

H40 The Effects of Skeletal Preparation Techniques on DNA From Human and Nonhuman Bone

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After attending this presentation, attendees will understand the negative and positive effects that standard and alternative methods for cleaning skeletal material have on the DNA found within human and nonhuman bone.

This presentation will impact the forensic community and/or humanity by encouraging anthropologists and others who analyze skeletal remains to refrain from using bleach cleaning techniques on bone, and instead adopt alternative methods such as the powdered detergent/sodium carbonate technique tested here. Subsequent DNA analysis of cleaned skeletal material will not be negatively impacted, and in fact may even be improved.

The goal of this presentation is to inform the forensic community about the effects bone cleaning techniques have on subsequent DNA analysis.

A common technique used by anthropologists and others who work with skeletal remains is to clean bone with adherent soft tissue by boiling it in a bleach solution. This method, while effective, causes damage to the exposed surfaces of the bone, which may have detrimental consequences in a forensic investigation. Furthermore, bleach, owing to its strong oxidative properties, is known to damage DNA, although the extent of which, if any, in a boiled bone sample, is not clear. In response to bleach's known detrimental effect on bone, alternative cleaning techniques have been proposed. A skeletal cleaning method is considered successful if it is as fast and effective as commonly used techniques, while being less destructive to the sample. The impact these procedures have on the DNA within bone is, however, more difficult to discern.

In the study presented here, an alternate tissue removal technique that consists of boiling bones in powdered detergent and sodium carbonate¹, a standard bleach boiling protocol, and a control of boiling bones in water, were examined. Animal bones with adhering tissue, as well as human bones from a forensic case, were cleaned using all three techniques. DNA was then isolated from each of the samples using a standard organic extraction, and DNA yields quantified using UV spectrophotometry or Real Time PCR. Gel electrophoresis or PCR amplification of progressively larger pieces of mitochondrial DNA were then used to determine how degraded mtDNAs were following each cleaning.

These experiments demonstrate that significantly less DNA is

recovered from bone samples cleaned in bleach than from water cleaned controls. Further, bleach cleaning was shown to cause DNA degradation. In contrast, the powdered detergent/sodium carbonate cleaning method¹ produced DNA for which quantity and quality were similar to or even exceeded the water control. The results from these experiments indicate that not only does bleach cause damage to the bone in addition to having a notable negative effect on DNA when compared to the water control, but the powdered detergent/sodium carbonate technique may protect DNA during bone cleaning procedures.

The results presented should encourage anthropologists and others who analyze skeletal remains to refrain from using bleach cleaning techniques on bone, and instead adopt alternative methods such as the powdered detergent/sodium carbonate technique tested here. Subsequent DNA testing of cleaned skeletal material will not be negatively impacted, and in fact may even be improved.

References:

1. Fenton TW, Birkby WH, J Cornelison. A Fast and Safe Non-Bleaching Method for Forensic Skeletal Preparation. *J Forensic Sci* 2003; 48(1):274-276.

Skeletal Remains, DNA Degradation, Bone Cleaning Procedures