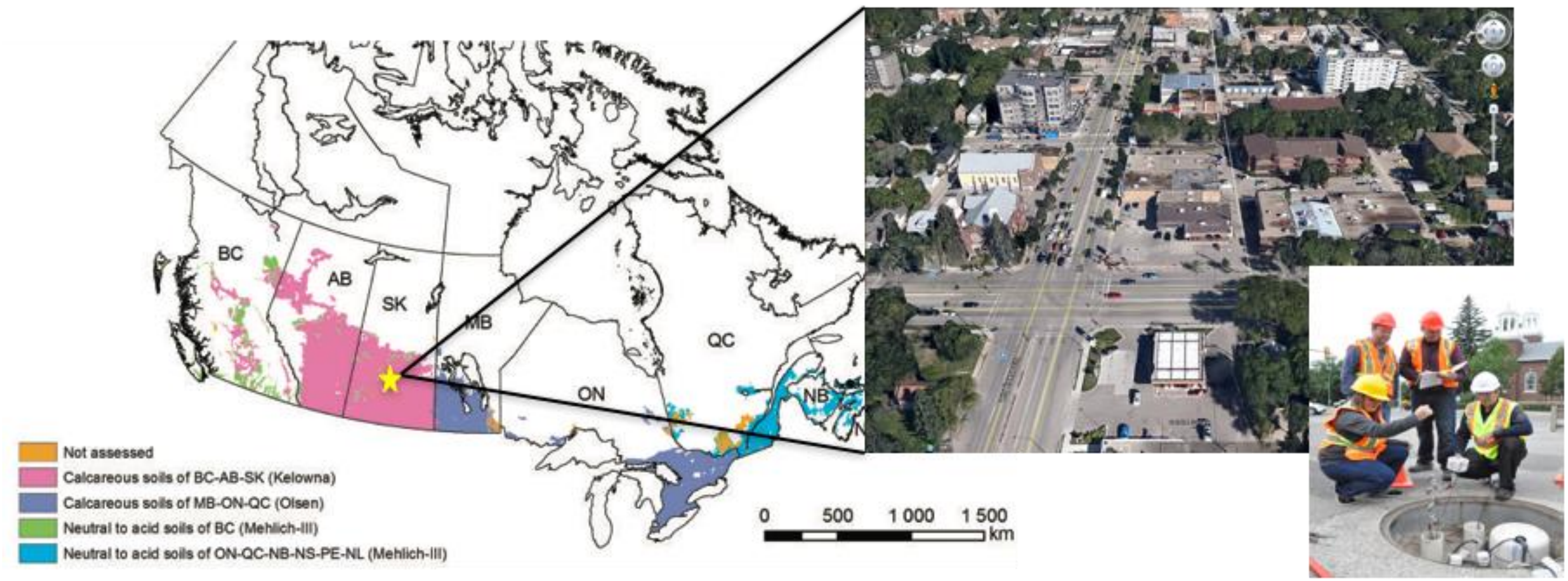
Where Innovation and Remedial Performance Meet – Findings From a Field Deployed Western Canadian In-situ Biostimulation Research Project



Derek Peak

The overall research project

Study 1: Soils were collected from 8 different PHC-contaminated sites of an industrial research partner, placed in permeable in situ reactors, and added to research wells at a contaminated site.

During this time, a nutrient solution (designed to be below bulk P solubility) was injected into the wells at a constant head. Soils were removed and analyzed.

Study 2: Citrate was added to the nutrient solution to evaluate the role of organic acids in biostimulation.

This work trained 2 M.Sc. Students and 3 Ph.D students

Installation of Research Infiltrators

Installation of in situ reactors

•Amendment 1mM HPO_4, pH 6, 0.011M $MgSO_4$ 20,000 L day^{-1}

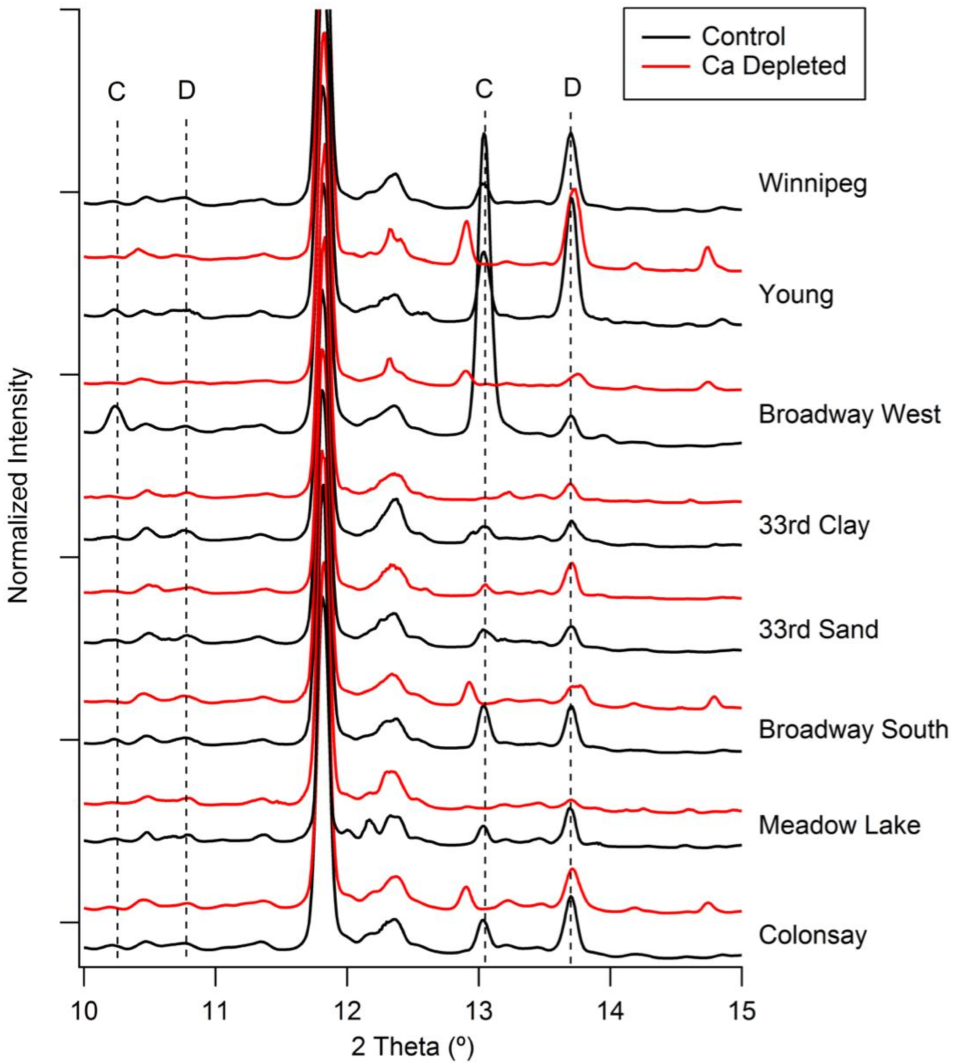
• Premade soil bags placed in the infiltrators August 22nd

Synchrotron powder XRD Results: Pretreatments

- One key difference in the samples is carbonate mineralogy:
  - Some are all calcite
  - Some have both calcite and dolomite
  - Some are mostly dolomite

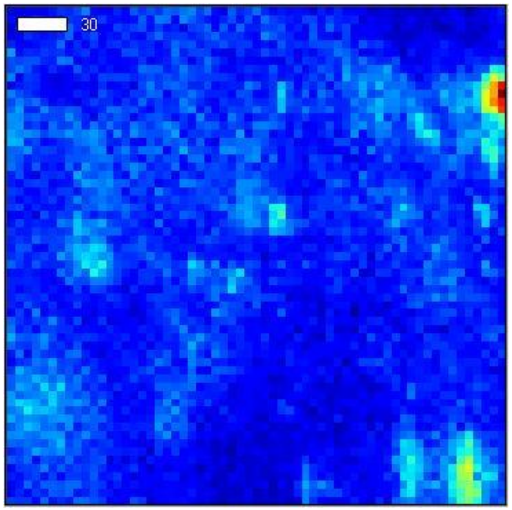
- Pretreatment with H_2SO_4 and $MgCl_2$ typically removed mostly calcite from soils.

- Pretreatment also changed smectites when present to Mg-pillared vermiculites.

