H41 An Examination of Postmortem Interval Relative to Microbial Biomass of Soil at the MSU Forensic Science Research Facility Plot

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After attending this presentation, attendees will be informed of the Mississippi State University (MSU) Forensic Science Research Plot and the interdisciplinary research taking place at the facility. The focus of this presentation will be on recent controlled decomposition studies examining the relationship of Postmortem Interval (PMI) to the microbial biomass of soil and bone.

This presentation will impact the forensic science community by contributing new data to the expanding knowledge base of microbial biomass in relation to PMI of various sample types (e.g., bone, tissue, and soil) and provide a new dataset to compare to recent studies at the Anthropological Research Facility in Tennessee as well as other research units across the United States.

The MSU Forensic Science Research Plot conducts interdisciplinary studies aimed at understanding PMI, decomposition, and taphonomic processes in Mississippi. Collaboration with the Mississippi Agricultural and Forestry Experiment Station (MAFES) allows access to non-euthanized porcine (*Sus scrofia*) samples and enables controlled research throughout the year.

To develop a baseline biomass reference sample, three porcine experiments were monitored throughout active decomposition to mummification/skeletonization. The pigs were enclosed in fencing to reduce scavenging which may alter soil composition. Basic observations and notes were maintained, as well as serial photography, and included into the Porcine Taphonomic Database. Two juvenile samples (approximately 25 – 30 pounds) were placed at the facility during the fall of 2011 and became mummified/skeletonized within two weeks due to high temperatures. The third sample (approximately 200 pounds) was placed in the winter of 2012 and became partially mummified and skeletonized throughout the spring. The high fat content, as well fluctuating temperatures, extended the decomposition interval. Prior to placing the pigs, control soil samples were collected. Daily soil samples were collected beneath the center of each specimen during active decomposition. As a final step, rib samples were collected and pulverized (200mg) for quantitative PCR analysis.

A quantitative real-time PCR for the evaluation of bacteria and fungi from each sample was performed. Standard quantification curves were created with universal primers targeting a 200-base pair fragment of the 16s rRNA gene from *E. coli* and a 300-bp fragment of the fungal ITS region from *Fusarium solani*. Five serial dilutions of known concentrations of PCR products were generated and run in triplicate for a single standard curve and used to estimate DNA concentration from the unknown samples.

With the same primers, test soils, bone specimens, and standard curve samples were analyzed. Asymmetrical cyanine dye was used for bacteria and a fluorescent DNA binding dye was used for fungi. Quantitative-PCR efficiencies of 94% and 77% were obtained for bacterial and fungal amplification, respectively.

Due to the climate in Mississippi, decomposition is generally accelerated and, therefore, bacterial growth is accelerated. Bacteria naturally reproduce rapidly (every 15 to 20 min) in soil and can be greatly influenced by nutrient availability and physiochemical environment, such as temperature and moisture. The first two pigs displayed a dramatic increase in bacteria within the first week of decomposition and then declined. Pig 1 became mummified and experienced a gradual decrease in bacteria while Pig 2 became skeletonized with a rapid decline of bacteria. The differential results may be due to lingering nutrients provided by the mummified remains. Pig 3 is currently being analyzed and the results are expected to mirror the results from Pig 1 and 2.

Although quantitative PCR has been established as a useful tool for measuring microbial biomass as a means for determining PMI, more regional studies are needed to better understand the relationship between bacteria and decomposition in different ecological settings. Researchers with the MSU Forensic Science Research Plot have collaborated with the National Museum of Health and Medicine to develop a baseline reference sample for Mississippi. Ultimately, understanding the rates of change in bacteria throughout the decomposition process will aid in determining PMI in future forensic cases.

Forensic Taphonomy, Postmortem Interval, Microbial Biomass

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