B170 Assessment and In Vitro Repair of Damaged DNA Templates

Ashley M. Hall, MS, and Jack Ballantyne, PhD, University of Central Florida, National Center for Forensic Science, PO Box 162366, Orlando, FL 32816-2366*

Attendees will learn methods for the assessment and repair of damaged DNA templates derived from forensically relevant samples.

At the present time, little is known about the damage done to the DNA in biological stains exposed to various exogenous insults. This presentation will impact the forensic community and/or humanity by presenting the first comprehensive study of this damage at the molecular level and intoduce methods for the repair of the damaged DNA, facilitating the recovery of a genetic profile.

DNA extracted from biological stains is often intractable to analysis. This may due to a number of factors including a low copy number (LCN) of starting molecules, the presence of soluble inhibitors or damaged DNA templates. Remedies may be available to the forensic scientist to deal with LCN templates and soluble inhibitors but none presently exist for damaged DNA. In fact, knowledge of the biochemical nature and the extent of DNA damage in physiological stains is rudimentary at best. Also unknown is the point at which the damage inflicted upon a particular sample precludes the ability to obtain a genetic profile for purposes of identification. Therefore, the primary aims of this work were first ascertain the types of DNA damage encountered in forensically relevant stains, correlating the occurrence this damage with the partial or total loss of a genotype, and then to attempt the repair of the damage by means of *in vitro* DNA repair systems.

The initial focus of the work was the detection of damage caused by exogenous, environmental sources, including factors such as UV irradiation, heat, and humidity. By incorporating various lesion specific enzymes, a set of assays, both PCR and gel-based, have been developed which describe the type and extent of damage inflicted upon DNA, both in a hydrated and dehydrated state. Using these procedures, the major causes of damage have been identified and their effects on genetic profiling assessed.

Armed with this knowledge, the next focus was the repair of the damage by means of *in vitro* DNA systems. Efforts have been concentrated on base excision repair, single strand gap repair, and translesion synthesis assays. By modifying the assays and employing various combinations of the systems, a genetic profile has been obtained from previously intractable samples.

DNA Damage, In Vitro DNA Repair, Lesion Specific Endonucleases

 ** Presenting Author*