ARPHA Conference Abstracts 6: e108263
doi: 10.3897/aca.6.e108263

Conference Abstract

Rhizobiome of ‘Ōhi‘a Lehua (*Metrosideros polymorpha*) Offers Insight into Plant-Microbe-Invertebrate Interactions in the Subsurface

Annette Summers Engel[‡], Mireille Steck[§], Audrey T. Paterson[‡], Amir Van Gieson[§], Megan L. Porter[§], Rebecca Chong[§]

‡ Department of Earth & Planetary Sciences, University of Tennessee, Knoxville, TN, United States of America
§ School of Life Sciences, University of Hawai‘i at Mānoa, Honolulu, HI, United States of America

Corresponding author: Annette Summers Engel (aengel1@utk.edu)

Received: 19 Jun 2023 | Published: 17 Oct 2023

Citation: Engel AS, Steck M, Paterson AT, Van Gieson A, Porter ML, Chong R (2023) Rhizobiome of ‘Ōhi‘a Lehua (*Metrosideros polymorpha*) Offers Insight into Plant-Microbe-Invertebrate Interactions in the Subsurface. ARPHA Conference Abstracts 6: e108263. https://doi.org/10.3897/aca.6.e108263

Abstract

Roots are common features in basaltic lava tube caves on the island of Hawai‘i. For the past 50 years, new species of cave-adapted invertebrates, including cixiid planthoppers, crickets, thread-legged bugs, and spiders, have been discovered from root patches in lava tubes on different volcanoes and across variable climatic conditions. Assessing vegetation on the surface above lava tube passages, as well as genetic characterization of roots from within lava tubes, suggest that most roots belong to the native pioneer tree, ‘ōhi‘a lehua (*Metrosideros polymorpha*). Planthoppers are the primary consumers of sap at the base of the subsurface food web. However, root physicochemistry and rhizobiome microbial diversity and functional potential have received little attention. This study focuses on characterizing the ‘ōhi‘a rhizobiome, accessed from free-hanging roots inside lava tubes. Using these results, we can begin to evaluate the development and evolution of plant-microbe-invertebrate relationships.

We explored lava tubes formed in flows of differing elevations and ages, from about 140 to 3000 years old, on Mauna Loa, Kīlauea, and Hualālai volcanoes on Hawai‘i Island. Invertebrate diversity was evaluated from root galleries and non-root galleries, in situ fluid physicochemistry was measured, and root and bare rock fluids (e.g., water, sap) were

collected to determine major ion concentrations, as well as non-purgeable organic carbon (NPOC) and total nitrogen (TN) content. To verify root identity, DNA was extracted, and three sets of primers were used. After screening for only *Metrosideros* spp., the V4 region of the 16S rRNA gene was sequenced and taxonomy was assigned.

Root fluids were viscous and ranged in color from clear to yellow to reddish orange. Root fluids had 2X to 10X higher major ion concentrations compared to rock water. The average root NPOC and TN concentrations were 192 mg/L and 5.2 mg/L, respectively, compared to rock water that had concentrations of 6.8 mg/L and 1.8 mg/L, respectively. Fluids from almost 300 root samples had pH values that ranged from 2.2 to 5.6 (average pH 4.63) and were lower than rock water (average pH 6.39). Root fluid pH was comparable to soil pH from montane wet forests dominated by ‘ōhi‘a (Selmants et al. 2016), which can grow in infertile soil with pH values as low as 3.6. On Hawai‘i, rain water pH averages 5.2 at sea level and systematically decreases with elevation to pH 4.3 at 2500 m (Miller and Yoshinaga 2012), but root fluid pH did not correlate with elevation, temperature, relative humidity, inorganic and organic constituents, or age of flow. Root fluid acidity is likely due to concentrated organic compounds, sourced as root exudates, and this habitat is acidic for the associated invertebrates.

From 62 root samples, over 66% were identified to the genus *Metrosideros*. A few other identifications of roots from lava tube systems where there had been extensive clear-cutting and ranching included monkey pod tree, coconut palm, *Ficus* spp., and silky oak.

