B68 The Rate of DNA Degradation in Fragmented Body Parts

Eleanor A.M. Graham, PhD, Victoria Bowyer, MSc, and Guy N. Rutty, MD, Forensic Pathology Unit, RKCSB, Leicester Royal Infirmary, Leicester, LE2 7LX, UNITED KINGDOM*

The goal of this presentation is to provide quantitative and qualitative data to describe the rate of DNA degradation observed over a twelve month period in human body parts exposed to differing environmental conditions and will relate this data to disaster victim identification projects.

The presentation will impact the forensic DNA typing and disaster victim identification communities by demonstrating that full DNA profiles can be obtained from heavily infested and decomposed body parts, which may not usually be considered useful for DNA analysis. It will also provide data to compare the sensitivity and accuracy of two DNA quantification techniques and demonstrate the usefulness of miniSTR analysis when template DNA is severely degraded.

The rate at which soft tissues decompose can impact DNA based identification of whole or disrupted corpses. Environmental factors such as temperature, humidity, and pH along with additional factors such as, infestation, submersion in water or burial will affect the rate at which DNA contained in soft tissues degrades. In order to determine the effect of temperature, humidity and submersion donated human limbs will be sampled once a day for two weeks and assessed for DNA quality and quantity recovered.

Ethical approval was gained (LREC: 06/Q2501/17) before this project was undertaken. Amputation patients at the Leicester Royal Infirmary were consented for donation of post-operative tissue samples. The donated limbs were then exposed to differing environmental conditions for two weeks. This was carried out over a 12 month period. Small tissue samples and photographs were taken each day during the two week period. DNA was extracted from tissue samples using the QIAamp DNA mini kit following the tissue protocol. Extracted DNA was quantified by Spectrophotometry and Real Time PCR. DNA quality was assessed by 1% agarose gel electrophoresis. DNA profiling was carried out on all samples using the SGM Plus PCR Amplification kit, initially at 28 cycles. Samples displaying drop-out or no amplification were re-amplified at 34 cycles and were additionally amplified using the MiniFiler™ PCR Amplification kit. PCR products were analyzed on a 3130 Genetic Analyzer and were analyzed using GeneMapper ID v. 3.2.

Sampling for the full 14 day period could not be undertaken in 8 of 16 limbs due to advanced infestation. The results of gel electrophoresis indicate that DNA degradation to fragments below 10kb begins within 1-4 days. The rate of DNA degradation is affected by the ambient temperature, with limbs sampled during the winter months showing slower degradation than warmer months. The results also show that rate of DNA degradation for samples submerged in water are similar to that of samples on dry land. The results of Real Time DNA quantification indicate that the amount of DNA recovered from samples decreases as decomposition progresses and that PCR inhibitors were not present in the extracted samples. The results of DNA profiling demonstrate that full SGM Plus profiles can generated even from highly infested remains by use of a standard 28 cycle protocol. When partial profiles were observed, the missing DNA profile components could be recovered by re-amplification at 34 PCR cycles. Additional and corroborative DNA profile information could also be obtained by amplification with the MiniFiler™ kit.

DNA profiling is an extremely useful tool for DVI, and in many cases, such as body fragmentation, represents the only primary identification technique applicable. The data presented here will demonstrate the rate of DNA degradation in amputated limbs over the period of one year, carried out in Leicester, UK. The data presented will also demonstrate that it is possible to produce full SGM Plus DNA profiles, using a standard 28 cycle protocol, from highly infested and decomposed body parts, which may not usually be considered as suitable for soft tissue sample collection.

DNA, Decomposition, Degradation

 ** Presenting Author*