BERKELEY LAB

LAWRENCE BERKELEY NATIONAL LABORATORY

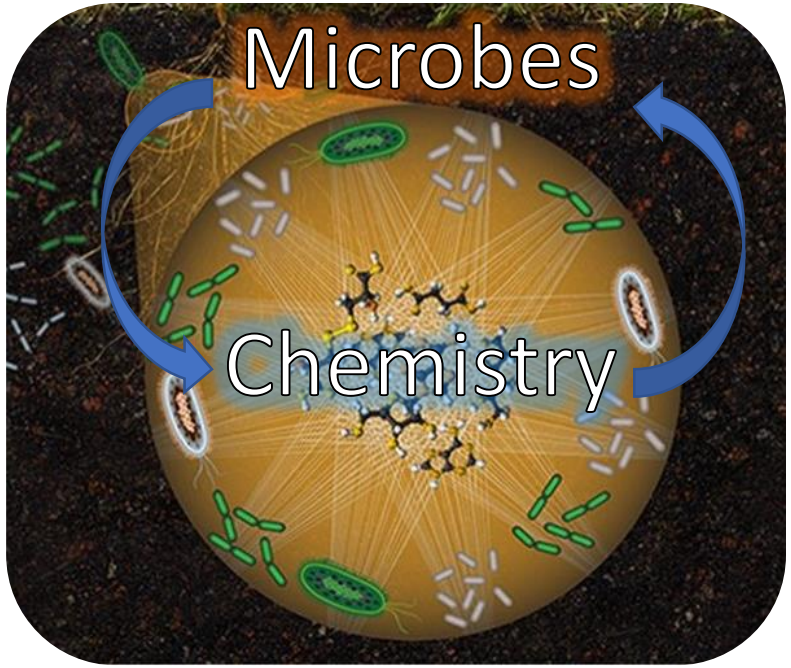
U.S. DEPARTMENT OF ENERGY

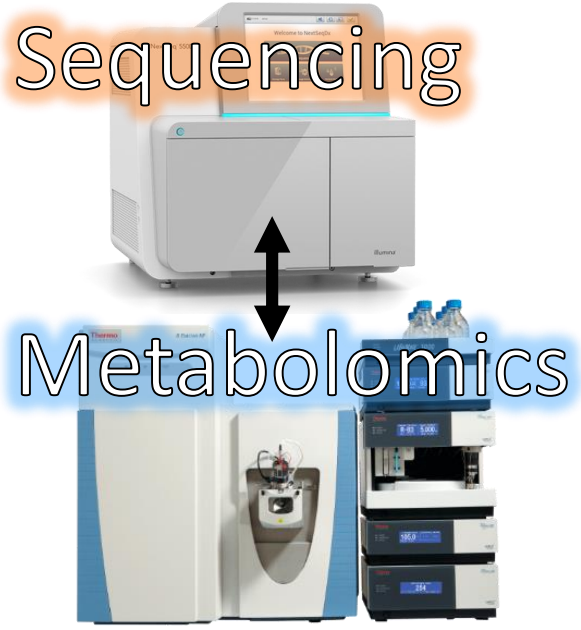
Trent Northen, Berkeley Lab

BER Advisory Committee meeting
October 25th, 2019 Bethesda MD

Deconstructing the metabolic webs of microbiomes within biological soil crusts

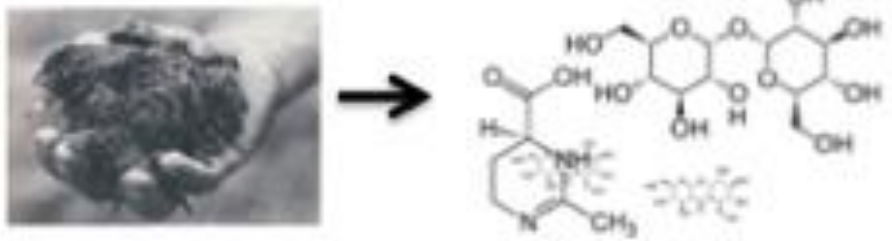
Overall Goal: This project will develop metabolomics approaches to link soil biochemical activity with microbial phylogeny and metabolic potential, complementing sequencing efforts to inform the development of next generation biogeochemical and climate models.



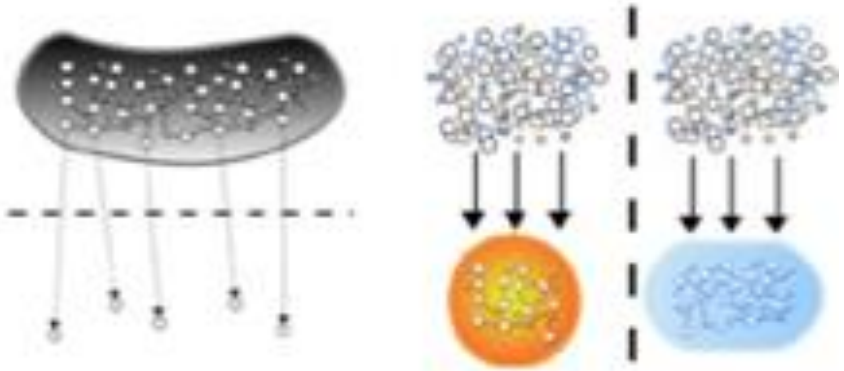


Specific Aims

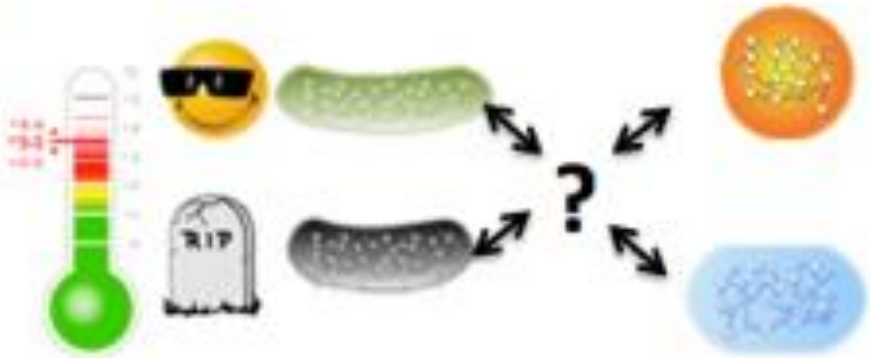
A1—Development of soil metabolomics approaches



A2—Metabolomics to identify bacterial uptake and release of soil metabolites



A3—Test approach to predict community responses across an environmental gradient



Too much to cover in 30min

1. Vandehey, Nicholas et al **Environmental Science & Technology Letters** (2014), 1 (10), 393-398. DOI: 10.1021/ez500209c
2. Nunes da Rocha et al. **Frontiers in Microbiology** (2015), 6:277. DOI: 10.3389/fmicb.2015.00277
3. Swenson, T**.** et al. **Soil Biology and Biochemistry** (2015), 80, 189-198. DOI: 10.1016/j.soilbio.2014.10.007
4. Baran R. et al. **Nature Communications** (2015), 6 (6): 8289. DOI: 10.1038/ncomms9289
5. Swenson, T**.** et al. **Soil Biology and Biochemistry** (2015), DOI: 10.1016/j.soilbio.2015.07.022
6. Swenson, T**.** et al. **Soil Biology and Biochemistry** (2015) DOI: 10.1016/j.soilbio.2014.10.007
7. Couradeau, E et al. **Nature Communications** (2016), Jan, 7:10373. DOI: 10.1038/ncomms10373
8. Baran R. **ACS Chem. Bio.** (2017) DOI: 10.1021/acschembio.6b00890
9. Swenson T. et al**. Nature Communications** (2018) 9:19. DOI: 10.1038/s41467-017-02356-9.
10. Swenson T. et al. Plant and Soil (2017) Karaoz U. **mBio** (2018) 9:e01366-16. doi.org/10.1128/mBio.01366-16.
11. Jose N. et al. **Biogeosciences,** 15, 2219-2229, 2018. doi.org/10.5194/bg-15-2219-2018
12. Couradeau E.et al. **Frontiers in Environmental Sciences**. 2018 doi.org/10.3389/fenvs.2018.00065.
13. Kosina, S. et al. **BMC Microbiology**. 2018 doi: 10.1186/s12866-018-1256-y.
14. Porcar, M et al. **Frontiers in Microbiology**. doi.org/10.3389/fmicb.2018.03043
15. Roux, S.et al. **Peer J** (2019) e6902. doi: 10.7717/peerj.6902
16. Morton, J. et al. **Nature Methods** (In Press).
17. Van Goethem M,et al. **mBio** (In Press)

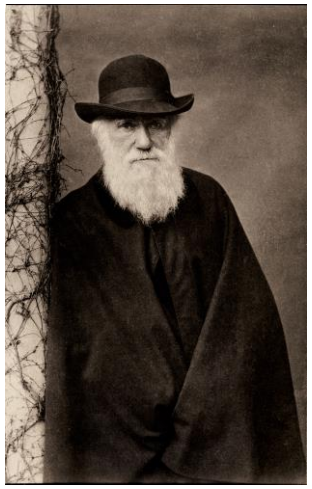


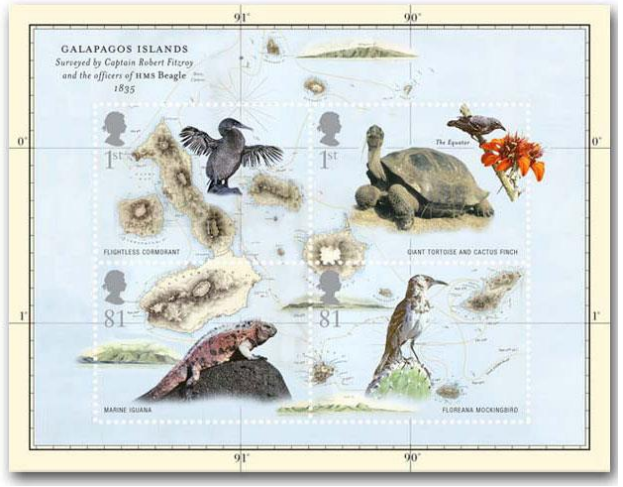




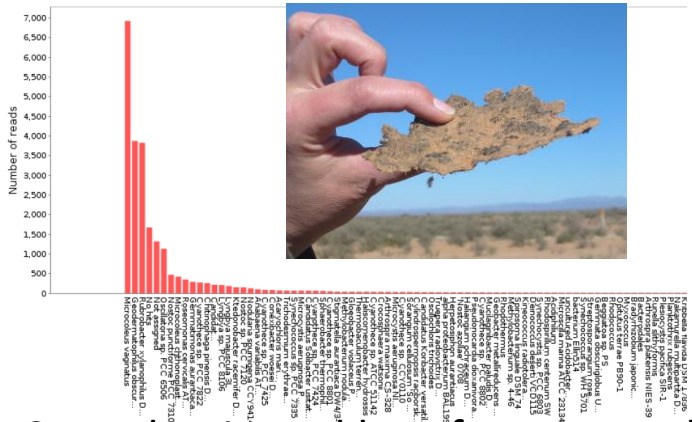
Choosing the soil community

A gram of soil likely has 1000s-millions of microbial taxa

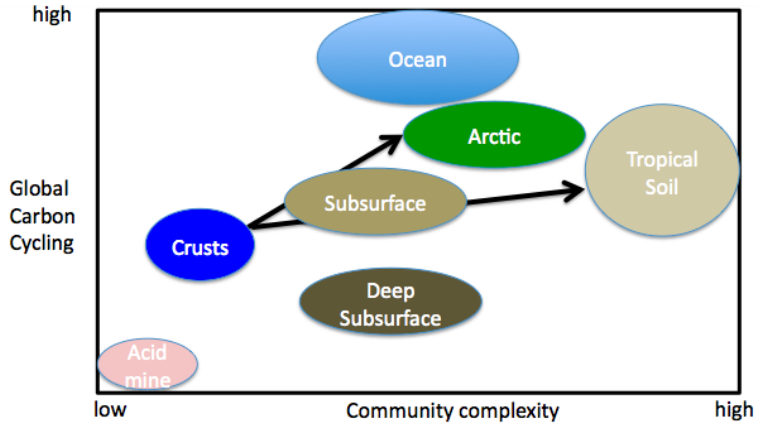




Importance of model systems that can be extended to more complex systems



Crust dominated by a few taxa and can be probed as an intact soil community



Extend understanding/approaches to more complex systems

Biocrusts fix carbon and stabilizes soil in arid lands

Arid Lands: 40% of Earth's terrestrial surface

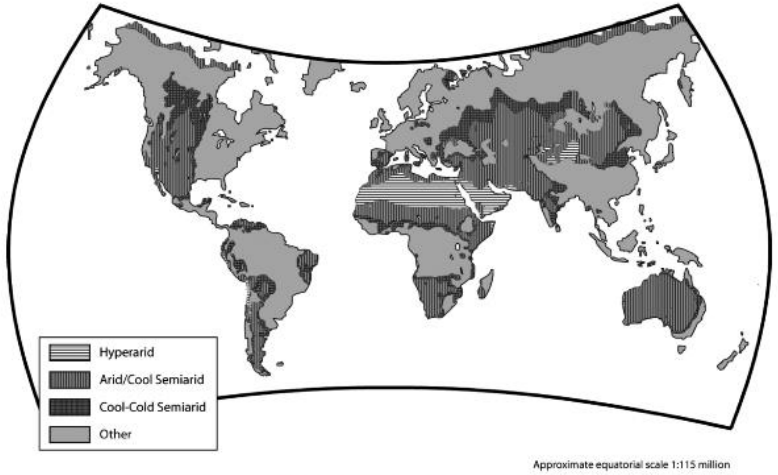
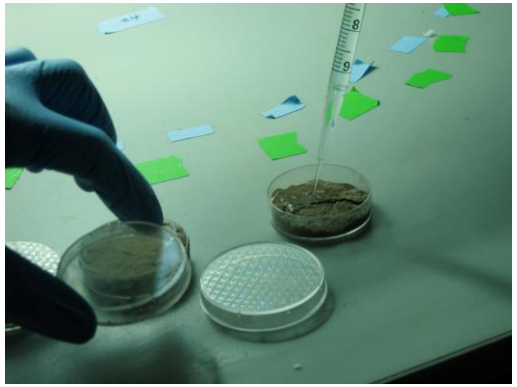


Figure 1. Patterned areas represent regions where biological soil crusts are likely to influence local hydrologic processes. Cumulatively, these lands represent 40% of the Earth's terrestrial surface

