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Application of genomics to develop a monitoring tool for stormwater treatment wetlands

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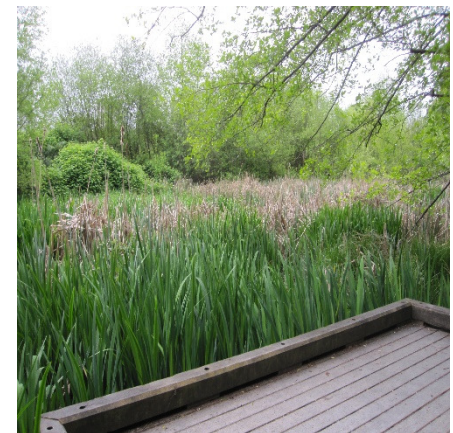
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Applying Genomics to Monitor the Performance of Urban Stormwater Treatment Wetlands



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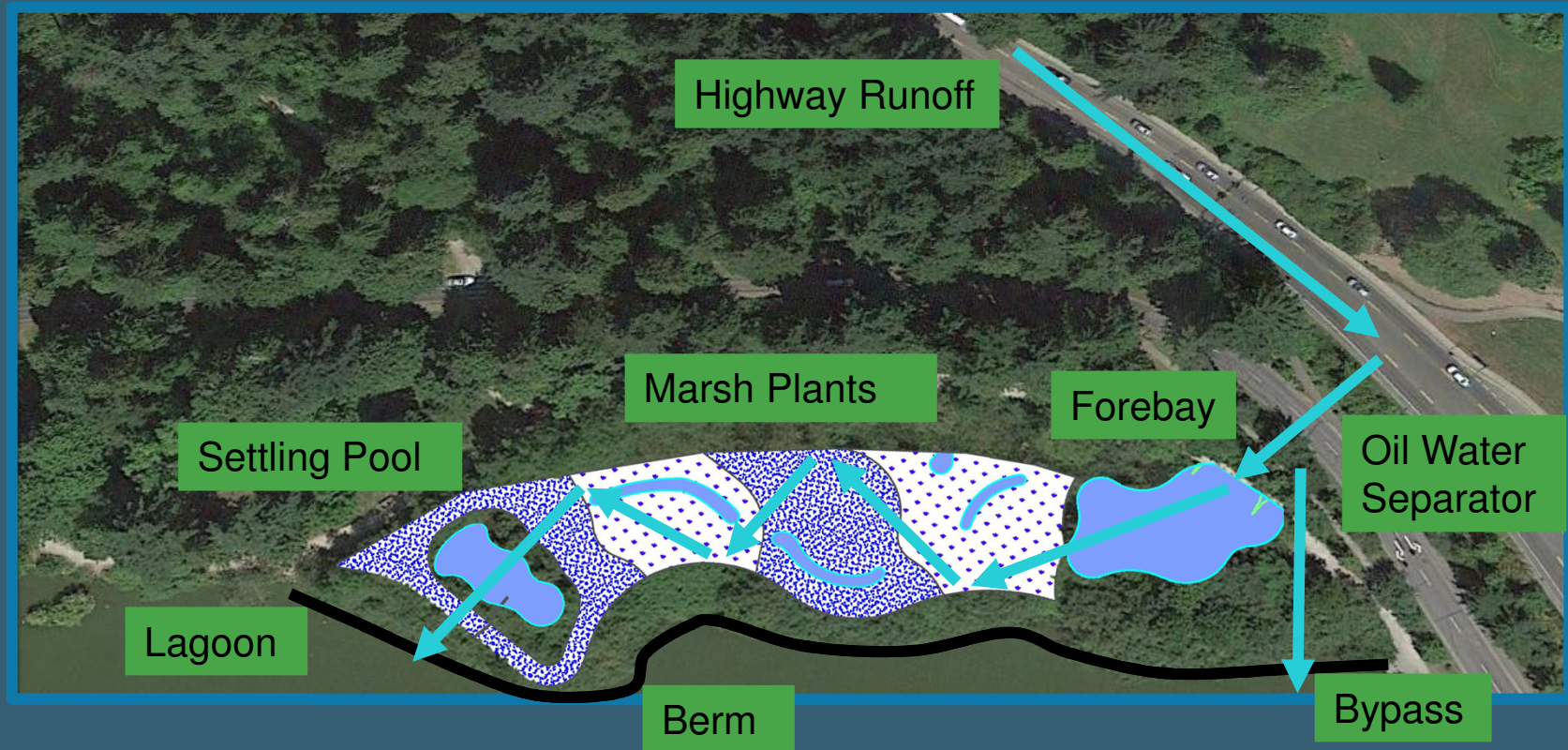


May 16, 2018

+ Wetlands for stormwater treatment

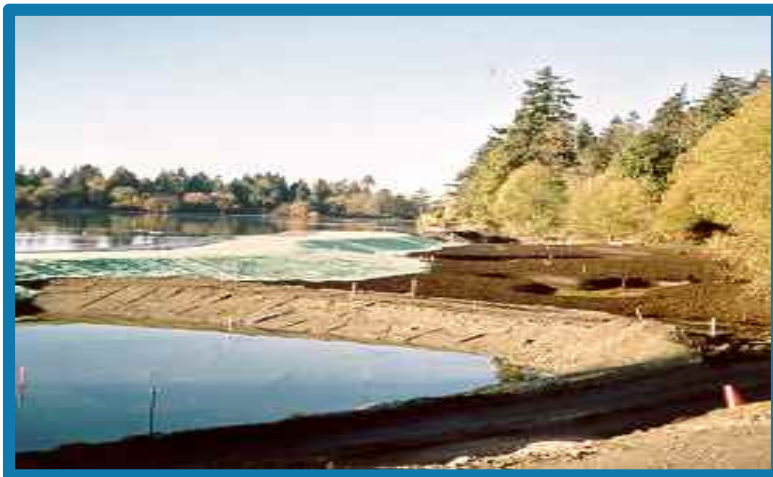


+ How do wetlands treat stormwater and what makes a `good` wetland ?



