(Fig. 4).
- Many detailed studied strains were added to **Genbank** and used to resolve NGS data sets (Fig. 2) and current **phylogeny** (Fig. 4 A)
- Most species were isolated only from single islands/seamounts - only one species was isolated from two sampling locations (Fig. 4 B)

Conclusion

Our integrated analyses comprising live observations, cultivation techniques, physiological and morphological studies as well as individual molecular and metabarcoding studies indicate that benthic protist **communities change significantly with increasing depth**. While some morphotypes and OTUs were found at all depths, others were only detected in sublittoral or lower bathyal depths, indicating that **several taxa are limited in their distribution by depth** whereas others should be able to **distribute over different habitats**. The high proportion of unidentified specimens and OTUs which increased with depth points towards a **high number of so far undetected species in the deep sea**.