NSF Workshop Report

Microbiome Soil Sensors Workshop Final Report

August 22-23, 2018

Executive Summary

This workshop provided an environment for a robust discussion among experts from microbiological, computational and technological fields that contribute to microbiome soil sensors. The workshop was hosted by the University of Chicago and was held on August 22-23, 2018 in La Jolla, California. The outcomes of the workshop included a broad consensus on the need for context when addressing microbiome soil sensors, including: what to measure, when to measure it, and how frequently (in space and time) it should be measured. These concepts were addressed in 3 broad themes: sensing microbial communities in context, sensing at scale, and technology, that build on a previous successful 2017 NSF-funded workshop: The Subterranean Macroscope (https://ime.uchicago.edu/subterranean_macroscope/). The current workshop was organized to provide more specific focus on capturing the dynamic composition and activity of the soil microbiome.

Sensors should be built to reflect the needs of both the research community to enable discovery science at the point of need for uncovering novel trends, interactions and dynamics, as well as for the translational needs of soil stakeholders (e.g. growers, land-users, climate action groups). Both research and translational end-points have specific requirements, which parallel those of research and clinical implementation in medical practice.

Sensors that detect key environmental parameters, microbiome structure or metabolic functioning could transform our fundamental understanding of microbial processes in soils. Sensors that accurately, repeatedly, and robustly measure specific parameters (e.g. a keystone microbe, a gene, a transcript, a protein, a metabolite) that drive important soil processes could provide actionable insights that enable precision soil management. For example, sensors that enable improved spatial and temporal resolution of soil fertility could enable precision fertilizer applications.

Discovery of those markers is still very much ongoing, and as such, no definitive list can be provided. However, it is possible to outline key overlaps between the needs of the research and translational communities. To organize the discussion of these needs, the workshop was structured around the following three themes.

Theme One: Sensing microbial communities in context. Soil biodiversity reflects the complexity of organisms living in soil, including the myriad metabolic, signaling, and physical interactions that shape their growth and activity. Microbial interactions with each other and their environment shape the emergent properties of these ecosystems that impact plant and animal health and can affect local and global climate.

Despite a considerable body of research and practice in understanding and managing soil microbial processes, there are still vast gaps in understanding of how microbial interactions are governed by changes in the physicochemical and biological dynamics of soil, and importantly how changes in the microbial process dynamics shape these physical, chemical and biological properties of soil. Research can help to elucidate these processes, and quantify the dynamics that underpin mechanistic impact, which can then be translated into soil sensors for agricultural and industrial activities. Therefore, there is a need to produce more accurate and frequent measurements of soil microbial processes in order to improve our understanding of these dynamics.

Soils sensors could rapidly improve understanding of microbial community interactions. However, soils are a particularly dynamic and complicated ecosystem in which to study

microbes. Therefore, technology must be developed that can robustly monitor key microbial community traits in this environment. Sensors need to consider not only which microbes are present, but how their metabolic and signaling pathways are responding to environmental change and to each other. For example, using a synthetic microbial cell *in situ* to sense chemical signals passed between bacterial cells, could help to quantify particular metabolic responses to shifts in redox potential that could shape climate active gas cycling in these systems.

Such sensors would need to be able to provide researchers with information on which microbes are playing an active role in the physical, chemical, and biological transformations and cycling in soils at any given time, to allow us to predict responses to particular perturbations. In the workshop, the question *what fundamental biology could be elucidated through the integration of microbiome studies and sensing technology that cannot be answered separately* was continually raised by participants. The question was not fully answered, due in part to the contextual complexity of answering it. While there are many potential answers to this question, the main impact of sensing technology of microbiome studies is one of improved resolution and scaled integration, which was explored in theme 2.

Theme Two: Sensing at scale. Microbial communities act on different scales, scale which must be sensed if we are to identify the traits and characteristics that should be reliably translated into commercial application. There is a need to understand microbial communities at different scales including: spatial, temporal, and molecular organization scales, which were outlined by Jizhong Zhou. Spatial scales for soil include microaggregates, macroaggregates, plots, fields, villages, counties, provinces, countries, and global scales. Temporal scales include minutes, hours, days, weeks, months, seasons, years, and decades. Examples of molecular organization scales include diversity (taxonomic, phylogenetic, functional), organismal (viruses, bacteria, archaea, fungi, protists, nematodes), and biochemical (DNA, RNA, proteins, metabolites, process rates).