+ Background

Motivation - Lost Lagoon Wetland



+ Background Monitoring Challenges



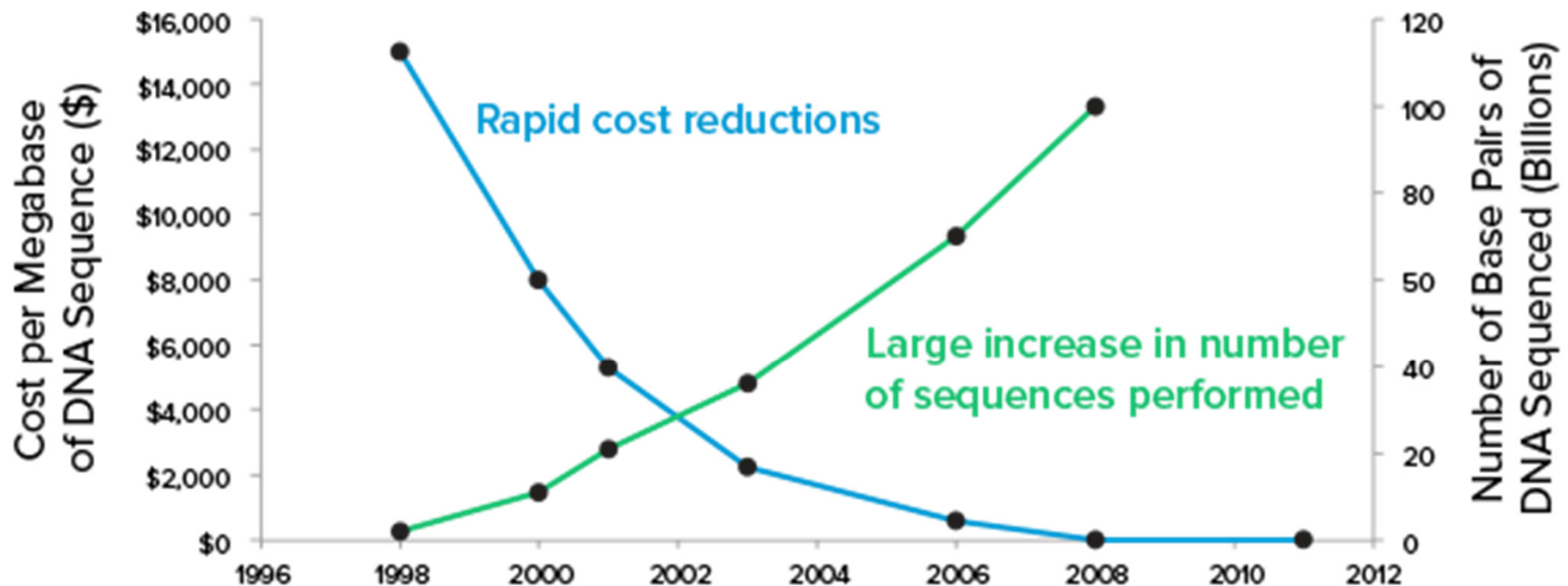
- Wetlands are challenging to monitor
 - Two-year sampling plan.
(Erickson et al 2013)
- Municipalities unlikely to prioritize extensive monitoring



+ Background

Genomics and DNA

“The branch of molecular biology that is concerned with the structure, function, evolution, and mapping of genomes, or the complete set of DNA within a single cell of an organism.” (Oxford University Press 2016)

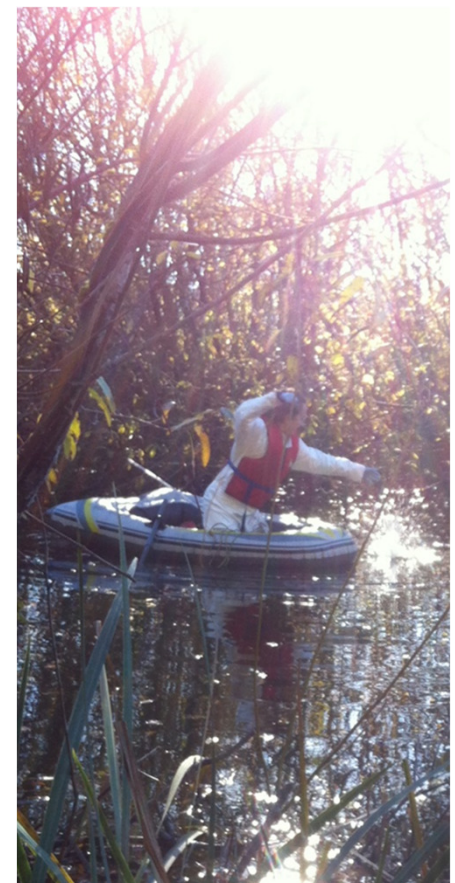
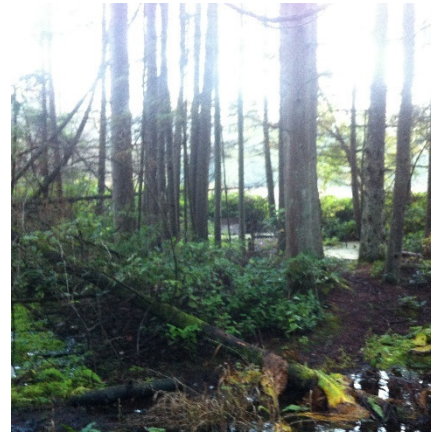


