

DNA EXTRACTION FROM BOVINE BONE: A TRAINING METHOD WHICH DETECTS HUMAN CONTAMINATION

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A training protocol was developed for the extraction and analysis of DNA from bovine bone which allows the sensitive detection of bovine mitochondrial DNA (mtDNA) as well as human mtDNA contamination.

The analysis of mtDNA from bone has widespread utility in forensics. The methods of amplifying the extracted DNA, e.g., Polymerase Chain Reaction (PCR), are so sensitive that strict adherence to proper technique is required to avoid compromised data. A major concern is the contamination of the test sample with DNA from the technician doing the extraction.

The protocol we have developed allows the simultaneous amplification of bovine and, if present, human mtDNA sequences. Minor changes were made to the Armed Forces DNA Identification Laboratory's (AFDIL) protocol "DNA Extraction from Tissue and Fresh Bone" (version 2.1) for the extraction of DNA from cow bones (available from local butchers). One set of PCR primers ("humbov1F&R"), derived from hypervariable region 1 of human mtDNA, will amplify bovine mtDNA efficiently but will preferentially amplify human DNA if present. The PCR products from the two mtDNA sources differ in size and can easily be distinguished on standard ethidium bromide stained agarose gels. Thus, bovine bone and these PCR primers are an extremely instructive pair of tools for the training of personnel in the art and science of mtDNA extraction and analysis.

The bovine bone extraction and analysis method is currently being used to train biotechnology students at Massachusetts Bay Community College (Wellesley, MA). It challenges students to take the necessary technical measures required to properly perform mtDNA extraction from bone as well as PCR amplification and analysis. A detailed description of the protocols will be available.

Special thanks are extended to Ed Huffine, DNA Coordinator at Physicians for Human Rights, and former Chief DNA Analyst at AFDIL, for generously sharing protocols and assisting with troubleshooting.