The 16S rRNA gene sequence surveys revealed that root bacterial communities were dominated by few groups, including Burkholderiaceae, as well as Acetobacteraceae, Sphingomonadaceae, Acidobacteriaceae, Gemmataceae, Xanthobacteraceae, and Chitinophagaceae. However, most of the reads could not be classified to a specific genus, which suggested that the rhizobiome harbor novel diversity. Diversity was higher from wetter climates. The root communities were distinct from those described previously from ‘ōhi‘a flowers and leaves (Junker and Keller 2015) and lava tube rocky surfaces (Hathaway et al. 2014) where microbial groups were specifically presumed capable of heterotrophy, methanotrophy, diazotrophy, and nitrification. Less can be inferred for the rhizobiome metabolism, although most taxa are likely aerobic heterotrophs. Within the Burkholderiaceae, there were high relative abundances of sequences affiliated with the genus *Paraburkholderia*, which includes known plant symbionts, as well as the acidophilic genera *Acidocella* and *Acidisoma* from the Acetobacteraceae, which were retrieved predominately from caves in the oldest lava flows that also had the lowest root pH values. It is likely that the bacterial groups are capable of degrading exudates and providing nutritional substrates for invertebrate consumers that are not provided by root fluids (i.e., phloem) alone.

As details about the biochemistry of ‘ōhi‘a have been missing, characterizing the rhizobiome from lava tubes will help to better understand potential plant-microbe-invertebrate interactions and ecological and evolutionary relationships through time. In particular, the microbial rhizobiome may produce compounds used by invertebrates nutritionally or that affect their behavior, and changes to the rhizobiome in response to

environmental conditions may influence invertebrate interactions with the roots, which could be important to combat climate change effects or invasive species introductions.

Keywords

subsurface, rhizobiome, *Metrosideros polymorpha*, cave, bacteria, invertebrate

Presenting author

Annette Engel

Presented at

2nd Joint Symposium of Environmental Biogeochemistry and Subsurface Microbiology

Acknowledgements

The endemic Hawaiian cave fauna exist nowhere else on Earth. The lava tubes at the center of our research are on the ancestral lands of the Native Hawaiian people of the sovereign Kingdom of Hawai'i, which is illegally occupied by the United States. As settlers and guests, we are grateful to work and study in Hawai'i, and are thankful for the opportunity to access the lava caves. We acknowledge that the Native Hawaiians are the original caretakers of the land, but we are committed to practice mālama ʻāina with aloha with them to improve and advance conservation efforts to protect these sacred places. Mahalo to the Cave Conservancy of Hawai'i, the Hawai'i Speleological Survey, and the HICAVES team for assisting in the field work. We also thank our collabortion with The Nature Conservancy and funding from the National Science Foundation (DEB-2204671 to A.S.E. and DEB-2204670 to M.L.P. and R.A.C.).

Conflicts of interest

The authors have declared that no competing interests exist.
Disclaimer: This article is (co-)authored by any of the Editors-in-Chief, Managing Editors or their deputies in this journal.

References

- Hathaway JM, Garcia M, Balasch MM, Spilde M, Stone F, Dapkevicius ME, Amorim I, Gabriel R, Borges PV, Northup D (2014) Comparison of Bacterial Diversity in Azorean and Hawai'ian Lava Cave Microbial Mats. Geomicrobiology Journal 31 (3): 205-220. https://doi.org/10.1080/01490451.2013.777491

- Junker R, Keller A (2015) Microhabitat heterogeneity across leaves and flower organs promotes bacterial diversity. FEMS Microbiology Ecology 91 (9). https://doi.org/10.1093/femsec/fiv097
- Miller J, Yoshinaga A (2012) The pH of Hawaiian precipitation a preliminary report. Geophysical Research Letters 8 (7): 779-782. https://doi.org/10.1029/gl008i007p00779
- Selmants P, Adair K, Litton C, Giardina C, Schwartz E (2016) Increases in mean annual temperature do not alter soil bacterial community structure in tropical montane wet forests. Ecosphere 7 (4). https://doi.org/10.1002/ecs2.1296