Biocrusts estimated to account for 7% of CO_2 fixed by terrestrial ecosystems

Sensitive to physical disturbance

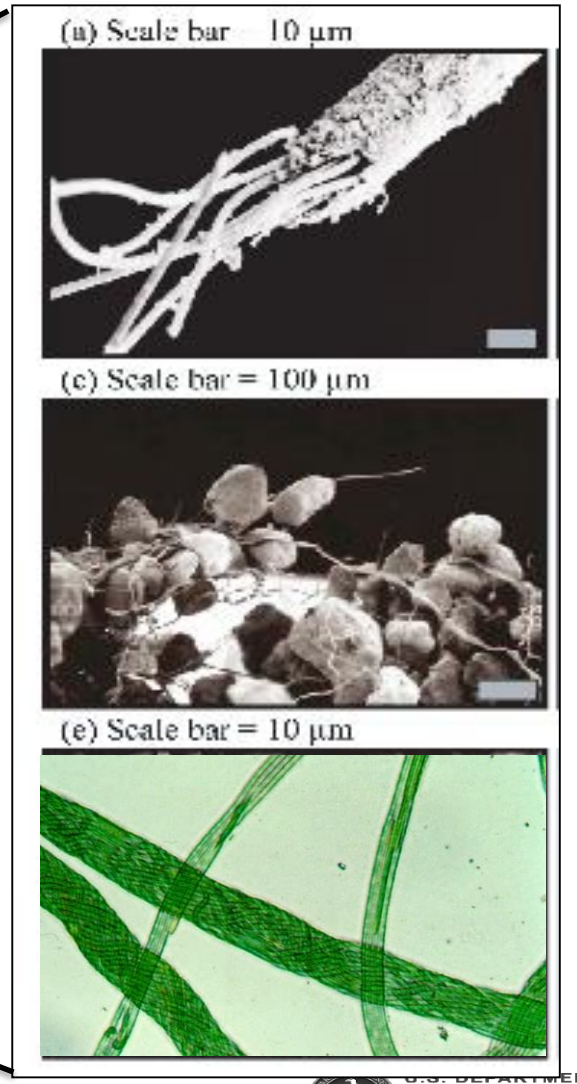
Experimentally tractable



Large-scale Biocrust inoculation

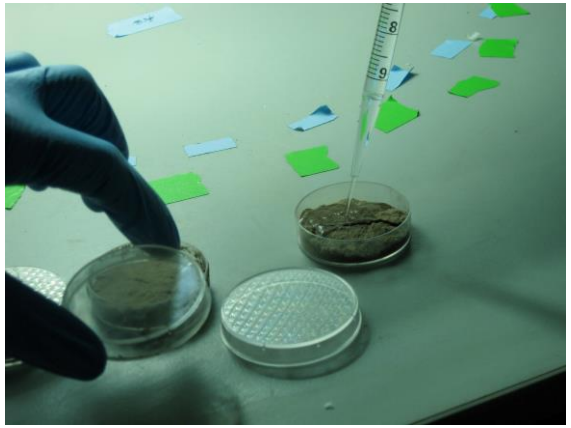
Biocrusts help prevent erosion

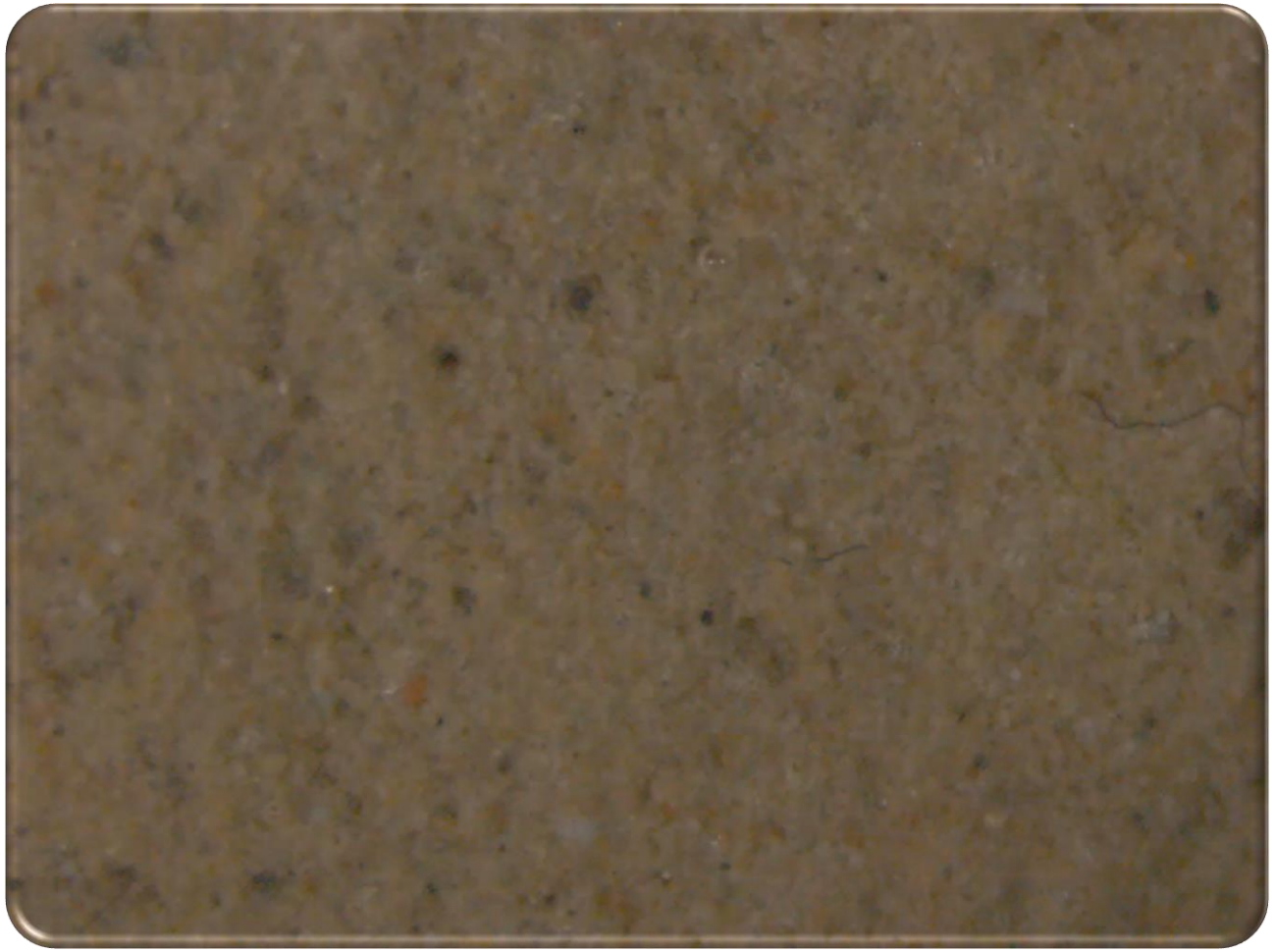
Microcoleus sp. Filamentous Cyanobacteria that fix atmospheric carbon dioxide and bind the soil forming these biocrusts



Exometabolite webs when biocrusts resuscitate from dormancy

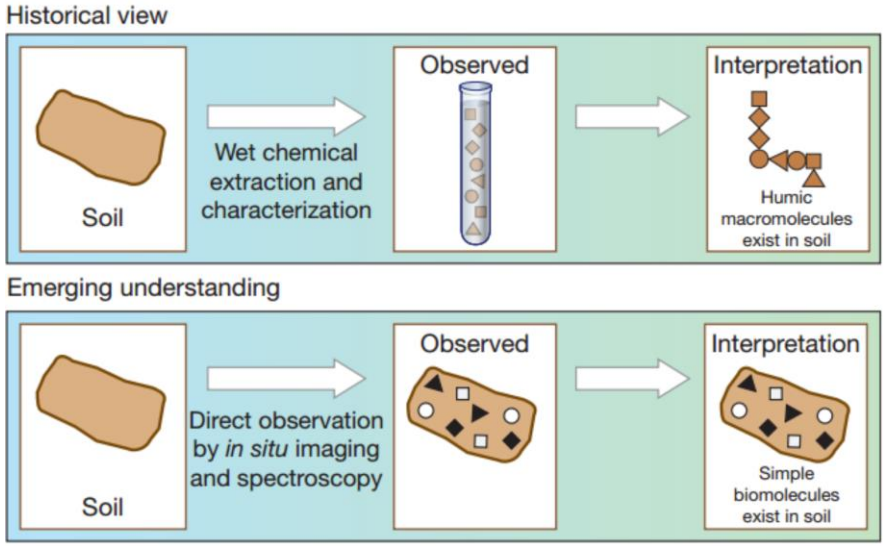
Laboratory Wetting Event



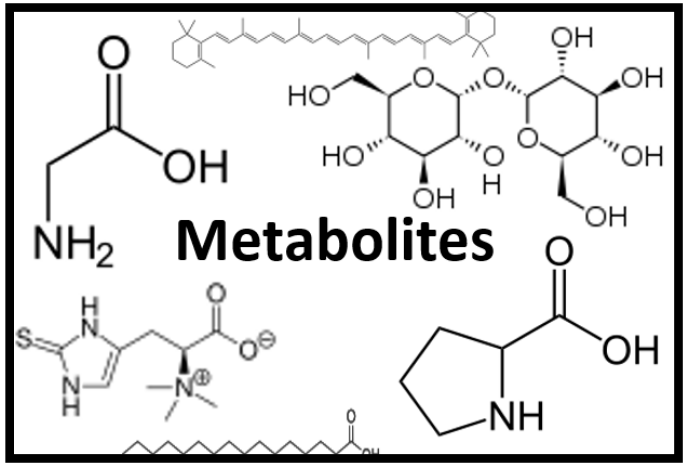


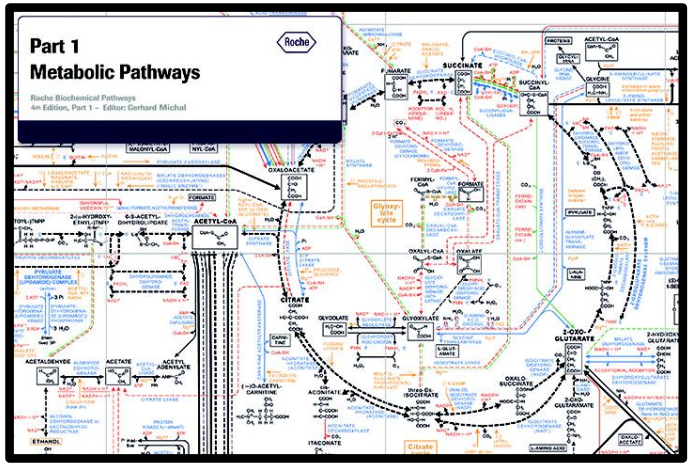
What is soil carbon?

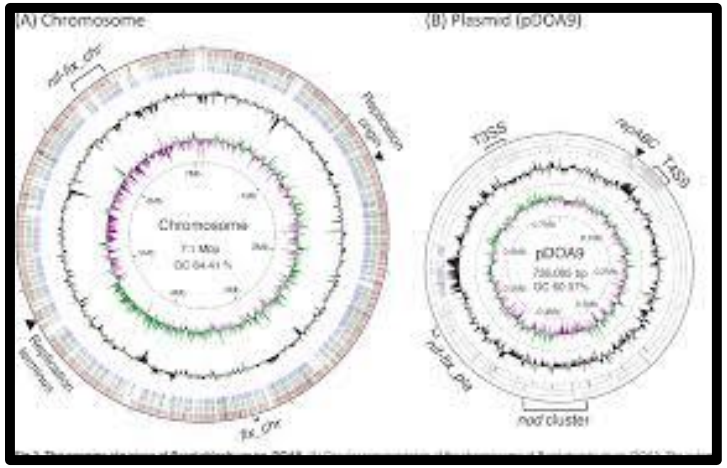
Emerging view: Microbial Metabolites



Schmidt, Torn, Nature 2011

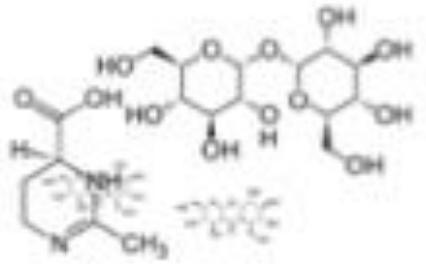


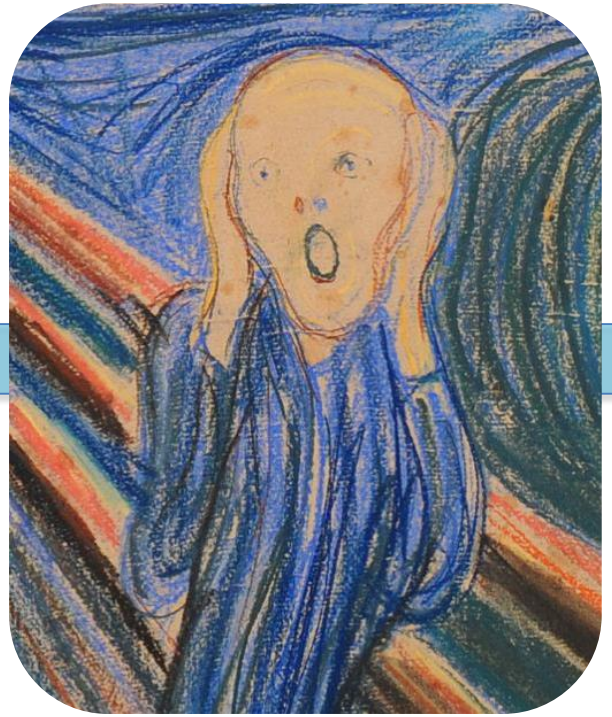




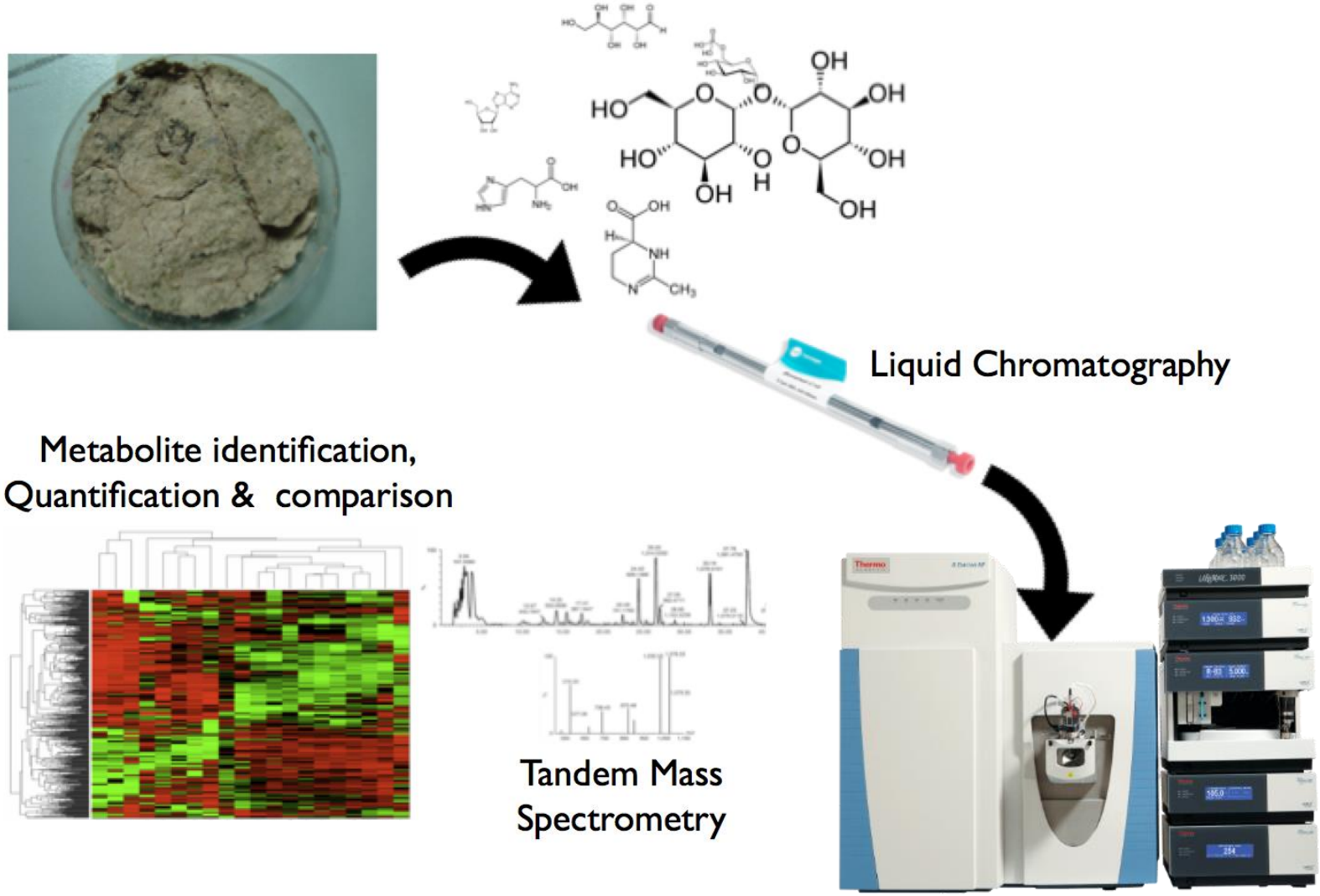
Application of metabolomics to soils?

A1—Development of soil metabolomics approaches



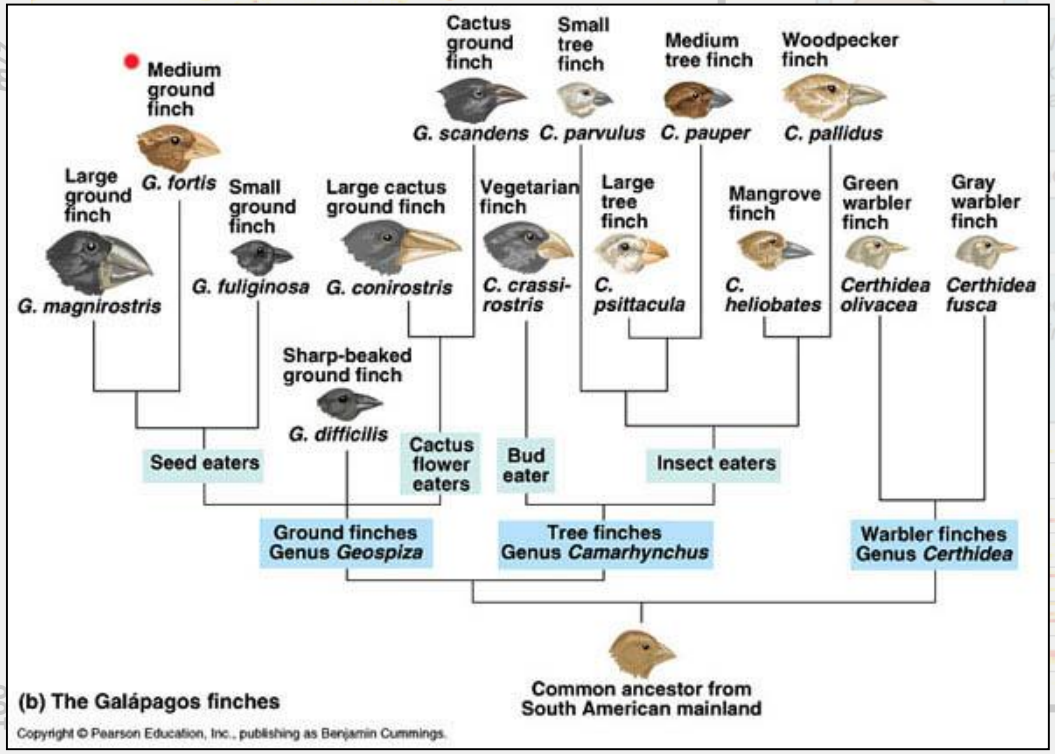


Developed and published the first LC/MS soil metabolomics methods



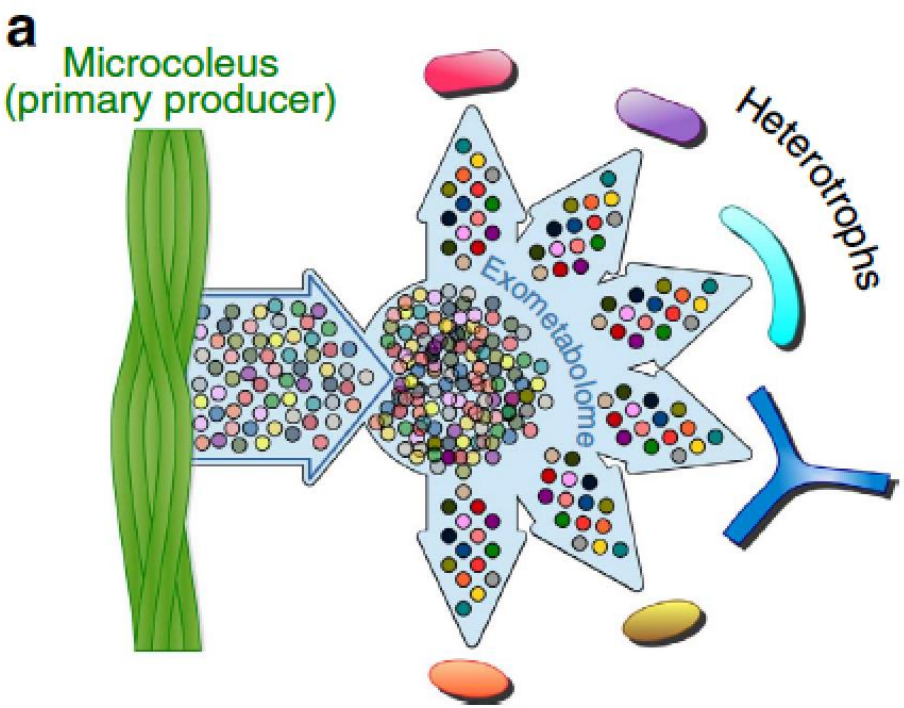
Swenson et al, Untargeted Soil Metabolomics Methods for Analysis of Extractable Organic Matter. **Soil Bio. Biochem**. 2015

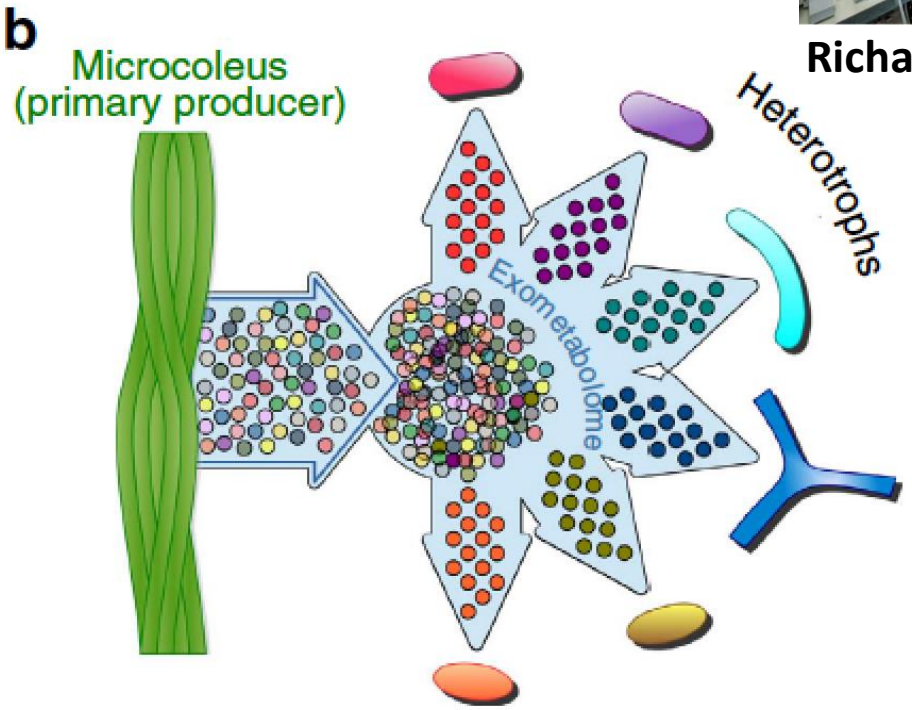
What is the connection between microbial diversity and resource diversity (diversity in soil carbon)?