Data from the National Human Genome Institute, Data from the National Center for Biotech Information

+ Project Overview

Purpose

Begin to develop DNA-based monitoring tool for engineered stormwater treatment wetlands



+ Project Overview

Objective 1 – Opportunities

- Demonstrate that the wetland is meeting water quality treatment guidelines
- Identify knowledge gaps and opportunities for complimentary data analyses using genomics



+ Project Overview

Objective 2 – Design



- Shifts in the bacteria and genes
- Correlations between the contaminant levels and the bacteria and genes observed
- Opportunities to expand and pursue DNA-based analyses at other sites

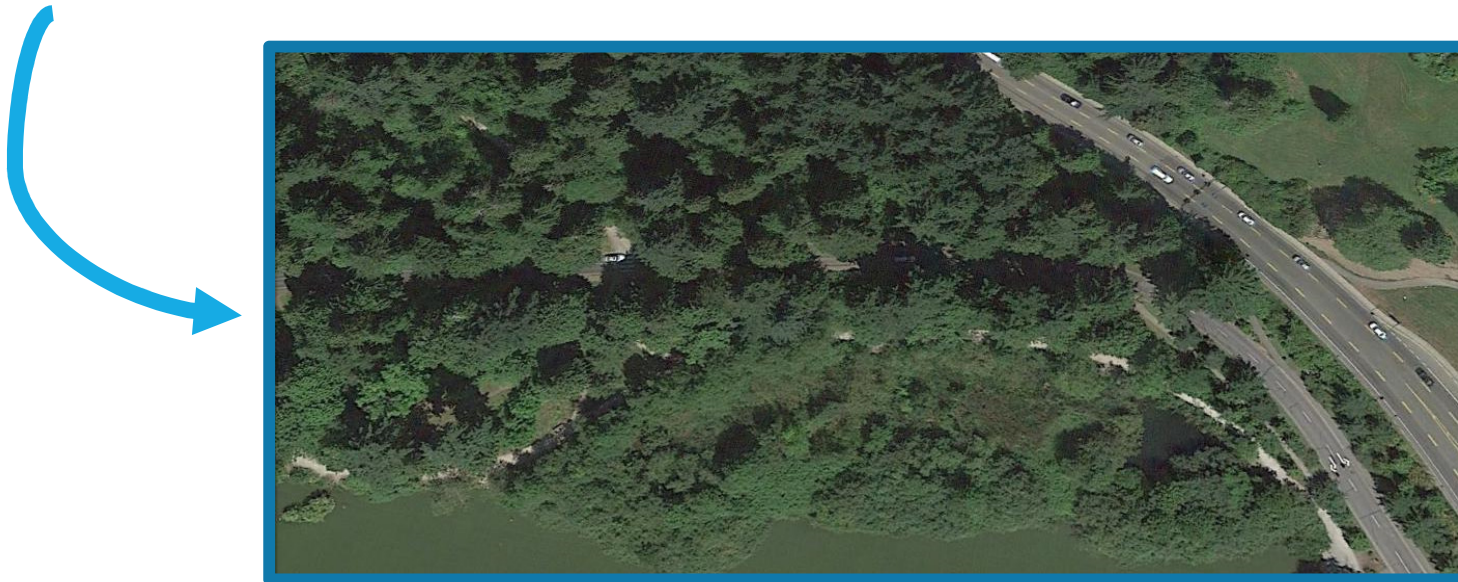


+ Research Activities

Field Study - Overview

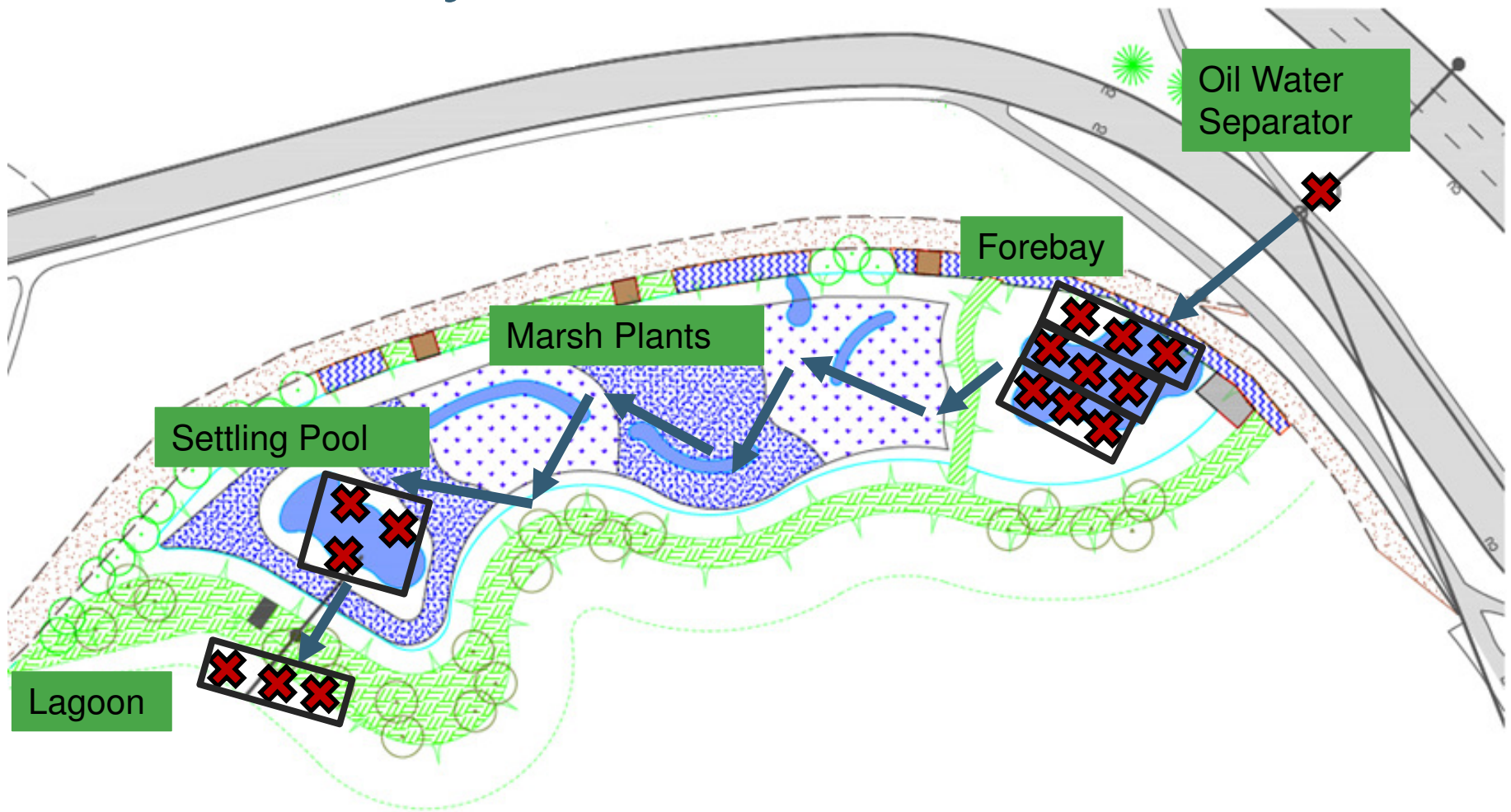


- “Textbook example” stormwater treatment wetland



- 8 sampling days between July and December 2015
- 90 core sediment samples, 12 grab sediment samples, and 105 water samples

+ Research Activities Field Study - Site



+ Research Activities

Laboratory Study - Overview



- Apply stormwater to an uncontaminated soil and monitor the response of the microbial community over time
 - 4 gal buckets with 10 kg soil
 - 11 columns fed stormwater and 6 columns fed distilled water