One sensor tool cannot cover all scales, but these scale groups represent targets for obtaining information at appropriate levels. As identified at the November 2017 workshop, new approaches are needed in computer science to both model complex environments like soil and microbial communities, and to integrate large quantities of complex data. The creation of sensor data at multiple scales will require new models that mathematically integrate different types and scales of data at nested levels. One example provided at this workshop, was that if we were able to accurately model the averaged rate kinetics, moisture level and distribution, redox potential and nutrient concentrations for a 100-micron scale cube of a particular soil type, could we determine the multiplication equations that would allow us to scale up this one cube to a cubic kilometer of soil? Computational scaling becomes extremely important to address these types of questions.

Theme Three: Technology. Current approaches to sensing and engineering have enabled a new class of sensors including those specifically designed for subterranean settings and those with various applications. As indicated in the final report from the November 2017 workshop, there are clear needs for research in both the wireless protocol domain and device level implementation, as well as in subterranean sensor networks. This workshop advanced those recommendations by dealing specifically with what existing and new technological targets can be developed in academic and industrial settings to further basic and applied science goals.

Summary of Breakout Sessions

Two breakout sessions were held during the workshop to synthesize presentations, identify future research directions, and capture key questions. One breakout session addressed two themes: microbial communities in context and sensing at scale. The second breakout session addressed technology. Each workshop attendee participated in one of the breakout sessions.

Microbial Communities in Context and Sensing at Scale

The microbial communities in context and sensing at scale breakout session focused critical environmental determinants, the need for mechanistic understanding of distribution gradients, and the need for transformative understanding of the mechanistic and ecosystem levels of microbial communities and their interaction. Discussion led to the development of future research directions and key questions outlined in the "Future Research Directions and Key Questions" section of this report.

Technology

The technology breakout session focused on the development of new sensors that are sensitive and provide for rapid monitoring of different metabolic and genomic information. An example of this was given by Xufeng Zhou's presentation on the technology optical ring micro-resonators to electronic FET transistor aptamer sensing to high throughput DNA melting sequencing (Argonne). The needs identified by this group were an integrated holistic system that could seamlessly handle data from sample prep to detection to modeling and data analytics. The greatest challenges identified were how to monitor metabolites' genomic information deep in the soil, especially the challenging measurement of redox at different depths. Future research directions and key questions emanated from the breakout session.

Motivation and Outcomes of Workshop

The previous workshop, Subterranean Macroscope: Sensor Networks for Understanding, Modeling, and Managing Soil Processes (https://ime.uchicago.edu/subterranean_macroscope/), held on Nov 1-2, 2017, focused on identifying the key grand challenges in soil and plant science and subterranean sensing. In addition to the needs of the community, the workshop discussed the underlying engineering and science challenges to create such a subterranean network. This workshop included participants from academia, industry, national laboratories, and government funding agencies.

One of the key topics discussed during the workshop was understanding soil, microbiome, and root interactions affecting plant productivity, water and nutrient efficiency, and soil degradation. The community emphasized the need to characterize soil microbiome capacity and expression, community diversity and function, and to understand how these interactions relate to crop production and other important environmental outcomes. Key sensor needs that were identified included: CO_2, O_2, N_2O, NO_3, P, H_2O, pH, temperature, as well as measurement of total soil C and fractionated soil carbon forms.

Critical questions around microbiome and sensing in soil were raised during the 2017 workshop included:

- What are the microbial determinants of plant phenotype and performance?
- What are the relevant spatiotemporal scales to measure the concentration and movement of soil water and nutrients to predict microbiome functioning?
- How can we develop subterranean sensors to measure these characteristics and processes?
- Can we use compact, chip scale polymerse chain reaction (PCR) with stable reagents for in-field soil microbial sensing?
- What are the energy and hardware needs for these sensors?
- Could we use alternative fuel cells to generate energy for remote sensing needs, such as microbial fuel cells?

Based upon these discussions, the workshop participants identified the need for a subsequent workshop focused on creating a roadmap to develop subterranean sensors for soil microbial activity. In order to address this need, the current, August 2018 workshop, was planned to bring together experts in sensing, soils, microbiome studies and analysis, and modeling.

Workshop Participants

This workshop included a diverse audience with a range of backgrounds, including: academic experts, stakeholders from industry and the national laboratories, and federal agencies who will be collaborators and eventually the end-users of this technology. This small workshop was organized to foster a focused discussion among approximately 20 participants. Workshop participants were selected using nominations from the scientific advisory committee members. All proposed participants were discussed and evaluated by the committee.

There were 22 attendees in total, of which 7 (32%) were female and 15 (68%) were male. Participants included experts from 11 different institutions/organizations in the fields of soil science, microbiology, computational biology, sensors and nanotechnology.

An additional goal of the workshop was to include junior researchers such as postdoctoral scientists and graduate students. These students came from organizations across the country such as the University of Illinois at Chicago, Argonne National Laboratory, and University of California Los Angeles. Names and affiliations of all participants are provided in Appendix A.

