



Positron Emitting Isotopes for Spatial Resolution of Microbial Activity in Soils: Applications and Future Directions in In Situ Bioremediation

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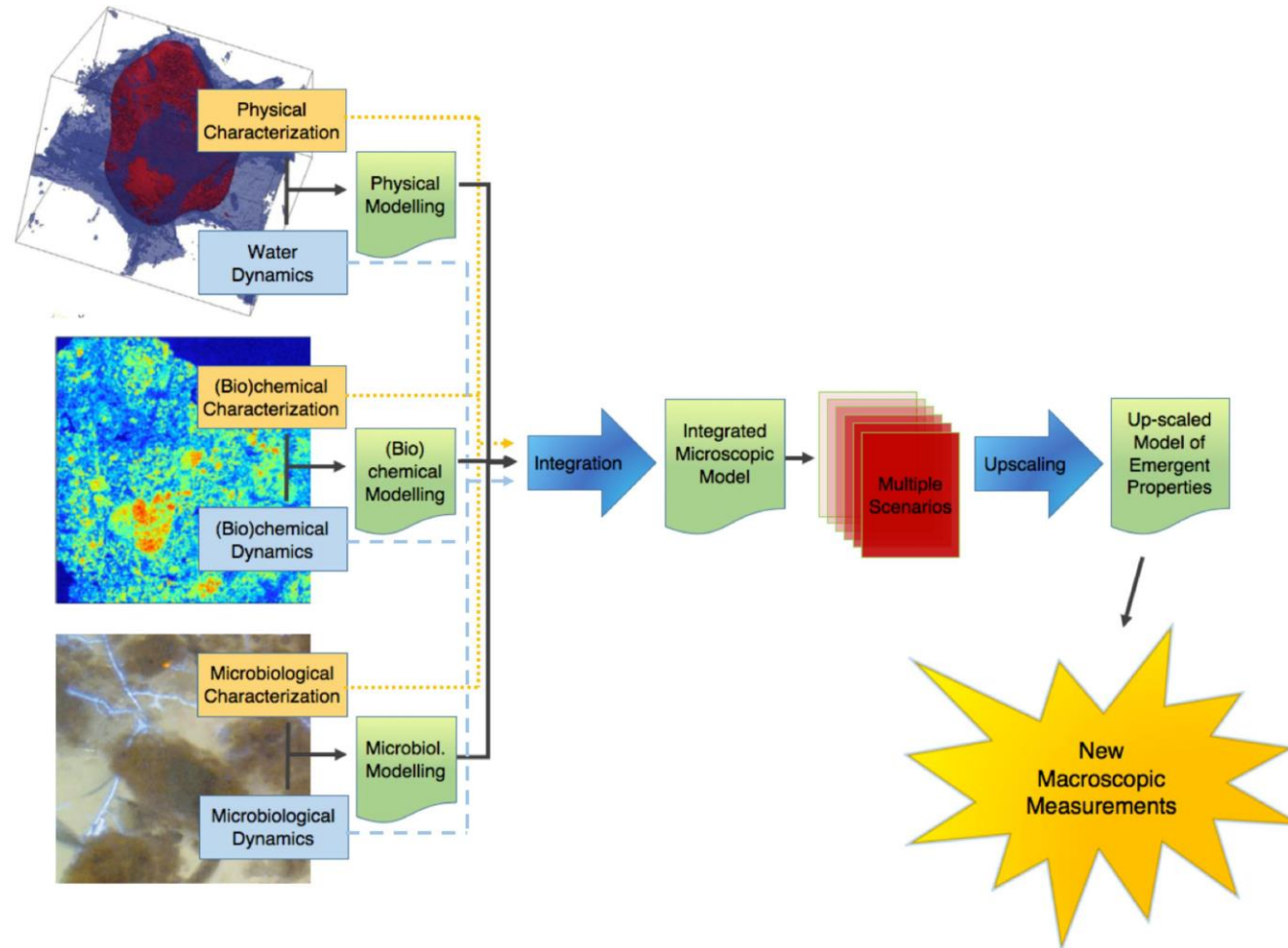
University of Saskatchewan



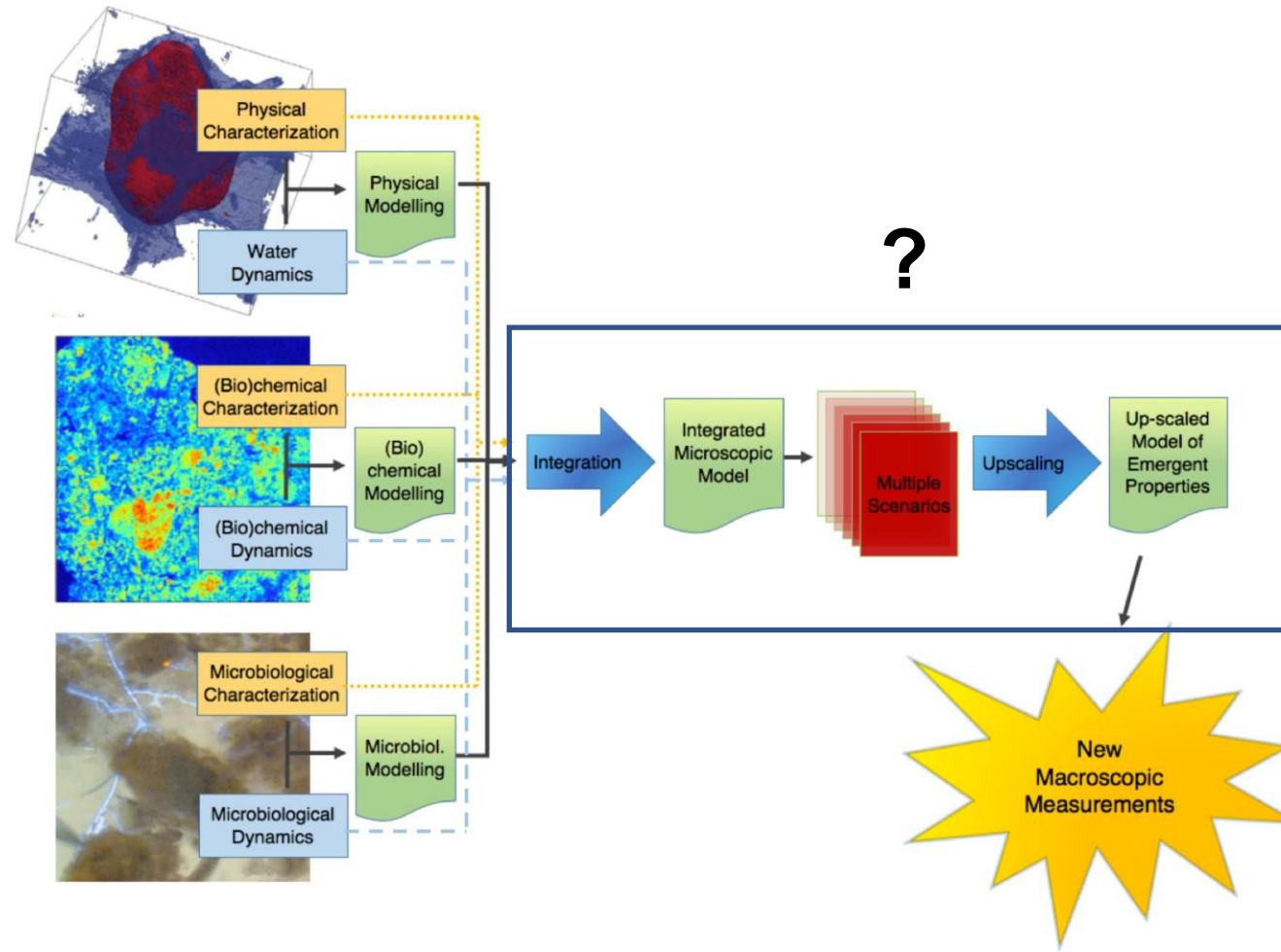
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Soil heterogeneity and microbial activity

