

A15 Application of Modified STR Amplification Protocols to Commingled Remains From the USS Oklahoma

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After attending this presentation, attendees will have learned about the use of aggressive amplification strategies to obtain nuclear data from degraded skeletal remains in the case of the USS Oklahoma.

This presentation will impact the forensic science community by demonstrating how additional genetic information can assist with the re- association and identification of commingled remains in missing persons cases.

Four hundred and twenty-nine crewmembers were lost when the USS *Oklahoma* capsized and sank during the attack on Pearl Harbor on December 7, 1941. Over the next two years, the *Oklahoma* was righted and eventually towed to dry dock. Unknown remains recovered throughout the salvage process were buried in individual or common graves at the Nu'uanu and Halawa cemeteries in Hawaii. In 1947, the unidentified remains from the USS *Oklahoma* were disinterred. After three years of controversy surrounding the segregation of the grossly commingled skeletons, the approximately four hundred unknowns were separated into sixty five caskets and reburied in the National Memorial Cemetery of the Pacific. In 2003, one of the *Oklahoma* caskets was exhumed by the Central Identification Laboratory (CIL). This casket was believed to contain the remains of five individuals; however, anthropological analyses determined that many more were represented. Subsequently, 177 skeletal elements from the casket were submitted to the Armed Forces DNA Identification Laboratory (AFDIL) for mitochondrial DNA (mtDNA) typing. The 95 distinct mtDNA sequences recovered confirmed the suspicions of the CIL anthropologists.

Although the identification of degraded skeletal remains at AFDIL is primarily achieved through mtDNA typing, the forensic utility of this data is often limited by the molecule's uniparental inheritance and lack of recombination. Additionally, mtDNA testing requires either direct or maternal references for evidentiary comparison, and in some cases these types of references are unavailable. In the case of the USS *Oklahoma*, reference material has been collected for only fifty three of the nearly four hundred missing individuals. Further, the sometimes low power of discrimination of mtDNA is evident in this case as sequences from several skeletal elements and references share matching control region haplotypes. In one particular instance, four *Oklahoma* samples possess a

common mtDNA haplotype shared by two families and the use of coding region data was unable to provide any resolution despite past successes.¹ When specific limitations of mtDNA testing such as these are encountered, data from alternative DNA markers in the nuclear genome can benefit the overall identification effort. Unfortunately, the poor quality and limited quantity of nuclear DNA present in degraded skeletal remains has historically restricted the use of short tandem repeat (STR) markers. However, aggressive STR typing protocols² have recently shown great promise on the degraded skeletal elements typically encountered at AFDIL³, particularly when the modified amplification is coupled with an improved DNA extraction.⁴

To provide additional genetic information, STR amplification protocols were applied to the four *Oklahoma* samples that share a common mtDNA haplotype. As in most cases processed at AFDIL, the degraded skeletal elements from the Pearl Harbor battleship yielded too little DNA to produce usable data under standard amplification conditions. Therefore modifications were made to the suggested protocols of two commercially-available amplification kits, one targeting markers on the Y-chromosome and another containing autosomal STRs with reduced-size amplicons. In addition to the commercial kits, two multiplex panels developed at AFDIL were utilized to type 15 X- chromosomal STR loci. Since aggressive parameters were used to amplify the low DNA quantity samples, data authenticity was confirmed by performing triplicate amplifications with only duplicated alleles included in a finalized, consensus profile.² Nuclear data generated with all three marker systems enabled the sorting of the four *Oklahoma* samples. Additionally, kinship analyses were performed using genetic data derived from the various skeletal elements and family reference specimens in order to assess the confidence in the presumptive identifications based upon odontological, anthropological, and contextual findings. The successful re-association of the commingled remains and genetic support for identification in this example from the USS *Oklahoma* demonstrates the practical utility of modified STR typing strategies in missing persons cases.

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References:

- ¹ Just R, Leney M, Barritt S, Los C, Smith B, Holland T, Parsons T. The use of mitochondrial DNA single nucleotide polymorphisms to assist in the resolution of three challenging forensic cases. J Forensic Sci 2009; 54(4): 887-91.
- ² Gill P, Whitaker J, Flaxman C, Brown N, Buckleton J. An investigation of the rigor of interpretation rules for STRs derived from less than 100 pg of DNA. Forensic Sci Int 2000; 112: 17-40.
- ³ Irwin J, Leney M, Loreille O, Barritt S, Christensen A, Holland T, Smith B, Parsons T. Application of low copy number STR typing to the identification of aged, degraded skeletal remains. J Forensic Sci 2007; 52: 1322-7.
- ⁴ Loreille O, Diegoli T, Irwin J, Coble M, Parsons T. High efficiency DNA extraction from bone by total demineralization. Forensic Sci Int Genet 2007; 1: 191-5.

Degraded Skeletal Remains, Short Tandem Repeats, Missing Persons