

Fungi inhabiting the microbialites of the alkaline lake in Pantelleria island (Italy): biodiversity across a submerged-emerged transect

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The Bagno dell'Acqua alkaline lake in Pantelleria island, Italy, is **characterized by** CO, emissions, alkaline waters with a pH of around 9, and strongly oxidizing conditions. The lake is home to actively growing microbialites rich in calcium carbonates and silica precipitates. The lake's sediments are dominated by Mgsmectite clays associated with K-feldspars and Ca-carbonates, and the presence of volcanic glass and minor phases like pyroxene and hematite has been observed.



The island is located in the Strait of Sicily, Mediterranian Sea, between Sicily and Tunisia. The National Park of Pantelleria was established on July 28, 2016, and is the first one in Sicily. It has the UNESCO World Heritage status and is part of the Natura2000 network, recognised as a Special Protected Area and a Special Area of Conservation under the European Habitats Directive.

The microbialites from the lake were studied by Ingrassia et al (2023) using X-ray diffraction (XRD), scanning electron microscopy (SEM) and energy dispersive Xray spectroscopy (EDS). A close association has been demonstrated between Mg-smectite/carbonate minerals, which are the primary components of the stratified deposits at the lake margins, and the presence of **exopolymer**producing microorganisms (EPS), capable of precipitating carbonates and clay minerals (Mazzoni et al., 2024).

These singular conditions could impact the fungal diversity in the sediments and microbialites, offering insights into the adaptation of fungi to high pH values. A transect from the lake's margins to the line of vegetation was used as a sampling scheme to evaluate the impact of alkaline water on the fungal communities in the first 5 cm of sediment depth (C0 only emerged, C1-2 water-front, C3-4 submerged), a further submerged sample (C11) was sampled on the lakeshore opposite the transect.



High-throughput sequencing of the **fungal internal transcribed** spacer 2 (ITS2) molecular marker region, performed with the Illumina NovaSeq PE250, was applied to compare the alpha and beta biodiversity of the microbialites' samples.

Fungi were also isolated using **classic culturing** methods, with the lake sediment serving as the culture medium. Identification was based on Sanger sequencing of molecular markers (ITS1-4) and morphology.

The strains isolated from the submerged sediments are different from those obtained from the microbialites that were exposed to air at the time of sampling, suggesting an overlap of two distinct fungal communities: those that colonise the surface material (airborne species) and those that seem to inhabit the submerged plant material. The latter appeared poorly pigmented compared to the species isolated from the vegetation line.

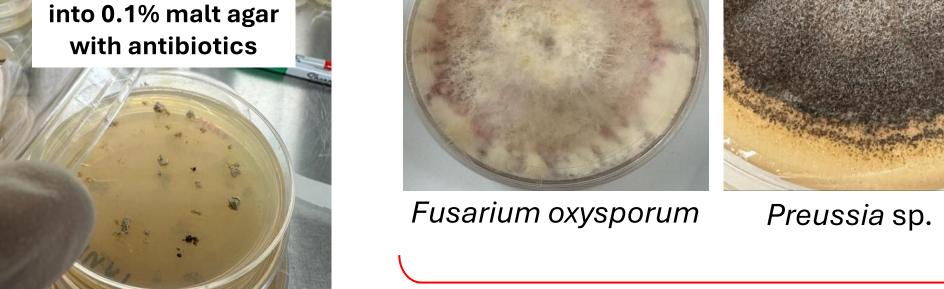
Microbialites

Inoculation of

sediment crumbles

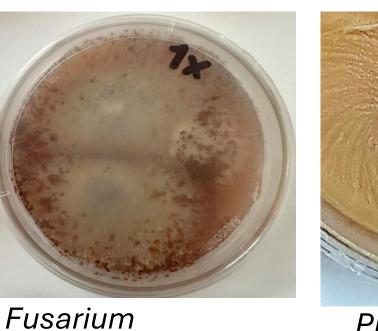
European Union, Copernicus Sentinel-2 imagery; ttps://www.copernicus.eu/lt/node/28487