- Soils are highly heterogeneous media, variety of microenvironments in a given sample
- Microbial activity (e.g. contaminant transformation) favors certain microenvironments over others based on physical and biogeochemical characteristics



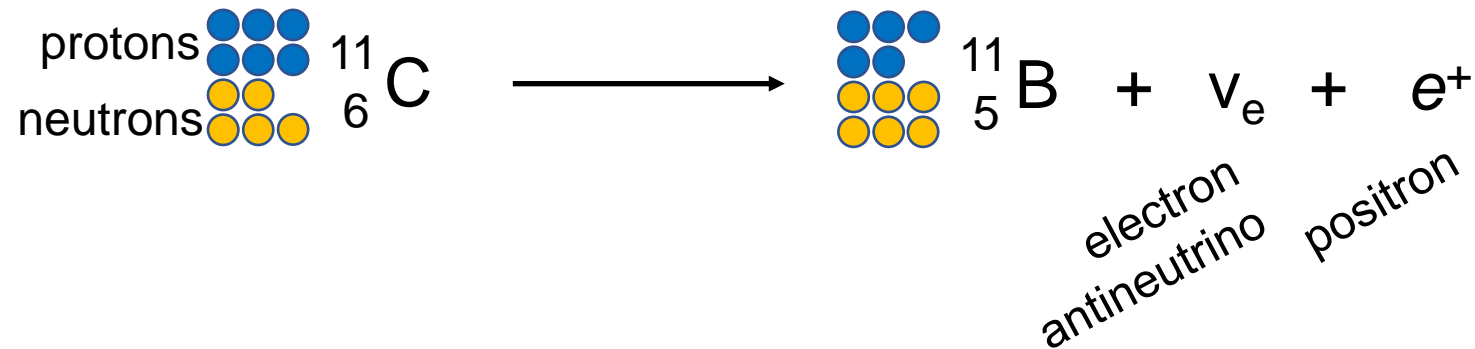
Soil heterogeneity and microbial activity



- Bulk measurements of these properties often do not correlate well with activity
- Unraveling spatial relationships between activity and local environment key to better understanding microbial processes in soils, including bioremediation
- Positron imaging may reconcile these points

What is positron imaging?

Positron: a subatomic particle with the same mass as an electron and a numerically equal but opposite charge

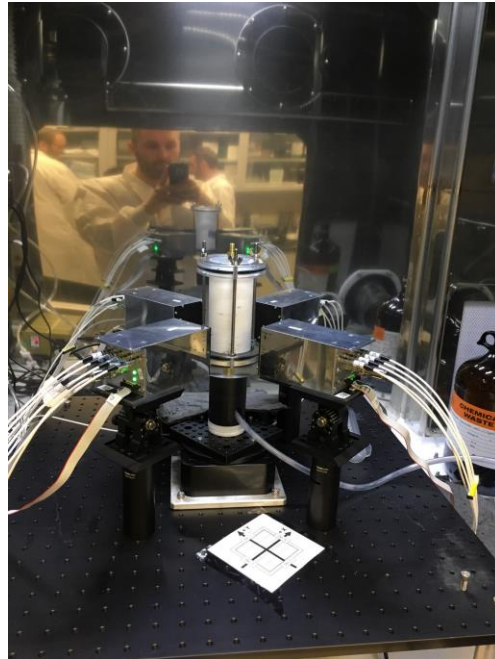
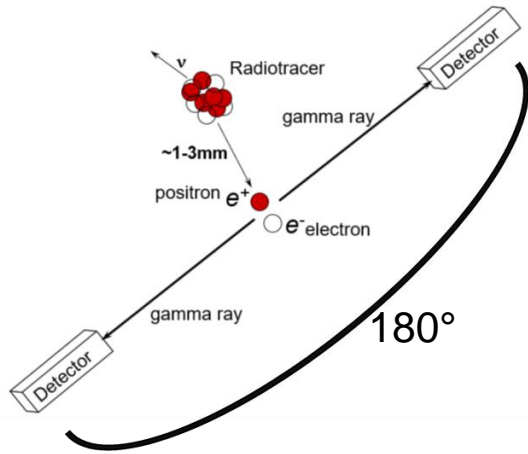


Positron-emitting
Radionuclides produced
at cyclotron facilities

Positron emitting isotope	Half life
^{11}C	≈ 20 minutes
^{13}N	≈ 10 minutes
^{15}O	≈ 2 minutes
^{18}F	≈ 119 minutes

