

Research Article

# Evaluation of a semi-automated, magnetic bead-based DNA extraction method for genetic fingerprinting of forensic casework samples

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## Abstract

In order to cope with the demanding workload for DNA profiling of forensic casework samples a concept for a semi-automated processing system was developed at the Landeskriminalamt (Office of Criminal Investigation) Baden-Württemberg, Germany [K. Vollack, et al., Implementation of a semi-automated processing system for DNA profiling of forensic casework samples, this issue]. The applied magnetic bead extraction method is based on ChargeSwitch<sup>®</sup> Technology (CST) from Invitrogen and was established on a liquid handling workstation Freedom EVO<sup>®</sup> 150 from Tecan.

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## 1. Introduction

During the last years the demand for DNA analysis in forensic investigations continuously increased. Extraction of DNA from human cells is the crucial initial step within the process of DNA analysis. To guide samples faster through this process a semi-automated method was established on a liquid handling workstation.

## 2. Materials and methods

For manual DNA extraction the First-DNA All-Tissue DNA Extraction Kit (GEN-IAL) was used. Establishing the semi-automated DNA analysis the DNA extraction was performed using the ChargeSwitch<sup>®</sup> Forensic DNA Purification Kit. Volumes were decreased to 50% of the manufacturer's protocol. Cross-contamination tests were carried out by DNA extractions of positive and negative control samples arranged in checked pattern. DNA-quantification was determined by real-time PCR using the AB Quantifiler<sup>™</sup> Human DNA Quantification Kit [1] (Applied Biosystems). For differential extraction of mixed sperm/vaginal secretion stains, samples were incubated 10 min at 56 °C and isolated from the trace carrier. Subsequent

centrifugation separated the vaginal secretions from sedimented sperms.

## 3. Results

The application was adapted on a Freedom EVO<sup>®</sup> 150 liquid handling workstation equipped with an eight-channel liquid handling arm, two Te-MagS<sup>™</sup> magnetic bead separation modules and a PosID<sup>™</sup> barcode sample identification device (Tecan).

It takes less than 2 h to extract DNA from 88 samples. To avoid DNA cross-contaminations liquid handling arms do not travel across open wells or tubes other than their targets. Using the Te-MagS<sup>™</sup> modules instead of a shaking device for mixing additionally prevents the formation of potentially contaminating aerosols. Routinely carried out cross-contamination tests reveal that up to now more than a thousand positive control samples were tested without any contamination detected. One routine protocol was optimized with regard to lysis conditions (time, DTT) to cover the whole range of biological sample types like saliva, blood, semen and skin particles. Overnight incubation of proteinase K lysis (56 °C) and the presence of DTT (2.6%) were shown to significantly improve DNA yield of different sample types (see Fig. 1). Finally we developed a protocol to separate sperm-heads from vaginal secretions and other epithelial cells by differential lysis. In comparison to our manual extraction method (GEN-IAL) the protocol for

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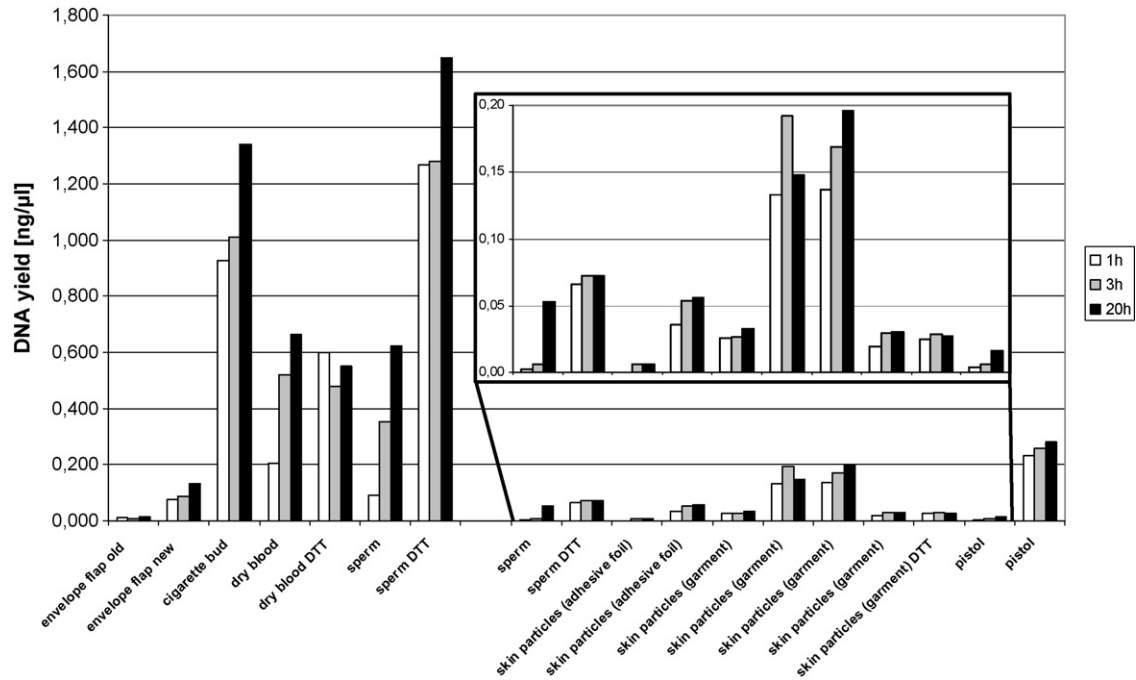


Fig. 1. Comparison of DNA extraction by CST for different crime scene sample types: optimization of the incubation duration and by the addition of dithiothreitol.

automated CST extraction showed similar results in respect of sensitivity and DNA yield with stains limited of human cell material. DNA yield is restricted by maximum binding capacity of CST beads. Concerning the PCR of short-tandem repeats (STRs) comparable results were achieved by both methods with diverse sample types.

#### 4. Discussion

The semi-automated magnetic bead-based extraction method was successfully established to routine and embedded into the automated processing system for DNA profiling of forensic casework samples developed by the Landeskriminalamt Baden-Württemberg, Germany [2]. The extraction system could handle about 40,000 samples per year and processes nearly all types of forensic crime scene samples.

Concerning the forensic relevance (STR/Y-STR analysis) the limited DNA yield of CST exhibits its only disadvantage, extracting mixed stains with enormous excess of female DNA because the male part could be lost. The reduced requirement of labour frees up personnel for other tasks. The automated

processing also minimizes operational errors and therefore improves the quality of data.

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#### Conflict of interest

None.

#### References

- [1] R. Schwenzer, et al., Implementation of a robotized real-time-PCR setup for the use of the AB Quantifiler™ Human DNA Quantification Kit, *Forensic Sci Int. 1* (2008) 68–70.
- [2] K. Vollack, et al., Implementation of a semi-automated processing system for DNA profiling of forensic casework samples, *Forensic Sci Int. 1* (2008) 83–85.