Sediment core



Chao1

100 -



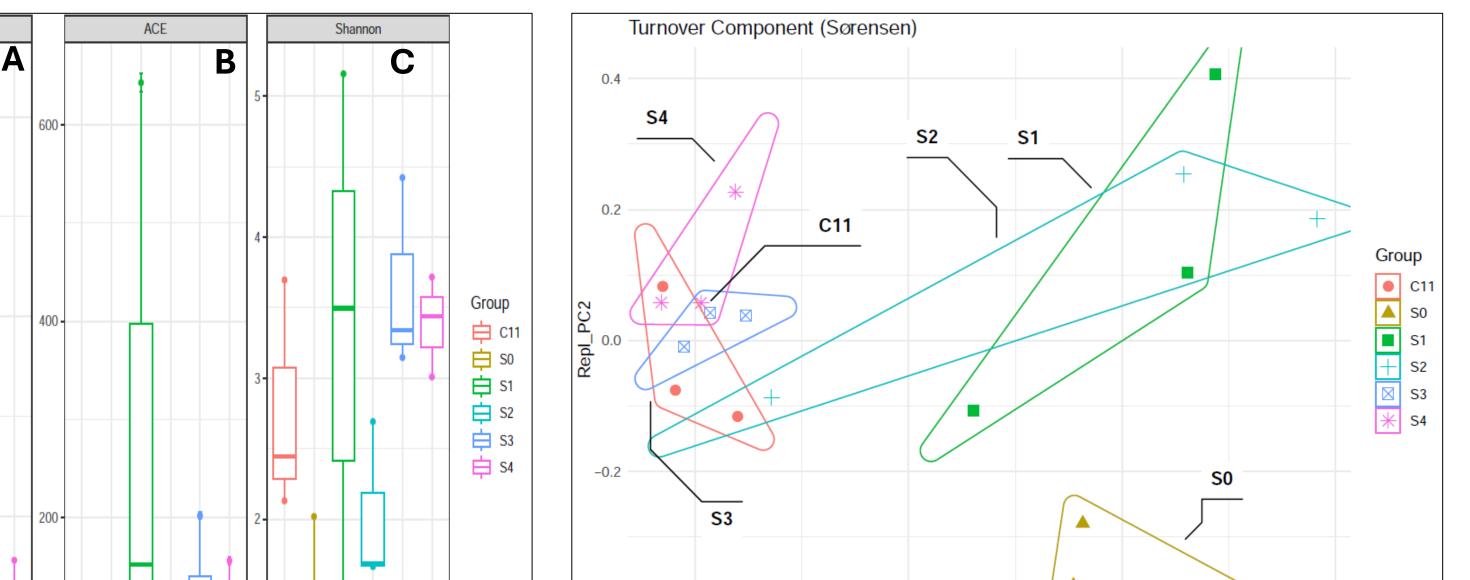




Plectosphaerella neocosmosporiellum pauciseptata

Cladosporium herbarum

Some of the most recurring fungal isolates identified with morphology and ITS1-4 barcoding



Transect from vegetation to shallow water Sterile water + sediment inoculum **Separation of** different isolates on agar with selective pH (from 5 up to 9) Incubation + monitoring at 25°C

Entirely submerged sediment fungi (4) **Fungi from entirely** include species that emerged sediment (0) are not pigmented and colonized by plants include several pigmented species





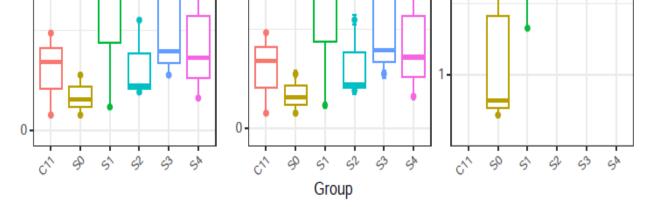
Gradient with respect to emersion from water

References

- Ingrassia M. et al. (2024) *Minerals*. 14(10):1013
- Mazzoni C. et al. (2024) Front Microbiol. May 22;15:1391968.

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Sample S1 (waterfront) exhibited the highest alpha diversity, but also showed variability between replicates. In the boxplot the values obtained for **Abundance-based coverage estimator Chao1** diversity (A), (ACE) index (B), Shannon diversity (C).

-0.4 0.25 Repl_PC1

Submerged samples (S3, S4, C11, partially S2) showed higher overlap. The emerged sediment (S0) has its own fungal community. This is a measure of community dissimilarity (βsim) based on presence/absence data, calculated using the Sørensen index. The turnover component quantifies the degree to which species are replaced between locations. A high βsim value indicates a large amount of species turnover, suggesting that different locations have very different species compositions.

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at all

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