 - 3 columns analyzed each month

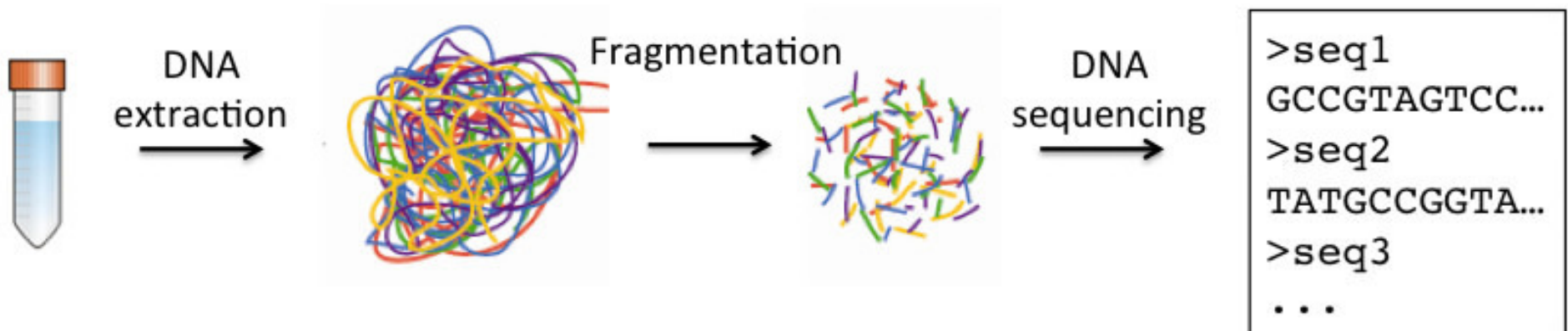


+ Research Activities

Analytical Methods - Sequencing



- To compare the composition of bacteria species in each sample:
 - Sequence a specific 'signature gene' fragment of the DNA known as the 16S rRNA gene, which is unique to each species
- To compare the composition of functional genes within the bacteria in each sample
 - Sequence all of the DNA and prepare what are known as metagenomes



+ Research Activities

Analytical Methods – Data Analysis



```
>seq1
GCCGTAGTCC...
>seq2
TATGCCGGTA...
>seq3
...

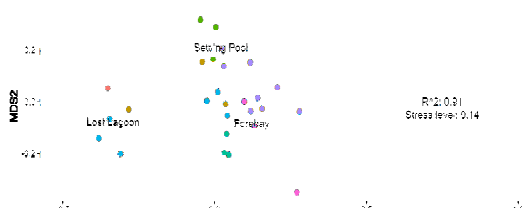
```

Assemble, bin and compare to a reference library

	Sample No.							
	1	2	3	4	5	6	7
<i>Acidobacteria</i>	15	4947	15	6806	0	0	15	
<i>Actinobacteria</i>	10	5674	10	7141	1	1	20	
AncK6	1	802	1	896	0	0	2	
<i>Bacteroidetes</i>	16	1487	15	1305	3	678	21	
<i>Chlamydiae</i>	0	0	0	0	0	0	0	
<i>Chloroflexi</i>	35	1763	34	1902	1	1	18	
<i>Cyanobacteria</i>	0	0	0	0	0	0	3	
<i>Firmicutes</i>	7	120	1	1	1	1	5	
<i>Gemmatimonadetes</i>	8	879	7	503	0	0	7	
<i>Lentisphaerae</i>	0	0	0	0	0	0	0	
<i>Nitrospirae</i>	2	163	1	272	0	0	2	
OD1	0	0	0	0	0	0	0	
PAUC34f	4	3759	4	2634	0	0	4	
<i>Planctomycetes</i>	5	13	4	15	0	0	8	
<i>Poribacteria</i>	6	3726	5	2137	0	0	5	