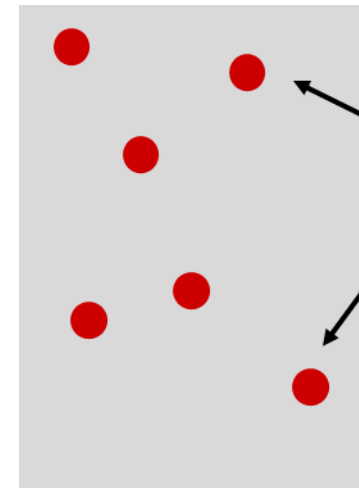
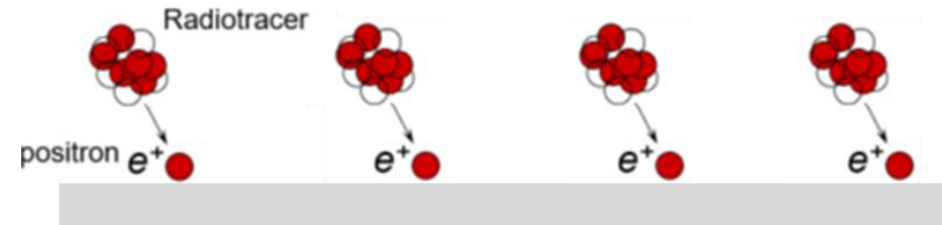
What is positron imaging (PET and autoradiography)?

Positron emission tomography (PET)



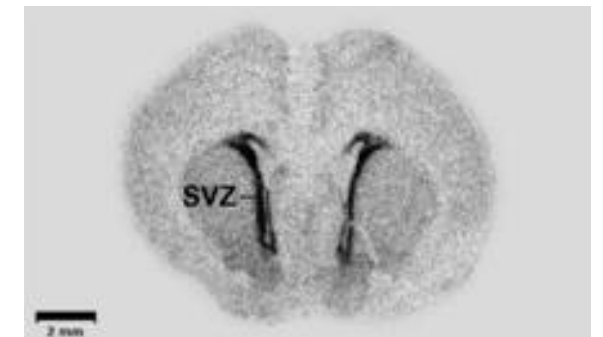
Using detector arrays, three dimensional tracer distribution accessible

Autoradiography



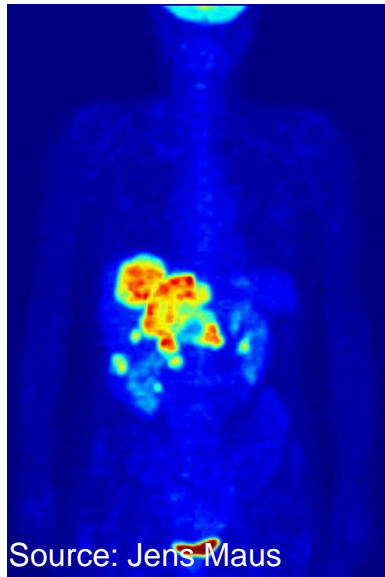
Regions exposed to radiotracer

Tracer distribution in rat brain



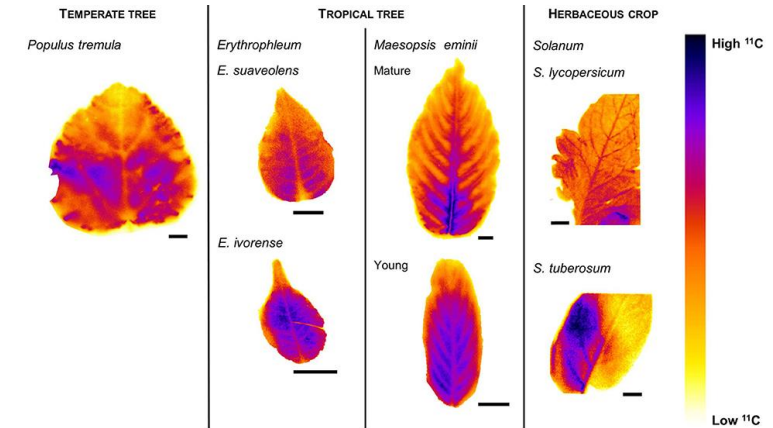
What are the benefits of positron imaging?

Medical diagnostics



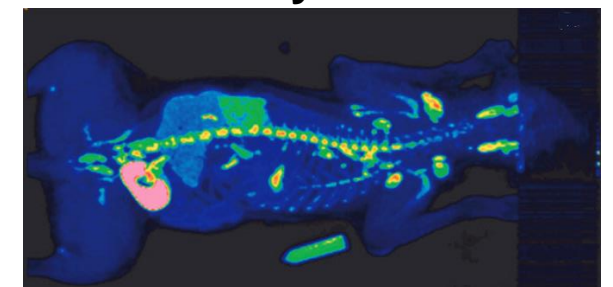
Property of Positron imaging	Benefits
Radiation penetrates well in many media	Visualization in opaque media
Numerous isotopes and chemistries available	Tracers tailored to many systems
Uses synthetic isotopes	What we see is what was added (i.e. no natural background)
Short half-life of radiotracers	Active regions visualized
Able to detect low tracer levels (sub-picomolar concentrations)	Minimal disturbance to sample chemistry and biology
Repeated scanning/imaging	Time-resolved visualization of tracer movement