Future Research Directions and Key Questions

The workshop participants identified critical research questions and future opportunity areas from each of the breakout sessions. These questions and areas are summarized below.

Microbial Communities in Context and Sensing at Scale:

Question One: Can we sense the critical environmental determinants of the assembly and function?

- Overall requirements are in-ground, real-time, inexpensive sensors that would allow for a plethora of sensors to be deployed to allow for temporal and spatial resolution. This greater sensor coverage would allow for soil microbial processes to be integrated with a mechanistic level of understanding of the complex nature of soil processes. This transformative understanding could have applications to precision agriculture.
- To have a mechanistic understanding, we need to have distributions of gradients, not just an average. One example of the distribution gradient would be from the root to the bulk soil of nutrients, oxygen and capture how theses gradients change temporally. This may require a facility (or facilities) to instrument soils with a dense array of sensors and imaging technology.
- The challenge is that soil is a complex matrix of microbial communities interacting between members of that communities and also with the environment.
- The first step to prioritize what to sensor would be a mapping exercise to guide environmental drivers in a test bed facility.

Question Two: How does microscopic behavior impact or govern large-scale ecosystem properties?

- The scale would be at the multiple meter scale that would capture plants interacting at the ecosystem level
- Integrated models would be needed to bridge across scales (see Question 1).
- Sensors would want to capture gradients not averages of water, O_2, CO_2, and roots, as examples.

Technology:

The Technology Session identified three additional questions for further exploration that focused on the best approaches for the detection of microbes in soil and environmental settings. Multiple technical approaches to monitoring specific soil sensors were discussed. Approaches were discussed for sensing DNA, RNA, and metabolites.

Question Three: There is a need for several types of sensors such as those for DNA, Metabolite and RNA.

- Sensors need to provide low cost, low power, real time measurement was although there were different opinions about where to put resources on improving current technology (e.g. PCR) for field work or for developing new approaches.
- Mass spec remains important for aerial sensing that can lead to targeted subsurface sensing.
- Some of the challenges of this approach raised was how to select for and prioritize detection. One way to avoid this issue is through the use of machine learning techniques which can comb through vast datasets and identify patterns and important information.

Question Four: Can we develop a holistic system from sample prep to detection and beyond that is able to adjust in real time?

- From sample prep to detection and then to analysis, we need a system that is able to be adjusted for different environments and on different temporal scales.

- There is a need for algorithms to integrate the various types of information that can lead to decisions about soil treatment in order to be able to interpret metabolic pathways.
- It is also important to have automatic integration and adjustment of the system in a way that is non-invasive to the environment.

Question Five: What is important to sense in soil? Shall we look for an overall profile or something more specific?

- Ideally, multiple approaches are needed so that researchers can look for small molecules as indicators of health.
- At the same time mass spec imaging can provide large amounts of data that can be integrated. Data integration remains a challenge in this complex approach and provides significant opportunity for collaboration.

Appendix

Agenda

Day 1 – Wednesday, August 22			
12:30 PM	**Workshop Check-In & Lunch**	**60 min**	**Location: Grande Room**
1:30 PM	**Welcome & Introduction of Workshop Focus Areas**	**15 min**	**Location: Magnolia** **Supratik Guha, Professor, Institute of Molecular Engineering, University of Chicago; Director, Nanoscience and Technology Division, Argonne National Laboratory**
1:45 PM	**Plenary: Microbial Communities (Theme 1)**	**30 min**	**Mary Firestone, Professor of Soil Microbiology, University of California, Berkeley**
2:15 PM	**Plenary: Sensing microbial communities in context (Theme 2)**	**30 min**	**Chuck Rice, University Distinguished Professor, Kansas State University**
2:45 PM	**Plenary: Technology (Theme 3)**	**30 min**	**Sufi Zafar, Research Staff Member IBM TJ Watson Research Center**
3:15 PM	**Questions**	**15 min**	
3:30 PM	**Break**	**30 min**	
4:00 PM	**Theme 1: Microbial Communities**	**45 min**	**A. Peyton Smith, Assistant Professor, Soil and Crop Sciences, Texas A&M University** **Matt Wallenstein, Associate Professor, Department of Soil and Crop Sciences,**

			Colorado State University; Co-founder and Chairmen, Growcentia, Inc. **Mike Ricketts, Graduate Student, Ecology & Evolution, University of Illinois at Chicago**
4:45 PM	**Panel Discussion**	**30 min**	**Moderator: Chuck Rice**
5:15 PM	**Day 1: Wrap Up**	**15 min**	
5:30 PM	**Day 1 Adjournment**		
6:00 PM	**Welcome Dinner**	**90 mins**	**Location: Adobe**