Biocrust exometabolite webs of microbes

Richard Baran





They all eat everything:

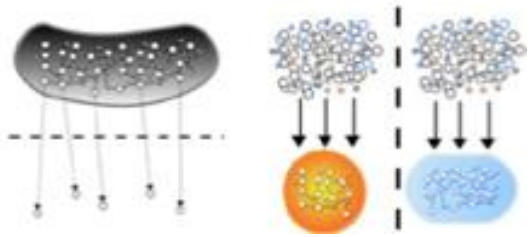
Metabolites do not support diversity

They target specific metabolites:

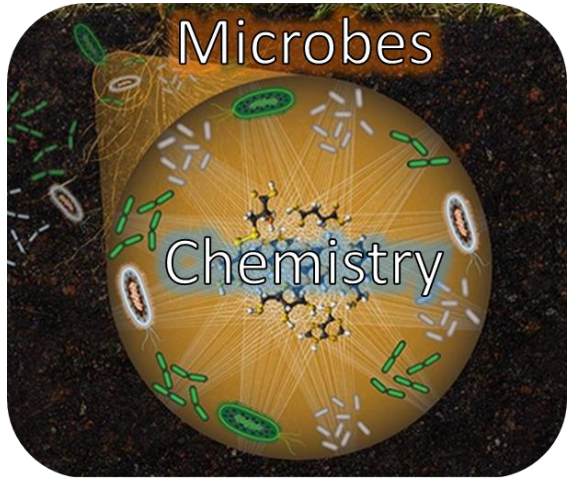
Exometabolite niche partitioning

Developed exometabolomics approaches

A2—Metabolomics to identify bacterial uptake and release of soil metabolites



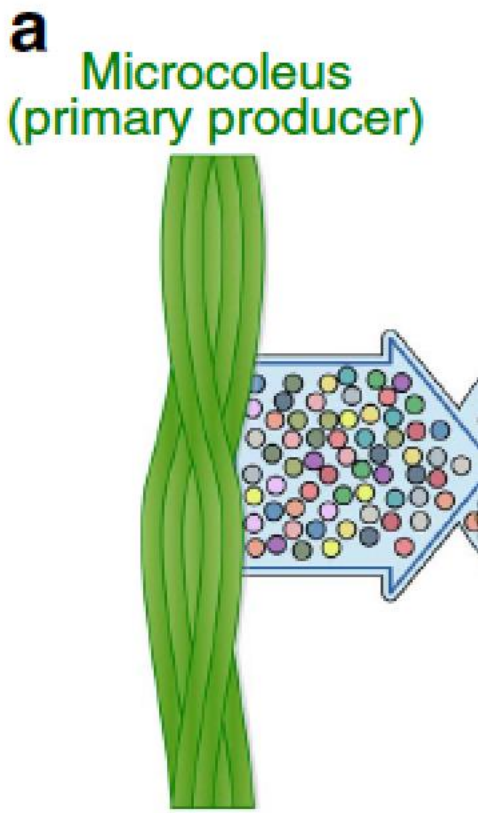
Use LC/MS to measure th changes in metabolites

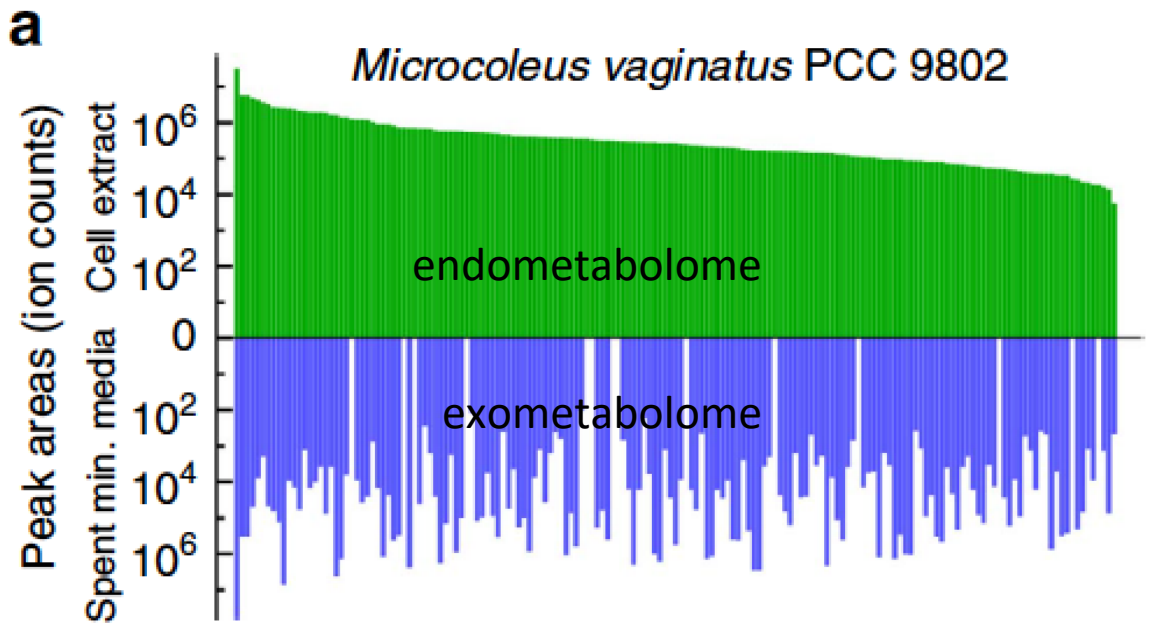


Time 0

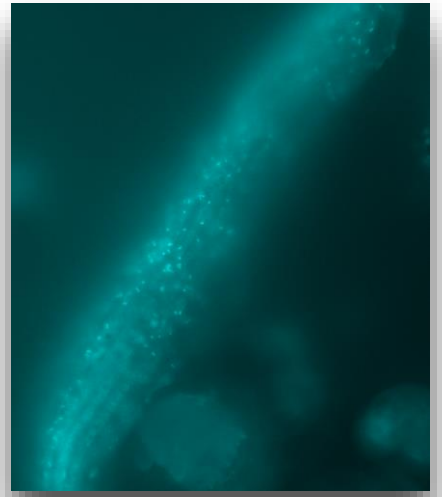
T final

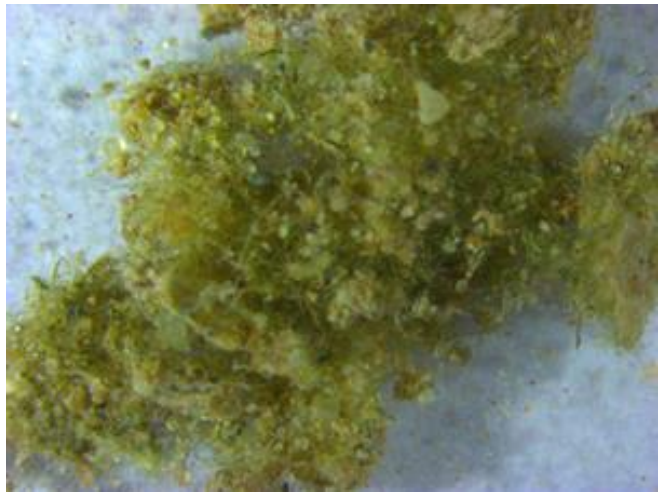
Microcoleus releases a large fraction of its ‘metabolome’

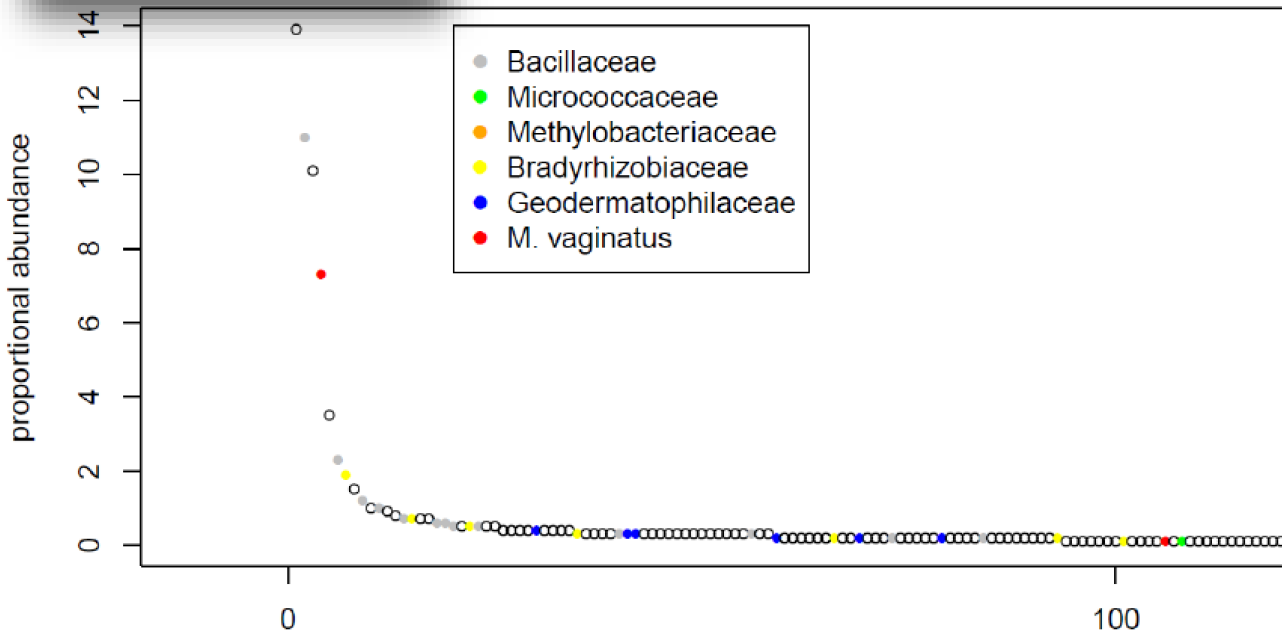


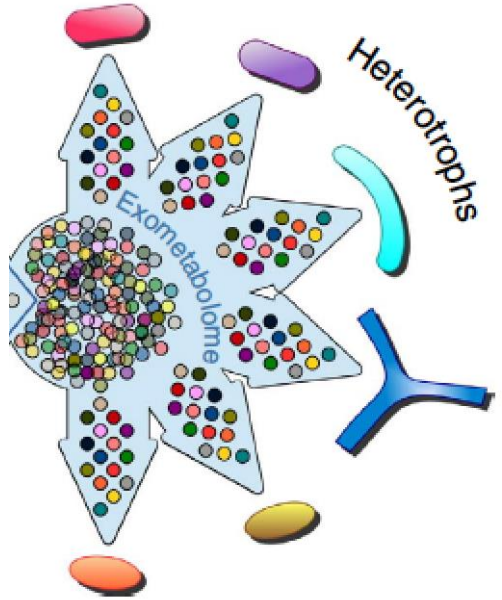


Resource utilization by cyanosphere heterotrophs



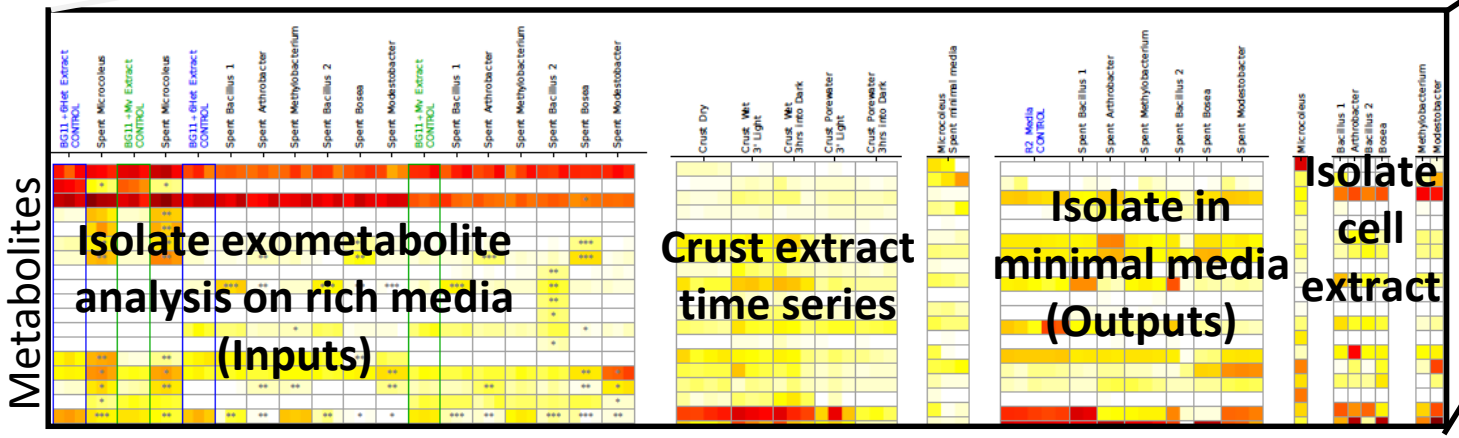


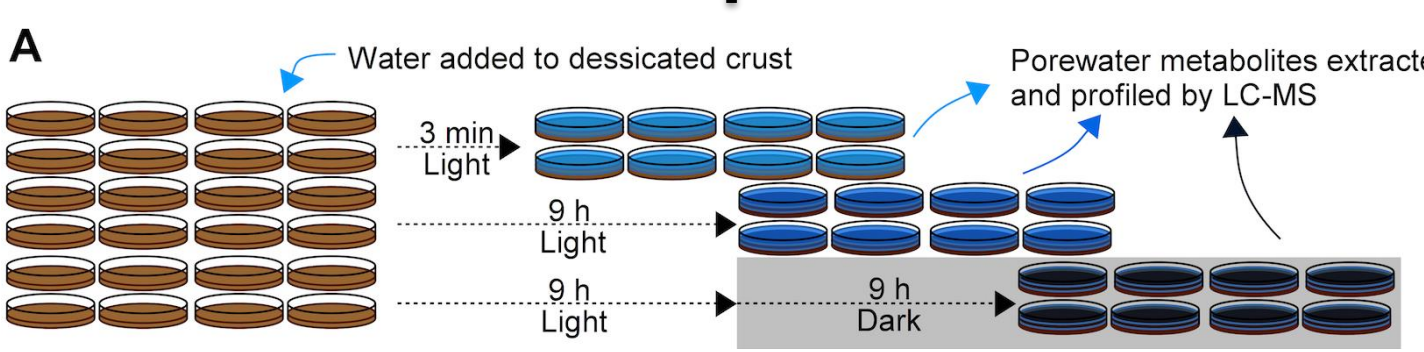


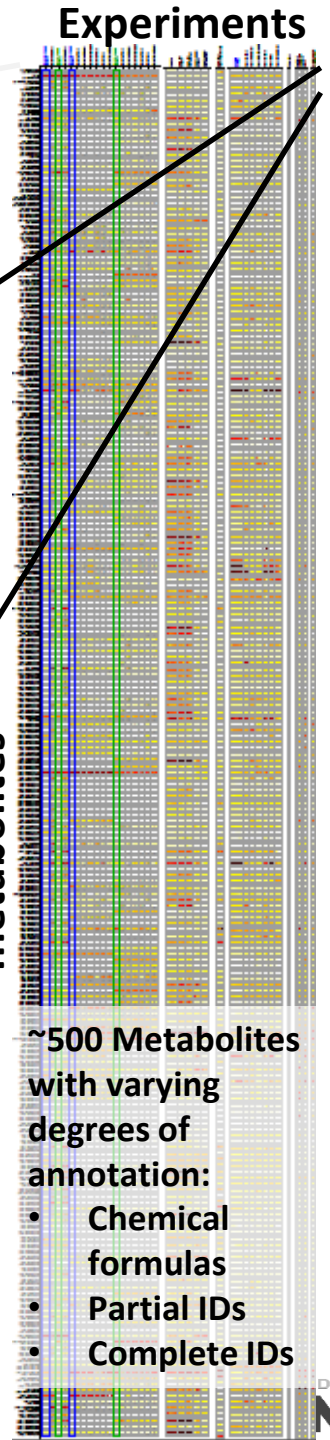


Nunes da Rocha ***Frontiers in Microbiology*** (2015), 6:277.

