B5 Developing a Simple Method to Process Compromised Bone Fragment for Forensic DNA Isolation

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The goal of this presentation is to introduce a new method for processing compromised bone fragments prior to DNA isolation. The advantage of this method over conventional methods is in applying the proteinase solution that will omit the step of a physical cleaning procedure, such as sanding.

This presentation will impact the forensic community and/or humanity by demonstrating data which suggests that this method could be used for 1) initial sample preparation for cleaning the outer surface of compromised human skeletal fragments, and 2) could be adapted for automated DNA isolation for bone fragment in the near future.

Skeletal remains have been challenging biological samples for DNA isolation. Bones are more difficult for preparing and sampling prior to DNA extraction. One of the labor-intensive and time-consuming steps in DNA isolation from bone fragments is the initial cleaning and sampling of the bone. Due to the potential of having co-mingled remains, adhering inhibitors and bacterial contamination, the outer surface of the bone fragment has to be cleaned by a current method like sanding. However, to avoid cross-contamination between samples, the bone dust that is generated during the sanding of the bone must be removed. Additionally, safety protection equipment and procedures are necessary to protect lab workers and technicians from exposure to blood-borne pathogens.

To address this issue, a simple processing method has been developed using proteinase solution prior to DNA isolation. In this study, the use of proteinase solution requires much less labor than a physical method such as sanding. By incubating with the proteainse solution, the soft tissue and outer surface of the bone fragment sample is removed. The processed bone fragment or a portion of the fragment can then be used for DNA isolation. The characterization of the effect of the proteinases on the cleaning of bone fragments was performed. Proteinases were screened and the best candidates were selected. Additionally, the optimum incubation condition (concentration of proteinase, incubation temperature and pH) for the proteinase was determined. DNA from processed bone sample was isolated. The results demonstrated that this method is effective for removing those soft tissues attached to bone samples and the outer surface of bone fragment samples. The data suggest that this method could be used for 1) initial sample preparation for cleaning the outer surface of compromised human skeletal fragments, and 2) could be adapted for automated DNA isolation for bone fragment in the near future.

Bone, Challenging Samples, DNA Isolation

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