

# **The Development of an Experimental Setup and Recovery of Biological Evidence from Bullets for DNA Analysis<sup>1</sup>**

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After attending this presentation, attendees will gain insight into the challenges that face DNA recovery from fired bullets.

This presentation will impact the forensic science community by providing a protocol for evaluating biological evidence on bullets.

The recovery of DNA profiles from firearm ammunition is of specific interest to law enforcement and the criminal justice system. Swabbing fired projectiles can be of high importance in the systematic and hypothesis-driven reconstruction of shooting scenes.

In incidents of mass shootings or when the crime scene scenario is in question, bullets bearing biological samples from “touch” or from penetrating wounds can become lodged into structures such as walls of various compositions, seat cushions, flooring, and other strata. DNA on fired bullets endures a severe environment of heat and friction, and inhibitors from swabbed bullet surfaces can have a negative impact on the amplification efficiency. Therefore, the recovery of DNA from bullet surfaces may be in question due to the quantity and the quality of recovered biological tissue. To address these challenges, the parameters that can significantly impact the persistence and recovery of biological samples on fired bullet surfaces were considered. Priority was given to factors such as bullet composition and calibers, the nature of impact surfaces, and the effect of muzzle-to-target distance.

To initiate this long-term and multi-stage study, an experimental setup compatible with current human Short Tandem Repeat (STR) analysis platforms was devised. This setup consisted of biological tissue targets made of human blood in ethylenediaminetetraacetic acid (EDTA) and 2% agarose mixture at a ratio of 1:40 (blood to agarose). After cooling the agarose, the agarose-blood mixture was prepared and poured into a plastic container to form a 10.5cm-diameter target approximately 1cm in thickness. A portion of the 10cm x 12cm piece of sterile gauze was submerged in the mixture, leaving approximately a 10cm length of gauze exposed for removal. In ~10 min, the target set and adhered to the gauze inside the container. Then, using the extra length of gauze, the targets were removed from the container just before the bullets were fired. Next, one wall section was prepared using one piece of plasterboard and three pieces of dressed yellow pine. One piece of yellow pine was 3.8cm x 23.5cm x 2.44m and one piece of yellow pine was 1.9cm x 23.5cm x 2.44m. The plasterboard was 1.5cm x 23.5cm x 2.44m. The wall section was made by attaching the plasterboard to the pieces of yellow pine with screws and the plasterboard and the two yellow pine pieces were clamped together using C-clamps for transporting the wall section to the firing range. At the firing range, the biological targets were attached by stapling the extra length of gauze to the plasterboard side of the wall section.

Handguns were used to fire .22 caliber, 9mm, and .38 caliber bullets into the targets at a distance of 1m. This distance prevented a significant portion of gunshot residue and gases from entering the target. The wall sections were disassembled and the bullets recovered for DNA analysis. Most of the .22 caliber bullets disintegrated with fragments <1mm in size. Consequently, there was insufficient surface area for swabbing from that caliber. The 9mm and .38 caliber bullets were collected and swabbed with COPAN® 4N6 FLOQSwabs™ in order to recover biological samples for DNA analysis. DNA samples were extracted using the COPAN®

Nucleic Acids Optimizers (NAO), a semi-permeable basket, which retains fluid until centrifuged, with the PrepFiler Express™ and PrepFiler Express BTA™ Forensic DNA Extraction Kits. DNA quantitation was carried out using the Quantifiler® Human DNA Quantification Kit. The AmpFLSTR® Identifiler® Plus PCR Amplification Kit was used for Polymerase Chain Reaction (PCR) and the 3130 Genetic Analyzer™ was used for analysis.

The quantitation results show that over 90% recovery of biological sample was achieved from the 4N6 FLOQSwabs™ swabs. While several swabs from bullets resulted in detectable DNA profiles, none of the profiles were attributable to the target with human blood. The results illustrate the utility of the experimental setup, reveal the challenges of biological evidence recovery from fired bullets, and pave the way for further testing.

<sup>1</sup> Abstract for paper presented at the 66<sup>th</sup> American Academy of Forensic Sciences Conference, Seattle, Washington, February 2014