Comparison of isolate metabolite use with intact biocrusts



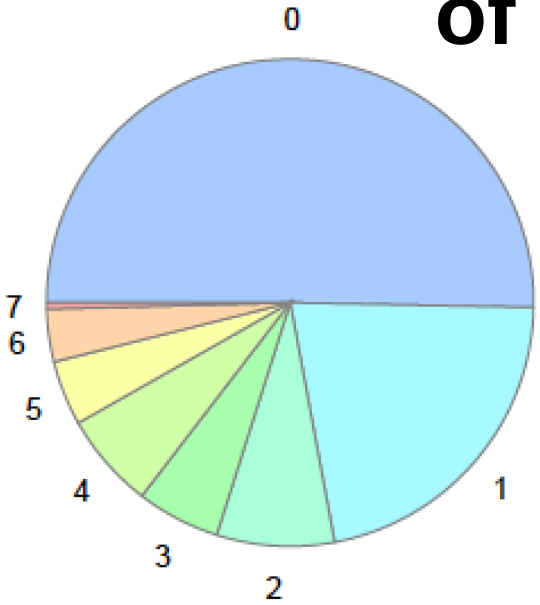


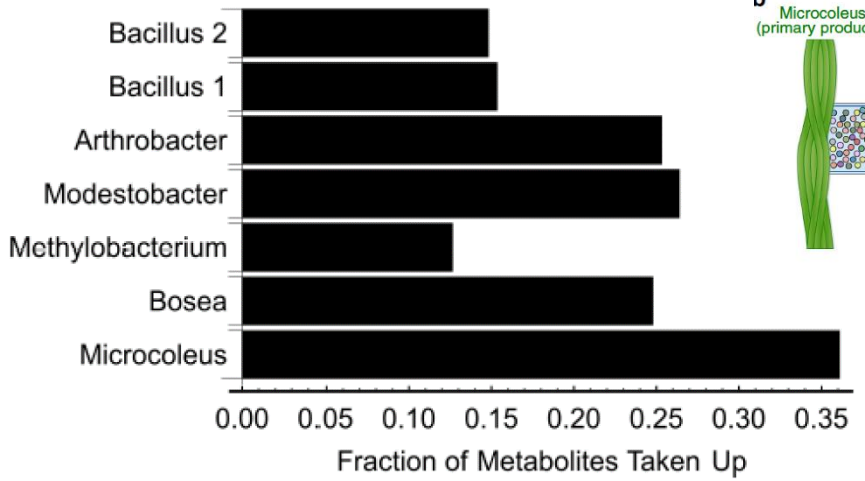


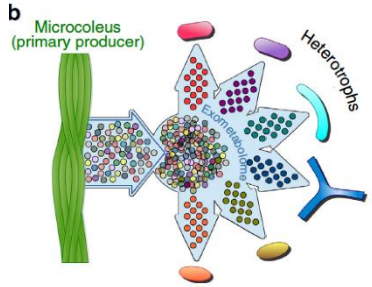
~500 Metabolites with varying degrees of annotation:

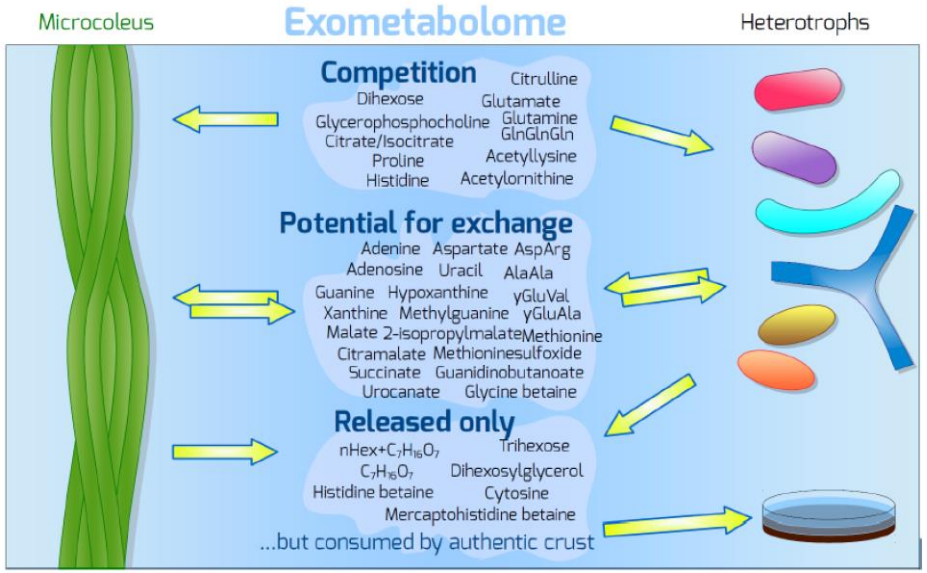
- Chemical formulas
- Partial IDs
- Complete IDs

Isolates show specialization for uptake of specific metabolites



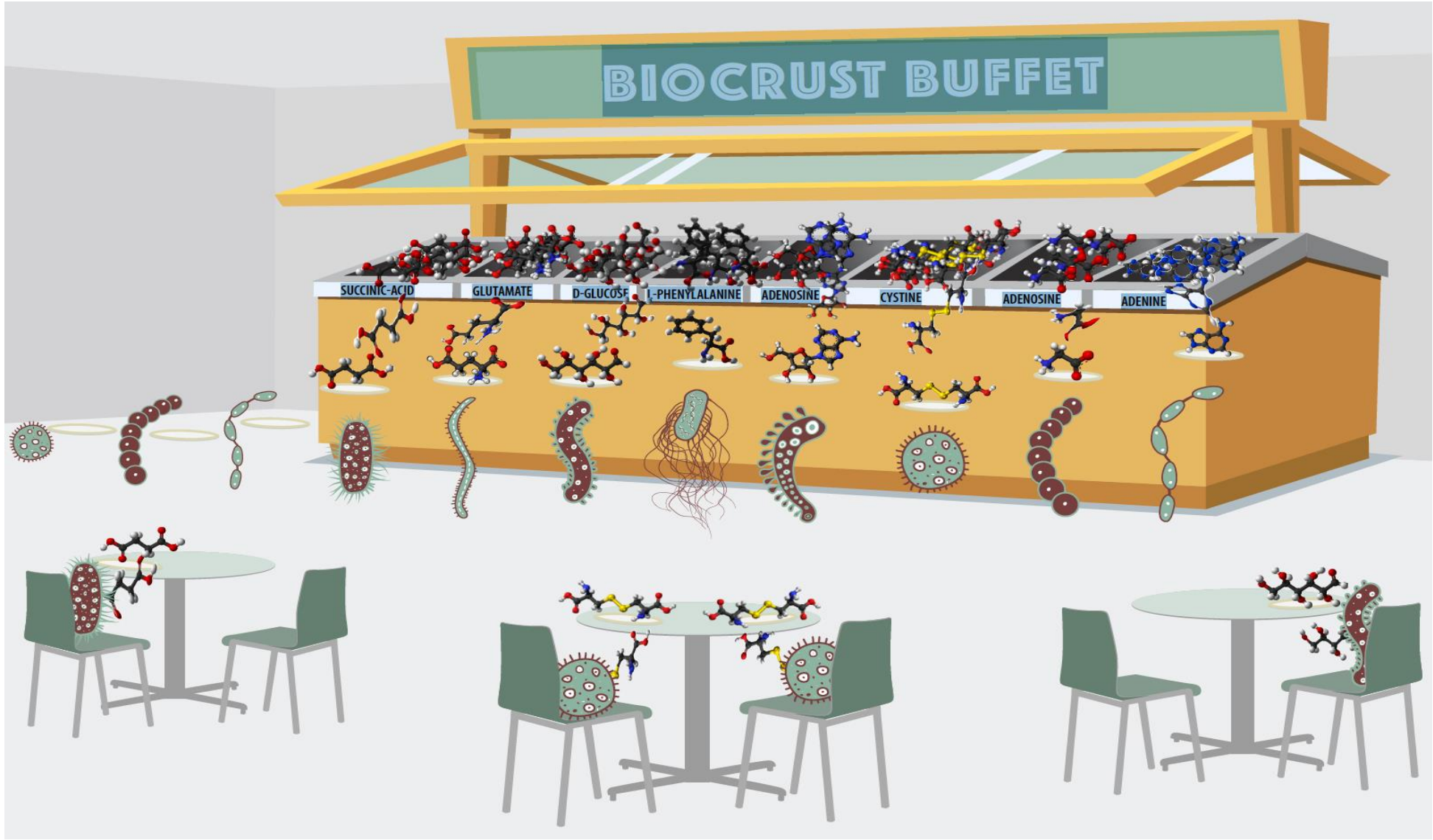




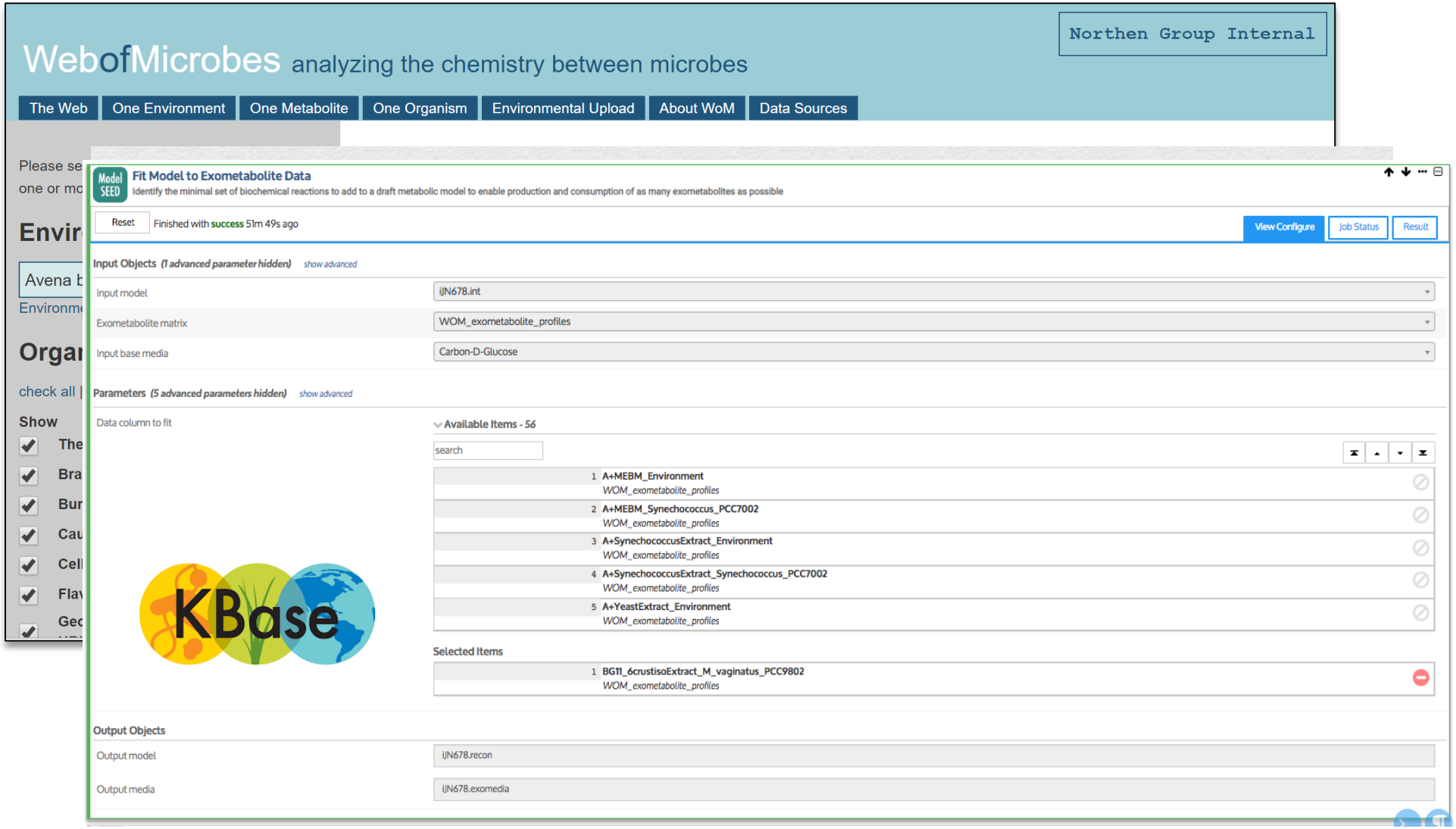


Baran et al ***Nature Communications*** 2015 DOI:10.1038/ncomms9289

Exometabolite niche portioning couples microbial diversity to chemical diversity



Built Webofmicrobes.org to predict interactions

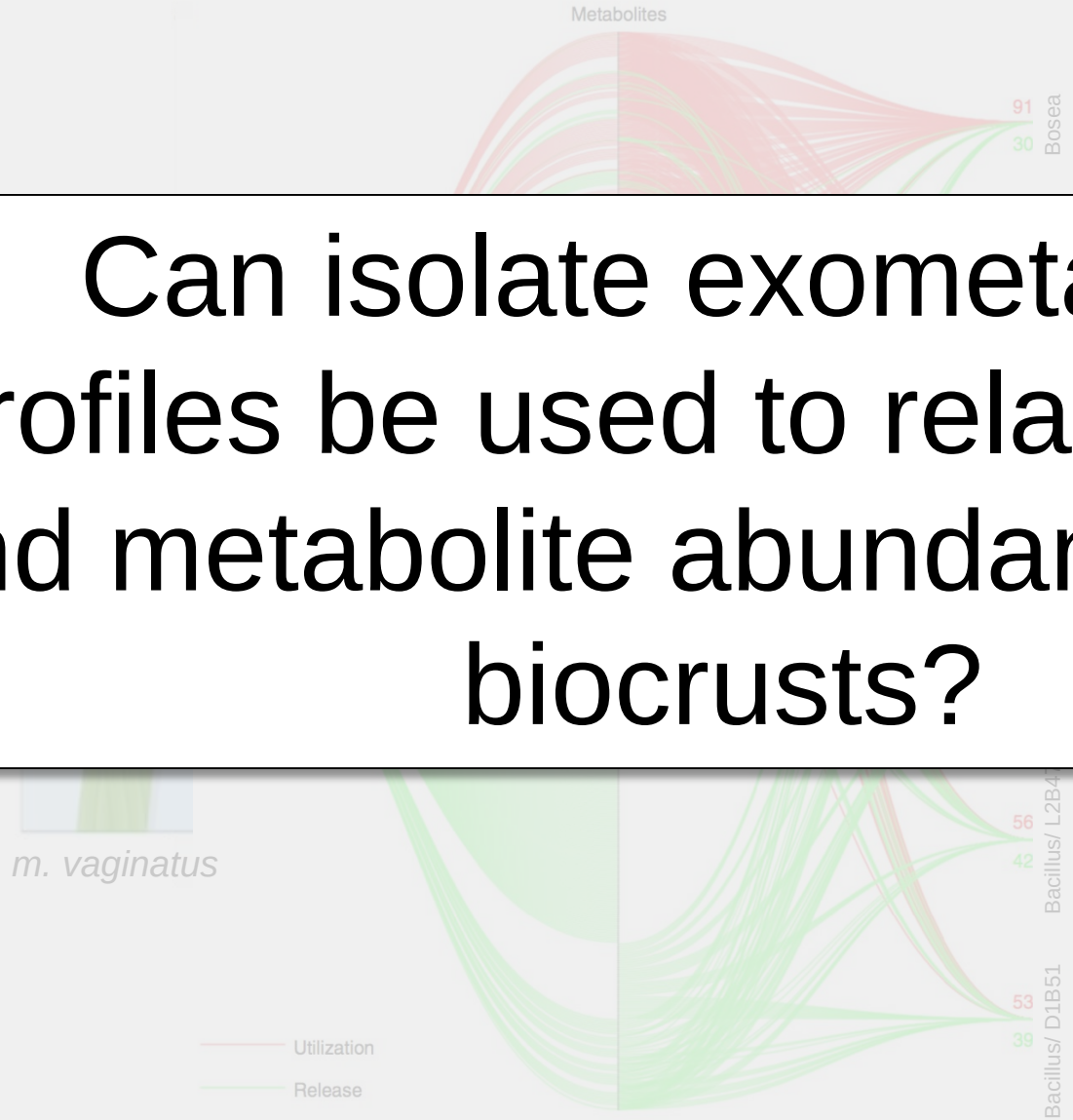


Suzie Kosina (LBNL)

Annette Greiner (NERSC)

Kosina et al **BMC Genomics** 2018

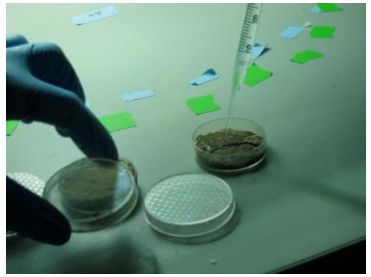
Exometabolite data predicts metabolic webs

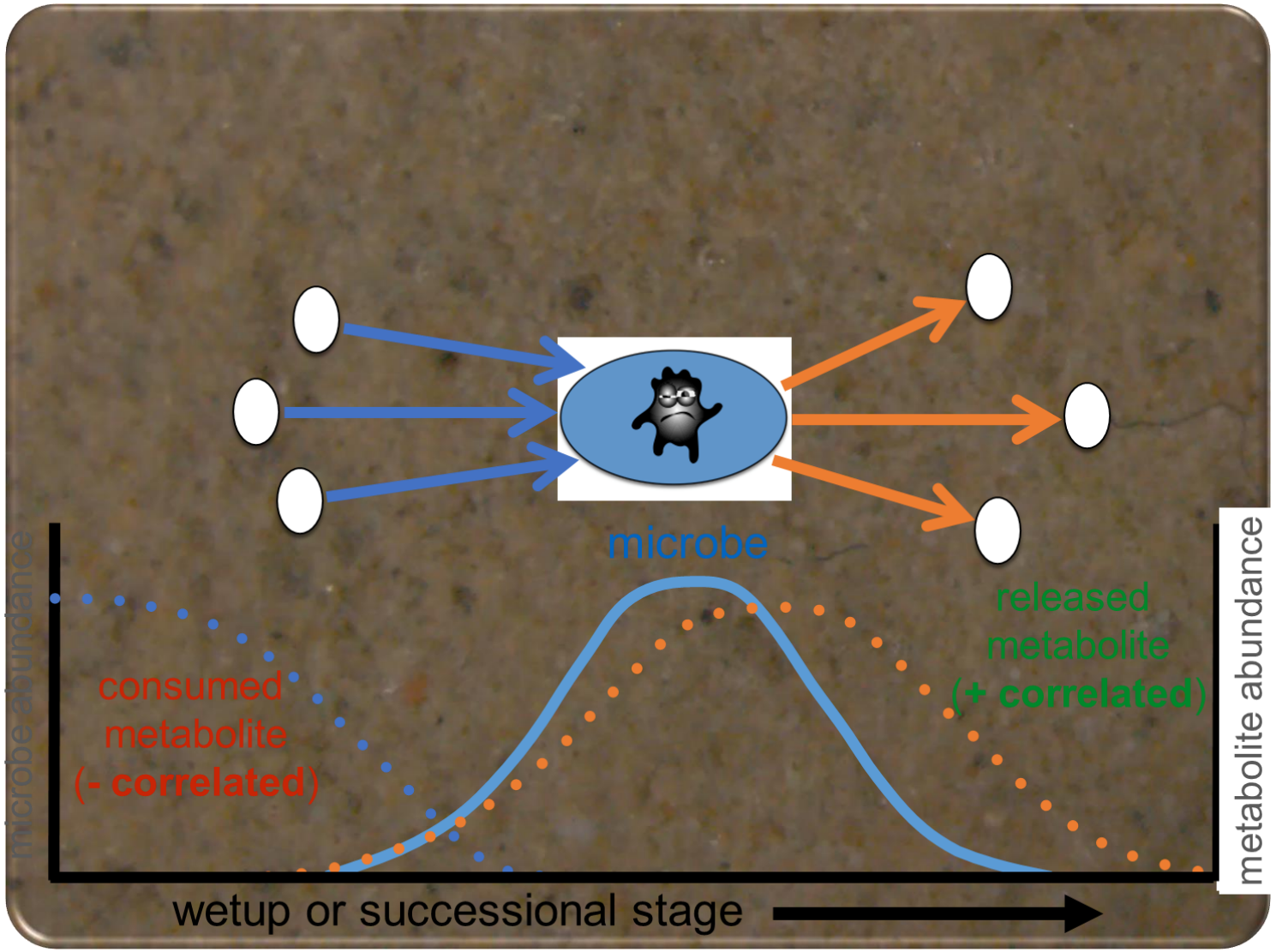


Can isolate exometabolite profiles be used to relate isolate and metabolite abundance within biocrusts?

