

BLIND STUDY USING CYTOCHROME B PRIMERS FOR PROCESSING ANCIENT SKELETAL REMAINS

Michelle F. Perella, Timothy P. McMahon, Sarah L. Bettinger, Ryan E. Vachon, Amanda Coute, and Louis N. Finelli

Armed Forces DNA Identification Lab, 1413 Research Blvd., Rockville, MD 20850, USA

The Armed Forces DNA Identification Laboratory (AFDIL) aids the Joint POW/MIA Accounting Command -Central Identification Laboratory (JPAC-CIL) with the identification of military service members missing from previous conflicts (World War II, the Korean War, and the Vietnam War). As the time span between war and recovery increases, the ability to determine whether fragmented and degraded skeletal remnants are human or animal on the basis of morphology alone becomes increasingly difficult. This can lead to increased amplification failures if non-human bone fragments were processed, since AFDIL uses human specific primers for amplification and sequencing. In the event that these primate specific primers fail to amplify extracted DNA, a validated set of vertebrate specific primers that amplify a small, variable region among species, such as the cytochrome B (cyt b) gene, would be helpful for targeting causes of amplification failure. Towards this end, AFDIL has validated the use of Parson et al.'s (2000) vertebrate cyt b primers for use in species identification. The cyt b primers amplify a 358 base pair region that contains species-specific information. Previous work optimized amplification conditions such that 1pg of input genomic DNA was efficiently amplified from high copy DNA sources and that non-redundant BLAST searches of the generated sequence can identify samples to the Family level 100% of the time and to the species level 90% of the time. To assess the ability of the cyt b primers to identify the biological origin of DNA from ancient skeletal remains, a blind set of twenty bones collected from various locations and environmental conditions were sent to AFDIL from JPAC-CIL. The samples were extracted, amplified and sequenced using the cyt b primers and analyzed on the AB 3100 and 3130xl ® Genetic Analyzers. The genus or species of the bone specimen's sequences were determined using the non-redundant BLAST function in the GenBank nucleotide database, which houses any published cyt b gene sequence. The results from the 20 bone blind study will be discussed here. The views expressed in this abstract are those of the author and do not reflect the official policy of the Department of the Army, the Department of Defense or the U.S. Government