Linear algebra and coding software for analysis

Example – convert to two dimensions using similarity between samples and prepare a plot to visualize the samples



+ Environmental Analysis

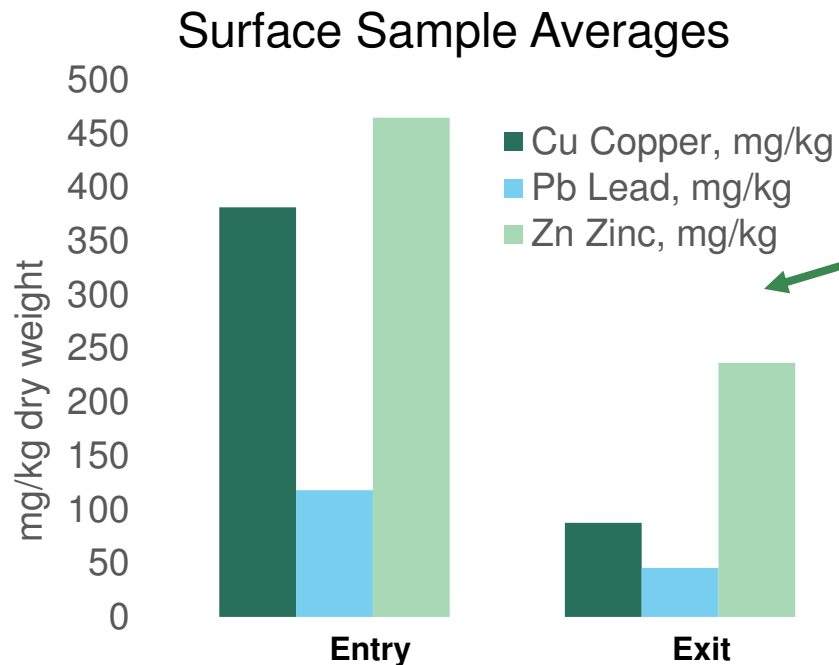
Hypothesis 1



1. The wetland removes heavy metals



+ Environmental Analysis Results



- Stormwater metals show high concentrations at the wetland entry
- Cu, Pb, and Zn had the most significant reductions (p.val <0.05 using Wilcoxon Rank test)
- High variability between sampling dates even at the same location

Hypothesis 1

The concentrations of metals associated with stormwater decrease along the length of the wetland.

