EVALUATION OF STABILITY AND STORAGE TREATMENTS FOR FORENSIC DNA SAMPLES

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DNA is the biometric of choice for forensic purposes as it can identify individuals with high confidence, be collected after an event, and be used to infer kinship. After a DNA sample has been extracted into aqueous solution, it must be kept frozen or refrigerated to prevent degradation. However, lack of refrigeration space or an unreliable power supply can result in loss of irreplaceable forensic evidence. There are significant logistics challenges and increased costs associated with shipping purified DNA from a point of origin overseas while refrigerated. FedEx states that boxes without temperature control may reach temperatures of 60°C during certain times of year. Once DNA samples are received stateside, forensic laboratories are faced with similar storage issues. The ability to store DNA for extended periods of time without refrigeration would greatly improve the logistics of handling of DNA evidence.

Many studies have shown the detrimental effects of heat, oxygen, and water on DNA stability (Lindahl, 1993). DNA damage that occurs during storage of biological samples includes base modifications, mispairs, cross-linked nucleotides, and breaks in the DNA. Double-stranded breakage of DNA causes longer sequences to be fragmented into shorter ones, and is the most common physical change associated with DNA degradation. These breaks prevent the DNA from being amplified and identified by current forensic technologies.

Examination of anhydrobiotic organisms has lead to the exploration of trehalose and similar chemicals for purposes of stabilizing DNA in dehydrated form. (Ivanova NV, 2013). In recent years, several new technologies for the storage of DNA have been developed. At the time this study was conducted there were two commercial kits available for stabilization of purified DNA samples: DNAstable (from BioMatrica Inc., USA, also licensed as QIASafe from Qiagen) and GenTegra (from Integenex, USA). Both rely on a pre-lyophilized chemical provided in a tube to which the sample is added and then dehydrated. DNAstable uses a synthetic polymer with qualities similar to trehalose, which presumably stabilizes DNA through hydrogen bonding with the minor groove and when dehydrated, forms a thermo-stable barrier between the sample and the outside atmosphere. (Lee, Howlett) GenTegra is an inorganic mineral matrix that provides oxidation and anti-microbial protection to the DNA when dehydrated. Life Technologies' PureLink was on the market until March of 2014 but was discontinued immediately prior to the beginning of this study.

Several other studies have examined the performance of one or the other of these kits (Frippiat, 2011, Lee 2012, Howlett 2014, Wan 2009), but none of them have compared DNA preserved with both kits to untreated aqueous controls stored at each temperature, and to DNA stored in dry form with no additives. We established a baseline for stability of purified aqueous DNA at both ambient and elevated temperatures, and assessed the performance of both kits over the course of months. We also examined the stability of DNA from two sources, as well as the stability of DNA dehydrated without preservatives.

We saw no significant change in quantification or sign of degradation (measured by STR amplification) for buccal-derived DNA stored untreated at -20°C, 4°C, room temperature

(~22°C) and 37°C for 11 weeks. We saw only partial degradation after two weeks at 50°C. In contrast, partial degradation of untreated commercially-acquired genomic DNA was observed after 6 weeks at 37°C, and complete degradation was observed after 2 days at 50°C. While Biomatrica's DNAstable kit and IntegenX's GenTegra kit completely preserved the DNA from degradation, dehydration in the absence of preservatives provided the same level of stability.

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