



### A55 STR Profiling of Bloodstained T-Shirts Submerged in a Freshwater Lake

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After attending this presentation, attendees will have a greater understanding of how bloodstains are affected by submersion in freshwater environments for variable periods of time.

This presentation will impact the forensic science community by providing data relevant to the effects water submersion has on the appearance, DNA quantities, and STR profiling of bloodstains submerged in freshwater environments for variable periods of time.

The success of DNA analysis is affected by the environment and substrate from which the sample is recovered. The rate of DNA degradation depends on light exposure, water content, temperature, and the presence of microorganisms that may result in physical, chemical, and biological degradation of the genomic DNA. Extreme environments, such as submersion in water, can greatly reduce the likelihood of successful short tandem repeat (STR) genotyping. Submerged samples are exposed to the contents of the water, which may introduce inhibitors, or degrade the bloodstain itself. Another factor acting on the bloodstain is the mechanical action of water removing cellular material from the substrate, washing away valuable evidence. To date, some research has focused on developing methodology for extracting and typing DNA from corpses, tissue, and bone that have been immersed in water; however, little has been done to explore the possibility of testing bloodstained clothing recovered from an aquatic environment. Bloodstains found on submerged clothing may offer investigators information regarding the identity of the victim and foreign DNA that may belong to a suspect or witness to the crime.

DNA extraction and DNA typing of bloodstains immersed in water have not been extensively studied. It can be envisioned that in certain circumstances where a body is absent and only circumstantial evidence, such as bloodstained clothing, may be the investigators' only piece of biological evidence. Dried bloodstains have been extensively examined for successful extraction and typing. However, these same studies have not been performed on bloodstains following submersion in water.

The goal of this study is to determine the effect water submersion has on the recovery and amplification of DNA from bloodstained T-shirts submerged in a freshwater lake for periods ranging from two minutes to 90 days. Based on the success of previous research on submerged samples, this study evaluated and compared two extraction methods, a phenol-chloroform method and the manufacturer's protocol for a commercial, silica-column-based kit. This study will serve as a baseline, utilizing whole blood and a cotton substrate to minimize degradation and possible inhibitors other than what is introduced by the aquatic environment.

All bloodstains recovered from the lake, deionized water controls, positive controls, and negative controls were divided and extracted using two methods: a commercial, silica-column-based kit, and an organic extraction method. DNA was quantified using real-time PCR, and STR profiles were generated using a commercial multiplex kit.

Preliminary results suggest that bloodstains submerged for up to 24 hours are viable for obtaining quantifiable amounts of DNA capable of producing full Short Tandem Repeat (STR) genotypes. Findings have also shown that the appearance of the bloodstain following recovery from the lake does not necessarily correlate to the success of DNA analysis. The preliminary results from bloodstains submerged in deionized water indicate greater quantities of DNA from the same time periods, suggesting that either the water currents or contents impact the amount of DNA retained by the substrate.

#### **DNA Analysis, Bloodstains, Submerged**