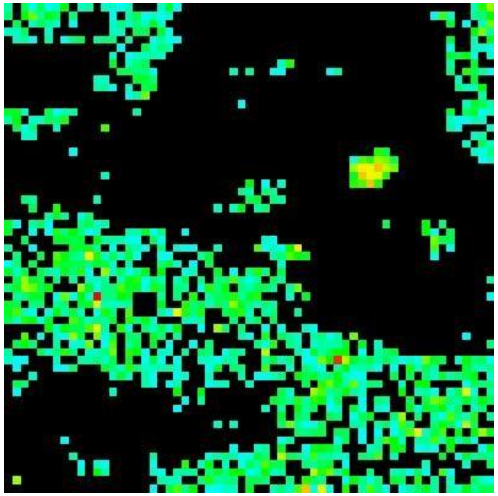


Direct Imaging of Carbonate Minerals with Synchrotron Microprobe

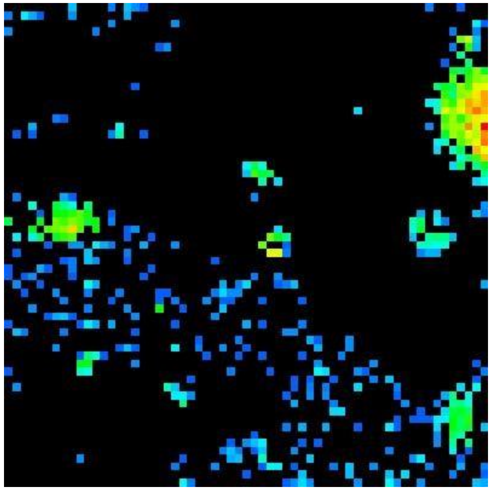
Ca XRF



Dolomite XRD



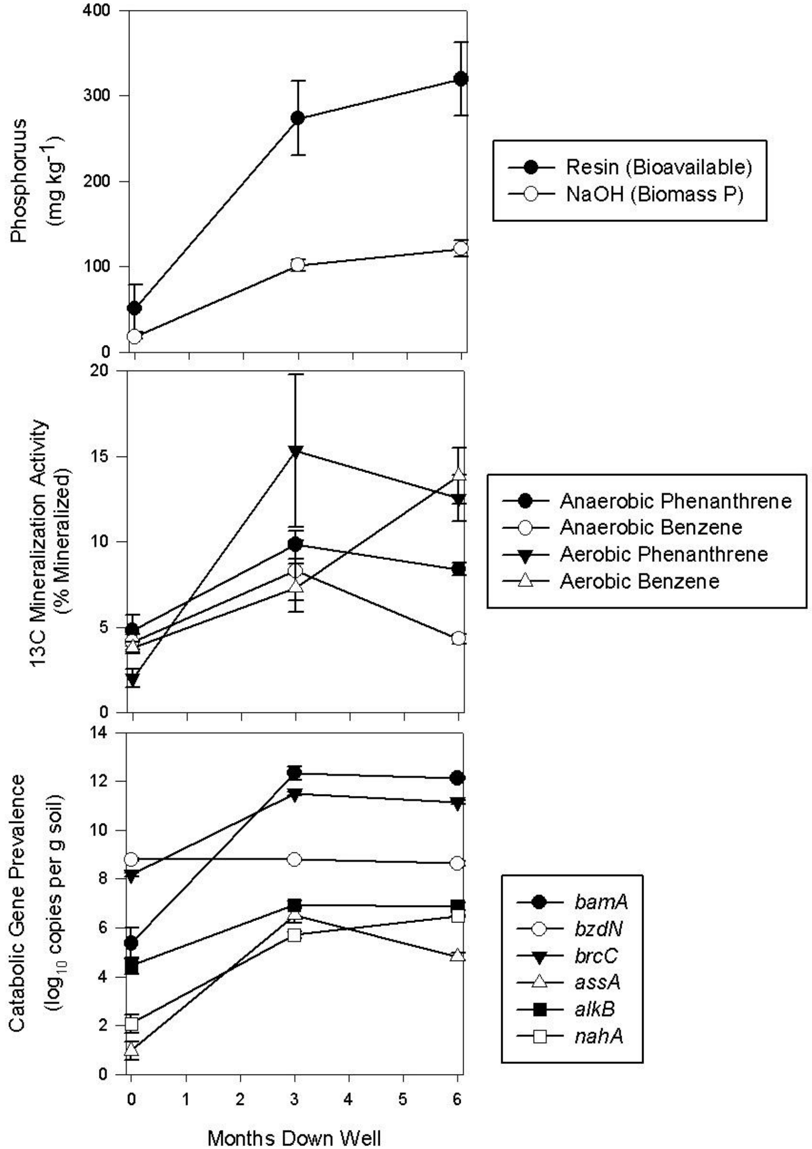
Calcite XRD



- It was typical for calcite to be found in large particles (50-500 microns) whereas dolomite often observed in much smaller diffuse particles.
- Parent material vs. secondary phases?

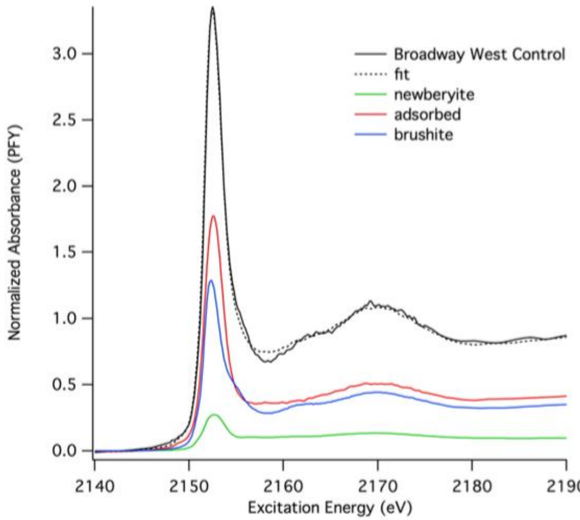
Overall Changes in the Soil Reactors

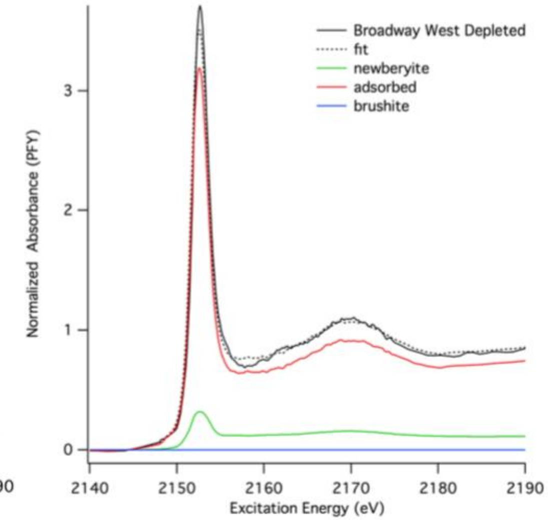
- Phosphorus fractions, mineralization activity, and catabolic gene prevalence of 16 soils incubated for 0, 3, and 6 months inside duplicate large bore injectors all increased over time (n=50)
- Both aerobic and anaerobic genes present in the soil reactors and both increased over time.
- Only slight differences between 3 and 6 months.

