After attending this presentation, attendees will understand the principles of dcDOP-PCR, and how it can be used to obtain genetic profiles of DNA found on spent shotgun shells.

This presentation will impact the forensic science community because there is a need for validation of a technique that can analyze touch DNA that is severely degraded.

It is recommended that 0.5-2.5 ng of template DNA be used for commercially available STR multiplex amplification kits, however many samples processed consist of low copy number or severely degraded DNA. The goal of this research project was to evaluate a new low copy number technique, dcDOP-PCR, using fired shotgun shells as would be found at a crime scene. Specifically, the dcDOP-PCR technique will be compared to traditional PCR analysis.

According to the FBI in 2005, shotgun crimes accounted for 5% of homicides by firearm. This is second only to homicides by handguns. Typically when investigators arrive at the scene of the crime, the only evidence related to the gun is a spent shell casing. When the shells are handled and loaded into the shotgun, DNA is transferred through the loss of epithelial cells. It has been shown previously that transfer DNA can yield DNA profiles from spent bullet casings from hand guns. It is hypothesized that profiles can also be obtained from spent shotgun shell casings. However, fewer epithelial cells may be shed in the loading of shotgun shells as opposed to bullet casings because much less pressure is needed.

It is difficult to obtain STR profiles from fired shotgun shells due to their limited handling and the high temperatures to which the DNA is subjected. A modification of Whole Genome Amplification will be used known as dcDOP-PCR. This method uses a 10-N degenerate oligonucleotide primer in order to pre-amplify the sample. This produces a greater copy number of template DNA to be used in future PCR reactions and genetic profiling.

A group of twenty subjects consisting of ten males and ten females were selected to load and fire three shotgun shells with a 12-gauge shotgun. The shells were then collected post-ejection with a sterilized wooden stick and placed in a paper bag. Each individual shooter was assigned an identification number used to label all of their fired shell casings. The shotgun shells also underwent a colored coding to designate the first, second, and third loaded shotgun shells from each other. All of the labeling was performed by an individual who was not involved in the analysis of the shotgun shells, creating a blind study. A reference buccal swab was also collected from each individual that was labeled with their identification number. At no time was the subject’s name linked to their identification number during the processing of samples.

An equal number of males and females were used in this experiment in order to study the effect of gender on the completeness of the genetic profile that was obtained. High brass and low brass shotgun shells were also used in the study to determine if the type of shell affected the genetic profile. The two shells differ in the appearance of their outer brass covering. The brass covering extends along the sides of the shotgun shell with a smaller area of plastic exposed on high brass shells. Low brass shells have a small brass ring at the bottom and more plastic exposed on the sides of the shell. This difference in textures may cause a difference in the amount of epithelial cells shed onto the casing. Lastly, the order of loading was analyzed to see if there was a statistical difference between the completeness of the genetic profiles for the first, second, and third loaded shells.

A double swabbing technique was used on the collected shell casings using a 20% SDS solution. The DNA was then extracted using DNA IQ and then pre-amplified using the dcDOP-PCR method. The samples were then amplified using the AW12106, a miniplex for small base pair DNA (< 200 bp) that was developed at Duquesne University. AW12106 utilizes five loci including D8S1179, D16S539, D5S818, TPOX, and Amelogenin. The samples were then genotyped using the ABI 3100-Avant Genetic Analyzer. The genetic profiles from the spent shotgun shells were compared to the genetic profiles obtained from the reference samples. A partial profile was considered to have one to four amplified loci. The completeness of the genetic profiles were also examined in conjunction with the variables of gender, shell type, and loading order.

Reference:


Whole Genome Amplification, Degraded DNA, Fired Shotgun Shells