

HIGH-YIELD METHOD OF DNA EXTRACTION FROM OLD AND DEGRADED SAMPLES OF HUMAN SKELETAL REMAINS'

Hirofumi Fukushima^{1,2}, Kanako Nagasaki³, Hideki Asamura¹, Masao Ota¹, Yoshiya Fukuma³

¹*Dept. of Legal Medicine, Shinshu University School of Medicine, Matsumoto, Japan*

²*The Institute of Human Identification Science, Matsumoto, Japan*

³*Life Science Division, Hitachi Software Engineering Co., Ltd., Japan*

Human identification from forensic samples often presents difficulties due to poor quality and quantity of DNA caused by degradation. The purpose of our investigation is to evaluate the possibility of individual identification following detection of DNA from degraded human tissue, mainly old teeth and bone fragments discovered as skeletal remains. Those attempting identification from old and degraded human samples, such as remains buried at archeological sites and remains of war dead buried for a number of decades, have generally resorted to detection of mitochondrial DNA(mtDNA). Recently, however, new cases have been reported in which miniSTR technique has been successfully used to identify degraded samples of human remains. Our new high-yield method is designed to successfully detect STR, Y-STR, and mtDNA even if the subject remains samples are considered to be significantly degraded in quality. The extracted DNA is then amplified using AmpFLSTR® Identifier® and Yfiler™ Kit and genotyped using ABI PRISM® 3100 Genetic Analyzer. The results of the new method indicate that, DNA analysis is possible for almost any type of remains, as almost all samples yielded either a full or partial profile without use of miniSTR. In particular, the analyses of mtDNA extracted from human teeth were nearly all completed successfully. By utilizing high-yield method to retrieve markers including STR and Y-STR as well as mtDNA, it is possible to successfully complete a greater magnitude of human and kinship identifications relative to traditional methods. Our new extraction method has been successfully applied to a large amount of Japanese skeletal remains which had been buried in Russian territory during the 60 years since the end of World War II. These bone and teeth DNA were successfully analyzed for 15 STR loci and 16 Y-STR loci using commercially available kits and the control region in the mtDNA. Overall, the success rate for obtaining either a partial or full profile was over 70% without using short amplicon STR multiplex.