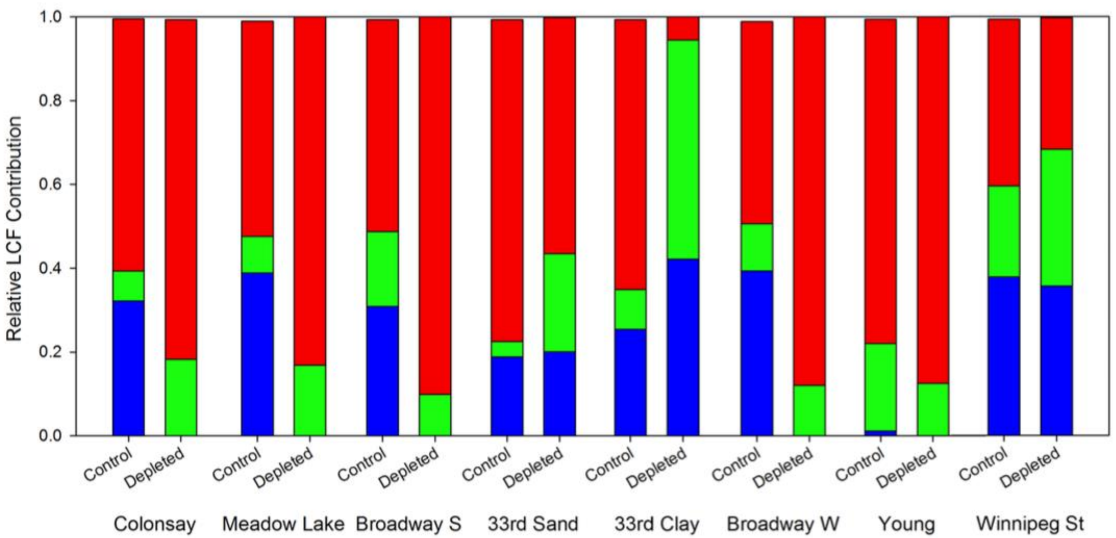


Quantitative XANES Analysis of 3 month Soil Samples

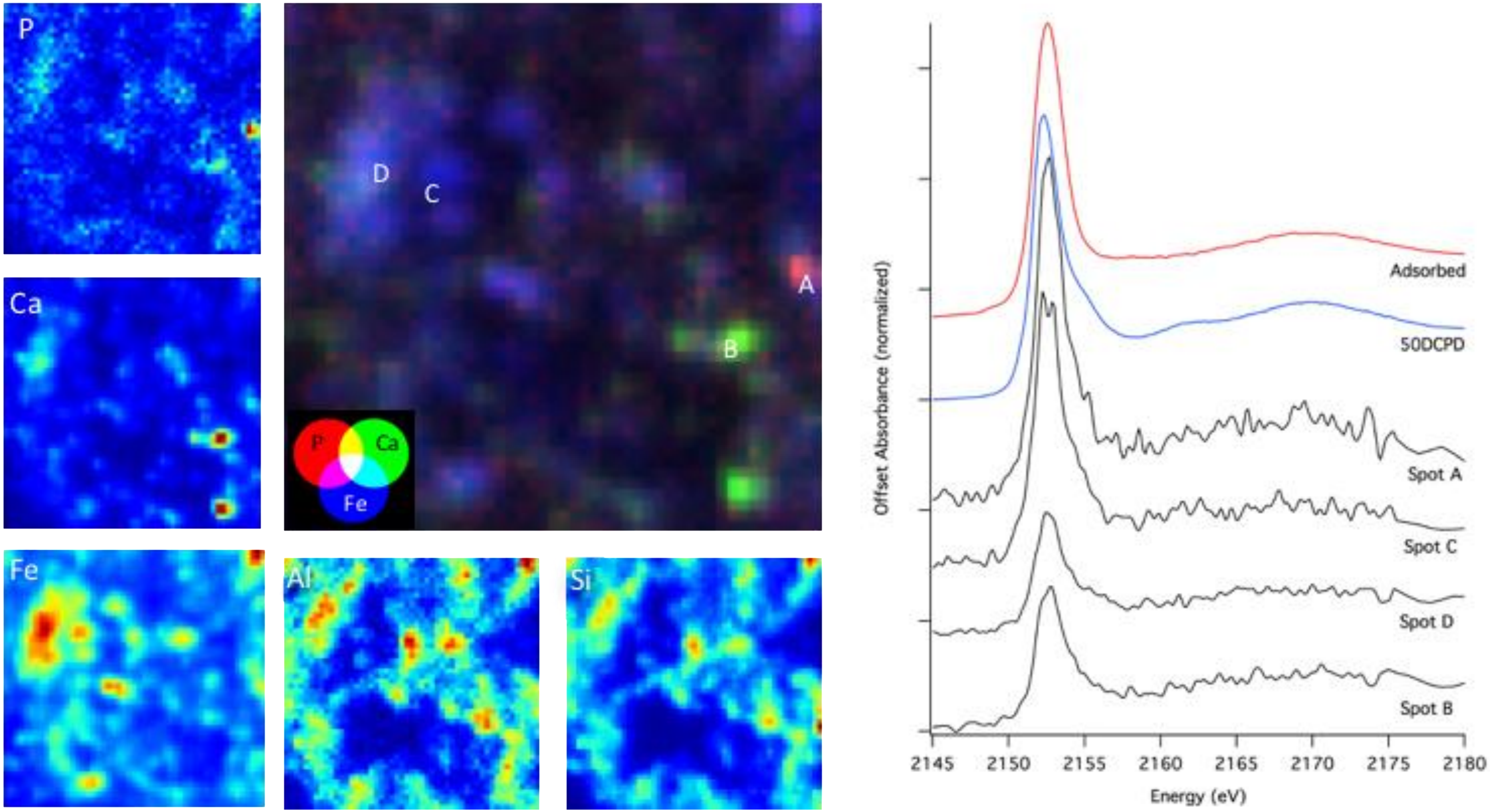
- Good fits with 3 components:
  - Adsorbed
  - Mg-rich brushite $CaHPO_4*xH_2O$
  - Newberyite ($MgHPO_4*xH_2O$)
- Clear trend: Depletion suppressed brushite and increased newberyite
- Adsorption the dominant species in almost all samples





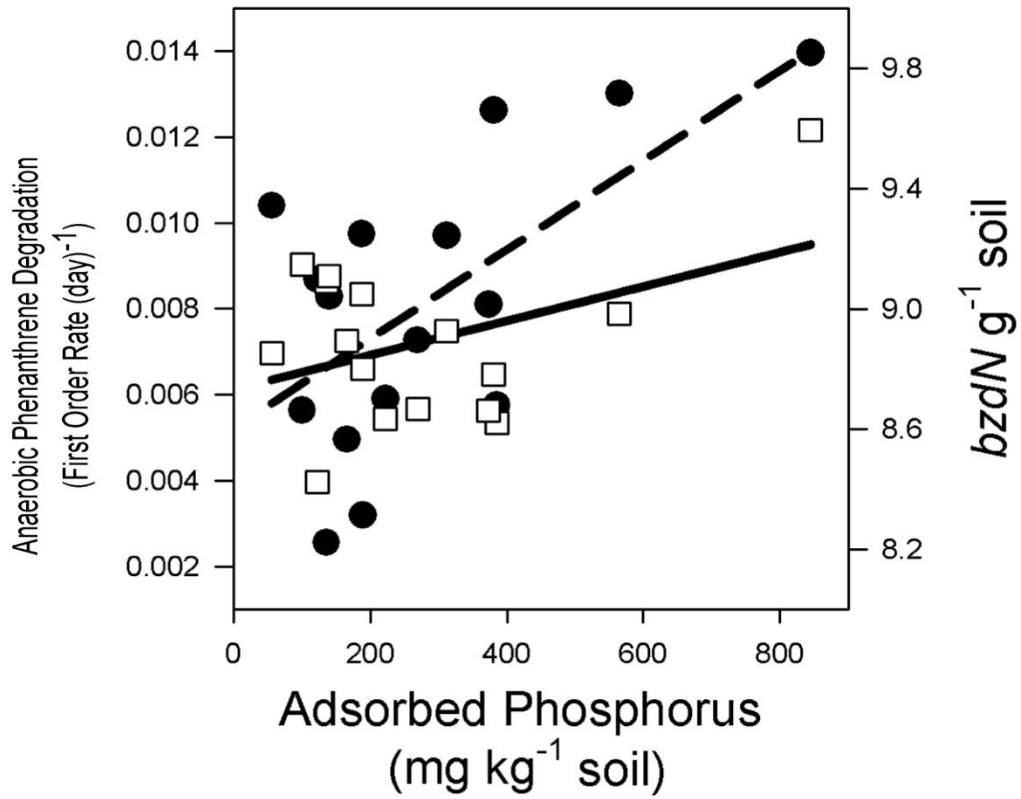


Microprobe: Broadway Control Soil



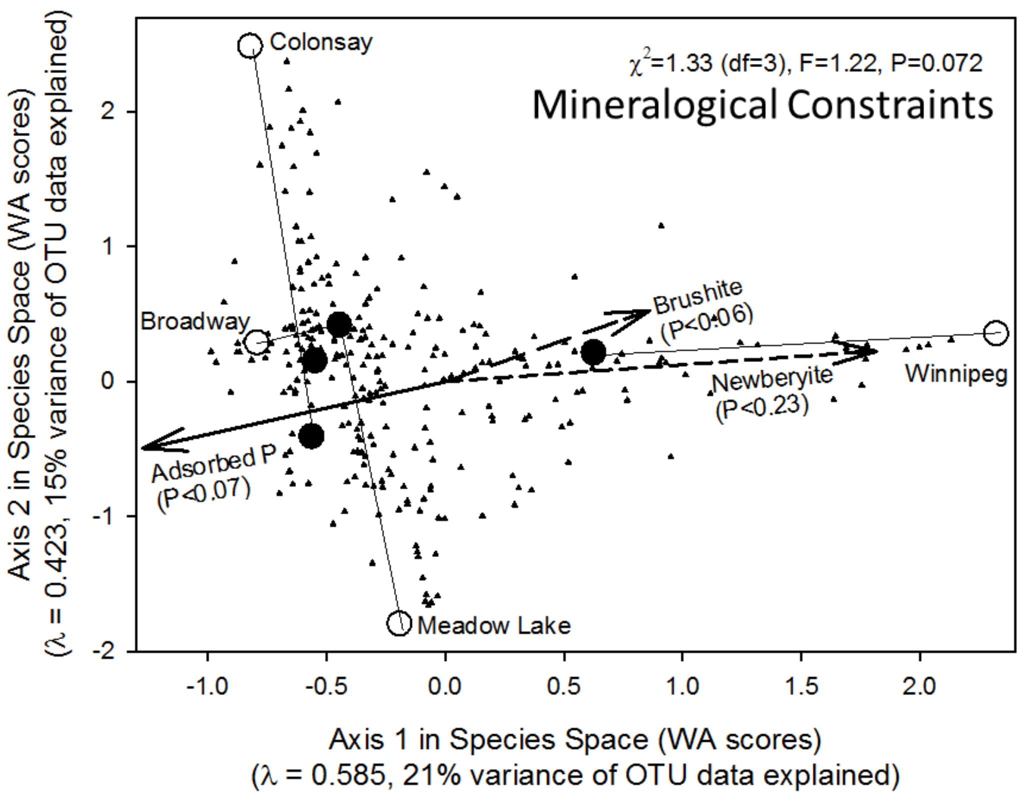
Relating P Speciation to Rates of Degradation

- Closed circles indicate anaerobic phenanthrene mineralization, and open squares indicate *bzdN* prevalence.
- The dashed line indicate a significant (P<0.10) correlative link between phosphorus speciation and anaerobic phenanthrene mineralization
- Solid lines indicate relationships between phosphorus speciation and catabolic gene prevalence.