Day 2 – Thursday, August 23			
7:15 AM	**Breakfast**	**45 min**	**Location: Grande Room**
8:00 AM	**Welcome & Session Information**	**15 min**	**Location: Magnolia**
8:15 AM	**Theme 3: Technology**	**90 min**	**- Xufeng Zhang, Assistant Scientist, Center for Nanoscale Materials, Argonne** **-Anindita (Oni) Basu, Assistant Professor, Genetic Medicine and Assistant Scientist, Center for Nanoscale Materials, Argonne** **-Todd Coleman, Professor, Bioengineering, University of California San Diego** **-Stephanie Fraley, Assistant Professor, Bioengineering, University of California San Diego**

			-<u>Thomas Young</u>, Graduate Student, California Nanosystems Institute, University of California Los Angeles **-<u>Ben Diroll</u>, Postdoctoral Scholar, Center for Nanoscale Materials, Argonne**
9:45 AM	**Panel Discussion**	**30 min**	**Moderator: Jack Gilbert, Faculty Director, the Microbiome Center, University of Chicago**
10:15 AM	**Break**		
10:30 AM	**Theme 2: Sensing microbial communities in context**	**45 min**	**-<u>Jizhong Zhou</u>, Professor of Microbiology, Director, Institute for Environmental Genomics, Oklahoma University** **-<u>Christopher Henry</u>, Computational Biologist, Argonne** **-<u>Joshua Ladau</u>, Scientist, DOE Joint Genome Institute**
11:15 AM	**Panel Discussion**	**30 min**	**Moderator: Tijana Rajh, Distinguished Fellow and Deputy Director, Center for Nanoscale Materials, Argonne**
11:45 AM	**Break**	**15 min**	
12:00 PM	**Working Lunch** **Concurrent Breakout Session Themes 1 & 2** **Concurrent Breakout Session Theme 3**	**75 min**	**Moderator: Chuck Rice** **Moderator: Tijana Rajh**

1:15 PM	**Report out from breakouts**	**45 min**	**Location: Magnolia**
2:00 PM	**Next Steps**	**15 min**	
2:15 PM	**Adjournment**		
2:30 PM	**Steering Committee Report Writing**		**Location: Magnolia**

Attendee List

Name	Institution	Title
Benjamin Diroll	Argonne National Laboratory	Postdoctoral Scholar
Brett Hansard	Argonne National Laboratory	
Christopher Henry	Argonne National Laboratory	Scientist
Chuck Rice	Kansas State University	University Distinguished Professor
Dave Knaebel	USDA Agricultural Research Service	National Program Leader - Soil Biology
Erin Lane	University of Chicago	Executive Director, Microbiome Center
Jack Gilbert	University of Chicago	Director, Microbiome Center
Jizhong Zhou	Oklahoma University	Professor of Microbiology, Director, Institute for Environmental Genomics
Joshua Ladau	Joint Genome Institute	Scientist
Julia Lane	University of Chicago	Executive Director of Research Development
Karl Rockne	NSF	Program Director Environmental Engineering
Mary Firestone	University of California, Berkeley	Professor
Matthew Wallenstein	Colorado State University	Professor and Department Head

Michael P Ricketts	University of Illinois at Chicago	PhD Candidate
Oni Basu	University of Chicago; ANL	Assistant Professor/Assistant Scientist
Stephanie I. Fraley	UC San Diego	Assistant Professor
Sufi Zafar	IBM T. J. Watson Research Center, Yorktown Heights, NY	Research Staff Member
Supratik Guha	Argonne National Laboratory	Director, Nanoscience and Technology Division
Thomas Young	University of California, Los Angeles	Graduate Student
Tijana Rajh	Argonne National Laboratory	Deputy Director, Nanoscience and Technology Division
Todd Coleman	University of California San Diego	Professor, Bioengineering
Xufeng Zhang	Argonne National Laboratory	Assistant Scientist

Scientific Advisory Committee Members

Jack Gilbert
Faculty Director, The Microbiome Center
Professor, Department of Surgery
The University of Chicago
Senior Scientist (Adjunct), the Marine Biological Laboratory
Group Leader in Microbial Ecology
Argonne National Laboratory

Supratik Guha
Professor, Institute for Molecular Engineering, University of Chicago
Director, Nanoscience and Technology Division and the Center for Nanoscale Materials
Senior Science Advisor to the Director
Argonne National Laboratory

Charles (Chuck) W. Rice
University Distinguished Professor
Mary L. Vanier University Professorship
Chair, Board on Agriculture and Natural Resources, National Academies of Science, Engineering, and Medicine

Kansas State University

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