Can exometabolite profiles link microbe and metabolite abundance *in situ*?

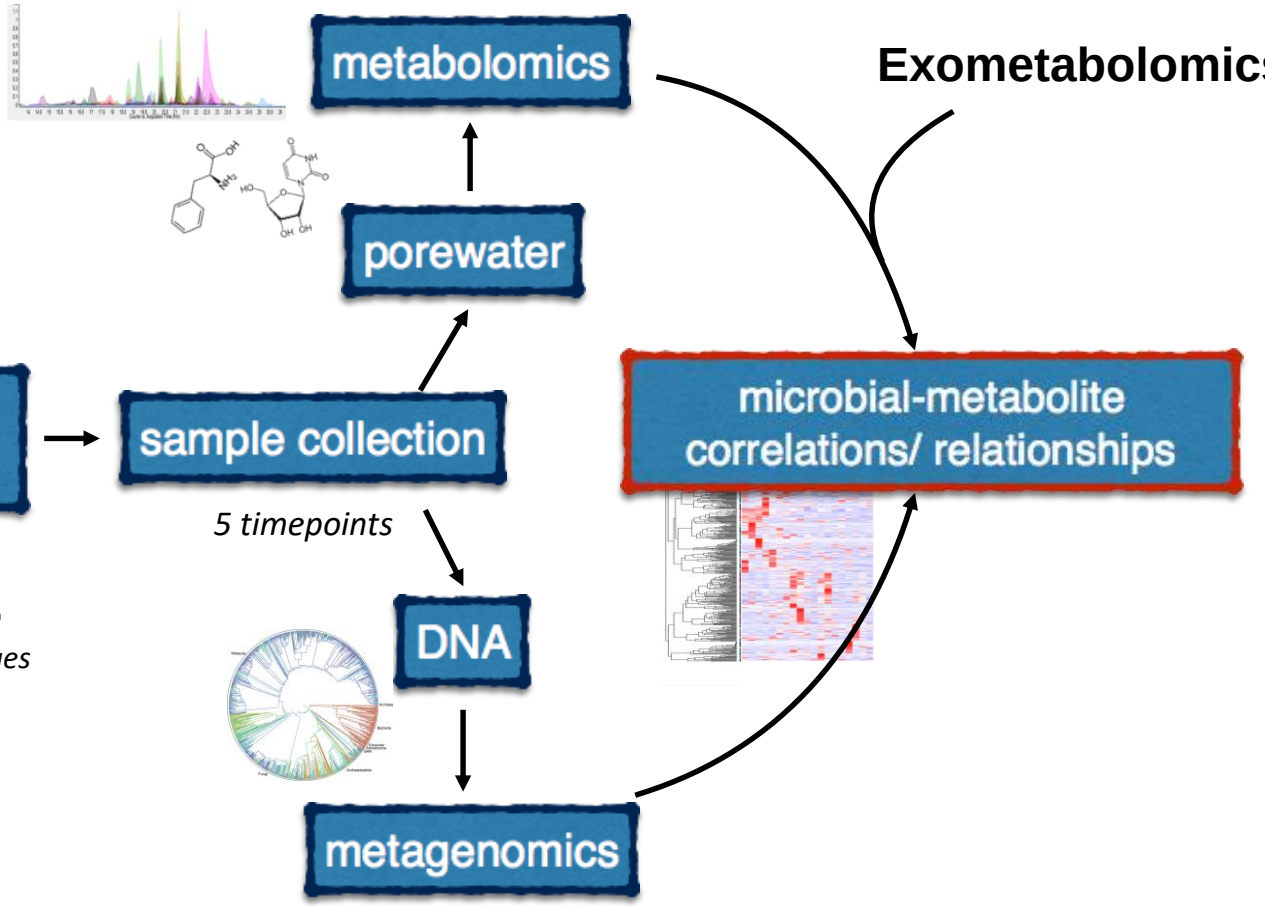
Laboratory Wetting Event





Approach

Tami Swenson

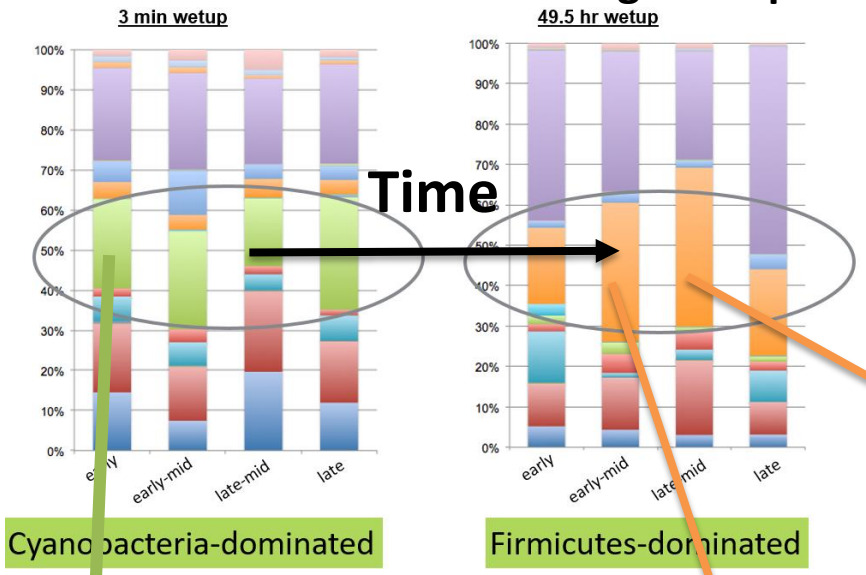


Metagenomics enables examination of the genomics of uncultivated microbes

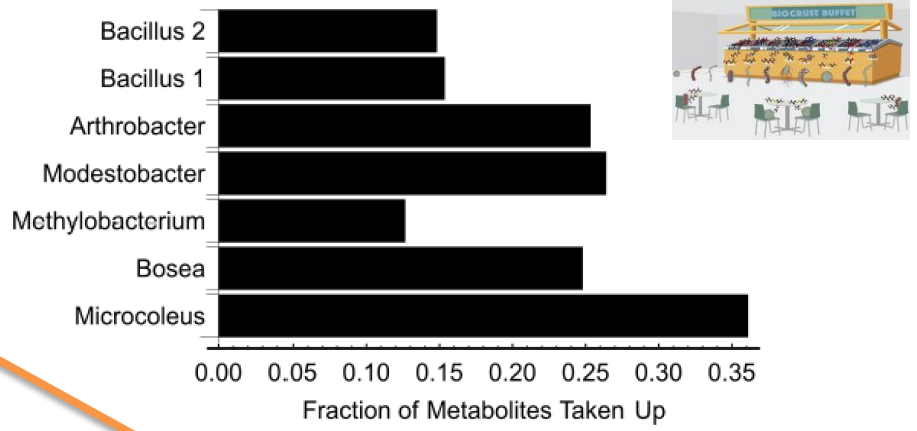
Swenson T. Linking soil biology and chemistry in biological soil crust using isolate exometabolomics. **Nature Communications** (2018) 9:19. DOI: 10.1038/s41467-017-02356-9.

Selection of abundant taxa that exhibit strong dynamics

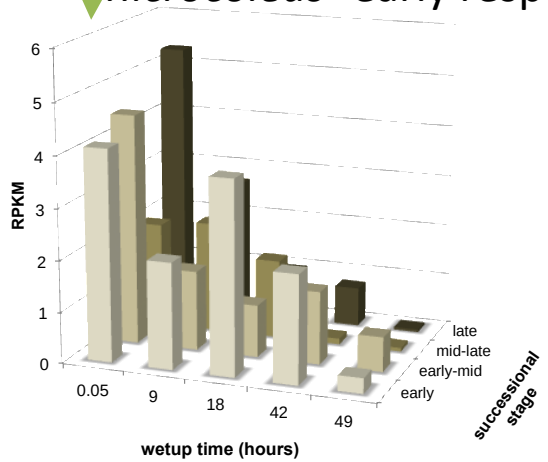
Increase in Firmicutes during wet-up



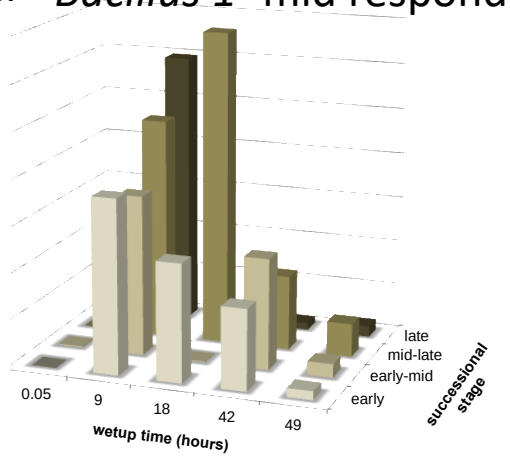
Exometabolomics niche partitioning results



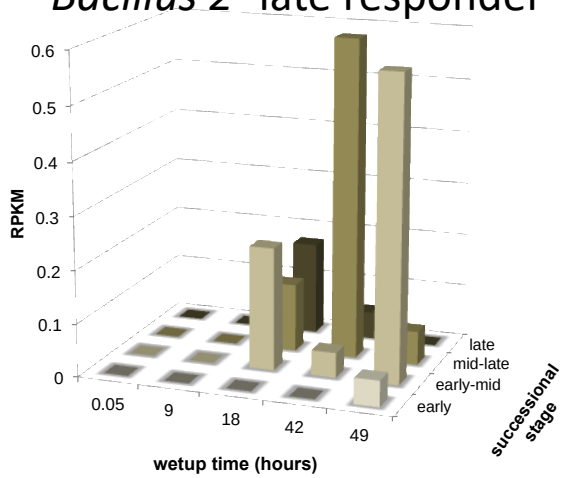
Microcoleus- early responder



Bacillus 1- mid responder

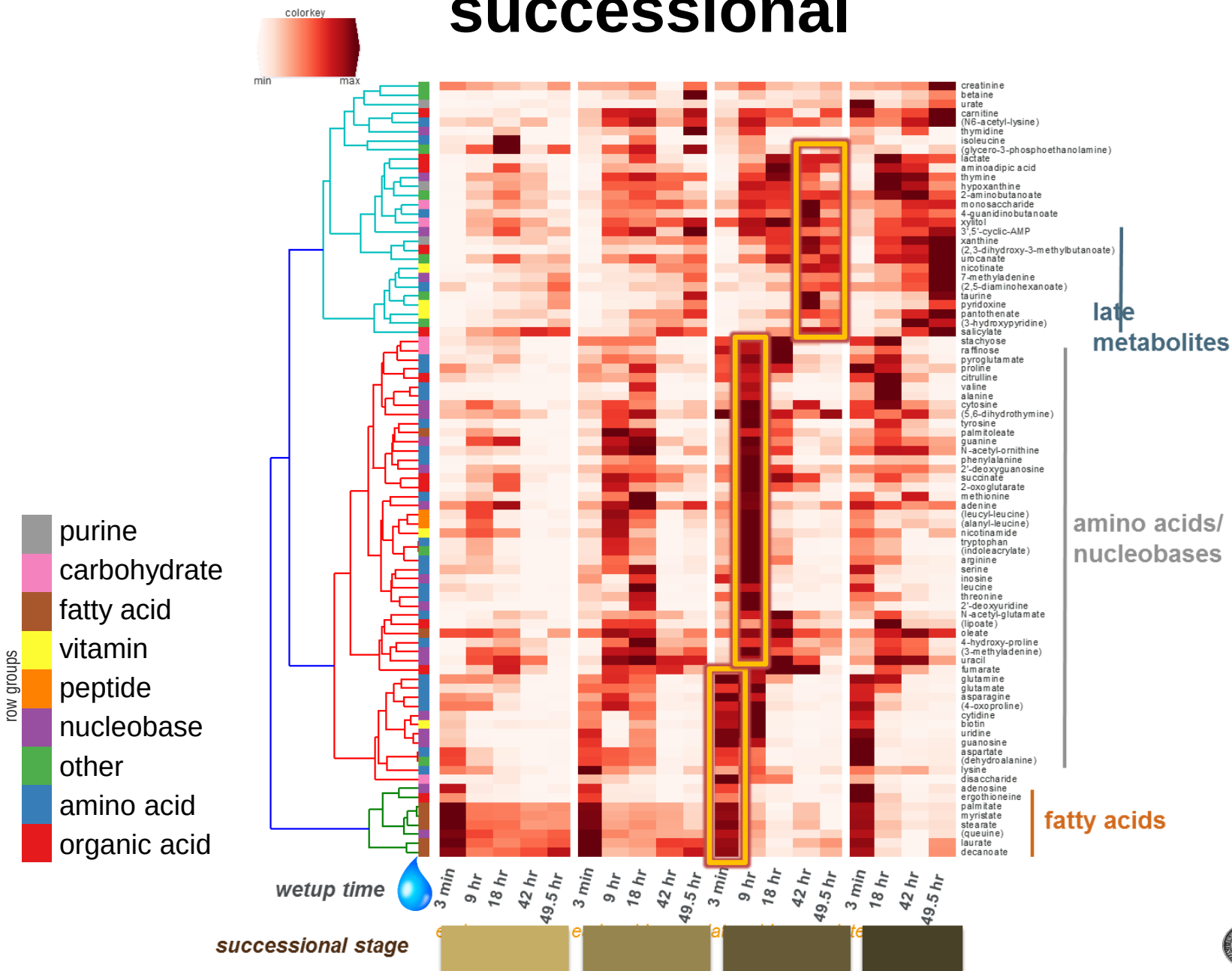


Bacillus 2- late responder

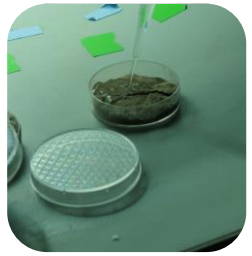


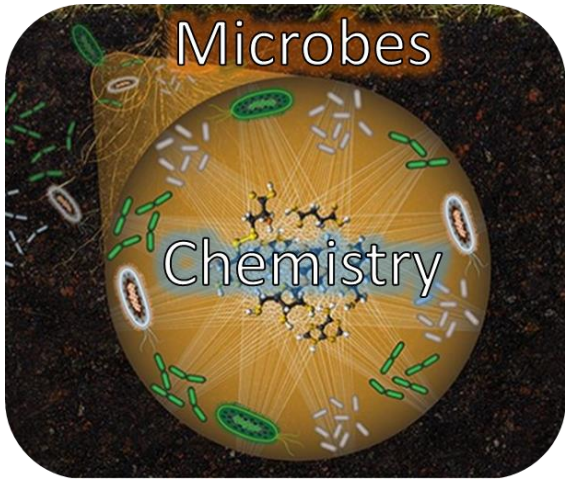
Karaoz U. **mBio** (2018) 9:e01366-16. doi.org/10.1128/mBio.01366-16

Metabolite cycling observed across wetup and successional

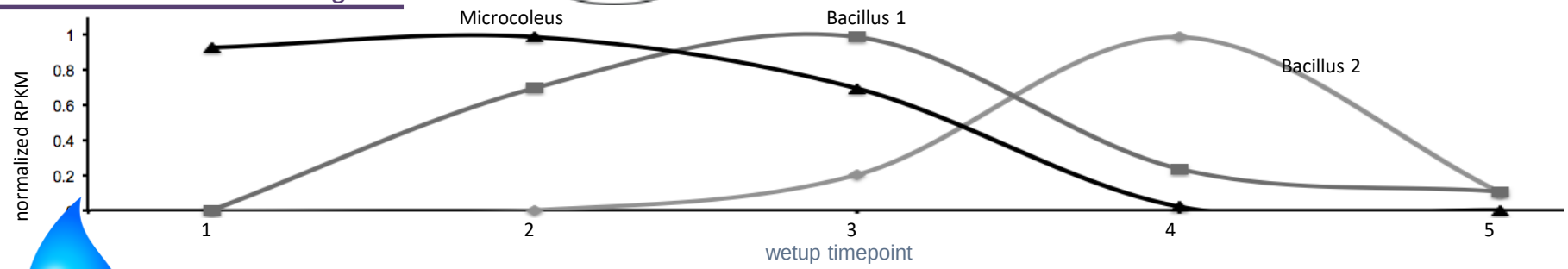


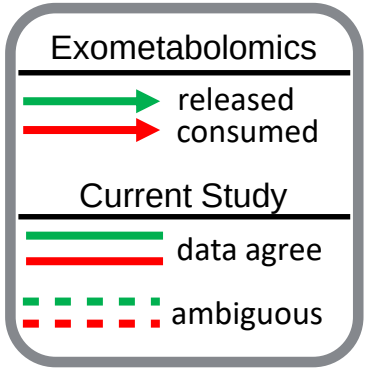
Isolate exometabolite profiles link microbial abundance to metabolite abundance





late-mid successional stage





wetup time

Swenson T. et al. **Nature Communications** (2018)

Morton, J. et al. **Nature Methods** (In Press)