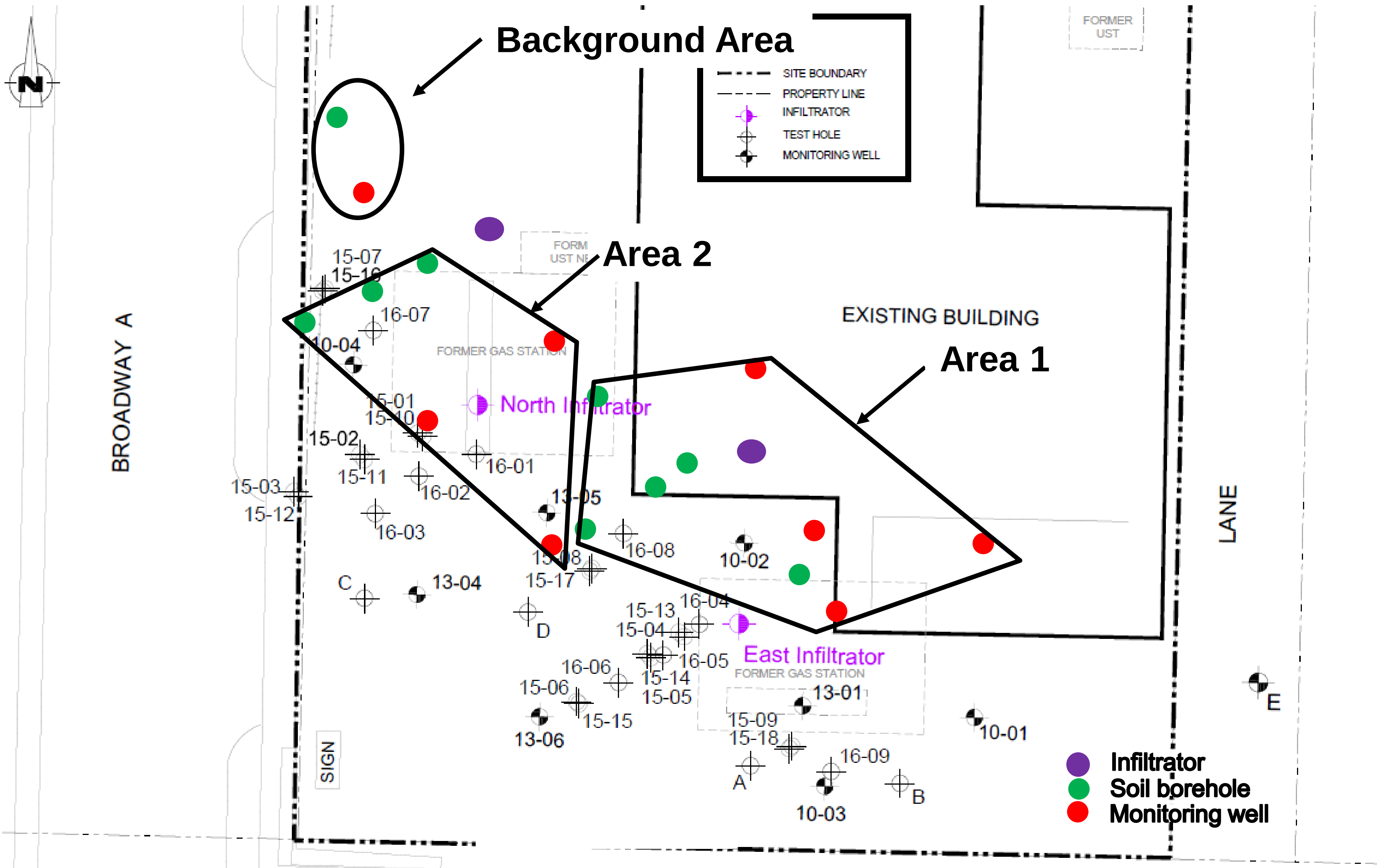


Relating P Speciation to Microbial Community Structure

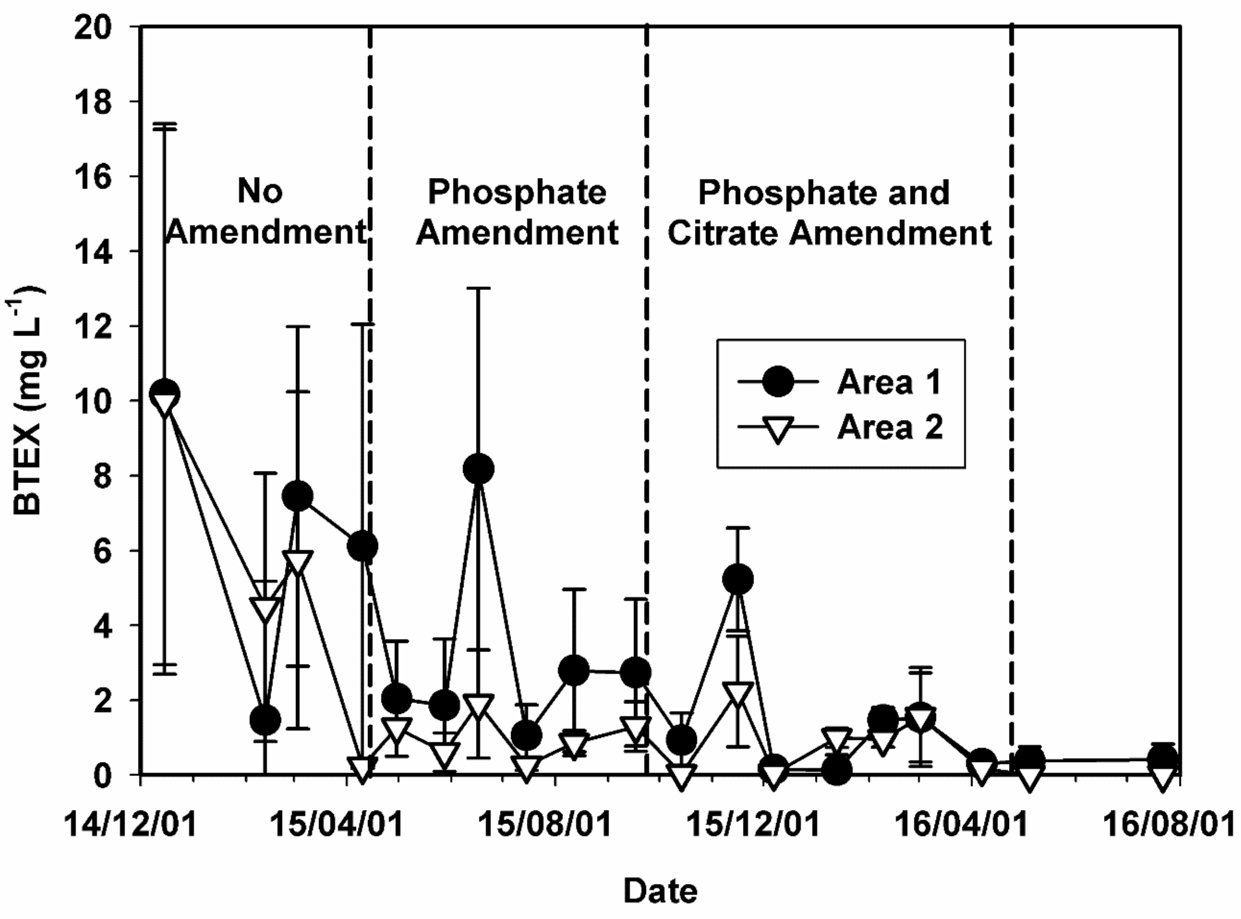
- Operational taxonomic units were identified by the EMIRGE protocol, and ordinated in species space by CCA
- Small symbols indicate OTUs, large closed circles indicate control, and open circles indicated calcium depleted sites.



Study 2: Role of LMOAs in biostimulation.



