



H128 A Survey of Bacterial Communities in Soil Associated With Porcine Remains

Denise Wohlfahrt, BS, Virginia Commonwealth University, Richmond, VA 23284; Kailey Babcock, BS*, Richmond, VA 23221; Shane Woolf, MS, Richmond, VA 23220; Tal Simmons, PhD, Virginia Commonwealth University, Richmond, VA 23284; Baneshwar Singh, PhD, Virginia Commonwealth University, Richmond, VA 23284

Learning Overview: After attending this presentation, attendees will better understand how porcine remains affect the soil microbiome. This information will assist forensic scientists in improving the estimation of the Postmortem Interval (PMI) based on microbial evidence.

Impact on the Forensic Science Community: This presentation will impact the forensic science community by providing information on the bacterial communities associated with carrion-deposited soil samples and their importance in forensics. This information will help attendees gain a better understanding of carrion resource utilization and improve the precision of PMI estimation using entomological and bacterial evidences.

Estimating the PMI is one of the crucial parts of a death investigation. Immediately after death, algor, rigor, and livor mortis are often used as a PMI estimation method. However, these methods often lose their reliability after 72 hours postmortem. In recent years, soil has become a useful tool in determining this timeline. As bodies are often buried underground or deposited on land, soil may serve as a logical means of estimating time since death. Soil acts as a sink or filter for water, gases, and other resources in the environment, potentially making it a new identifying factor in estimating the PMI. As decomposition progresses to the bloat stage, it releases nutrients and microbes into the underlying soil, altering the soil microbial community composition.¹ Understanding how the microbiome of soil is altered as time progresses will aid the forensic science community in enhancing PMI estimation methods.

This study characterized bacteria associated with carrion-deposited soil samples by using 16S recombinant DNA (rDNA) MiSeq® sequencing. Six porcine cadavers were left to decompose naturally on top of pristine soil. Soil samples were collected directly under the body and three meters away from the body. This repeated each day for a week, then once a week after that for a total of eight weeks. DNA was extracted from the soil samples using the DNeasy® PowerSoil Kit and was amplified and sequenced for Variable region Four (V4) of 16S rDNA using the dual-index MiSeq® sequencing strategy as described by Kozich et al.² Sequences were then analyzed using mothur version 1.39.4, and statistical analysis was performed using R version 3.4.0.^{3,4}

In general, the total DNA quant value obtained from soil under porcine remains was higher than the DNA quant value obtained from soil at the control sites. After all quality control steps, a total of 4,518,957 sequence reads (average read per sample=55,820) was generated from all samples. Preliminary results show that Acidobacteria and Actinobacteria are among the most prominent phyla observed in soil associated with porcine remains.

Reference(s):

1. Singh B., Minick K.J., Strickland M.S., Wickings K.G., Crippen T.L., Tarone A.M., Benbow M.E., Sufirin N., Tomberlin J.K., Pechal J.L. Temporal and Spatial Impact of Human Cadaver Decomposition on Soil Bacterial and Arthropod Community Structure and Function. *Front Microbiol* 2018;8:2616.
2. Kozich J.J., Westcott S.L., Baxter N.T., Highlander S.K., Schloss, P.D. Development of a Dual-Index Sequencing Strategy and Curation Pipeline for Analyzing Amplicon Sequence Data on the MiSeq Illumina Sequencing Platform. *Applied and Environmental Microbiology* 2013;79(17):5112–5120.
3. Schloss P.D. et al. Introducing mothur: open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Appl Environ Microbiol* 2009;75:7537-7541.
4. R: A language and environment for statistical computing. (*R Foundation for Statistical Computing*, <http://www.R-project.org>, Vienna, Austria., 2011).

Soil Microbiome, Necrobiome, 16S rDNA