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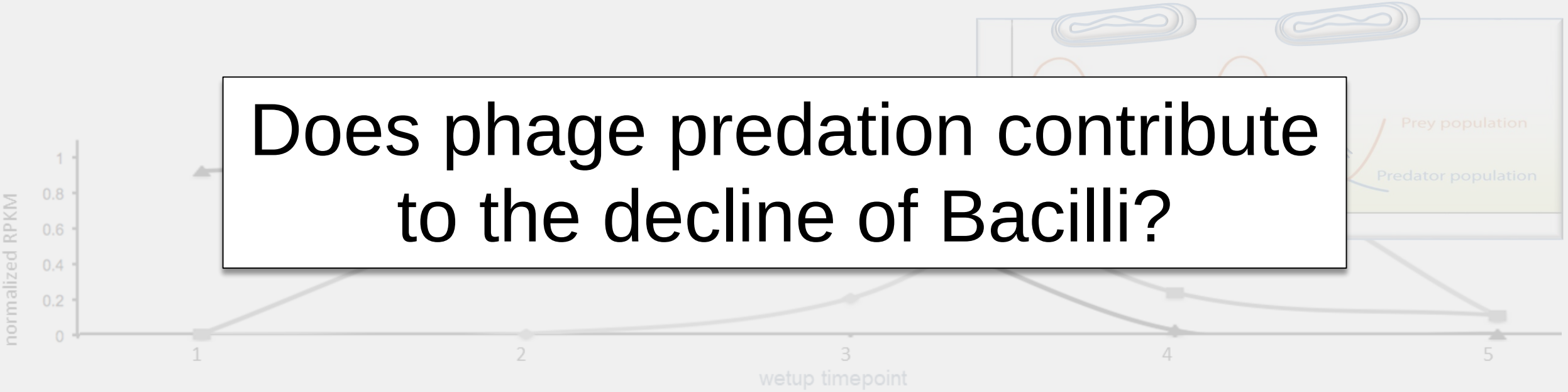
Phage haven't been studied in biocrusts

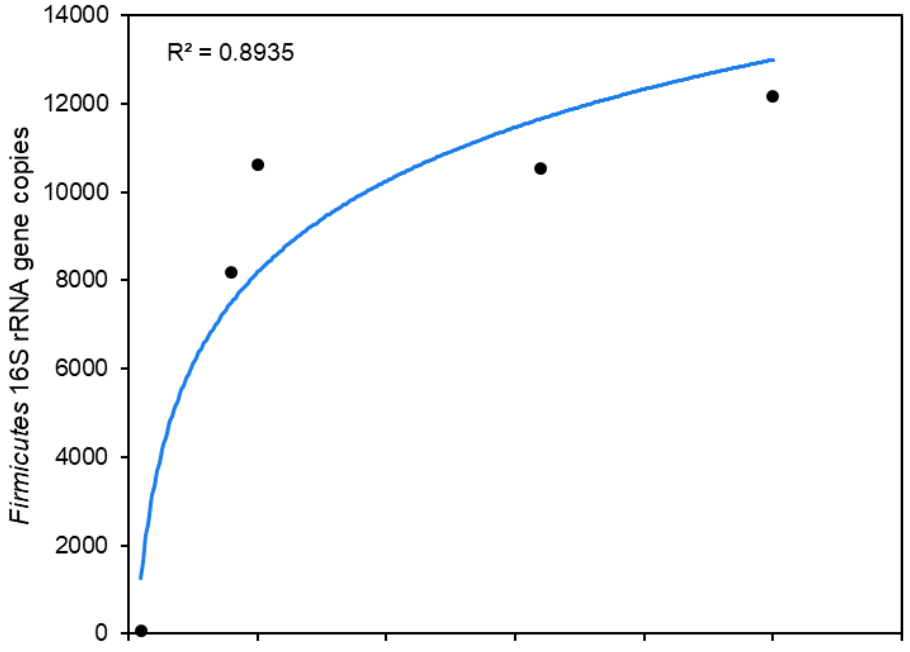


Does phage predation contribute to the decline of Bacilli?

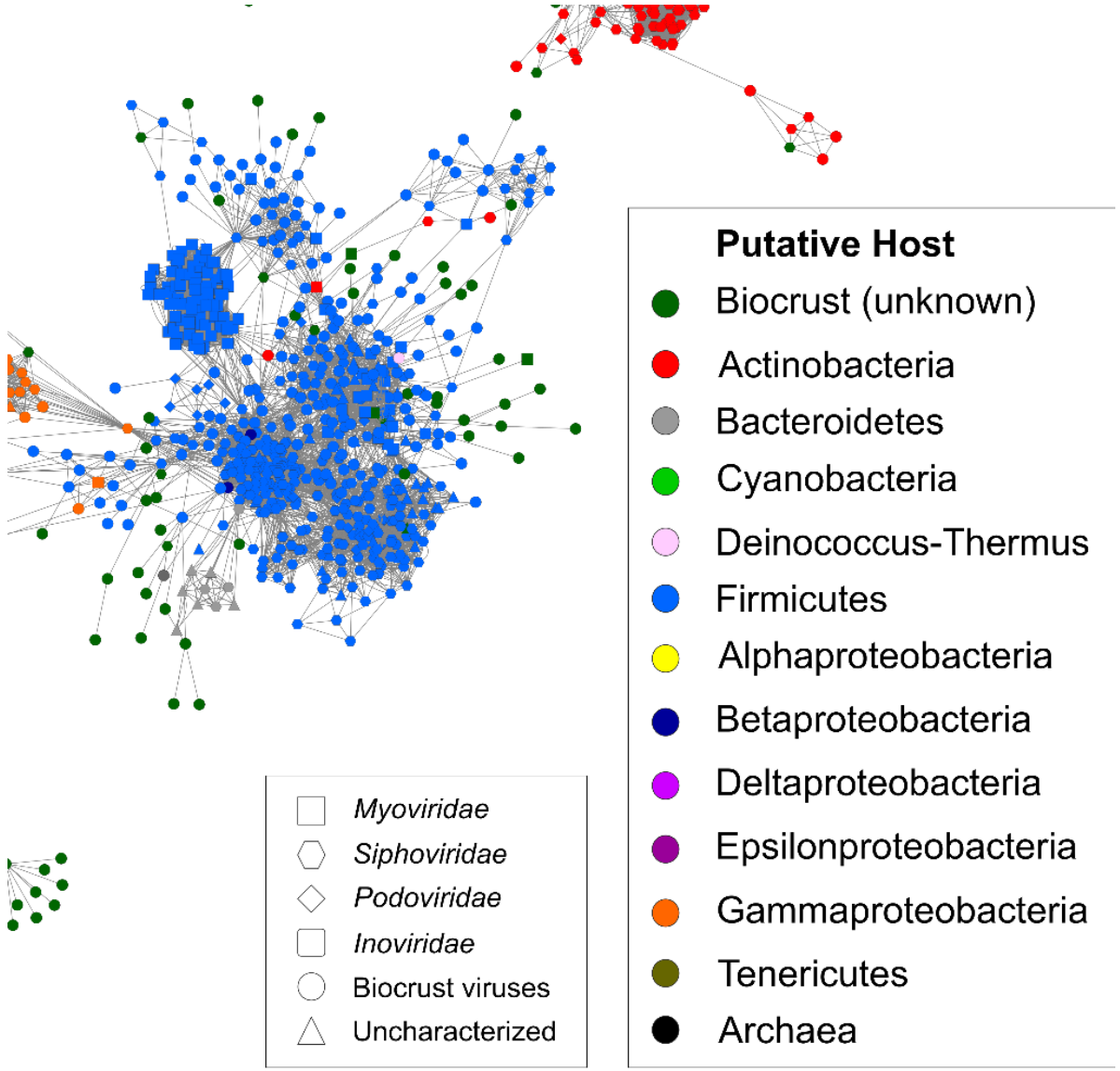
Observe a diversity of phage with putatively predating Firmicutes

Marc Van Goethem

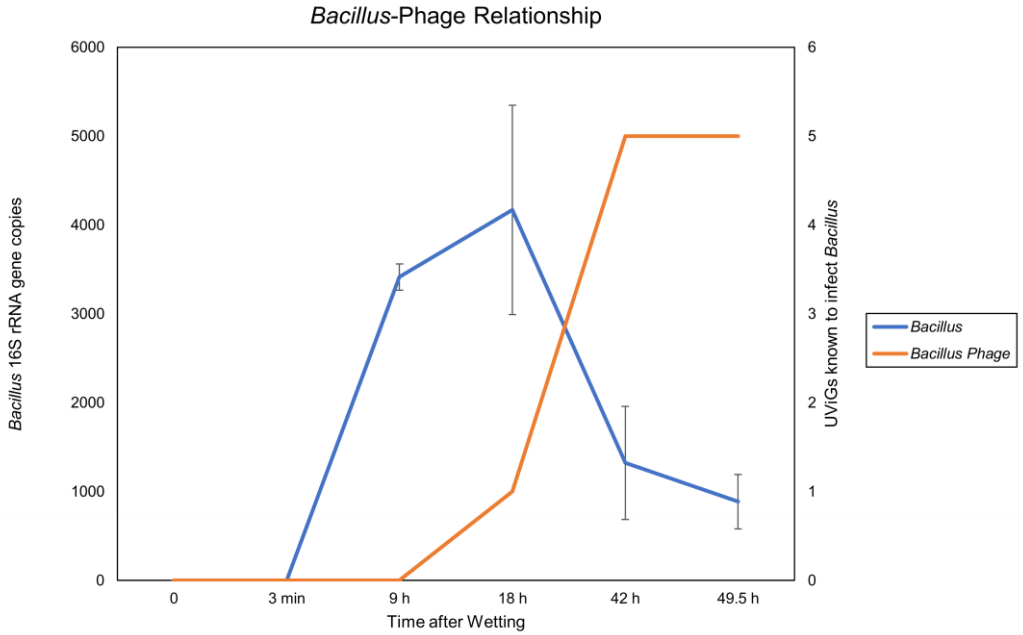
Simon Roux (JGI)



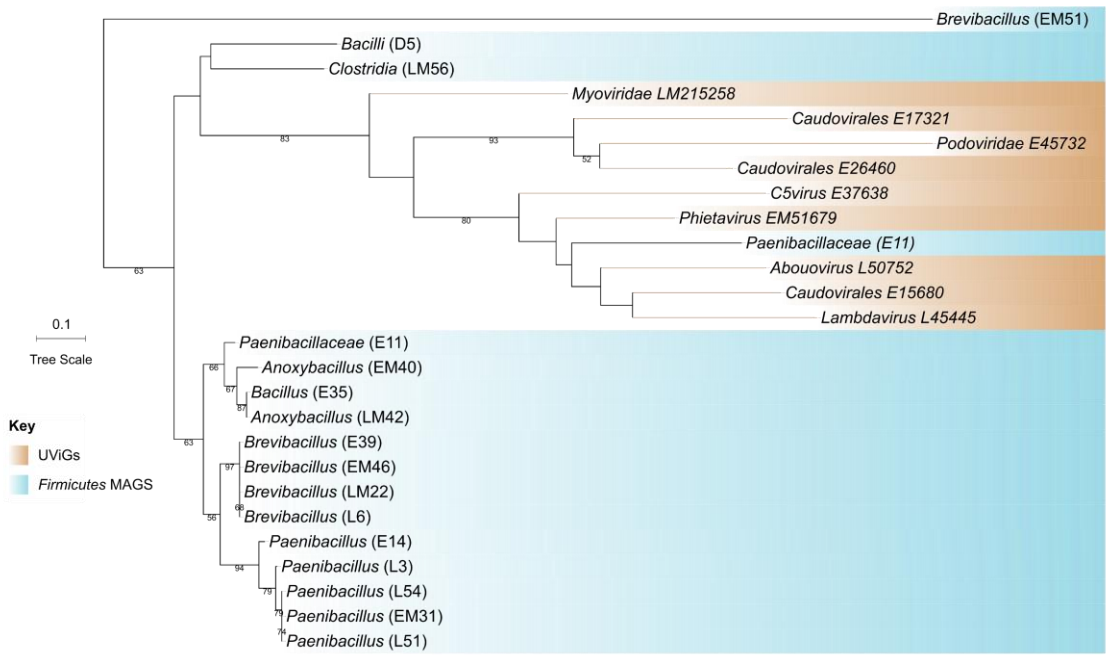
uncultivated viral genomes (UViGs).

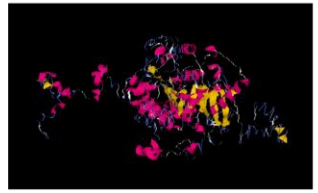


Observe predator prey relationships for some Bacillus and Bacillus phage and sporulation auxiliary metabolic genes



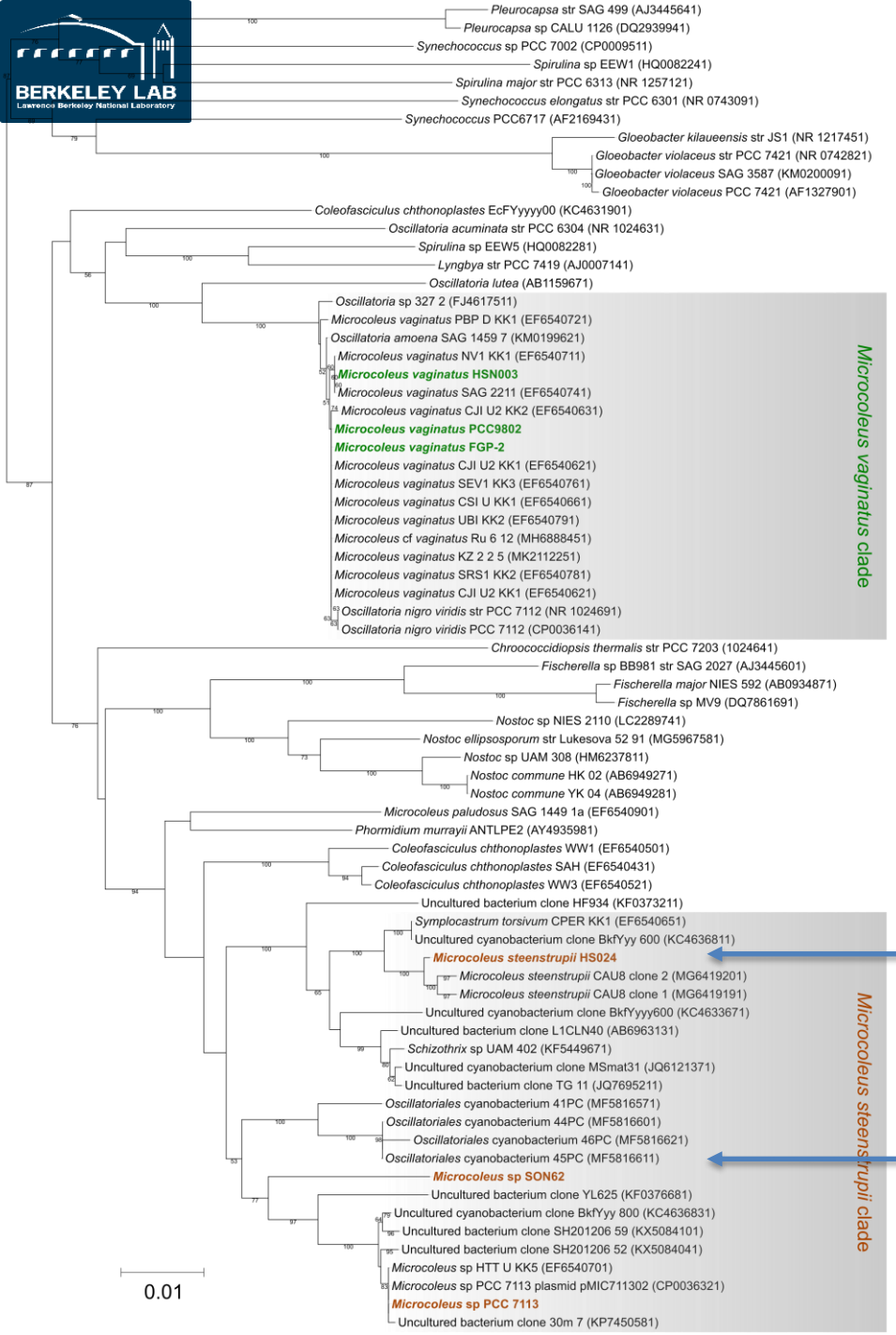
Phylogenetic placement of *spoIIIE* genes retrieved from UViGs with their nearest bacterial neighbors (from *Firmicutes* MAGs)





Van Goethem et al **mBio** (accepted)

PacBio sequencing of Microcoleus enrichments

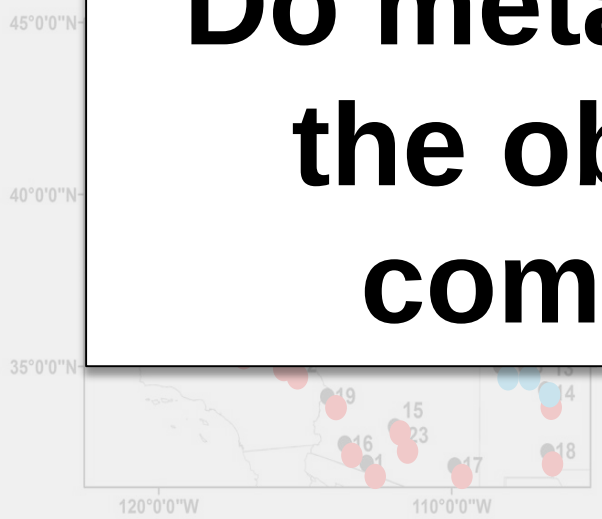


- Identified new Microcoleus species (average nucleotide identity 86 and 79 vs. known species).
- Genomic analysis suggests adaptations for life at higher temperatures, including higher base pairing in their 2D tRNA structures

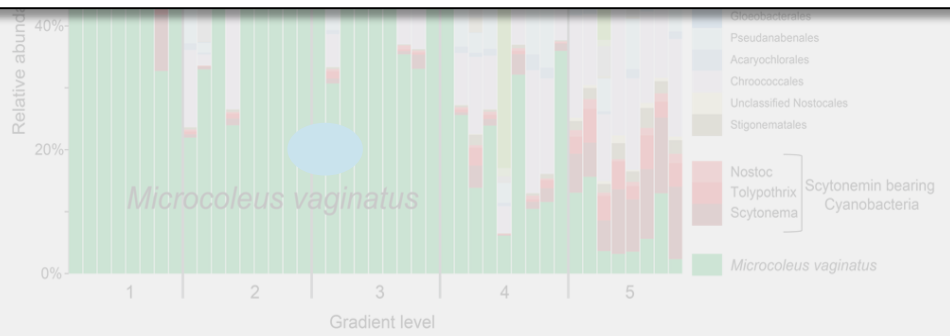
Collaboration with Ferran Garcia-Pichel

Temperature driven replacement of cyanobacteria at both the local and continental scales

A3—Test approach to predict community responses across an environmental gradient



Garcia-Pichel et al.
Science, 2013



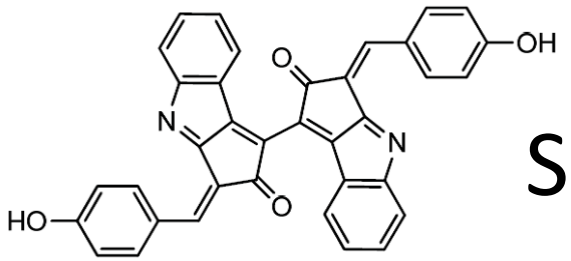
Do metabolites play a role in the observed changes in community structure?