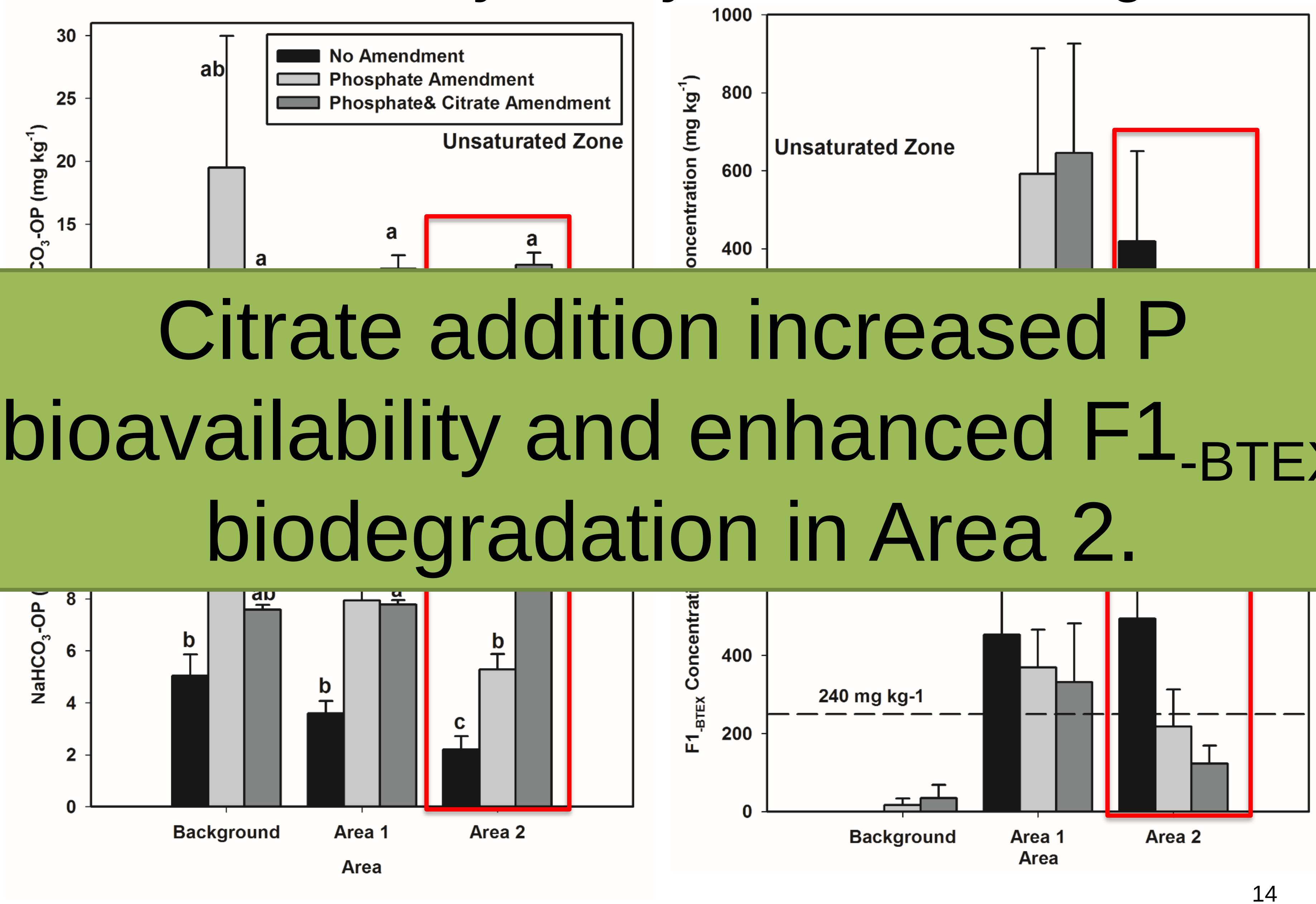
BTEX decrease in groundwater



Normalized to the Background Area
(minus Background BTEX concentration)

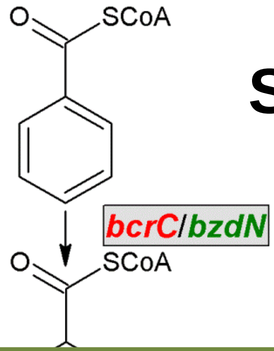
BTEX: benzene, toluene, ethylbenzene and xylene

Soil P bioavailability and hydrocarbon biodegradation



Citrate addition increased P bioavailability and enhanced $F1_{-BTEX}$ biodegradation in Area 2.

Soil functional microbial community in Area 2



ATP-dependent class I benzoyl coenzyme A reductase

Citrate addition selectively stimulated hydrocarbon-degrading anaerobes containing *bzdN* and increased culturable facultative hydrocarbon degraders.

bzdN ***bcrC***

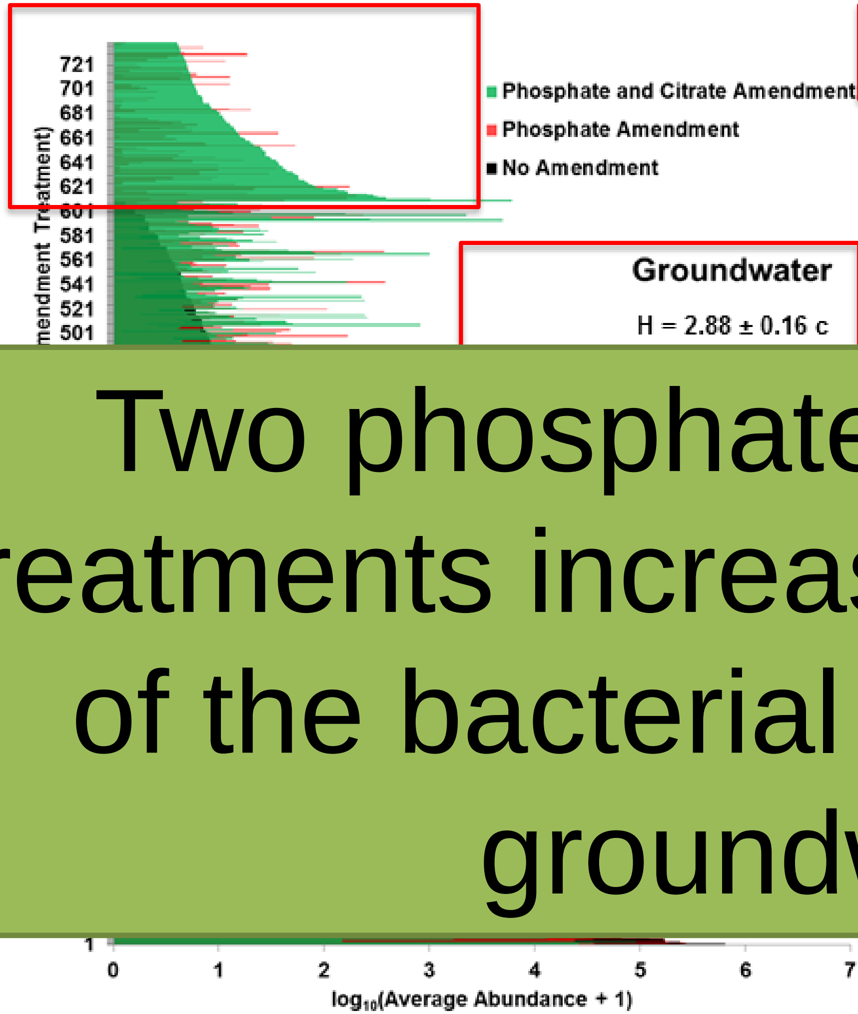
Functional Genes

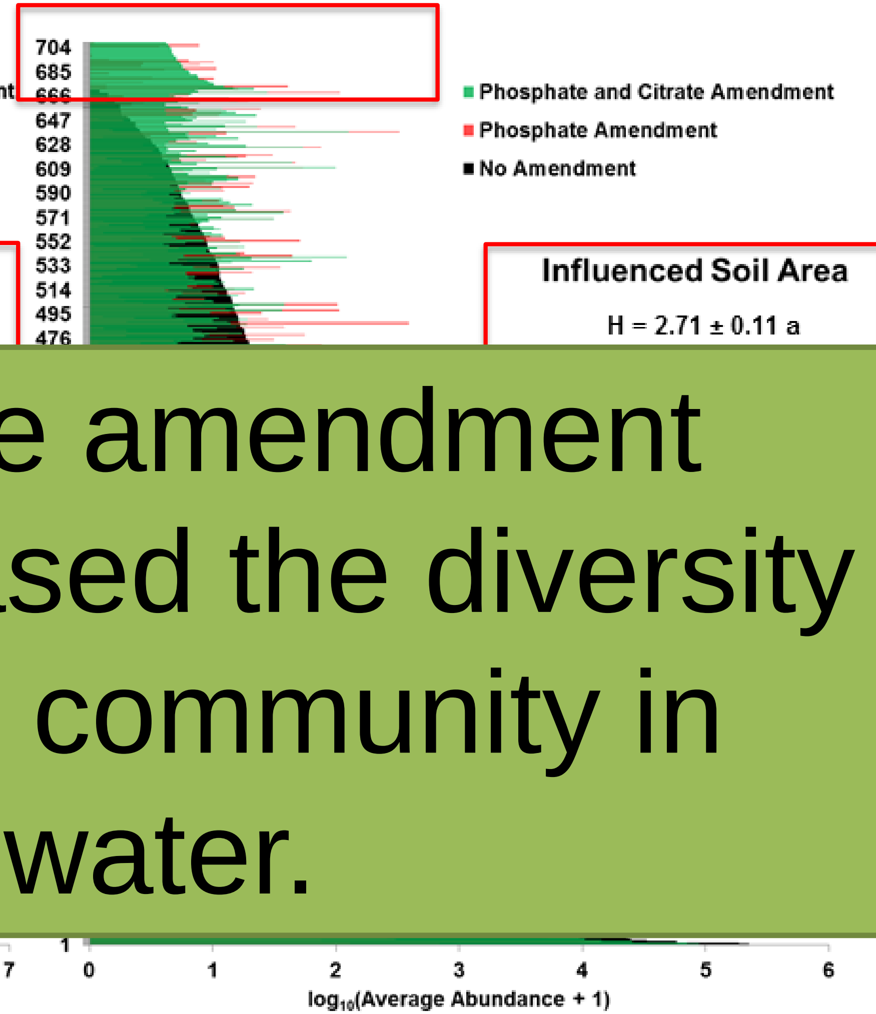
Phosphate Amendm...