Plant Science



Hubeau et al. *Front. For. Glob. Change*. 2019

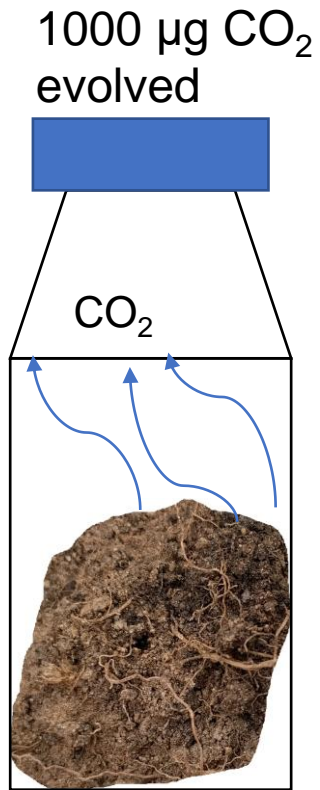
Veterinary medicine



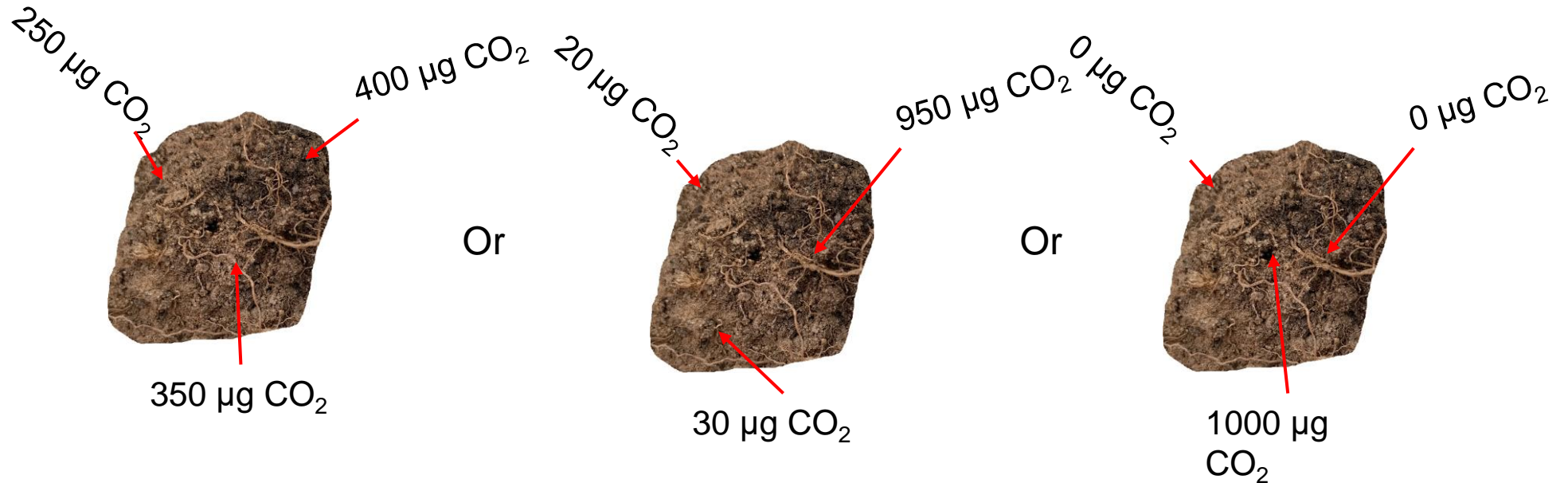
Rohren et al. *Vet. Comp. Oncol.* 2010, 8, 163-187

Some example projects from our lab
applying positron imaging to questions in
soil science

Visualizing microbial metabolism in soils

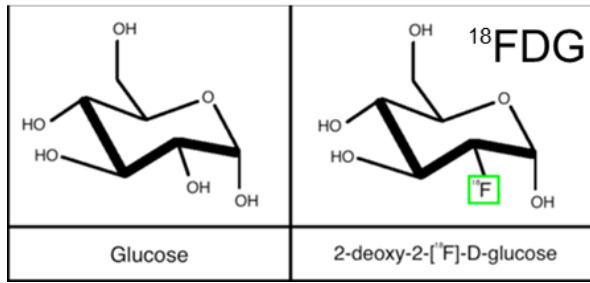


CO_2 evolution assay for microbial activity in soils



- Localized soil environments vary greatly in 3-dimensional space
- Many existing methods for measuring metabolism destructive and/or lack spatial resolution

Mapping microbial metabolism with PET

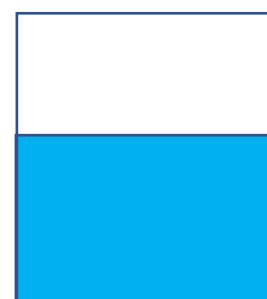
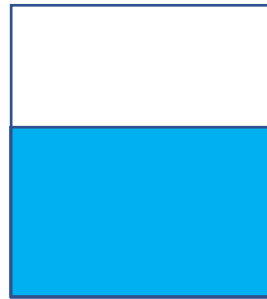