Estelle Couradeau

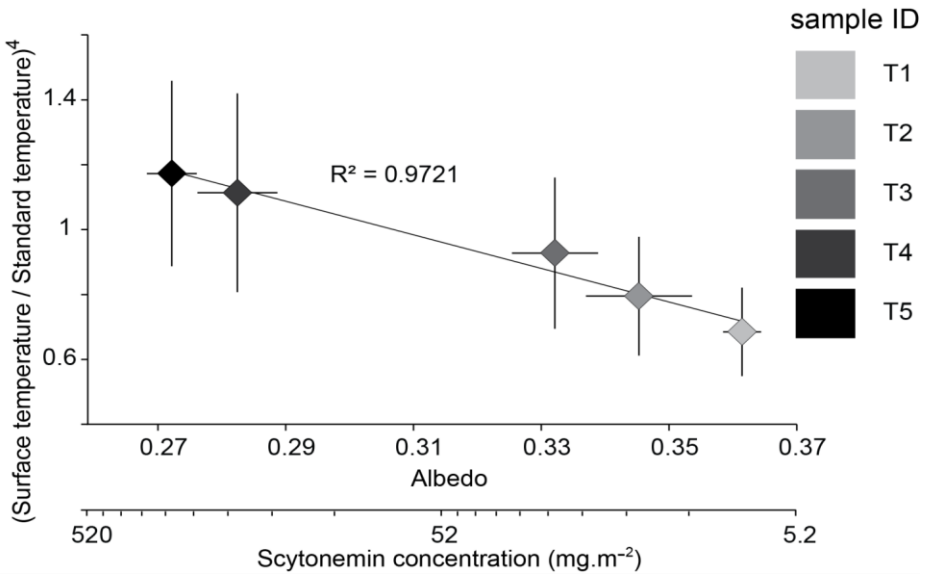
Eoin Brodie
Ferran Garcia-Pichel

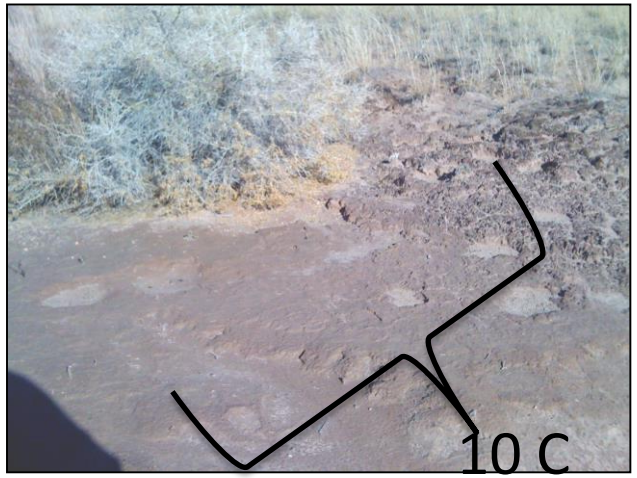
Couradeau et al **Nature Communications** 2016; doi:10.1038/ncomms10373

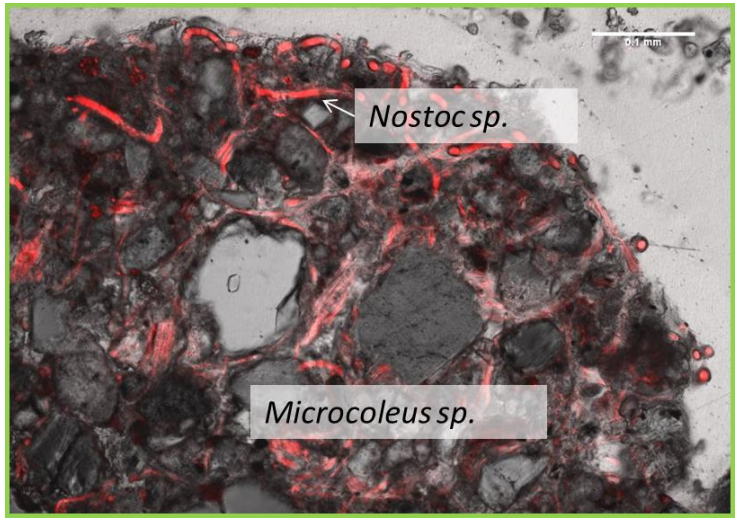
Changes in surface albedo explained by cyanobacterial secondary metabolite produced by Nostoc cyanobacteria

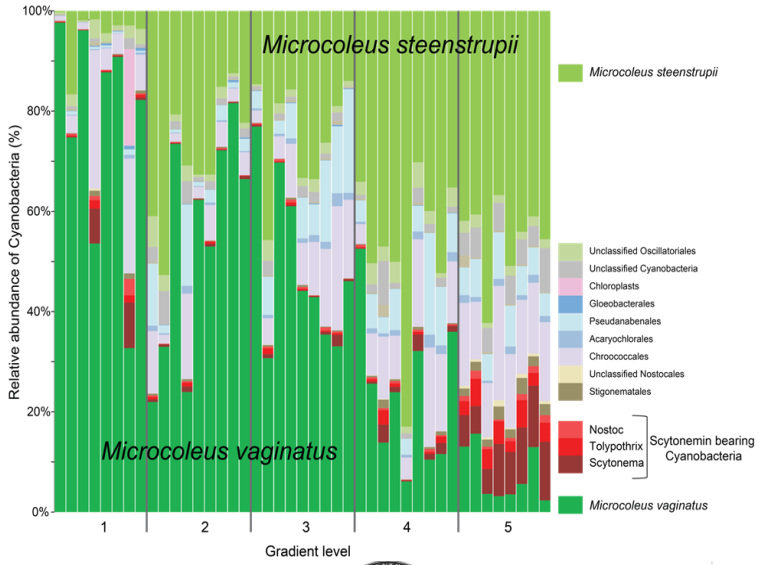


Scytonemin



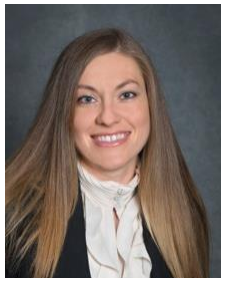




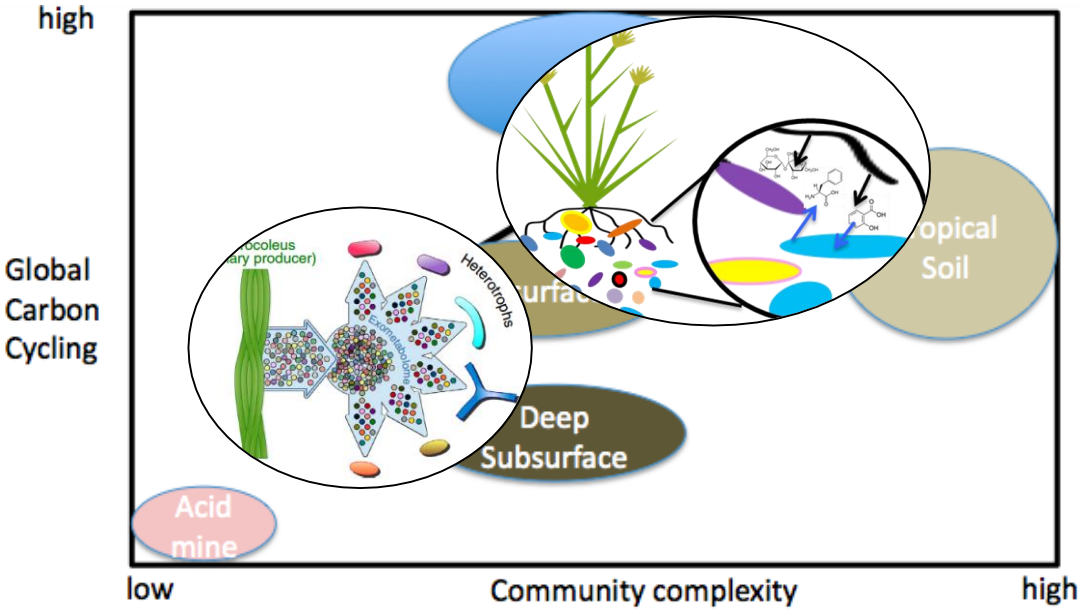


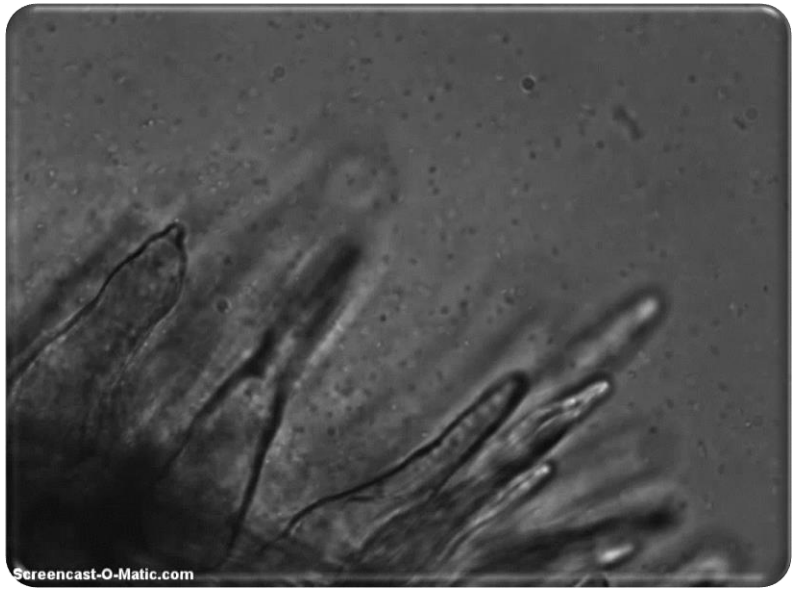
Couradeau et al **Nature Communications** 2016; doi:10.1038/ncomms10373

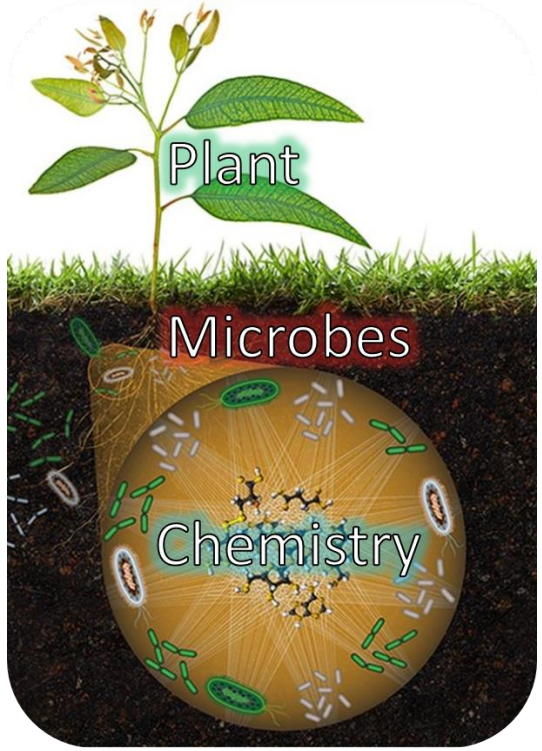
New direction: Using exometabolomics approach to study the role of exudates in structuring rhizosphere communities?



Kate Zhalnina
Brodie Lab
Firestone Lab



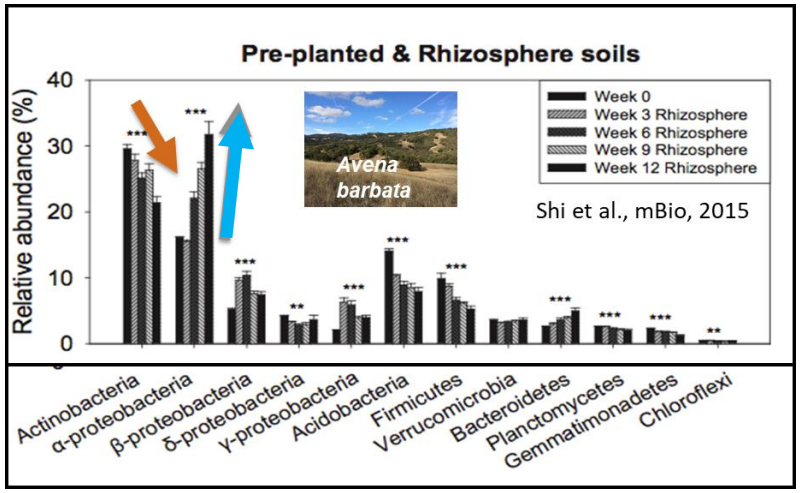




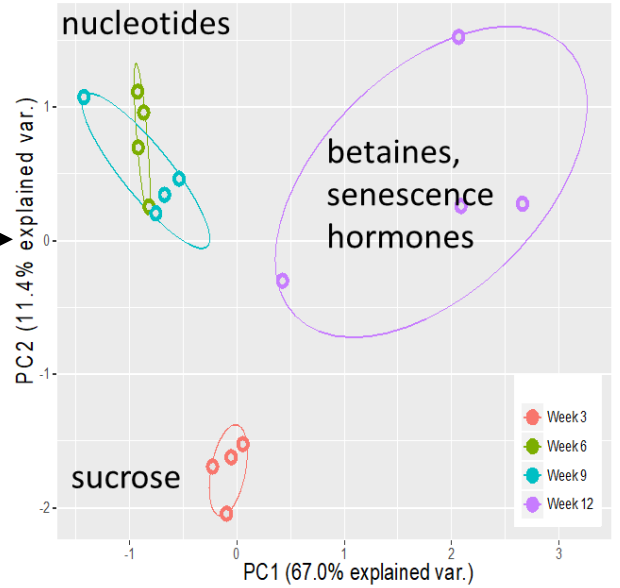
Extend understanding/approaches to more complex systems

~40% of fixed carbon is deposited from the roots to ‘pay’ beneficial microbes

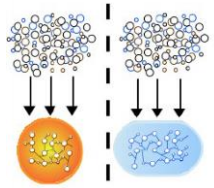
Extension of exometabolite analysis identified metabolites that may drive rhizosphere assembly



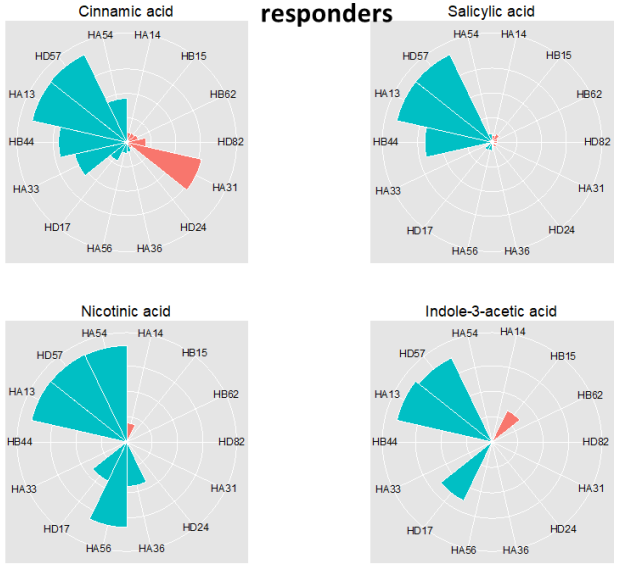
Aromatic organic acids, amino acids, nucleotides



Feed exudates to rhizobacteria



Phenolic acids are preferentially used by rhizosphere responders



Zhalnina et al **Nature Microbiology** 2018

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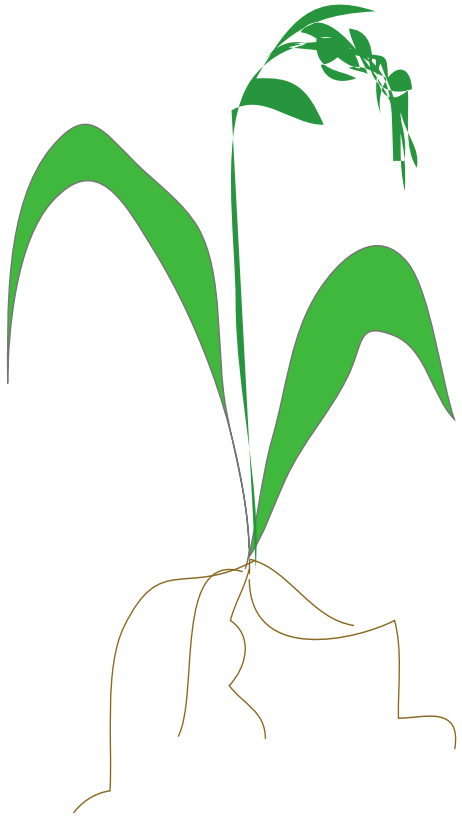
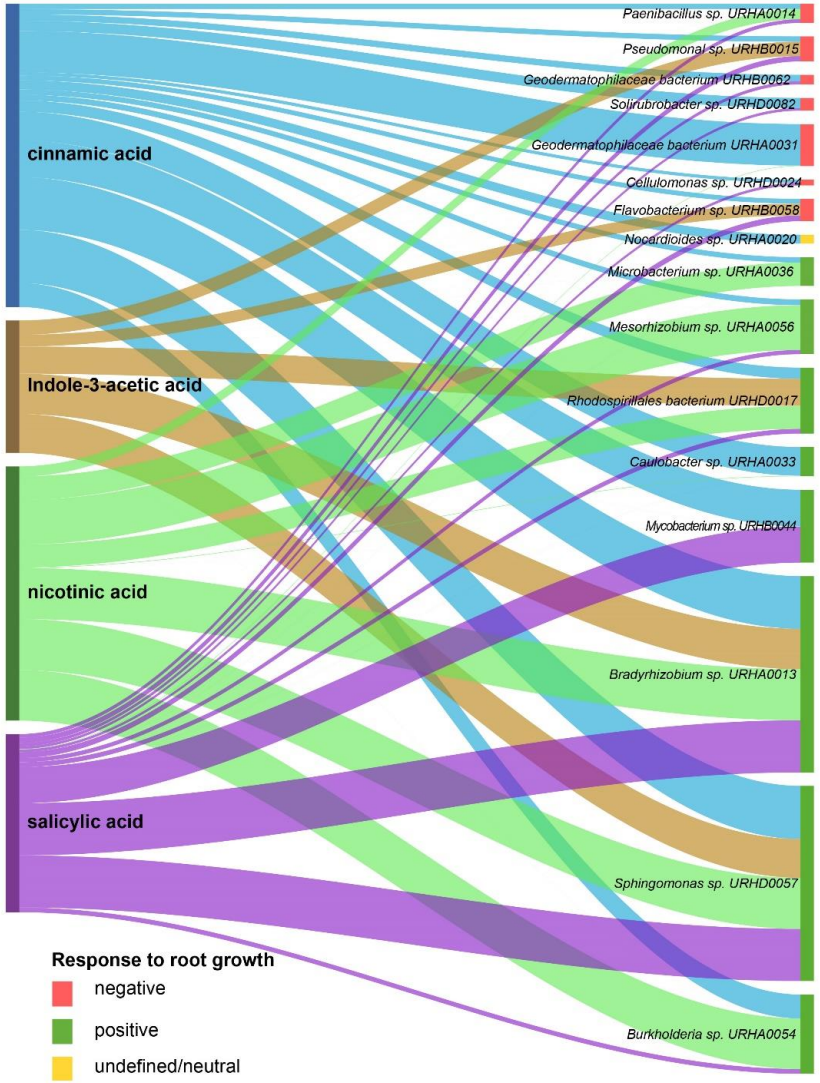
Suggests plant use exometabolite niche partitioning to manipulate microbiome composition