Phosphate and Citrate Amendm...

Treatment

16S metagenomic sequencing library

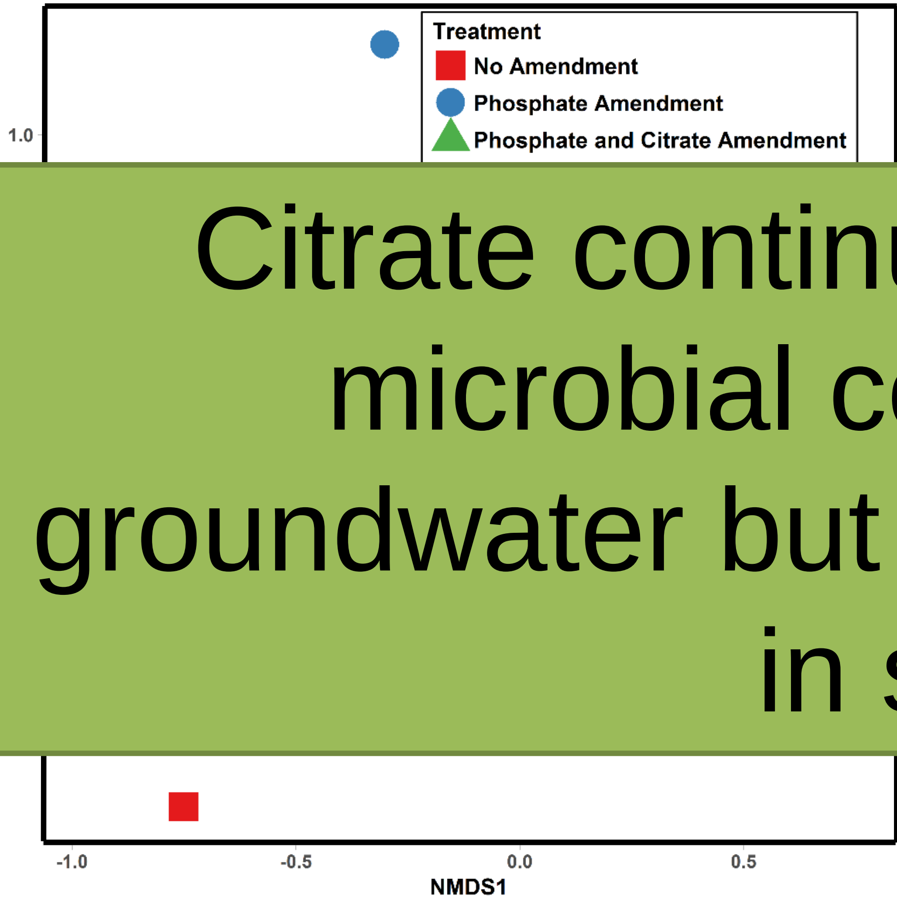




Two phosphate amendment treatments increased the diversity of the bacterial community in groundwater.

