

A192 Operation Invisible Dead: A Blind Study of Soil Identification Using a Microbiological Database

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After attending this presentation, attendees will understand more thoroughly the application of soil microbial community profiling and the potential use of a soil database to identify microbiological soil evidence.

This presentation will impact the forensic science community by exploring the use of Capillary Electrophoresis-Single Strand Conformation Polymorphism (CE-SSCP) to identify soil samples using a soil microbiological database.

Soils have been used as trace evidence for well over a century. As technology has advanced, so has the value of soil evidence. Most recently, soil microbial community profiles have been used to provide an accurate discriminating factor between soil samples of known and unknown origin. The use of microbiological physical evidence can detect the uniqueness or similarity of the soil evidence when compared to the collected reference soil samples from the crime scene. The current literature contains several papers that test the accuracy and limitations of these methods; however, no one has yet done a case study on how well soil microbial community identification works with a database. Thus, a blind study was set up with a database to test the sensitivity and reliability of the soil microbial evidence.

The blind study was set up where a "criminal" dug a shallow grave using a shovel. Upon completion of the shallow grave, the "criminal" then placed the shovel and one shoe into individual paper bags for evidence collection. The other shoe was worn back to a place of residence, and then placed in a paper evidence bag. The shovel and two shoes were allowed to air dry at room temperature for 48 hours. The soil was collected from these objects by a different undergraduate researcher. This analyst conducted all sampling and handling without knowledge of the location of the grave. DNA from the soil was extracted using a commercially available DNA extraction kit. Amplification of the extracted DNA was done with fluorescently labeled primers and a proofreading enzyme for the V3 region of the 16S ribosomal DNA. Using CE-SSCP the soil microbial community was characterized. This method yields electropherograms of the microbial community DNA specific to the bacteria present in the soil sample. The profiles of the soil microbial community were compared to a database of four soil plots in southeastern Nebraska over three different seasons. The database was made from a previous research project and holds 72 profiles. The samples in the database were processed with DNA extracted within 24 hours of removal of soil from the ground at a depth of 0cm to 5cm. DNA extraction, Polymerase Chain Reaction, and CE-SSCP for the samples in the database were same way as the samples from this study. It is understood that this is a relatively small database, but it can help to initiate the usefulness of a soil microbiological database.

The database has been made and the samples have been processed via CE-SSCP. The results of the study are two-fold. When visually examining the soil microbial electropherograms from CE-SSCP, the location of the grave was correctly identified. However, to ensure a reliable outcome is obtained, statistical analysis must be conducted. R software will be used to statistically compare and contrast the samples of interest to the database. **Soil, CE-SSCP, Microbial DNA**