B145 Optimized Recovery of DNA and Protein Components From Contact Traces on Fired and Unfired Cartridge Casings

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After attending this presentation, attendees will be aware of potential proteomic applications in forensic biology and realize that it is possible to extract DNA and protein from touch evidence without compromising yields for either type of biological material.

This presentation will impact the forensic science community by alerting crime laboratory scientists to a new tool for touch evidence analysis. This approach may ultimately improve source identification for contact traces on cartridge casings.

Current DNA typing success rates for spent cartridge casings have been reported as less than 10%, and recent studies have shown that metal surfaces, specifically brass, can be detrimental to DNA.[1-3] These types of probative but low-level samples would benefit from supplemental testing of protein polymorphisms, also known as Genetically Variable Proteins (GVPs), as has been described for keratin in hair samples.[4] Proteins are expected to be more abundant than DNA and may be less affected by heat and mechanical pressure. This data reveal that it is possible to extract DNA using a mass spectrometry-compatible lysis buffer and sequencing grade trypsin as the protease. The incubation buffer consists of 0.01% ProteaseMAX™ and 5mM DTT in 50mM NH_4CO_3. Extraction proceeds with 20 minutes of denaturation at 56°C, three hours at 37°C for trypsin digestion, and EMD Millipore™ MW100 membrane filtration. The concentrated DNA fraction will be above the membrane and mass spectrometry-ready peptides are in the flow through. Protein sequencing was performed by reversed-phase liquid chromatography using Easy-nLC™ 1000 High-Performance Liquid Chromatography (HPLC) and Q Exactive™ Orbitrap™ mass spectrometer. Polymerase Chain Reaction Short Tandem Repeat (PCR STR) testing utilized the Identifiler® Plus multiplex and a 3500 Genetic Analyzer. The method was first established using sebaceous-rich fingerprints on glass slides, then tested for fired and unfired 9mm nickel, aluminum, steel, and brass cartridges.

Trypsin digest DNA yields were similar to a standard proteinase K method and DNA was suitable for STR typing. Peptide fractions could be injected on the HPLC without further modification. Data revealed significant differences for protein and DNA recovery before and after firing and between the larger steel cartridges and the other three metals. Even samples with no detectable DNA still yielded peptide peaks, with the average of identified proteins ranging from 27-74 for unfired and 12-30 for fired casings. Extraction negatives were generally negative, but sampling uncleaned cartridges straight out of the box, without deliberate touching, showed high levels of protein background. This needs further investigation. Parallel sets of ten unfired touched brass cartridges were tested using dry collection with tape versus wet swabbing. While tape lifts on average did not recover more DNA, STR typing rates were slightly improved with six full or good partial profiles for tape-lifted samples compared to five for the swabbed samples. Supplemental peptide testing has the potential to increase the power of discrimination for low-level samples. The same proteomic data can also be used to infer body fluid and/or species attributions.

Reference(s):

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Cartridge Casings, Touch DNA, Protein Sequencing

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