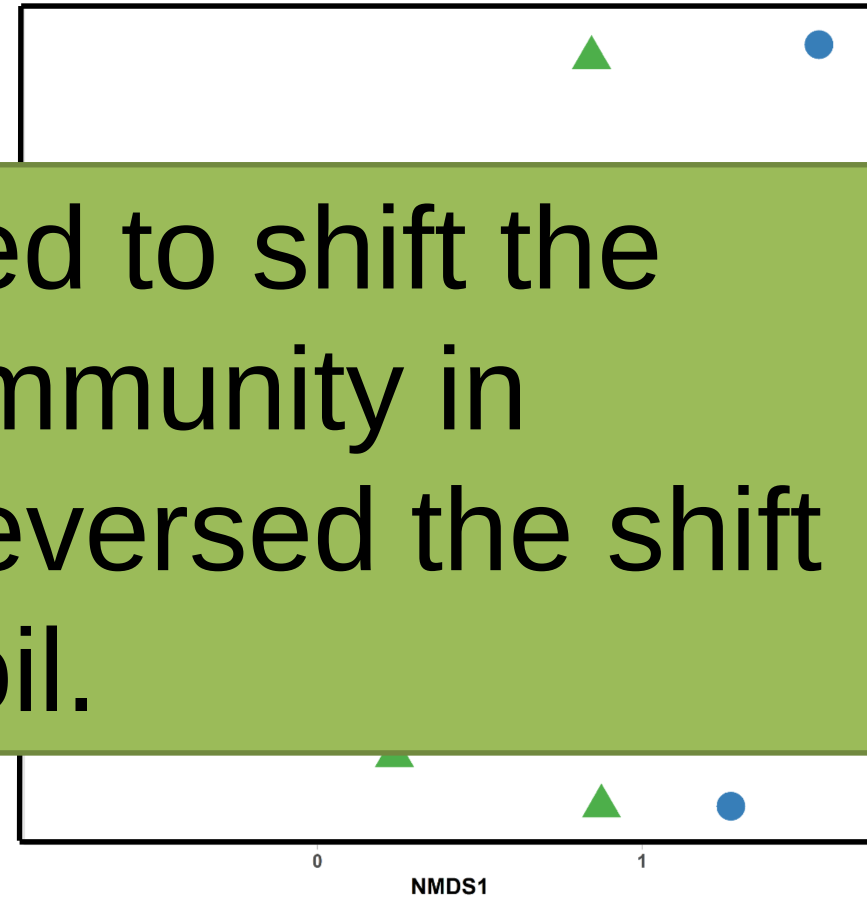
Microbial community structures

Groundwater



Stress: 0.09

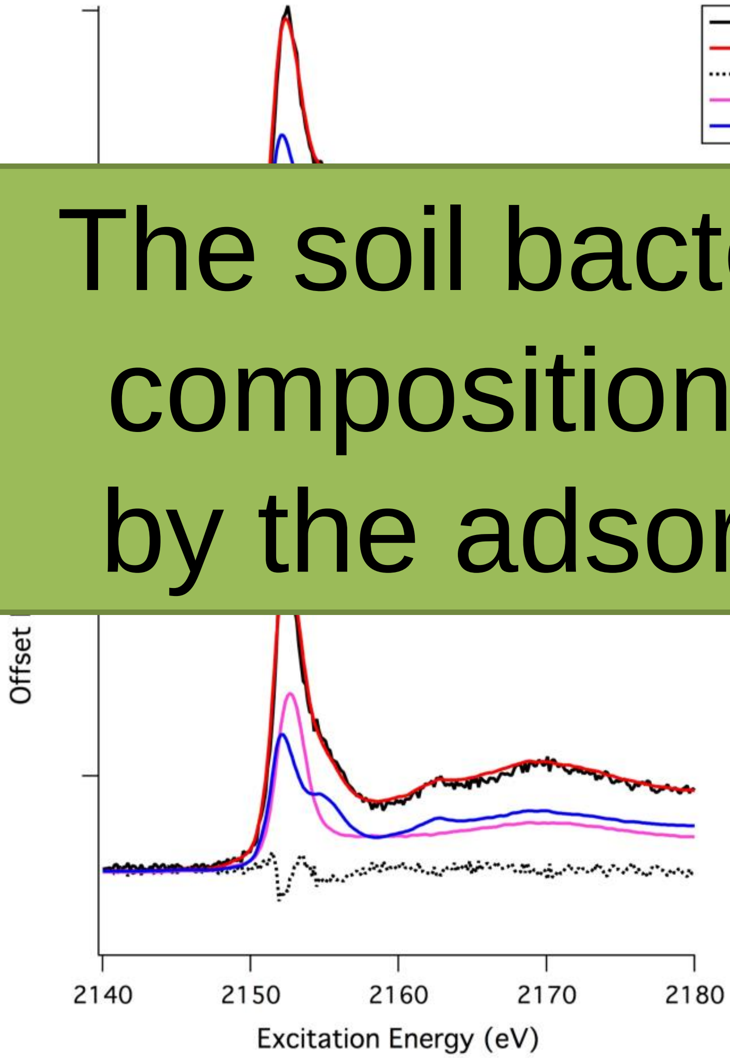
Influenced Soil Area

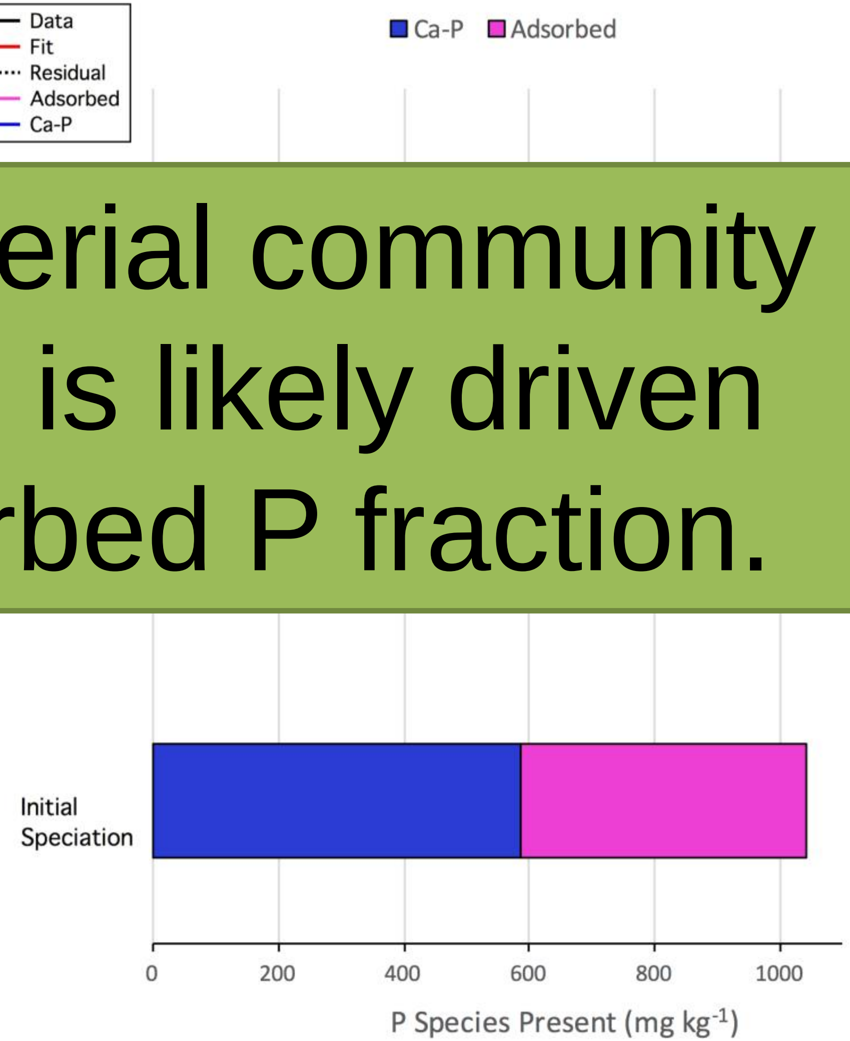


Stress: 0.08

Citrate continued to shift the microbial community in groundwater but reversed the shift in soil.

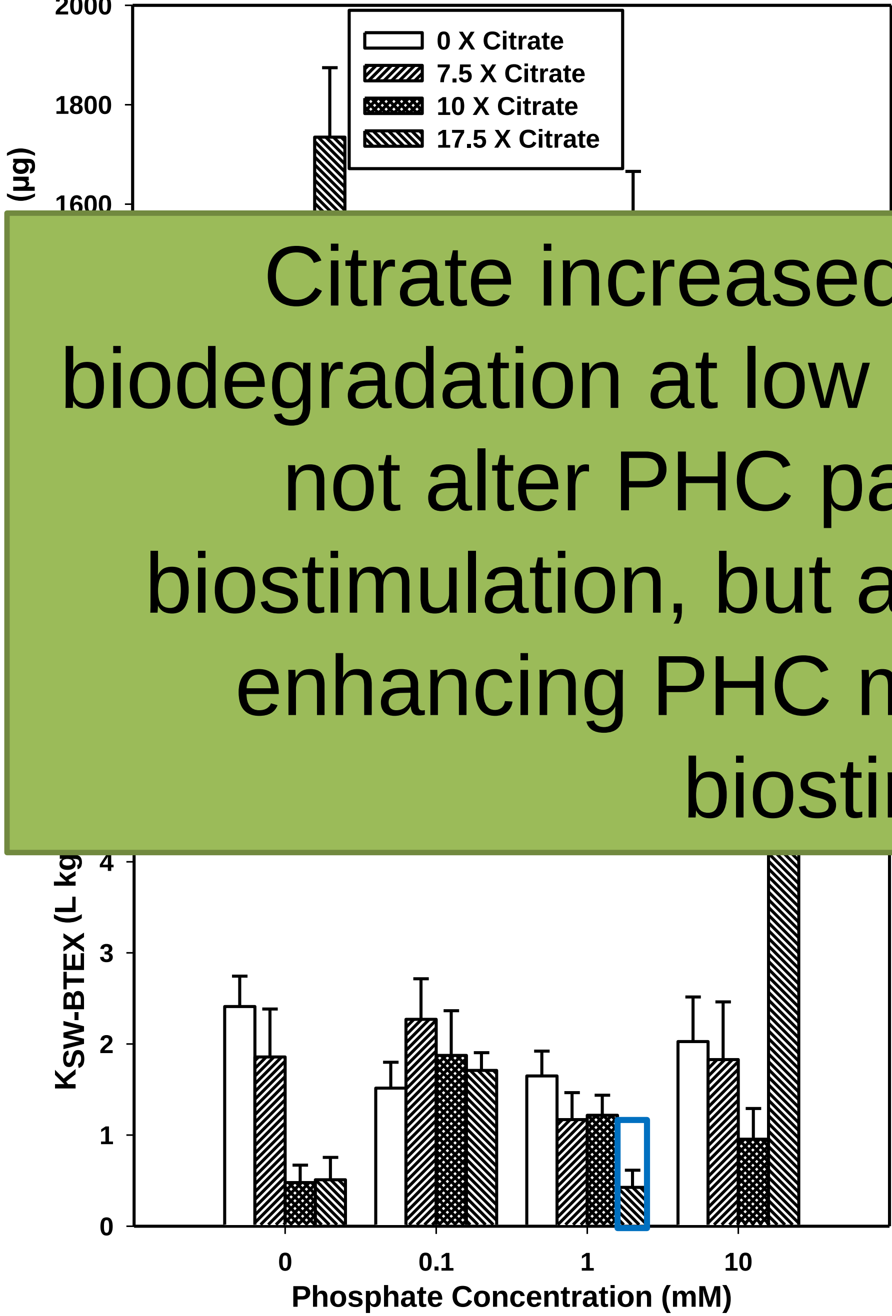
P K- edge XANES For Influenced Soil Area

