Methods in Extracting DNA: Using Phenol-Chloroform on Formalin Fixed Human Brain Tissue
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Genetic markers of diseases can be studied post-mortem with the isolation of DNA from tissues. While protocols exist for DNA extraction from fresh and formalin-fixed-paraffin-embedded tissue and formalin fixed hard tissues such as bone there are no published methods for formalin-fixed soft tissues, such as brain. Extractions from formalin-fixed soft tissues are more difficult due to the higher amount of cellular degradation that results from fixation. The development of efficient DNA extraction protocols for formalin fixed soft tissues will allow post-mortem study of genetic markers and aid in identification of genetic diseases. Existing techniques of phenol-chloroform extraction were modified to produce the most efficient method of DNA extraction. Tissue samples were obtained from 12 human cadaver donors preserved in 10% formalin solution. Samples were digested using 0.2mg to 2.0 mg proteinase K per 100mg of frontal lobe cerebral tissue under constant temperature of 55°C. Quantification of DNA was determined via agarose gel electrophoresis and ultraviolet spectrophotometer analysis. The quality of the purified DNA will be assessed by targeted gene amplification using standard polymerase chain reactions. Similar techniques will be utilized for the identification of specific genetic markers in disease, including those known in Alzheimer’s. This work will significantly expand the availability of tissues for genetic analysis post-mortem, thus contributing to genetic disease research.