¹⁸F-FDG
+
Growth
medium

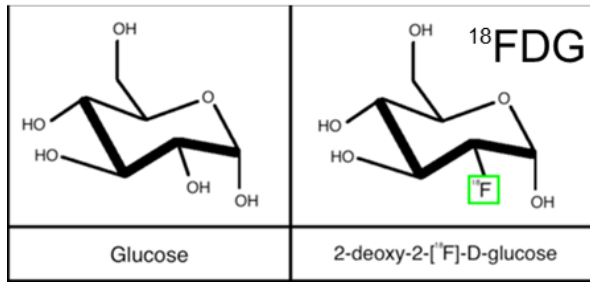
¹⁸F-FDG
+
Growth
medium

Column with active
P. fluorescens

Column with
autoclaved *P.*
fluorescens



Mapping microbial metabolism with PET

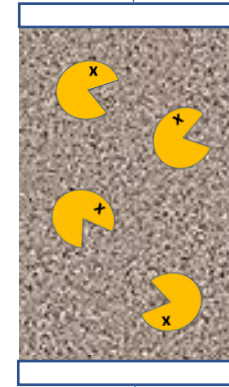


Column with active
P. fluorescens

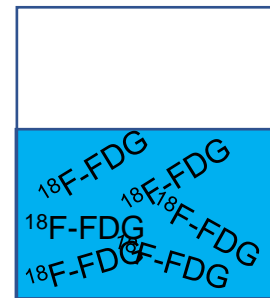
Growth
medium



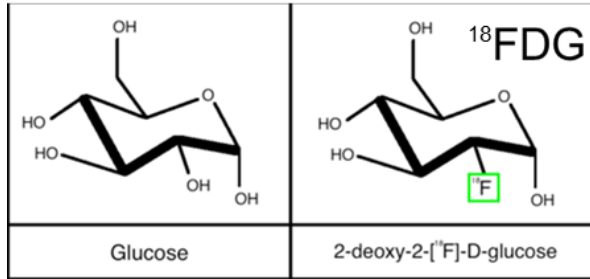
Growth
medium



Column with
autoclaved *P.*
fluorescens



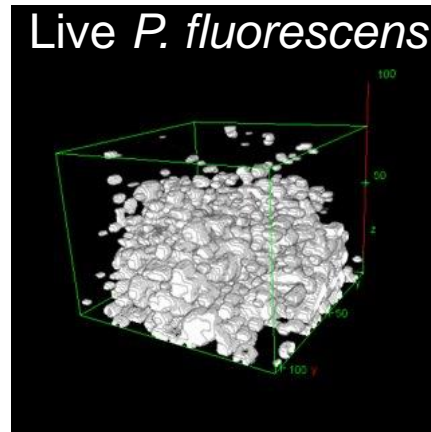
Mapping microbial metabolism with PET



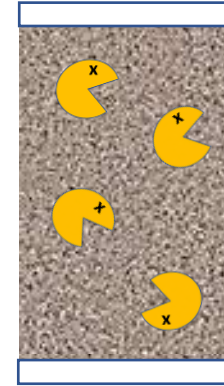
Column with active *P. fluorescens*



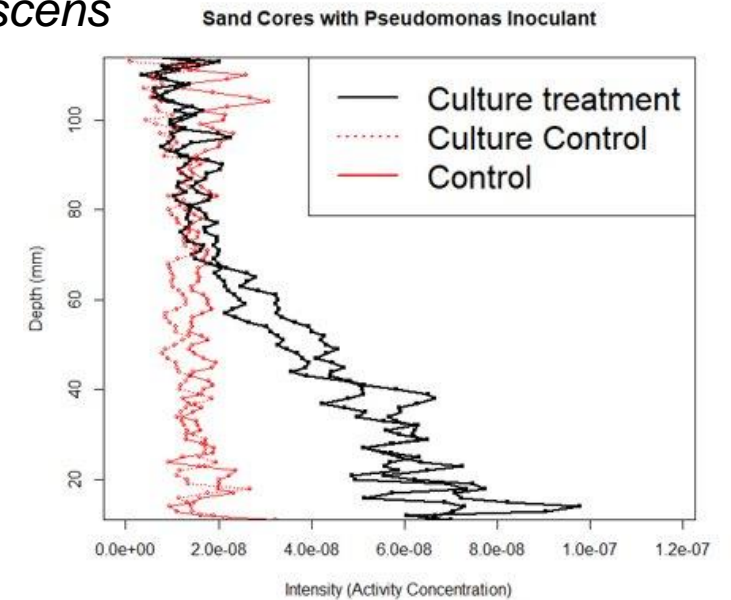
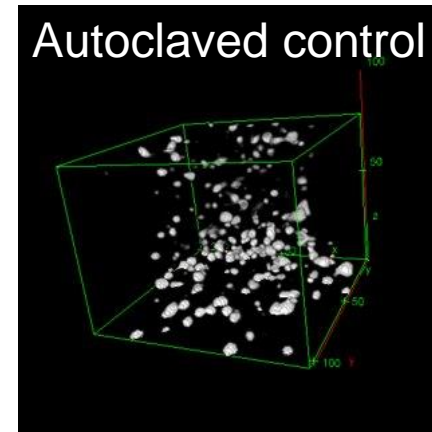
Live *P. fluorescens*



Column with autoclaved *P. fluorescens*

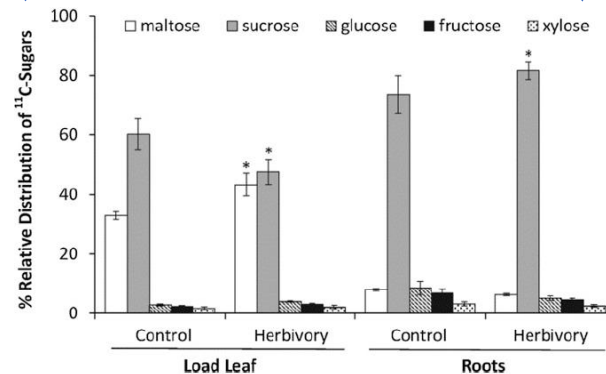
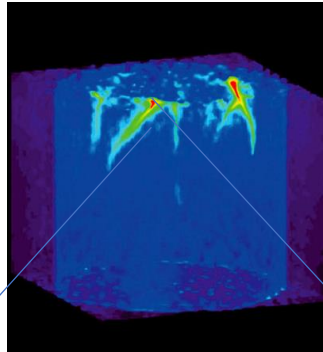


Autoclaved control



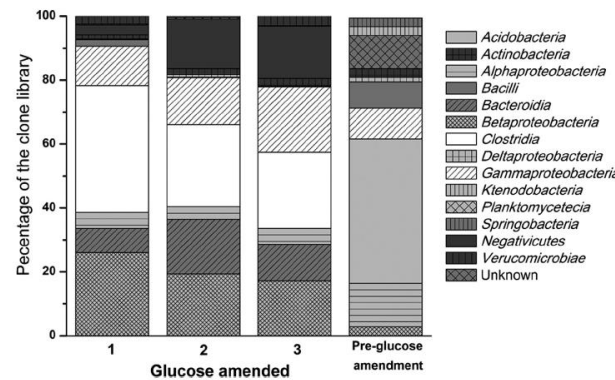
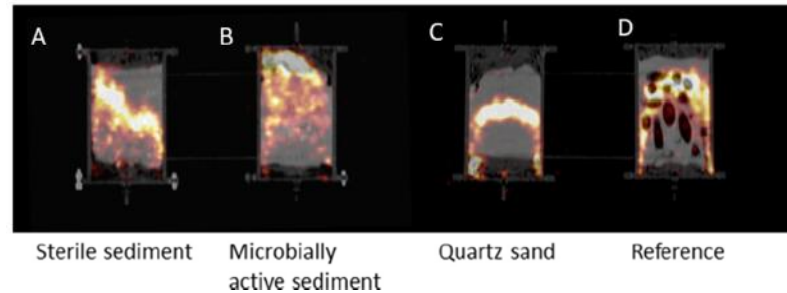
Positron imaging defines region of activity, then what?

