G23 Advanced Flow Cytometric DNA Degradation Analysis: Utility in Postmortem Interval Estimation

Erica Williams, MD, Hospital of the University of Pennsylvania, Department of Pathology and Laboratory Medicine, 6 Founder Building, 3400 Spruce Street, Philadelphia, PA 19104; Andrew Bantly, BS, Hospital of the University of Pennsylvania, Abramson Cancer Center Flow Cytometry and Cell Sorting Shared Resource, 3400 Spruce Street, Philadelphia, PA 19104; Jesse Chittams, MS, University of Pennsylvania School of Medicine, Department of Biostatistics and Epidemiology, 3400 Spruce Street, Philadelphia, PA 19104; Jonni Moore, PhD, Hospital of the University of Pennsylvania, Abramson Cancer Center Flow Cytometry and Cell Sorting Shared Resource, 3400 Spruce Street, Philadelphia, PA 19104*

After attending this presentation, attendees will be able to determine if application of a statistical model and objective computer modeling to DNA degradation data will yield reproducible, accurate results, and help in post-mortem interval estimation.

This presentation will impact the forensic community and/or humanity by providing preliminary results which indicate that more advanced analysis, including statistical evaluation and computer modeling, of DNA degradation data is possible. These methodologies could be then be applied to forensic autopsy samples to assess their validity in estimating a post-mortem interval.

This poster will show data used to develop a mathematical model for PMI estimation as well as juxtapose two different flow cytometric computer models in an effort to select the most reliable methods of estimation.

Over the past decade, since the first proposal by Cina that flow cytometry might be useful in post-mortem interval estimation by monitoring DNA degradation, several papers have been published attempting to establish a link. Using flow cytometry, previous studies on splenic and hepatic tissue have suggested that cellular DNA degradation increases with time, and could thus potentially be used as a tool for postmortem interval estimation. However, much of this previous work has been focused on selecting the best type of sample for analysis, rather than on perfecting the technique and analysis of the data obtained. In an attempt to delineate the best analytical method, this study involved the collection of easily obtainable blood samples, from normal living donors, which were stored at room temperature, then assayed for DNA content at varying times post collection (0-191.5 hrs) to determine the amount of DNA degradation. This amount of degraded DNA was quantified using two different computer programs: CellQuest™ v. 3.11 (Becton-Dickinson, San Jose, CA) and Modfit™ v. 3.0 (Verity Software, Topsham, ME, U.S.A). The first program allows for subjective analysis of the amount of DNA degradation, while the second program uses a computer model which can objectively assign the amount of DNA degradation, without user input.

After DNA degradation determination by both computer programs, the relationship of DNA degradation and sample age was plotted and further analyzed with a random coefficient statistical model to yield a population regression curve.

The validity of this curve was then tested using blood collected from another group of normal donors and analyzed at varying times in a blind study. In 20 of 28 samples, a correct 24 hour period was able to be assigned (71%). These preliminary results suggest that a mathematical model, combined with objective computer analysis, can be applied to the monitoring of DNA degradation of cellular material, and can potentially become a tool in determining post-mortem interval.

Post Mortem Interval, Flow Cytometry, DNA Degradation

 ** Presenting Author*