The microbial communities associated with the leaves and roots of turtle grass and manatee grass differ from seawater and sediment communities, but are similar

Kelly Ugarelli¹, Peeter Laas¹, Ulrich Stingl¹ Contact: kugarelli@ufl.edu, ustingl@ufl.edu; Phone: 954-577-6326

between species

1- Fort Lauderdale Research and Education Center, Institute of Food and Agricultural Sciences, University of Florida, Davie, FL, USA



Introduction

Seagrasses are vital members of coastal ecosystems providing services such as shelter, food and nursery to many species including economically important fish. Recent seagrass die-offs have occurred in Florida due to microbes that reduce sulfate into phyto-toxic sulfide, causing a cloud of sulfur as seen in the image below. Other microbes, such as algae, prevent sunlight from reaching the seagrass leaves. Investigating the microbial communities of seagrasses

can provide us with information about the key members of the seagrass core- microbiome. This information might allow us to explore the possibility of targeting specific microbes to help seagrasses endure environmental stress, and subsequently help the conservation of the ocean and fauna that inhabit the seagrass beds.



Results

Clustering of samples based on microbial composition

1.0

Comparison of the number of sequence variants per species and site

Phyllosphere



Rhizosphere

Cloud of sulfur surrounding dying and dead seagrass (FFWCC. Miami Herald).

Goals

- consistencies and/or differences in the identify То microbiome of two seagrass species in the same meadow
- To elucidate the possible functions of the abundant microbes for targeting in future studies.

Methods

Three replicates of turtle and manatee grass were collected from Hobie Island Beach Park in Key Biscayne, FL. Collection sites were between 0.1 and 1 km apart. We collected seawater and sediment from all three locations using sterile carboys and 50 ml Falcon tubes, respectively.



Fig 1. Cluster analysis of the microbial communities of the samples show that the roots, leaves, sediment phyllosphere communities resemble each and water microbes are distinct from one another. This a possible difference in symbiotic rhizosphere communities resemble each indicates relationships between the microbes and the seagrass at each part of the plant.

Relative abundance of the most common taxa in each sample





Fig 3. Comparison of sequence variants that make up > 0.01% of the total both sequences. species, In the other and not the water, while the other and the sediment. Several sequences are unique to each species.

Significantly distinct genes based on predictive metabolism





- 5. PCR amplification of the 16S rRNA V4 region followed by amplicon sequencing on the Illumina MiSeq platform
- 6. QIIME 2, SILVA taxonomy, R, and the MicrobiomeAnalyst Shotgun Data Profiling (SDP) module were used for bioinformatics

0.0-			
	K02030 - K02030 - K01958 - K00100 - K001286 - K01286 - K01286 - K01620 - K01620 - K00845 - K00845 - K00845 - K008309 - K07469 - K	K06872- K01043-	K05832 - K01989 - K05833 - K05833 - K05833 - K05833 - K05833 - K03465 - K03465 - K03465 - K03465 - K03968 - K039788 - K033300 - K033300 - K12254 - K122554 - K122554 - K122554 - K122554 - K122554 - K12254 - K122554 - K12254 - K12554 - K125564 - K125564 - K125564 - K125564 - K125564 - K125554 -
	Fe	atures	
KEGG ID	Function	KEGG ID	Function
K02029	polar amino acid transport system permease protein	K01043	undetermined
K02030	polar amino acid transport system substrate-binding protein	K05832	putative ABC transport system permease protein
K01958	pyruvate carboxylase	K01989	putative ABC transport system substrate-binding protein
K00100	butanol dehydrogenase	K05833	putative ABC transport system ATP-binding protein
K01286	D-alanyl-D-alanine carboxypeptidase	K03293	amino acid transporter, AAT family
K13954	alcohol dehydrogenase	K03465	thymidylate synthase
K01468	hutl, AMDHD1; imidazolonepropionase	K04762	ribosome-associated heat shock protein Hsp15
K01620	threonine aldolase	K00996	undecaprenyl-phosphate galactose phosphotransferase
K00845	glk; glucokinase	K08988	putative membrane protein
K08309	soluble lytic murein transglycosylase	K09788	2-methylaconitate isomerase
K08223	MFS transporter, FSR family, fosmidomycin resistance protein	K03300	citrate-Mg2+:H+ or citrate-Ca2+:H+ symporter, CitMHS family
K07469	aldehyde oxidoreductase	K12254	NAD-dependent aldehyde dehydrogenase
K06872	uncharacterized protein		

Fig 2. Some of the abundant taxa (> 1% of the sequences) are involved in biogeochemical cycles of Fig 4. The most abundant significantly sulfur (S oxidation: purple boxes; sulfate reduction: distinct genes per sample type are green boxes). Some taxa are more abundant on predominantly involved in basic metabolic different segments of the plants and others are more abundant on one species than the other.

functions. Seawater is more distinct than the other sample types.

References

- 1. Guide to Unique Wildlife of the Florida Keys. (2017) Marine Life Around the World. Key West Aquarium. (Background image).
- 2. Ugarelli, K.; Chakrabarti, S.; Laas, P.; Stingl, U. (2017)

Conclusions

The microbial community of the roots, leaves,

Future Directions

Metagenomic sequencing for analysis of the metabolic functions of the microbiome with

The Seagrass Holobiont and Its Microbiome. *Microorganisms*, 5, 81.

3. The dying seagrass feed a cycle of elevated sulfide in bay water. FFWCC (2016). Will Florida Bay survive the summer? Miami Herald.

4. 3 QIIME2 homepage: https://qiime2.org 5. 4 Quast C, Pruesse E, Yilmaz P, Gerken J, Schweer T, Yarza P, Peplies J, Glöckner FO (2013) The SILVA ribosomal RNA gene database project: improved data processing and web-based tools. Nucleic Acids Research 41, 590-596.

6. R Core Team (2013). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria.

sediment and water are distinct from one another

- Microbes involved in sulfur oxidation and nitrogen
- fixation were identified in the sediment and plant samples
- Some sequences are unique to each species while many overlap, indicating a possible core microbiome
- No significant differences in predictive metabolism

more replicates Nitrogen-fixing the analysis gene ot microbiome Effects of environmental factors on the microbiome Cultivation of the key members of the microbiome