

# 'Soil'ent witness: using grave soil microbial communities to aid forensic investigations

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After death, the human body becomes an active microbial ecosystem, undergoing a predictable succession that offers valuable clues for forensic investigation. This post-mortem shift, known as the thanatomicrobiome, unfolds as endogenous microbes migrate, proliferate, and respond to changing conditions within the cadaver. In a terrestrial environment, cadaveric fluids and microbial communities seep into the surrounding soil, forming a cadaver decomposition island that reshapes local chemistry and biodiversity. These interactions create the necrobiome—a dynamic ecological network whose temporal patterns can help estimate the post-mortem interval or time since death. Yet decomposition is profoundly influenced by temperature, moisture, soil type, insect activity, and other environmental factors, making standardisation a major challenge. As sequencing technologies advance and interdisciplinary research expands, decoding the post-mortem necrobiome and grave-soil microbiology offers a promising complement to the current forensic toolkit.

Prior to the 21st century, microbiology was seldom associated with forensic sciences. Fast forward to 2001, when letters laced with *Bacillus anthracis* spores were mailed across the United States. The anthrax attacks remain one of the most significant biological crimes in modern history, thrusting microbial forensics into the public consciousness and catalysing the development of a specialised field focused on tracing microbial agents used in biocrime, bioterrorism, and biowarfare.

Since then, research has increasingly turned toward harnessing the power of microbes to support traditional forensic investigations—the domain of forensic microbiology. Unlike microbial forensics, this branch isn't about identifying the source of a malicious pathogen. It's about using microbial communities as evidence to locate, to identify, to time, and to contextualise. Microbial evidence is not a replacement for DNA, fingerprints, or entomology. Instead, it complements them. The strength of forensic science lies in convergence—multiple independent lines of evidence pointing to the same conclusion. Microbiology adds another angle, another lens, another line of enquiry.

## Microbes as evidence: expanding the forensic toolkit

Microbiology can support a surprising range of forensic questions. Microbes can help determine the cause of death, identify bodily fluids, link individuals to locations, and even provide personal identification. Much of this stems from the concept of microbiomes—the communities of microorganisms that inhabit specific environments, from soil to seawater to the human body.

Diatoms offer a striking example (Figure 1). These microscopic algae vary dramatically between aquatic environments, making them useful for geolocation. Their presence in lung tissue can also help determine whether someone drowned, depending on whether they entered passively post-mortem or through active aspiration. The same principle applies to bacteria and fungi: soil microbiomes differ so distinctly across locations that soil from a shoe or tyre can be matched to a specific environment.