Avena barbata

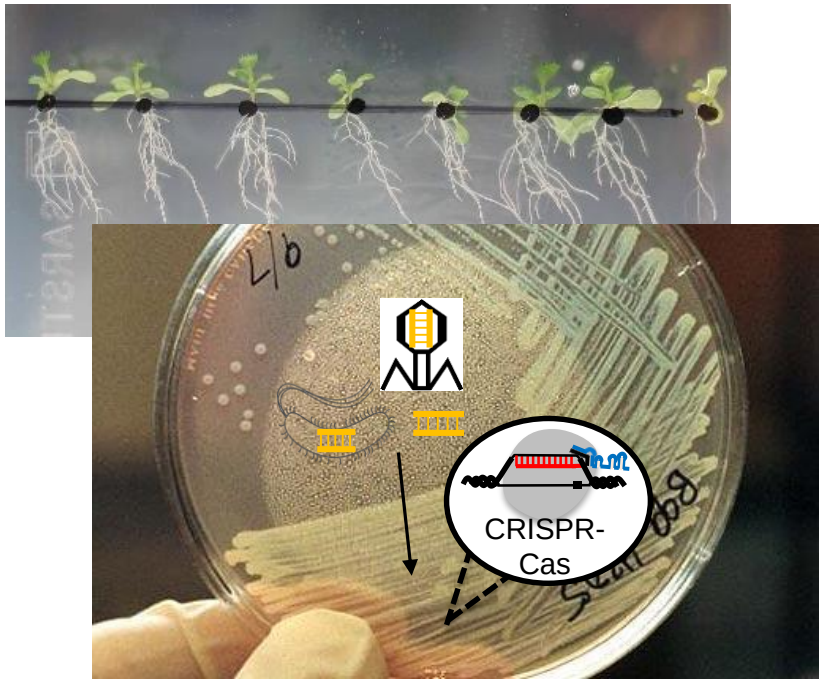




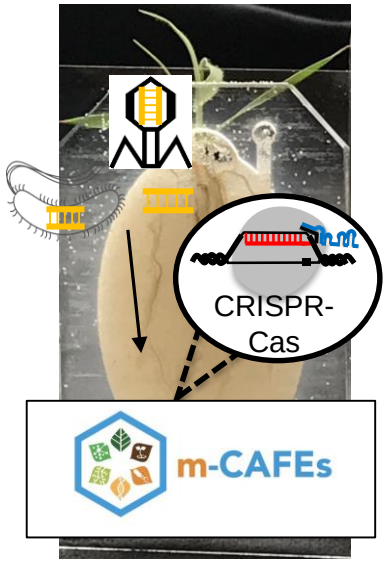
How can we test these predictions?

New capabilities are needed to achieve a predictive understanding of microbial communities

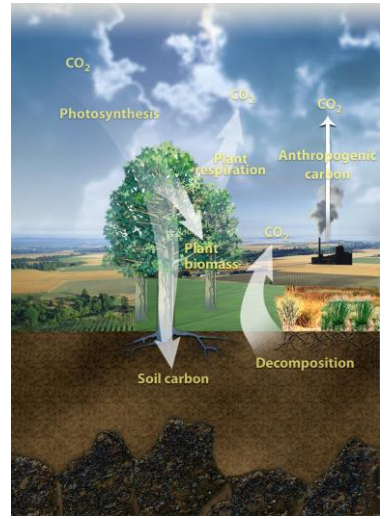
Cellular and organismal biology

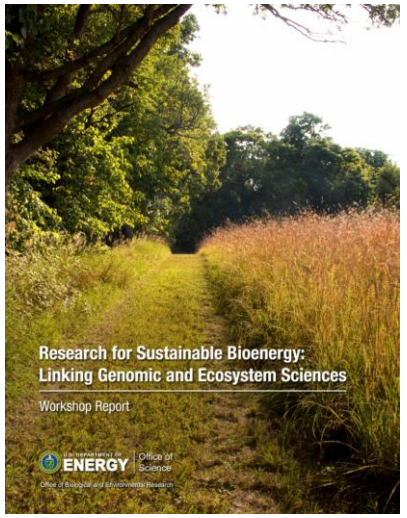


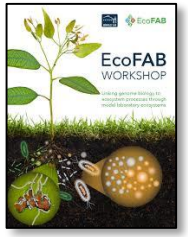
Controlled and reproducible fabricated ecosystems (EcoFAB) + perturbation technologies



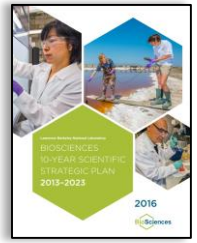
Field







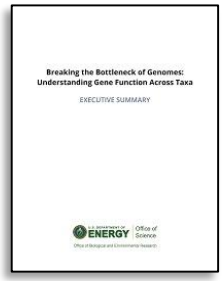
2016 EcoFAB workshop



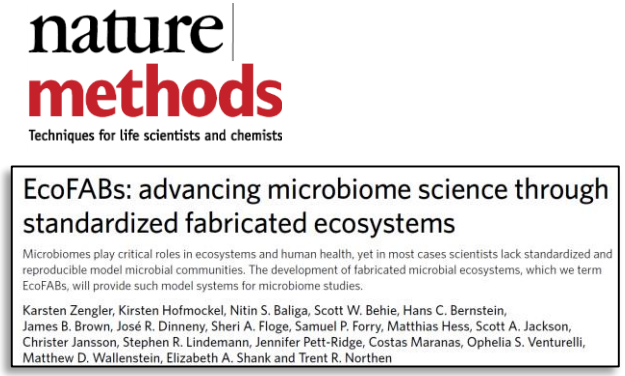
Biosciences strategic plan

2017 EcoFAB summit

2018 AAAS Session



2018 Breaking the bottleneck of genomes

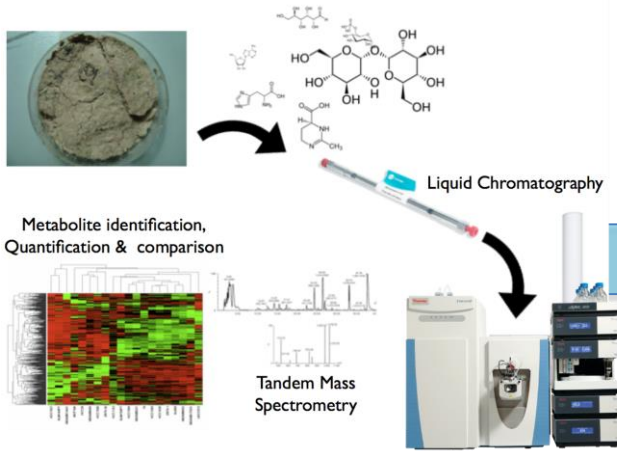


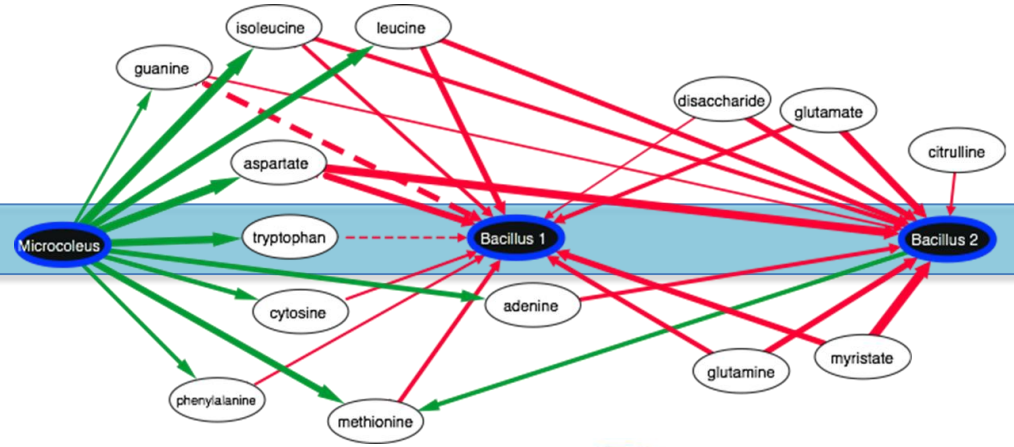
2018 EcoFAB paper

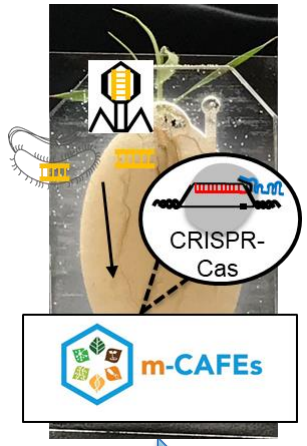
2020 AAAS Session

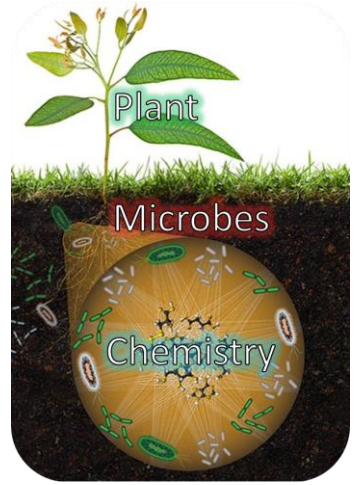
Dramatic positive impact of this project on my career

NorthenLab: Exometabolomics linking genomes with environments to understand how webs of microbes sustain biomes

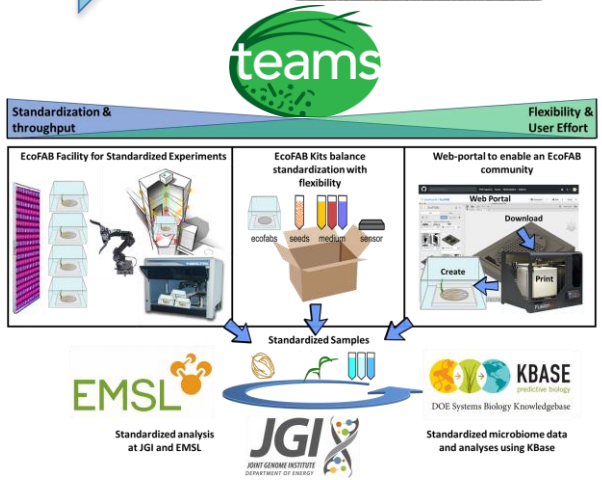








Webofmicrobes.org



JGI JOINT GENOME INSTITUTE

KBase

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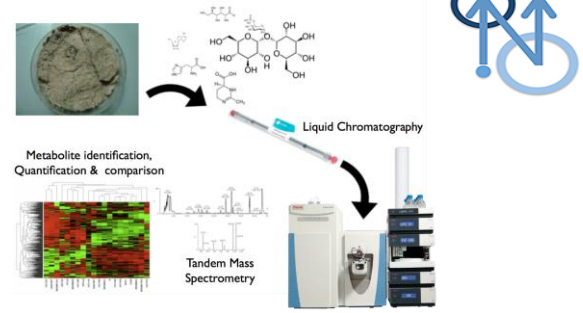
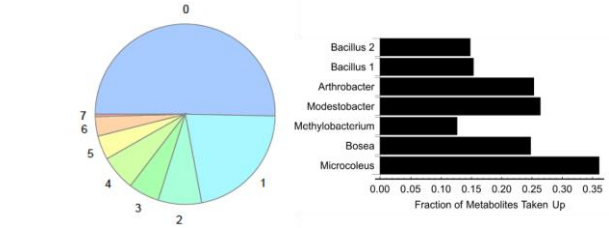
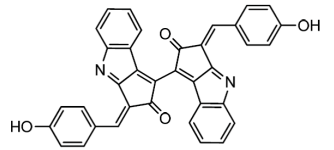
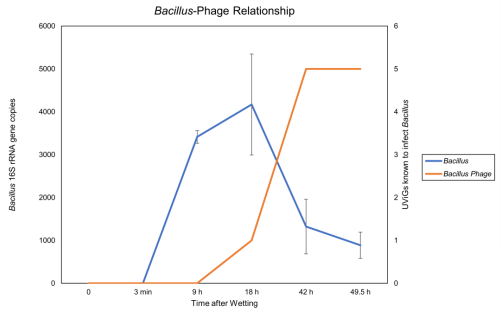
Summary

- Developed soil metabolomic methods to measure microbial substrates and products
- Developed exometabolomic approaches to measure microbial activities on environmental metabolites
- Observed exometabolite niche partitioning among biocrust isolates
- *In situ* biocrust microbe-metabolite correlations were consistent with isolate exometabolite profiles
- Identified that a bacterial pigment could explain changes in community structure along an environmental gradient
- Discovered evidence of phage predation on biocrust community structure and sporulation auxiliary metabolic genes









Acknowledgements

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Richard Baran
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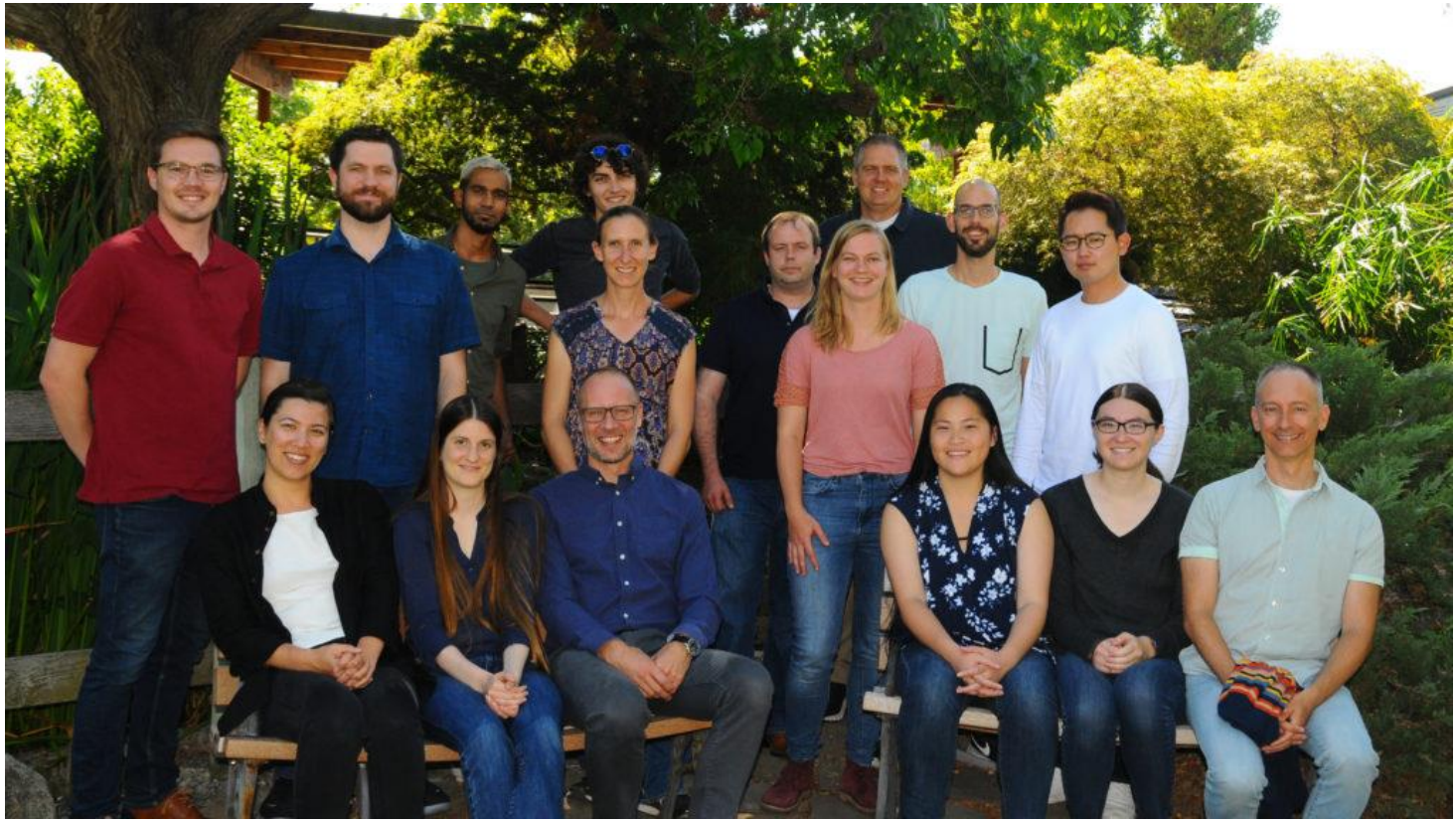
Rebecca Lau
Suzie Kosina
Annette Greiner
Ben Bowen
Ulisses Nunes da Rocha
Ulas Karaoz
Eoin Brodie

UC Berkeley
Mary Firestone

Arizona State Univeristy:
Ferran Garcia-Pichel Lab



DOE Office of Biological and Environmental Research Early Career Award

LBNL LDRD Program

THANK YOU!!!

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