USING A DNA BARCODING TECHNIQUE TO IDENTIFY DIATOMS FROM CARDIAC TISSUE SAMPLES FOR DROWNING INVESTIGATIONS

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Diatoms have been used in forensic investigations to assist in determining whether an individual has drowned. Diatoms are unicellular photoautotrophic eukaryotes ranging from 2 μm to 0.5 mm in size and can be found in almost all well-lit aquatic environments. The presence of diatoms in a person's blood stream and tissues, such as heart or lung tissues, indicate drowning; the absence of diatoms indicates that the person may have expired prior to being found in a body of water. The identification of diatoms can be carried out based on their morphology using light microscopy or scanning electron microscopy. However, the morphological methods can be labor-intensive and susceptible to subjective interpretation. To address these issues, DNA barcoding techniques for the identification of diatoms are potentially useful for drowning investigations. In this study, we tested the sensitivity of a DNA barcoding method in detecting the presence of diatoms in specimens. Samples were prepared by placing various amounts of diatom, *Thalassiosira pseudonana,* in 10 milligrams of cardiac tissue. Swine cardiac tissue, a useful model system because of its similarity to human tissue, was used for this study. Diatom DNA was extracted using DNA extraction kits currently used for forensic applications. The region of DNA barcoding used was the 18s rDNA. The presence of diatom DNA is verified by sequence comparisons using BLAST. Using this method, the limit of detection of diatoms is approximately $2.7x10^3$ per 10 mg of tissue. Additionally, a lesser amount of diatom DNA was recovered when homogenized cardiac tissue was present than when diatom alone was present. The results of this study indicated that the yield of diatom DNA extracted can be interfered by the presence of the tissue matrix. A DNA extraction kit with a high yield for diatom DNA should be sought and tested in future studies. Additionally, this study suggested that lung fluid, a simple matrix, would be a more suitable specimen for the DNA barcoding technique than tissues for the detection of diatoms in drowning cases. This hypothesis will also be tested in future studies.