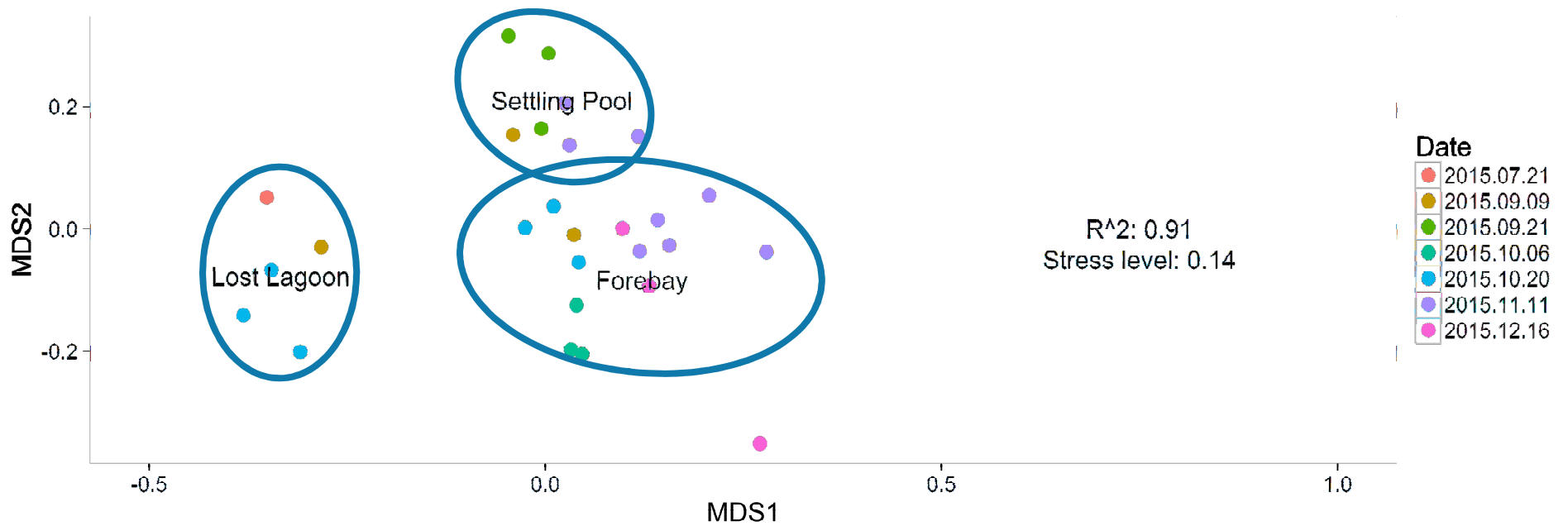
+ Microbial Analysis Hypothesis 2



2. The bacteria at the entry of the wetland differ from the bacteria at the exit of the wetland



+ Microbial Analysis Results – Surface Sediment

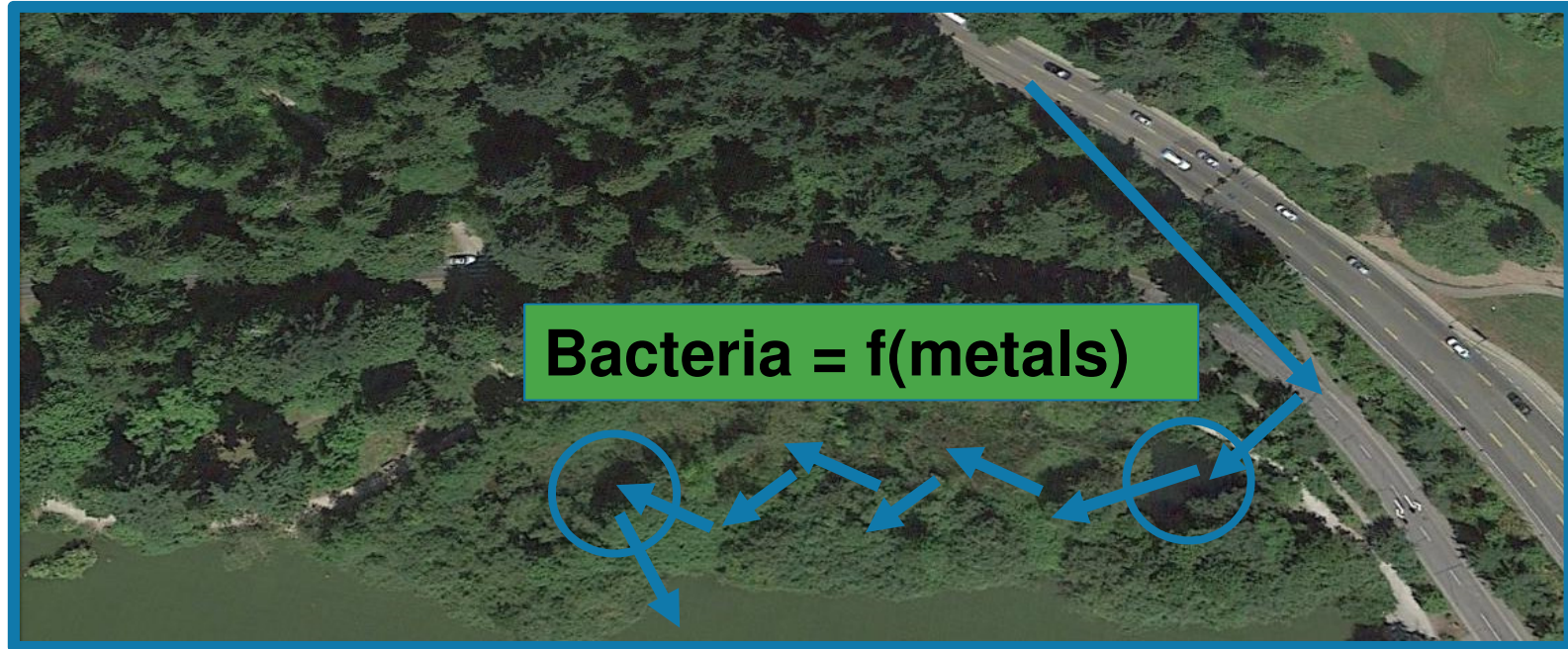


+ Microbial Analysis

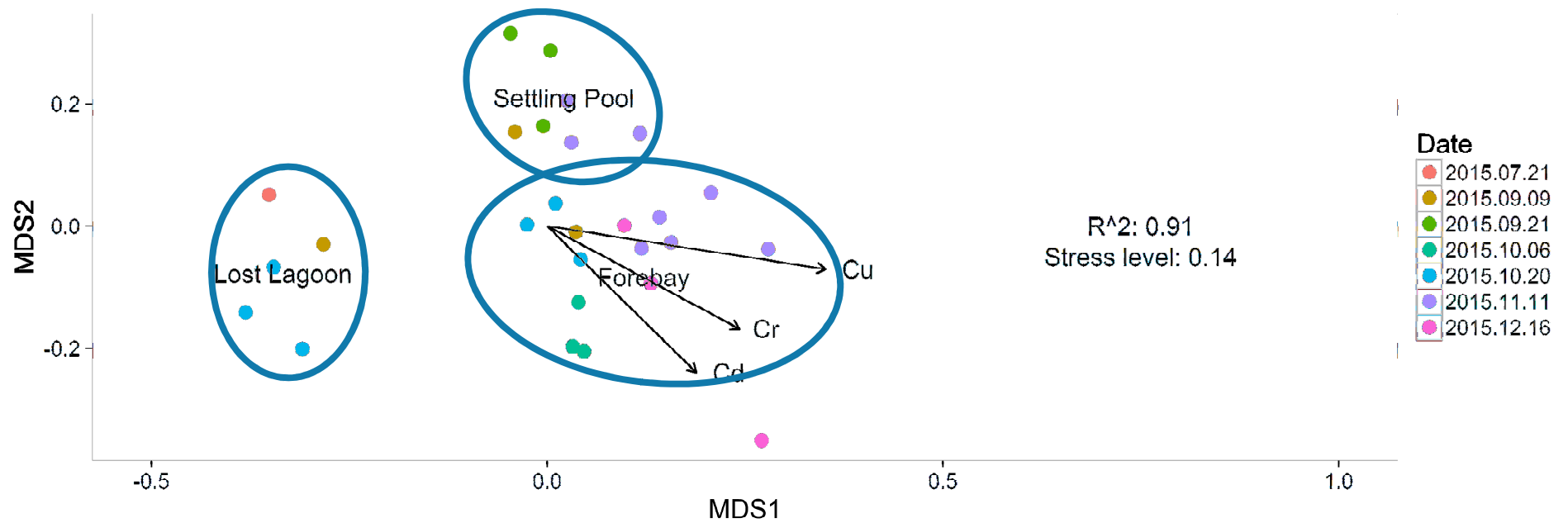
