Lectins:

Characterization of Extracellular Polymeric Substances in Hypersaline Cyanobacterial Mats and Mat-forming Cyanobacterial Isolates

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Introduction

Photosynthetic microbial mats are highly structured laminated microbial communities that can host all the major biogeochemical cycles in a few millimeters of depth and their activity likely dominated biogeochemical cycling throughout Earth’s history. More recently they have generated interest because of their ability to produce hydrogen gas. While most microbial life exists as biofilms, a few extracellular polymeric substances (EPS) are a major component, our understanding of nutrient exchange and energy flow through the EPS in these mats is not well understood.

In this work we designed methods to extract EPS from both natural mats from Elkhorn Slough in Monterey Bay, CA (Fig. 1) and a diazotrophic cyanobacterium isolated from these mats (Fig. 2) and profiled proteins present in the isolate EPS.

Results: Microscopy and Enzyme activity

1. Total homogenized cells (D0)
2. EPS extracted pellet (4°C)
3. OMP extracted pellet (4°C)

Alcaloid blue staining after each step of extraction:
- We remove some of the EPS, but not all
- Low percentage of lysed cells (also verified by GFP2H enzyme activity not detectable in EPS and OMP fractions)

Lectin-based probe staining in natural mat community and cultures. Green represents lectin binding, red represents autofluorescence and blue is DAPI DNA stain. Natural mat stains were done on homogenized fixed samples and were gravity stained. Culture stains were done directly in 6-well plates with 6 day old cultures.

- ESFC-1 lacks an outer sheath that many other filamentous cyanobacteria have, seen in the natural mat ConA stain.
- EPS fraction has a diverse set of enriched putative functions such as protein degradation and adhesion

Conclusions

- Fractionation of cultures and natural mats results in distinct fractions with different proteins in each fraction
- Lectin based stains provide visualization of EPS to compare natural mats to cultures
- Extracellular enzyme activity is more diverse in the natural mats
- Proteomics reveal many secreted proteins of unknown function in the EPS and OMP fractions indicating extracellular protein degradation

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