



SUCCESS STORY

NIJ and Multi-Institute Academic Team

Establishing a “Microbial Clock” to Improve Time of Death Prediction



“This research clearly demonstrated that there is potential to develop microbiome tools for estimating time of death. Developing connections with the right interdisciplinary collaborators was instrumental to the success of this project.”

*—Jessica Metcalf, Assistant Professor,
Colorado State University*

Problem and Solution Synopses

The postmortem interval (PMI) is the time that has elapsed since a person’s death. Determining the PMI at a crime scene may help to recreate the victim’s timeline and movements, support or disprove a suspect’s alibi, or corroborate evidence collected from autopsies. Several methods are used to estimate PMI at different stages of decomposition. Lack of postmortem changes can be used to infer a PMI up to approximately 24 hours postmortem, and forensic anthropologists may calculate the calcium-to-carbon ratio and bone volume to predict the general time of death based on decades old skeletal remains.

Forensic entomology, which relies on insect stage development, is commonly used to estimate PMI for remains that are several days to a year old. In rare cases, entomology can estimate PMI after numerous years. However, this method can be less reliable when the victim has extensive trauma or is in an area with significant weather fluctuations, including temperature and precipitation. Thus, determining the time of death on intermediate time scales (from days to months) is a significant challenge for the forensic community.

Dr. Rob Knight and colleagues developed a PMI estimation method based on the composition of microbial communities present on the body and within close-proximity soil samples compatible to intervals of days to

months. The team characterized temporal changes in microbial communities on the skin of mice pigs, and human cadavers at fixed PMIs. Knight and colleagues collected, sequenced, and classified 223 abdominal cavity, skin, gravesoil, and no-corpse soil samples, creating a dataset of over 2.9 million 16S rRNA sequences that represent 4505 Operational Taxonomic Units (OTUs, a unit used to represent microbial “species” at various taxonomic levels in the microbial community based on sequence similarity)¹. Next-generation rRNA sequencing demonstrated that groupings of microbial species change as a body goes through the various stages of decomposition.

The team used the data to develop a regression model for PMI estimation and concluded that microbial communities changed predictably as the corpse decomposed.

Key Benefits

- ▶ Demonstrates that microbial communities can accurately determine body PMI up to 48 days since death, with an average error of about 3 days.
- ▶ Relates “universal” microbial markers for determining PMI, despite the environment.
- ▶ Identifies environmental conditions, such as temperature, location, and season, that can affect microbial communities’ composition.
- ▶ Demonstrates the potential for microbial signatures to identify clandestine gravesites.



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Key References

[Science Journal Publication](#)
[eLife Publication](#)
[NCJRS Final Report](#)

More Information

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Disclaimer

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NIJ-Funded Research

NIJ support facilitated this interdisciplinary collaboration among anthropology, microbiology, biochemistry, and forensic science researchers from the University of Colorado-Boulder, Sam Houston State University, the University of Nebraska-Lincoln, and Chaminade University of Honolulu. The combined research team collaborated to build the regression model by leveraging field collections, laboratory connections, and resources (such as DNA sequencing instrumentation).

Bringing Research to Practice

- ▶ Conducted a feasibility study focused on mice and swine to gather initial results demonstrating universal decomposition stages across mammalian corpse species and social types. The team used 16S and 18S rRNA gene sequence analysis to discover the patterns of microbial community diversity.
- ▶ Validated the viability of human microbe communities with four cadavers across two seasons at Sam Houston State University. Confirmed that seasonal variation affects the microbial community composition, which will be an important focus for future research.
- ▶ Developed a regression model that predicted PMI using Random Forests with bacterial and microbial eukaryotic data, validating the use of microbes to estimate PMI.
- ▶ Communicated results in the journal *Science*, 10 other publications and books, and presentations at professional meetings (e.g., the American Society of Human Genetics).

The Future

- ▶ The team seeks to refine their model by understanding the extent to which environmental conditions, such as seasons and geographic regions, affect the ability to accurately predict PMI. They received additional research funding for an expanded feasibility study modeling 36 human bodies from outdoor sites in Tennessee, Texas, and Colorado.
- ▶ Dr. Pieter Dorrestein at the University of California-San Diego will research chemical compositions of the microbial communities.
- ▶ The researchers plan to test whether postmortem predictions can be made for longer time periods based on the microbes present in bones after an extended decay period.
- ▶ The team intends to construct an open-source reference database to store the models for future trainings and future work with law enforcement and crime scene investigators.

¹Metcalfe JL, Wegener Parfrey L, Gonzalez A, et al. A microbial clock provides an accurate estimate of the postmortem interval in a mouse model system. Kolter R, ed. *eLife*. 2013;2:e01104. doi:10.7554/eLife.01104.

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