A130 Comparison of Different Methods for DNA Isolation

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After attending this presentation, attendees will learn about the performance of different methods for DNA isolation and comparisons by application of test parameters such as yield, quality, cost, and time taken to complete the isolation process.

This presentation will impact the forensic science community by proving it is useful to periodically assess the performance of different

forensic science techniques as new ones become available. The present study provides knowledge on the outcome of testing and comparing different products for DNA isolation that may aid practitioners' evaluation of options for different analytical needs.

In forensic DNA testing, a crucial first step is the isolation of DNA from evidence samples for additional procedures. The performance of this first step enables the final results. Different methods and kits for the isolation of DNA are commercially available and may vary in different characteristics that are of importance to analysts such as time for processing, yield of DNA, integrity, purity, cost, and simplicity of process. Consequently, it can be useful to measure such parameters in comparison of different commercially available kits for DNA isolation. DNA analysts and educational institutions would thus have more bases for the selection of kits for particular needs.

Three different DNA isolation kits were initially obtained for study with others being later tested. The kits were QIAamp DNA Investigator from Qiagen, ZR Genomic DNA from Zymo Research and AccuPrepGenomic DNA Extraction kit from Bioneer. The kits were evaluated according to the following criteria: (a) purity of isolated DNA;

(b) integrity of isolated DNA; (c) yield; (d) time taken to process; and, (e) cost per sample. Purity and yield were assessed by using a Nanodrop spectrophotometer and integrity of DNA was assessed by agarose gel electrophoresis. DNA was isolated according to the different manufacturers' instructions for three different samples of whole blood for each kit.

The results showed differences in performance among the kits. Agarose gel electrophoresis of products isolated by each kit showed bands at the positions expected for genomic DNA in each case and no signs of degradation. Staining intensity was strongest for the DNA isolated by the Qiagen kit. Based on measurement of ultraviolet wavelengths absorbance by Nanodrop spectrophotometry, the yield of DNA from 200 microliters of blood was highest for the Qiagen kit (5.6 µg) followed by the Bioneer kit (2.74 µg) and the Zymo kit (2.39 µg). In regard to purity as measured by A260nm/A280nm ratio, the DNA isolated by the Qiagen kit had a ratio of 1.85 and was closest to the ideal ratio of 1.80, whilst the ratios were 2.0 and 1.03 for the isolates from Zymo and Bioneer kits. Assessment of costs gave values of $3.60/sample for Qiagen, $1.50 for Bioneer and $0.72 for Zymo kits.

Comparison of the time taken to isolate DNA by the different manufacturers' kits showed that the shortest time taken to isolate DNA was used by the Zymo kit which took 25 minutes. The Bioneer kit took 35 minutes for the isolation of DNA and the method that took the longest was the Qiagen kit which took 50 minutes. During the process of isolation, samples were transferred among three tubes for the Qiagen and Bionner kit while the Zymo isolation involved one tube.

In conclusion, it was found that each kit had advantages and disadvantages. While the Qiagen kit gave the highest yield and quality, it was also the most expensive and took the longest to complete, in addition, the Qiagen filtration columns used for DNA binding and the proteinase K used in the procedure must be stored at -20°C. The Bioneer kit was less expensive and took less time than the Qiagen kit for the completion of DNA isolation, but the yield of DNA was less and the products were more contaminated with ultraviolet light absorbing material. In addition, the procedure uses proteinase K which must be stored at -20°C. The Zymo kit was the least expensive and took the least time to complete DNA isolation. The DNA isolated by the Zymo kit was not as pure as that isolated by Qiagen based on ultraviolet light spectrophotometry. All the components of the Zymo DNA kit can be stored at room temperature.

These findings may be exploited by individual laboratories to select kits for DNA isolation based on the type of sample from which DNA is being isolated, the subsequent analytical steps for the DNA and the numbers of samples, and economic considerations.

The various parameters for the efficacy of DNA isolation will also be compared for additional kits and for other types of samples of forensic interest such as saliva, semen and hair.

DNA, Spectrophotometry, Electrophoresis

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