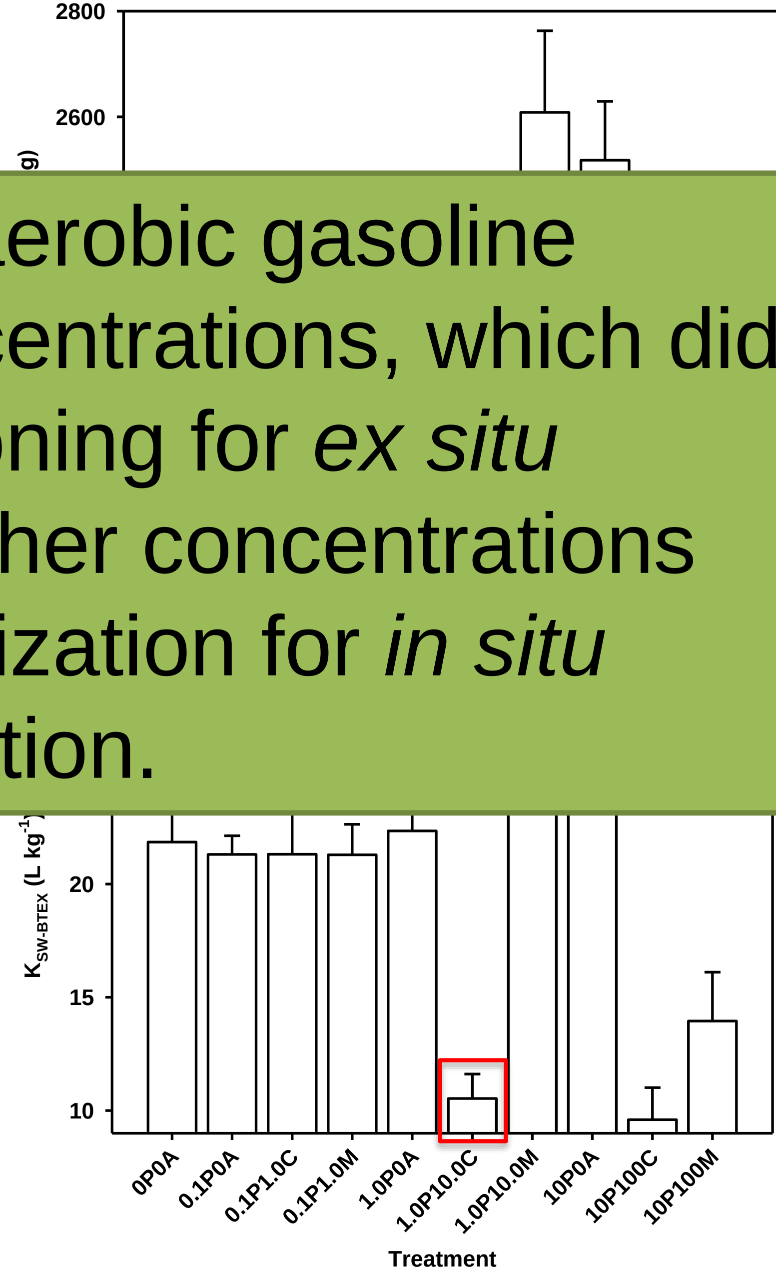




The soil bacterial community composition is likely driven by the adsorbed P fraction.

PHC bioavailability

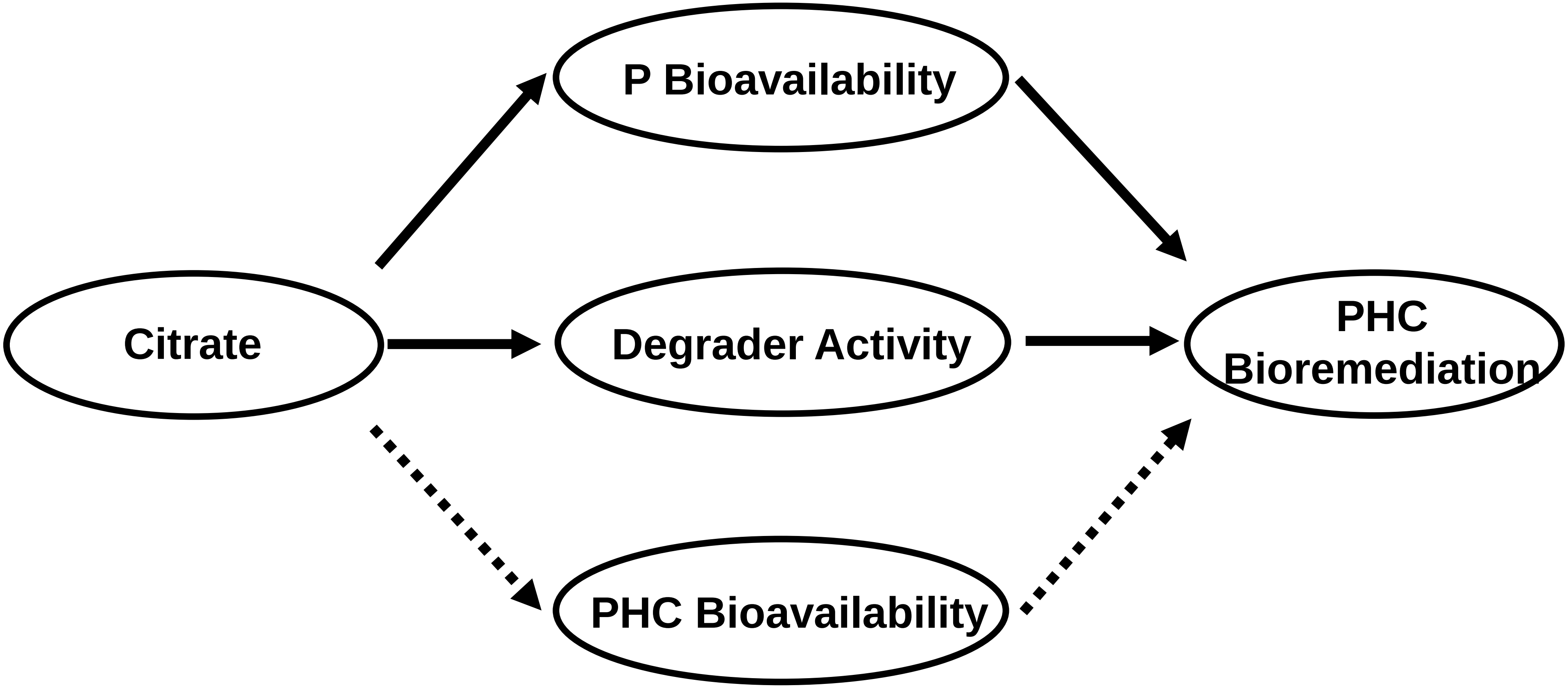




Citrate increased anaerobic gasoline biodegradation at low concentrations, which did not alter PHC partitioning for *ex situ* biostimulation, but at higher concentrations enhancing PHC mobilization for *in situ* biostimulation.

$K_{SW\text{-}BTEX}$: distribution factor between soil and water for BTEX

Summary

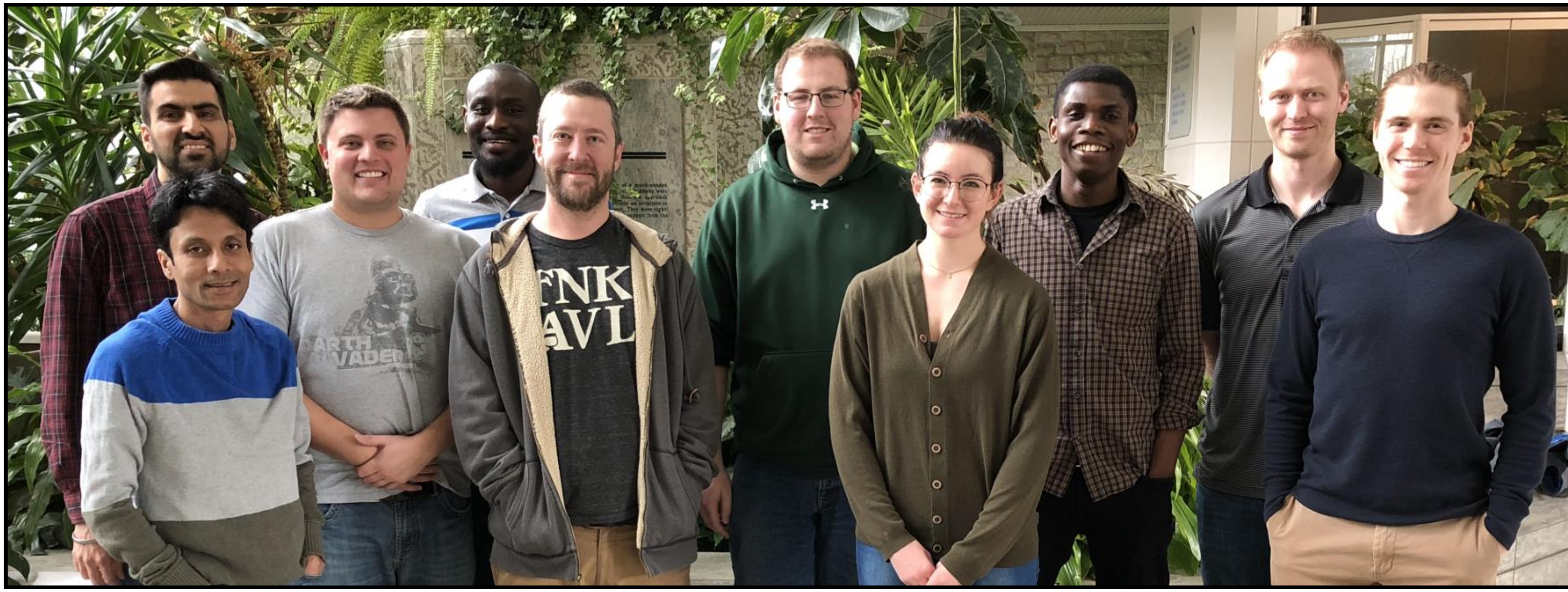

P Bioavailability
Citrate
Degrader Activity
PHC
Bioremediation
PHC Bioavailability

Overall Summary

- PHC on site moving rapidly below regulatory guidelines!
- XANES analysis showed best fits with 3 poorly crystalline components:
  - Adsorbed phosphate
  - Mg-rich brushite
  - newberyite
- Adsorbed phosphate positively correlated to biodegradation rates… this species is bioavailable to microbes!
- Clear differences in community of microbes occur when the soil P speciation changes.

Acknowledgements

- Environmental Soil Chemistry group:



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