Hypothesis 3



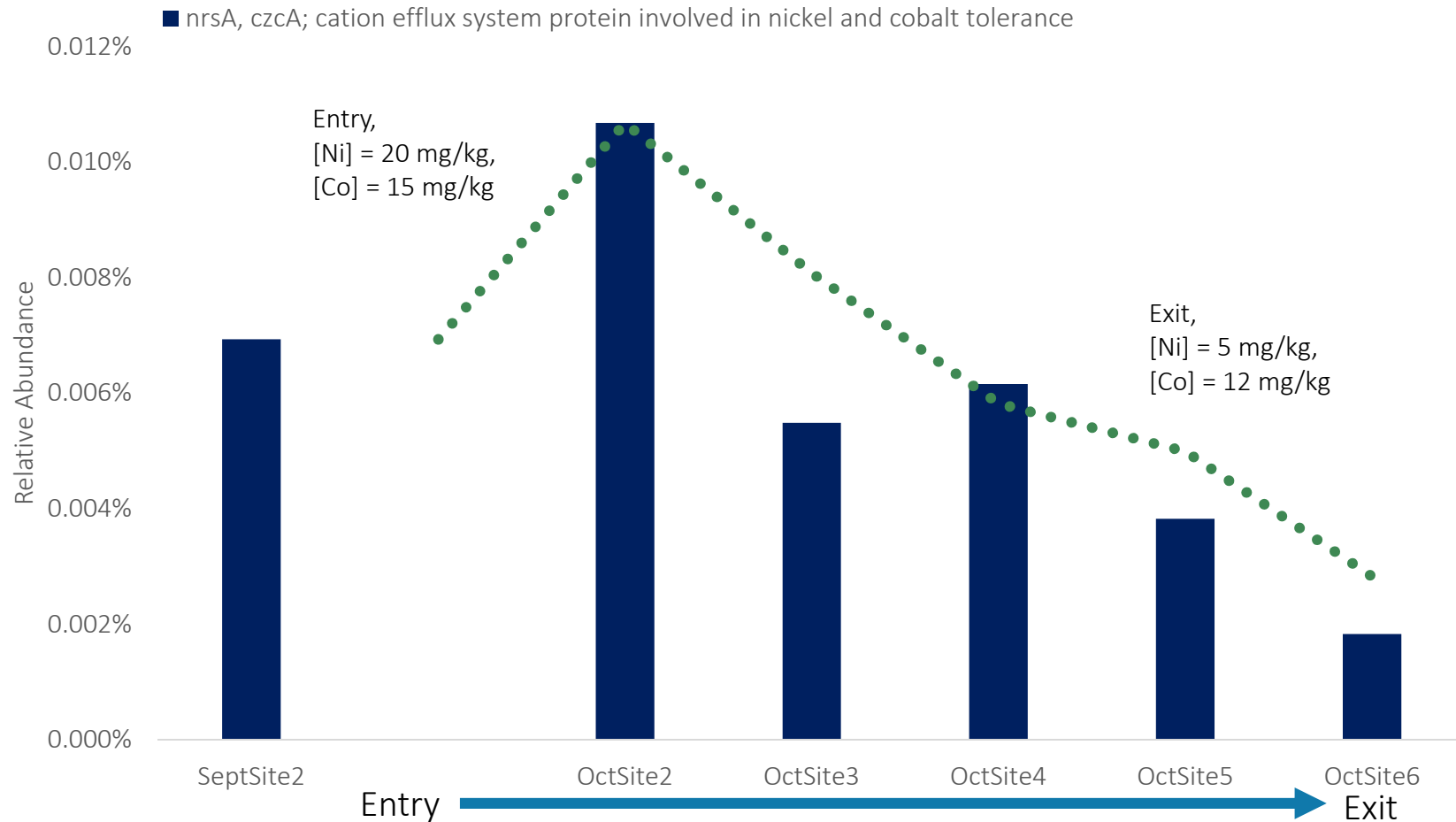
3. There is a link between metal removal and bacteria



+ Microbial Analysis Results – Surface Sediment



+ Functional Analysis Results – Surface Sediment



Relative Abundance of Functional Genes Associated with Nickel and Cobalt Measured in Field Samples

