



B151 DNA Profiling for Forensic Soil Comparisons

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After attending this presentation, attendees will have been presented with the preliminary finding of a novel approach to differentiating soil samples for forensic applications.

This presentation will provide the forensic community a basis for further research into the use of DNA profiling technology to identify and/or compare forensic soil samples.

This paper will present the results of a preliminary investigation that aimed to develop a reliable method of microbial DNA profiling to differentiate between surface soil samples. Samples collected from given locations with different ecological characteristics, such as uncultivated fields, forests, and sand dunes were compared, and the variation within and between ecologically different sites was determined. The implications of this research for forensic examination of soil evidence will be discussed.

Forensic soil comparisons can be used to associate a person found with soil on their clothes or possessions with a crime scene. Criminals often commit crimes, or deposit evidence, in isolated areas such as fields or forests. In most forensic cases where soil comparisons would be considered, a crime would have been committed in a known location (e.g., a field), and a suspect apprehended, who was found to have soil on their clothes, shoes, or possessions (such as a shovel or vehicle tyres). It would then be useful to compare these soil samples with control samples collected from the scene to determine if the suspect's samples could have originated from that location.

Most current forensic soil comparisons are based on geological properties. However, since the majority of forensic laboratories cannot afford an expert geologist, such analyses are rarely performed for routine casework, but are reserved for particularly high-profile, serious crimes. It has been suggested that DNA profiling of the microbial community in soil may be a potential method for soil comparisons. Currently, microbial DNA profiling techniques such as Amplified Ribosomal DNA Restriction Analysis (ARDRA), Denaturing Gradient Gel Electrophoresis (DGGE), Thermal Gradient Gel Electrophoresis (TGGE), Single Strand Conformation Polymorphism (SSCP), and Terminal Restriction Fragment Length Polymorphism (TRFLP) are being used to study the diversity of soil microorganisms. However, only ARDRA and TRFLP can be performed on the equipment normally available in the forensic laboratory. An ARDRA profile of a community with many different species tends to become too complex for easy interpretation, as one species can contribute 4-6 restriction fragments. However, TRFLP involves labelling the 5' end of the PCR primers with a fluorescent dye, so that only the terminal restriction fragments are detected following gel electrophoresis. This means that TRFLP yields less complex profiles where every visible band (fragment) represents a single 'ribotype.' Additionally, TRFLP analysis relies on the detection of fluorescently labelled primers, which is how human DNA profiling is performed. Therefore, all necessary equipment and expertise should be available in forensic laboratories, making this technique potentially more applicable. Moreover, the technical nature of TRFLP would allow high throughput of samples, as required for forensic applications.

As a pre-requisite for TRFLP analysis, various DNA extraction methods suitable for a range of soil types were evaluated. An appropriate procedure was developed for rapid and reliable extraction of DNA representative of the total microbial community. PCR was performed using fluorescently labelled universal primers targeting the 16S ribosomal RNA genes of the bacterial soil community. TRFLP analysis was then performed using the ABI Prism 373 gel electrophoresis system and GeneScan software. The resulting profiles were standardized and binary code was assigned to the peak profiles. Similarities between these binary codes were determined using the Jaccard coefficient to create similarity matrixes and dendograms were used to display the results of agglomerative hierarchical cluster analysis.

Preliminary results suggest that samples from within a specific ecological site (e.g., a field) show a higher similarity to each other than to those from other ecological sites (e.g., a forest). These results may allow the determination of characteristic profiles that will facilitate identification of ecologically different sites so that a given sample collected from a suspect could be identified as originating from, for example, a field, rather than a forest. Further research will explore small-scale spatial variation at various sites and the potential of these techniques to link a suspect's sample more precisely to its origin, and thus provide stronger circumstantial evidence.

Soil, Microbial DNA Profiling, TRFLP