Targeted biochemical analysis



Qu et al. *Plant Physiol.* **2016**, *172*, 776-788

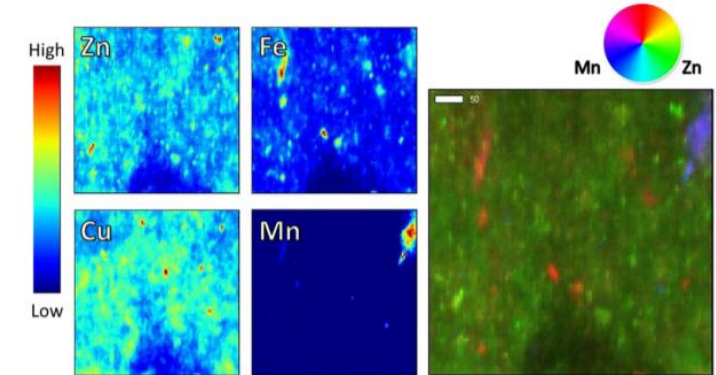
Targeted genetic analysis



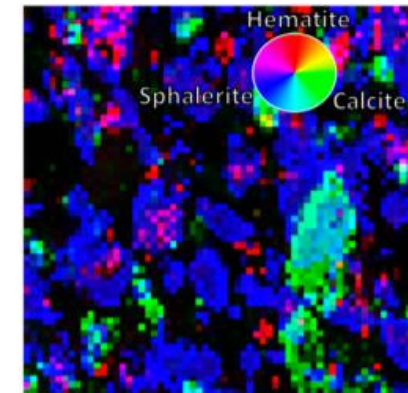
Thorpe et al. *Appl. Radiat. Isotopes* **2019**, *144*, 104-110

Focused chemical and physical imaging

X-ray fluorescence mapping



Microdiffraction studies



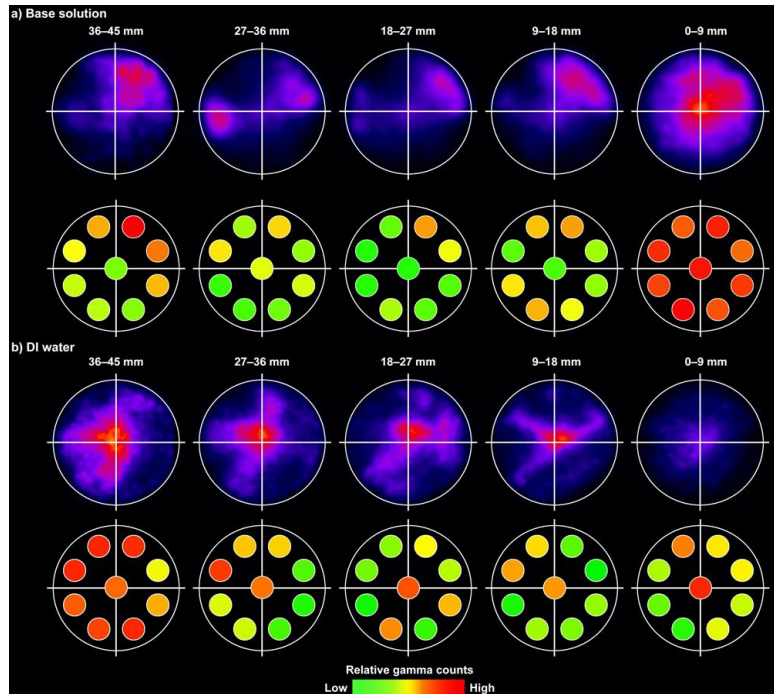
Hamilton et al. *JEQ* **2016**, *45*, 684-692 and Hamilton et al. *ACS Earth Space Chem* **2018**, *2*, 1161-1167.

... as well as some more examples from our lab

Visualizing heterotrophic activity in diesel contaminated soil with PET

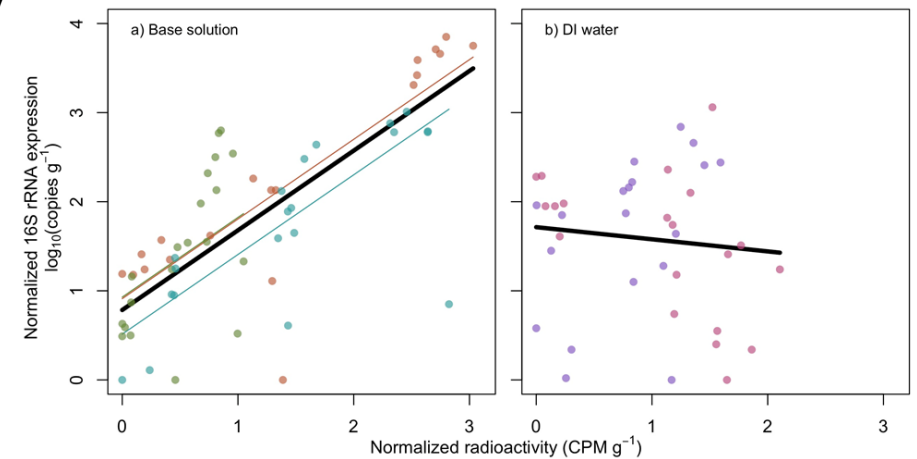
Soil core with biostimulatory solution

Soil core with water



Biostimulatory solution alters distribution of microbial activity locally

Targeted genetic analysis



Bulk measurements in system correspond with higher activity from PET

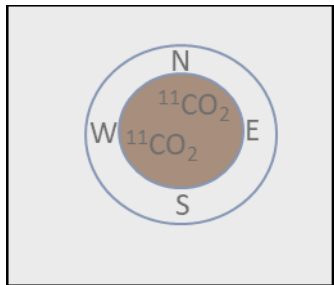
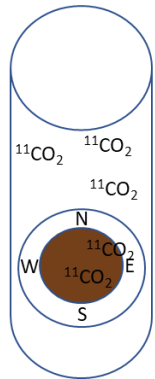
With biostimulatory solutions:

- ↑ Overall microbial activity
- ↑ Nitrogen uptake
- ↑ Hydrocarbon degradation rates