**Figure 1.** Examples of different diatom structures from a single sample under the microscope

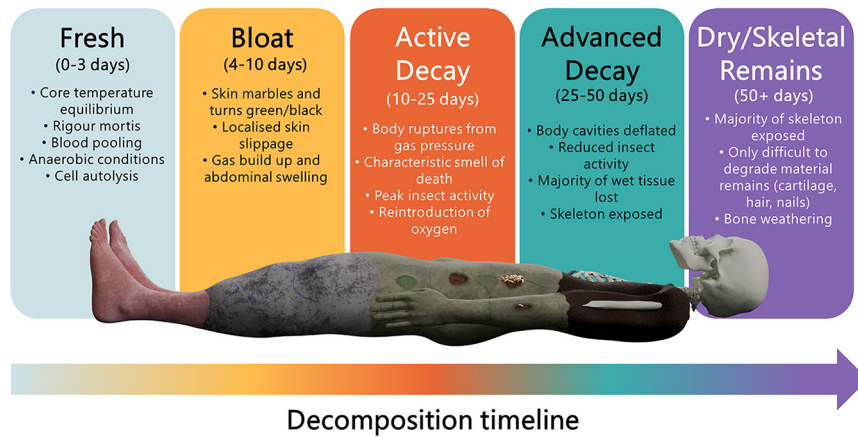


Figure 2. Schematic of the five categorised stages of mammalian decomposition

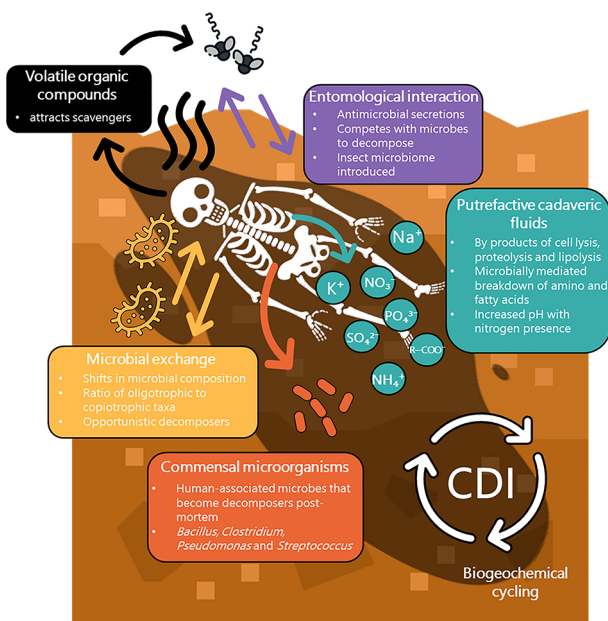


Figure 3. Schematic of the creation of the CDI

The human microbiome is even more individualised. Bacterial cells outnumber human cells by roughly ten to one, and our microbial communities are shaped by everything from diet to detergent choice. Under Locard's Exchange Principle—the idea that every contact leaves a trace—microbial transfer becomes a powerful tool. Even anatomical niches, such as the genital microbiome, have been used in sexual assault investigations to identify bodily fluids or link individuals.

Nevertheless, the most fascinating microbial clues emerge not during life, but after death.

### Life after death: the post-mortem microbial clock

The post-mortem microbial community within the cadaver is known as the thanatomicrobiome, encompassing internal organs, blood, and other tissues. Upon death, the previously unique personal microbiome begins to change, with microbial succession unfolding in a rather predictable manner.

When the heart stops, the body's internal environment changes instantly as fresh stage decomposition sets in (Figure 2). Oxygen

disappears, tissues become anaerobic, and endogenous gut bacteria such as *Clostridium* flourish, migrating into organs that were sterile ante mortem. As microbial metabolism intensifies, gases accumulate, and the body enters the bloat stage, swelling and marbling with by-products such as methane and hydrogen sulphide.

Eventually, pressure forces fluids and gases out through natural openings or unnatural ones—reintroducing oxygen and reshaping the microbial landscape in a process known as purge or rupture. This marks the onset of active decay, a period defined by rapid microbial turnover and competition with both insect scavengers and external microbial communities. As tissues break down and nutrients diminish, the process slows, giving way to advanced decay and finally skeletonisation. By this stage, only the hardest microbes remain, including taxa from the Firmicutes and Bacteroidetes phyla, capable of metabolising the last recalcitrant materials such as bone, skin, and hair.

A brief aside: the microbial community inhabiting the external surfaces of the cadaver—the skin, hair, and surrounding tissues—is known as the epinecrobium and follows its own successional trajectory.

Together, these ecological shifts act as a biological clock. Interpreting them can provide crucial insights into time since death, allowing investigators to estimate the post-mortem interval (PMI). PMI is a cornerstone of criminal proceedings, essential for establishing timelines, identifying victims, and validating or disproving suspect alibis.

### Beneath the surface: how bodies reshape soil

In terrestrial settings, when a body is deposited on or in the ground, it fundamentally alters the surrounding environment. Cadaveric fluids, rich in nutrients and microbes, seep into the soil, creating a localised ecosystem known as the cadaver decomposition island (CDI). These fertile islands induce temporal changes in the physical, biochemical, and—most crucially—ecological properties of the soil. This ecological landscape is referred to as the necrobiome.

The CDI beneath and around the corpse becomes enriched with nitrogen, carbon, lipids, and other by-products of decay and often increases in alkalinity (Figure 3). The first shift in grave-soil microbial communities is typically a transition from a broad, nutrient-limited oligotrophic community to one dominated by copiotrophs that thrive in nutrient-dense, anoxic, and pH-variable conditions. This means a reduction in soil phyla such as Acidobacteria and Verrucomicrobia and an increase in early coloniser genera like *Pseudomonas* and *Bacteroides*, native to both host and environment.

