

CEMENTUM AS A DNA SOURCE IN DEMINERALIZED POSTMORTEM TEETH FOR FORENSIC HUMAN IDENTIFICATION

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When there are doubts about the identity of an encountered human body, scientific methods for human identification are employed, of which DNA analysis is considered the gold standard. The objective of this study was to validate an alternative protocol for DNA extraction from postmortem teeth that targets the cementum. In our study, one to five teeth from each of 20 unidentified human bodies recovered from Midwest Brazil were analyzed. First, one tooth from each individual were placed in separate sterile containers and demineralized in 20 mL of EDTA solution (0,5 M, pH 8), changed daily. After seven to nine days, the final centimeter of the root apex (extremity of the root) was excised with the aid of sterile disposable scalpel blades. This portion of the sample was used for DNA extraction through a conventional organic extraction protocol using microcon membranes (YM-100). We added 300 µL of extraction buffer and 20 µL of Proteinase K to the sample. After overnight digestion, one extraction with 300 µL of phenol:chloroform:isoamyl alcohol, the aqueous phase was filtered and subsequently washed in 300 µL of ultrapure water. Elution volume was 40 µL. STR amplification was performed using Identifiler Plus, NGMSelect or NGM followed by capillary electrophoresis on a 3130 or 3500 platform. When multirooted teeth were processed, the apex of only one root was extracted. If the result was a poor profile, we performed extraction on another root or another tooth if there were no apexes left. For 60% of the bodies (12 of 20), a full genetic profile was obtained in the extraction of the first root. For 4 individuals, 2 teeth were processed, while for one individual, 5 teeth were analyzed. By the end of the analyses, full genetic profiles were obtained for 85% of the individuals (17 of 20), while the other three individuals showed an incomplete, a partial and a blank profile. In terms of quality, 2 profiles showed higher than normal stutter peaks (vWA and TH01); in 6 samples the peak balance of some loci was not good (PHR around 60% or lower), and drop-ins were observed for samples from 5 different individuals (allele 10 on D8S1179). These artifacts are believed to be due to the degradation level of the DNA in the samples and not a product of the protocol described. Nevertheless, the Forensic DNA Laboratory was able to confirm the identity of 80% of the bodies in this study. The results obtained show that this alternative protocol for postmortem teeth is capable of extracting DNA in sufficient quantity and quality, producing full genetic profiles in forensic human identification cases.