



## Pathology/Biology Section - 2016

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### H10 The Six Little Pigs: Estimation of Long-Term Postmortem Interval (PMI) Based on Bacterial Community Succession in Porcine Remains

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After attending this presentation, attendees will better understand porcine decomposition, the roles of bacteria and insects during the process, and how bacterial community succession associated with porcine remains can be useful for human PMI estimation.

This presentation will impact the forensic science community by providing further insight into the potential usefulness of changes in relative bacterial abundance and, subsequently, more accurate long-term PMI estimation.

Bacteria, like insects, drive the vertebrate decomposition process.<sup>1,2</sup> Previous studies have demonstrated changes in human and porcine bacterial community structure over time.<sup>1,3,4</sup> In this study, six porcine cadavers were placed in an open field at the Virginia Public Safety Training Center (VPSTC) in Hanover, VA. Primary arthropod colonizers (e.g., blow flies, flesh flies, beetles, etc.) had free access to all six carcasses. Minimum 10m distance was maintained between any two carcasses. Sterile cotton swabs were used for microbial sample collections from skin regions of each cadaver. Swabs were collected daily through day 7 (T7), every other day through day 15 (T15), and once weekly through day 61 (T61). DNA was extracted using a phenol:chloroform:isoamyl alcohol organic extraction method.<sup>5</sup> Two variable regions (V3-V4) of the 16S ribosomal RNA (rRNA) gene were amplified using barcoded primer pair 341F and 806R for MiSeq® sequencing.<sup>6</sup>

Preliminary assessment of primary insect colonization showed that adult flies from family Calliphoridae (*Phormia regina*, *Lucilia* spp., *Cochliomyia macellaria*) and beetles from families Silphidae (*Necrophila americana*, *Necrodes surinamensis*, *Oiceoptoma novaboracense*), and Staphylinidae (*Creophilus maxillosus*) were observed in large numbers during early decomposition stages (fresh and bloat). Egg masses, fly larvae, and a single rove beetle larva were observed and collected within 24 hours of pig placement. In addition to those aforementioned species, beetles from family Cleridae (*Necrobia rufipes*) were noted during later decomposition stages (active decay, advanced decay, and dry). Overall, *Pharma regina* and *Creophilus maxillosus* were the most prevalent fly and beetle species throughout this study. Maggot masses began migrating on or about days 3-4 with peak migration on days 7-8. Vultures were also observed at the test site on days 7 and 8.

In conclusion, this study will expand knowledge regarding bacterial community succession in porcine remains and will further elucidate the usefulness of bacterial succession associated with porcine remains in estimation of long-term PMI. This study will also help determine how bacteria associated with porcine remains change in different geographical regions, identify indicator bacterial species for each stage of decomposition, and clarify which species are common in all geographical locations.

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#### Bacteria, Forensics, Postmortem Interval

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