

Research article

Implementation of a semi-automated processing system for DNA profiling of forensic casework samples

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Abstract

The concept for a semi-automated processing system for DNA analysis of crime scene samples was developed at the Landeskriminalamt Baden-Württemberg (LKA BW) and comprises the extraction of genomic DNA from human cells by ChargeSwitch[®] magnetic bead technology (CST), quantification of purified DNA by real-time PCR, amplification of short tandem repeats (STRs) by PCR and DNA fragment length analysis of STRs by capillary electrophoresis. Three liquid handling workstations from Tecan, a real-time PCR device and a 16-channel capillary electrophoresis (CE) system, both from Applied Biosystems (AB), are linked via laboratory data network. Transmission and management of sample and analysis data is enabled by a Laboratory Information and Management System (LIMS). Suitability for a wide range of stain types, early exclusion of DNA-free samples, barcode sample identification and prevention of cross-contaminations guarantee efficiency and high quality standards.

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1. Introduction

As DNA profiling has become one of the most efficient and required tools in criminal investigations, forensic laboratories are challenged with increasing workloads. To step up sample throughput and analysis efficiency, we developed a concept for a semi-automated processing system with a medium throughput of 20,000 samples annually, covering the whole range of biological sample types (e.g. saliva, blood, semen, epithelial cells, buccal swabs). Highest analysis sensitivity and minimum DNA contamination risk should be guaranteed in consideration of economical aspects.

2. Material and methods

Operational steps and hardware components of the DNA analysis automation concept are listed in [Table 1](#). A workflow scheme is shown in [Fig. 1](#). Further details are described elsewhere [1,2].

3. Results

Sample identification by integrated barcode reading is performed with reference to work lists provided by the SQL server-based LIMS. Without ionic chaotropes (potential PCR inhibitors) and offering long-term stability of eluted DNA (double-stranded conformation), CST magnetic bead extraction of DNA is promising, especially for efficient STR analysis. The full range of different crime scene samples is covered by our CST protocol. The single tube format used for DNA extraction allows to work with samples of relatively large sizes and to handle the purified DNA individually further on. Besides the magnetic mixing function of the TeMagS module specially programmed travel pathways of the robots' liquid channel arms represent an essential measure to avoid cross-contaminations. Quantification data is used to normalize template DNA for STR PCR. Unpromising concentrations of human DNA below an empirically determined cutoff value are routinely not submitted to STR PCR amplification. The sample preparation for CE comprises a rearrangement of the plate positions of individual amplification products to avoid CE runs without PCR products. The complete analysis process from cell lysates to STR results is accomplished within 15 h ([Table 2](#)).

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Table 1
Operational steps and hardware components of the DNA analysis automation concept

Operational steps	Hardware components	Applications
DNA extraction	Tecan Freedom EVO [®] 150 liquid handling workstation I	ChargeSwitch [®] Technology (Invitrogen)
Real-time PCR setup	Tecan Freedom EVO [®] 150 liquid handling workstation II	Quantifiler [™] Human DNA Quantification Kit (AB)
Real-time PCR	AB 7500 Real-Time PCR System	SDS v1.2.3 (AB)
Normalization/negative selection		Special created Software tool (Tecan, ERL)
STR PCR setup	Tecan Freedom EVO [®] 150 liquid handling workstation II	Kits of various manufacturers
STR PCR	AB 9700 GeneAmp PCR System	
Amplification product pipeting/preparation for CE/rearrangement	Tecan MiniPrep [™] 75 liquid handling workstation III	Special created Software tool (Tecan, ERL)
Fragment analysis	AB Prism 3130x1 Genetic Analyzer	Data Collection Software v3.0 (AB)
Data analysis		Genemapper [®] ID S v3.2 (AB)

4. Discussion

To cope with the increasing demand for forensic DNA analysis capacity we established a semi-automated system for DNA profiling that was adapted to our specific demands and is now performing routinely the efficient, contamination free and high quality DNA analysis of diverse biological samples for routine casework at the LKA BW. To avoid errors by human failure, barcode sample identification and convenient transmission and management of sample and analysis data via LIMS were established. Our measures, especially the early pick out of DNA-free samples, were found to minimize significantly analysis time and costs. As a result personal capacity was set free for other time-consuming and labour-intense tasks like the microscopic preparation of perpetrators' skin particles from relevant case-work objects. Next, analysis data should be incorporated automatically via LIMS into reports and expertises.

Table 2
Time flow of the semi-automated process for DNA profiling of forensic case-work samples

Time flow (bulk of 88 samples)	
Manual separation of lysates from stain carriers	50 min
CST [™] extraction + transfer to plate (Tecan I)	2 h
Real-time PCR setup (Tecan II)	30 min
Real-time PCR (AB 7500)	1 h 45 min
Evaluation of Real-time PCR data	20 min
STR PCR setup (Tecan II)	45 min
STR PCR (AB 9700)	3 h 30 min
CE sample preparation (Tecan III)	1 h
CE (AB 3130x1)	4 h
Total	14 h 40 min

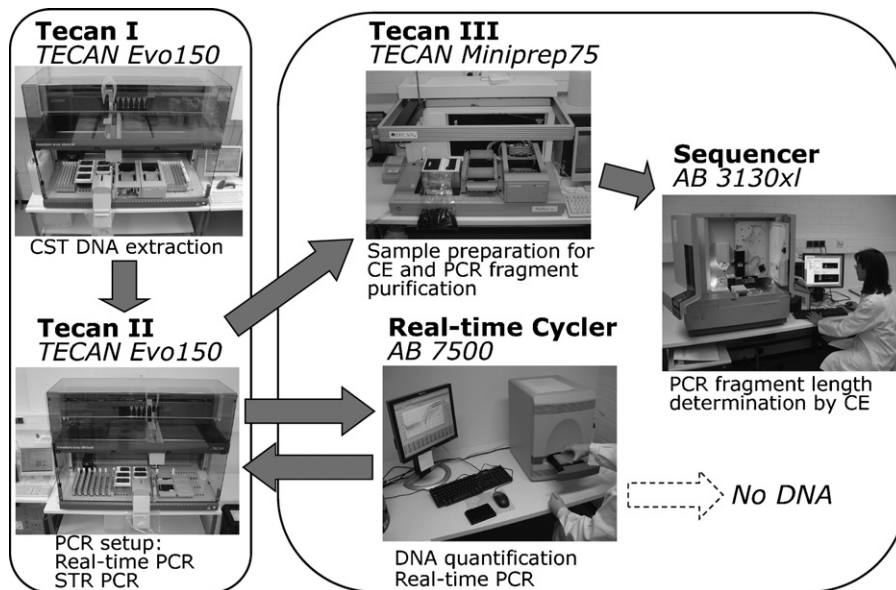


Fig. 1. Workflow scheme of the semi-automated DNA analysis of crime scene samples.

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Conflict of interest

None.

References

- [1] B. Haak, et al., Evaluation of a semi-automated, magnetic bead-based DNA-extraction method for genetic fingerprinting of forensic casework samples, *Forensic Sci. Int.* 1 (2008) 35–36.
- [2] 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.