+ Microbial Analysis

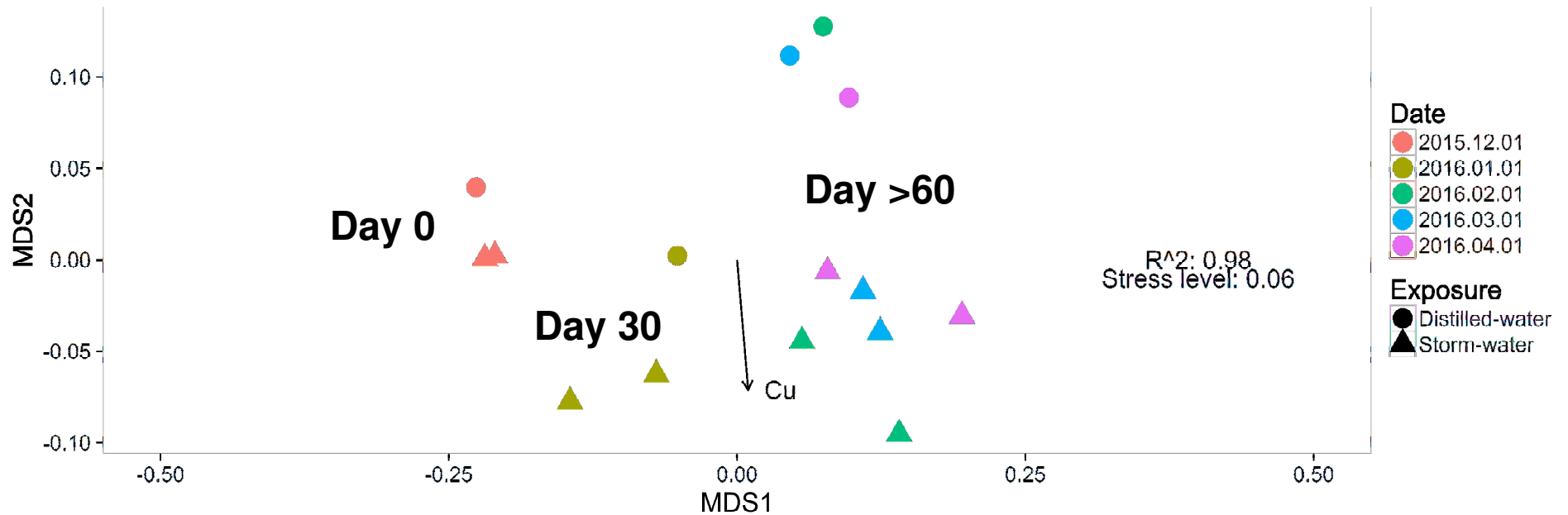
Hypothesis 4



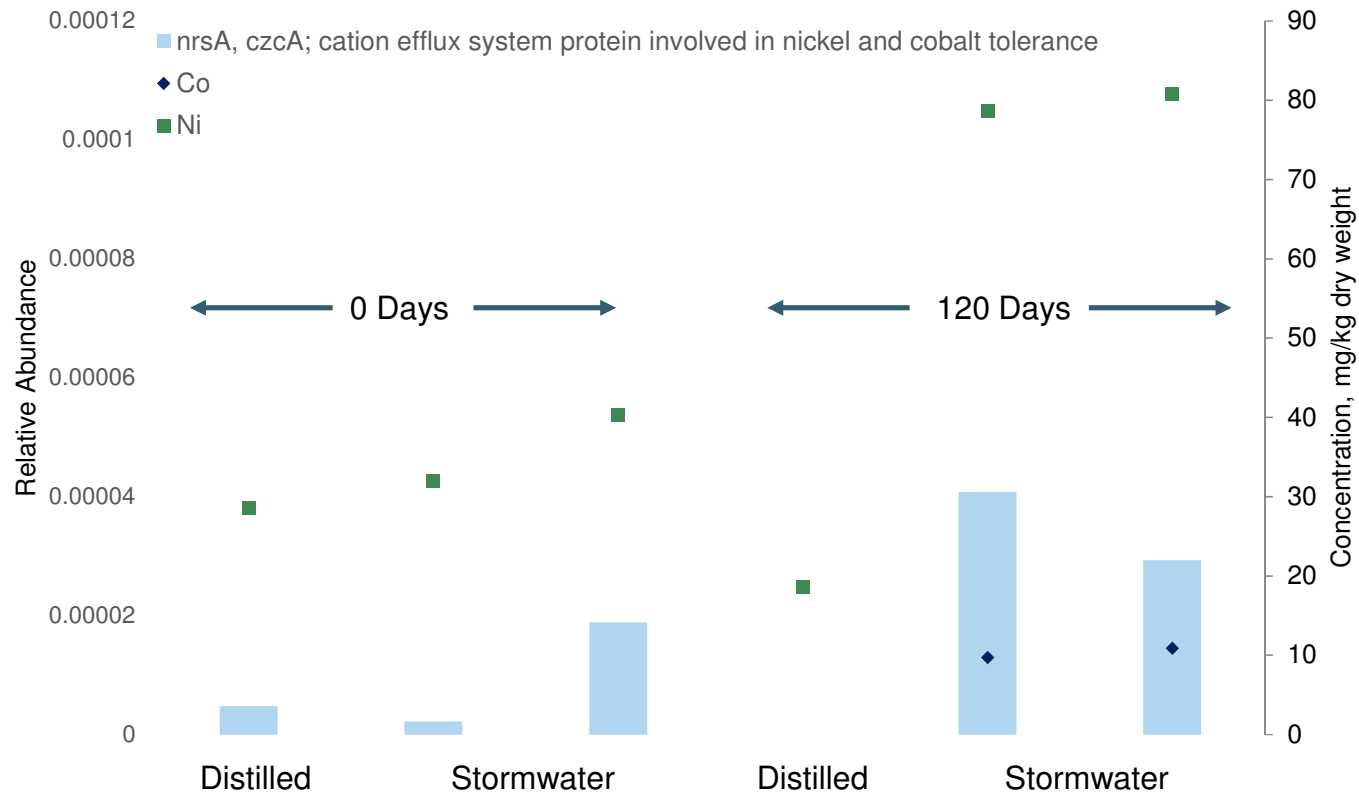
4. Stormwater exposure causes bacterial communities to form similar adaptations at other sites



+ Microbial Analysis Results – Surface Sediment



+ Functional Analysis Results – Column Surface Sediment



Relative Abundance of Functional Genes Associated with Cobalt and Nickel

+ Final Comments

- Data support hypotheses and interest in microbial results
- Results will support the Lost Lagoon wetlands
- Next step is to repeat this work at other sites



+ Acknowledgements

Thanks!



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ECOLOGY SOCIETY



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Questions??
Comments!!

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