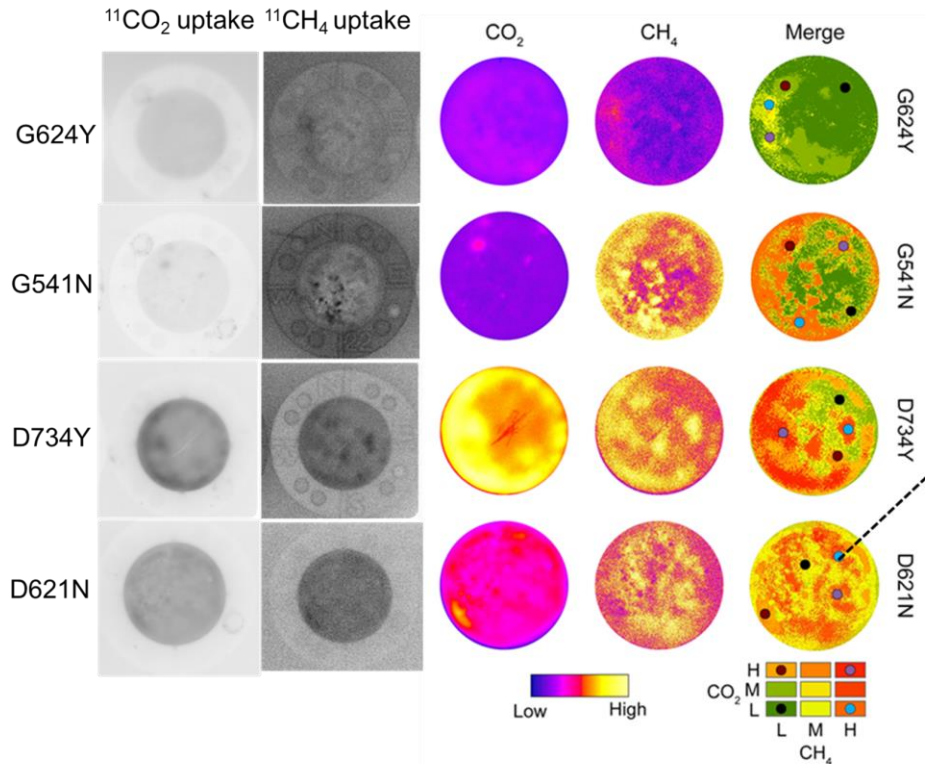
Radiographic imaging of methanotrophs and autotrophs in soils