As decomposition progresses from bloat to active decay, the grave-soil community transitions again, favouring taxa capable of metabolising the large quantities of organic matter released from the cadaver, such as *Sphingobacterium* and *Alcaligenes*. Finally, as remains skeletonise, the community shifts towards hardy, often spore-forming genera such as *Bacillus* and *Xanthomonas*.

While this is only the bare bones of the field, tracking these ecological interactions can provide PMI estimations with remarkable accuracy—sometimes within hours, even for remains up to a decade old.

## Filling the gaps: what research still needs to solve

So, everything is straightforward and works exactly like that? Perfect!

Except, of course, not; this is science—forensic science.

Mammalian decomposition is neither tidy nor linear. It is influenced by a dizzying array of factors, from corpse weight and condition to temperature, burial depth, and entomological accessibility. Microbes are extremely sensitive to their environment, and a wealth of research has focused on isolating the effects of such confounding factors on necrobiome accuracy. Warm conditions accelerate microbial activity, while cold slows it dramatically. Soil type and matric potential—the soil's ability to retain water—can lead to preservation in dry or extremely waterlogged conditions, still mediated by microbial activity.

Entomology adds another layer of complexity. Insects and microbes compete for resources, but insects also secrete antimicrobial compounds that can alter microbial succession. Although entomology remains the gold standard for PMI estimation, it falters in situations where insect access is delayed or prevented, such as burial, wrapping, clothing, exhumation, or scattering of remains. In these anomalous but common forensic scenarios, microbes may provide crucial insights.

Current research often relies on pigs as human analogues, but the extent to which these findings translate to human decomposition remains an open question. Soil is inherently heterogeneous, and sampling methods, sequencing pipelines, and analytical approaches vary widely. Standardisation is still in its infancy. To make microbial evidence courtroom-ready, the field needs consistent sampling protocols, harmonised sequencing and analysis pipelines, and large, geographically diverse microbial databases.

Technological advances are helping. Forensic microbiology has moved from culturing to PCR to high-throughput sequencing, and nanopore technologies now offer real-time data that could be transformative for forensic timelines. Yet cost, accuracy, and accessibility remain barriers. Artificial intelligence and machine learning offer promising tools for interpreting complex necrobiome data, but they require robust, standardised datasets that reflect real-world conditions. Many bottlenecks remain unresolved.

## Looking forward

Forensic microbiology sits at the intersection of ecology, technology, and justice. It is a field still finding its footing, but its potential is enormous. Microbes can offer insights when insects are absent, when DNA is degraded and when bodies are exhumed, burned, fragmented, or buried—though these applications still require further research. They can help reconstruct timelines, track movement, and reveal hidden interactions between the body and the environment.

The future of forensic microbiology is not about replacing traditional methods. It's about enriching them—adding another layer of evidence, another perspective, another way of accessing the stories of the 'soil-ent' witness left behind. Beneath our feet, in the soil, microbes have always been there, keeping time. We are only just learning how to read the clock.

## Further reading

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## Author biographies



**Phebie Watson** completed her undergraduate degree in biomedical sciences at the University of Liverpool in 2019, followed by an MSc in forensic bioscience at Liverpool John Moores University, which inspired her doctoral research in forensic ecogenomics, completed in 2026. As an early-career academic, her work focuses on post-mortem microbial ecology, remains exhumation and reburial, and the implications of land reuse for forensic and environmental practices. Alongside her research, Phebie is committed to embedding equity, diversity, and inclusion; widening participation; and opening research principles in teaching and scientific communication, particularly within the applied and interdisciplinary landscape of forensic science.



**Komang Ralebitso Senior** is a Reader in Microbial Ecology at Liverpool John Moores University. She completed a BSc in environmental microbiology at the University of Natal, South Africa, where she also obtained her doctorate. Her research centres on harnessing microbial community structure and function for sustainable environmental management and forensic ecogenomics, with particular interests in biochar-mediated soil bioremediation and post-mortem microbial ecology. She has supervised multiple PhD researchers across these areas. Komang is also Associate Dean (Diversity and Inclusion) for the Faculty of Health, Technology, Innovation, and Science, supporting equitable teaching, research, and enterprise practices.