B72 DNA Purification From Forensic Samples Using the BioRobot® M48

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The goal of this presentation is to educate investigators and analysts about the applicability of medium throughput automation for extraction of nucleic acid evidence from a wide range of difficult case work sample types.

This presentation will impact the forensic community and/or humanity by enabling forensic investigators to process case evidence with higher accuracy, reproducibility, greater consistency, and standardization. Data will be presented.

Optimized protocols for automated extraction of genomic DNA from forensic reference and casework samples have been developed. 6–48 samples can be processed in parallel, giving efficient processing of forensic samples and optimized workflow with minimal human contact.

Materials and Methods: The BioRobot® M48 system provides fully automated purification of DNA using silica-coated magnetic particles. The process does not involve any time consuming centrifugation steps. The easy-to-use workstation allows purification of 6–48 samples in a single run. Between processing runs, the worktable can be decontaminated using an integrated UV lamp to reduce the chances of environmental contamination. For this study DNA was purified from various forensic samples or from diluted DNA using fully automated protocols according to the new *MagAttract DNA Mini M48 Handbook* (www.qiagen.com/goto/M48ForensicHB).

Results and Discussion: Genomic DNA purification protocols were optimized for use with forensic samples. Fine tuning of bead concentrations and washing procedures lead to higher peaks and improved signal-to-noise ratios. This enabled highly sensitive detection and high performance in downstream applications. The application has been tested with difficult sample types known to contain high concentrations of inhibitors, such as cigarette butts. STR analysis shows higher peaks for more sensitive detection.

In addition to the standard trace protocol, a new, fully automated “tip dance” protocol was established, where the filter-tip moves back- and-forth relative to the worktable platform while pipetting. This enabled processing of solid materials, such as swabs, fabrics, blood discs, or cigarette butts, directly in the sample tube. There was no need for prior centrifugation to remove solid materials that could clog the tip. The “tip dance” protocol simplifies handling and, in some cases, increases DNA yield with more efficient extraction of the sample and minimal human contact with the sample.

New large volume protocols allow processing of sample volumes up to 500µl to enable higher yields and retrieval of more concentrated DNA for greater sensitivity in downstream applications.

A normalization protocol was developed where DNA yields could efficiently be limited to 150–250 ng by a precise control of the surface area provided in the DNA binding step. This protocol enables subsequent genetic analysis without the need to measure or adjust DNA concentration. This application is sought to be particularly useful for database sample preparation.

Conclusions: Forensic sample preparation on the BioRobot M48® system provides:

- **Efficient yields for sensitive analyses** — from casework samples
- **Normalized yields** – from database samples
- **High signal-to-noise ratios** — for sensitive downstream detection assays
- **Easy handling** — swabs, blood discs, cigarette butts, and other solid samples can be processed directly on the workstation
- **Newly optimized protocols** — including larger starting volumes (500 il) for increased sensitivity on dilute samples and normal-
  ization for uniform yields

Automation, Nucleic Acid Extraction, Casework Evidence

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