Soil dosing and imaging

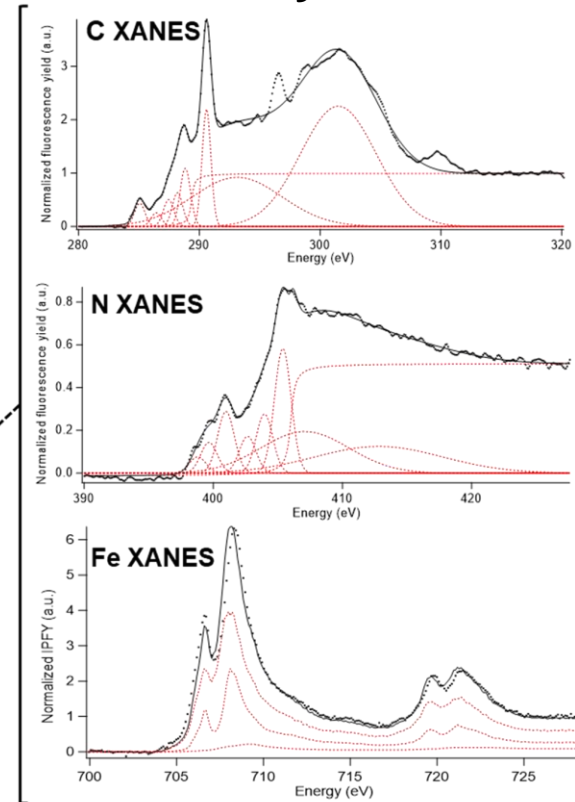


← Imaging film

Images of radiotracer uptake



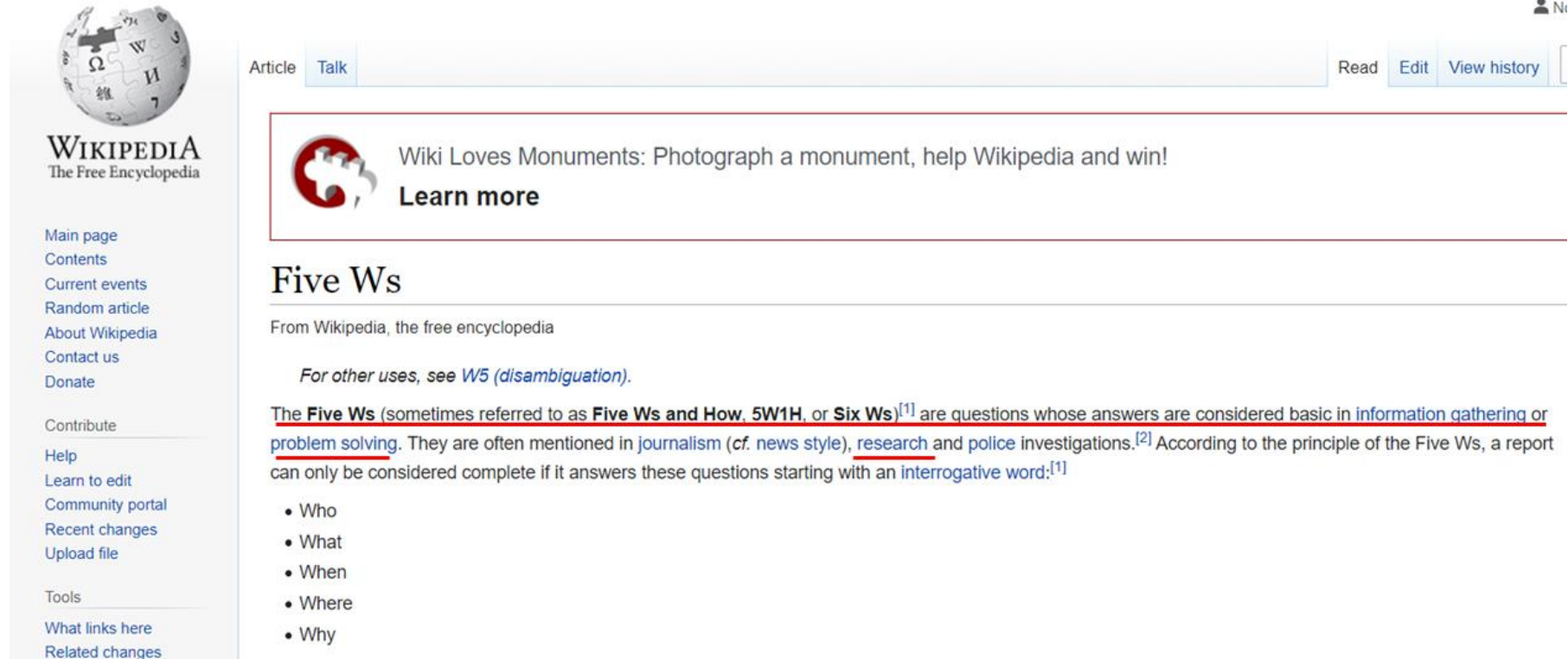
Targeted biogeochemical analysis



Regions of methanotrophic activity have greater proportion of inorganic N (nutrient controlled)

Regions of autotrophy associated with greater presence of carbonate minerals and shifts in Fe mineralogy (mineralogy controlled)

Summary of positron imaging applications in research

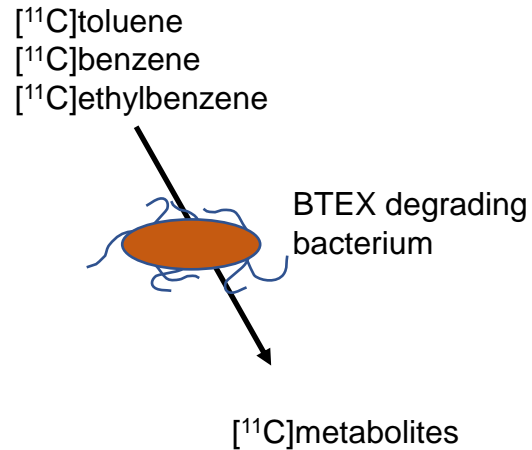


The screenshot shows the Wikipedia article for "Five Ws". At the top, there is a navigation bar with "Article" and "Talk" tabs, and buttons for "Read", "Edit", and "View history". Below this is a banner for "Wiki Loves Monuments: Photograph a monument, help Wikipedia and win!" with a "Learn more" link. The article title "Five Ws" is prominently displayed. Below the title, it says "From Wikipedia, the free encyclopedia" and "For other uses, see *W5 (disambiguation)*". The main text states: "The **Five Ws** (sometimes referred to as **Five Ws and How**, **5W1H**, or **Six Ws**)^[1] are questions whose answers are considered basic in information gathering or problem solving. They are often mentioned in journalism (cf. news style), research and police investigations.^[2] According to the principle of the Five Ws, a report can only be considered complete if it answers these questions starting with an interrogative word.^[1]" Below the text is a bulleted list: "• Who", "• What", "• When", "• Where", and "• Why". On the left side of the page, there is a sidebar with the Wikipedia logo and various navigation links such as "Main page", "Contents", "Current events", "Random article", "About Wikipedia", "Contact us", "Donate", "Contribute", "Help", "Learn to edit", "Community portal", "Recent changes", "Upload file", "Tools", "What links here", and "Related changes".

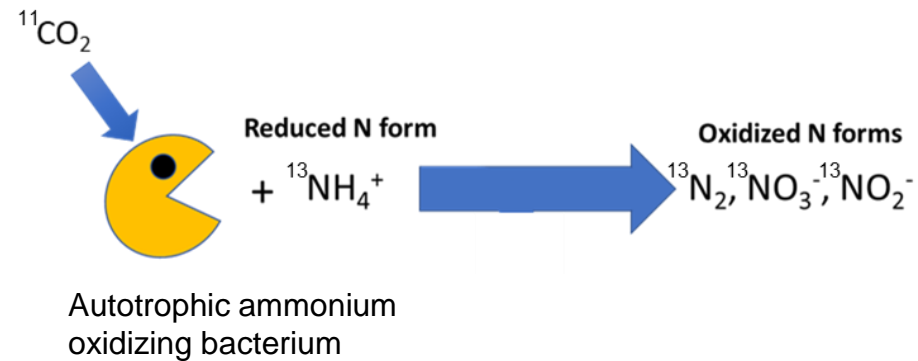
- Positron emitting radiotracers can follow molecule-specific activities (**what is happening?**)
- Positron imaging allows for time-resolved visualization of activity (**where did it happen?, when did it happen?**)
- Subsequent genetic analysis has potential to characterize communities (**who is there?**)
- Local biogeochemical speciation may help understand underlying factors (**why did it happen?**)

Future applications of positron imaging in bioremediation processes

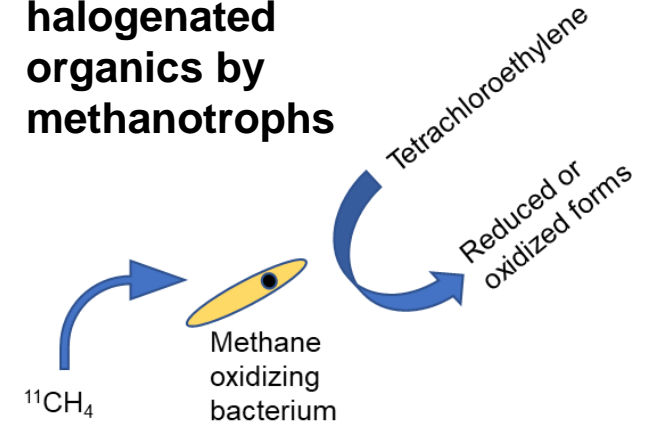
Remediation of BTEX compounds



Remediation of ammonium in soils



Remediation of halogenated organics by methanotrophs



Varying tracer chemistry can open up spatial visualization of many biodegradation/transformation processes (modify the “what”)



Acknowledgments

- Peak and Siciliano lab groups at U of S
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