International Congress Series 1288 (2006) 583-585





Evaluation of methodology for the isolation and analysis of LCN-DNA before and after dactyloscopic enhancement of fingerprints

Peter Leemans^a, André Vandeput^b, Nancy Vanderheyden^a, Jean-Jacques Cassiman^a, Ronny Decorte^{a,*}

 ^a Laboratory of Forensic Genetics and Molecular Archaeology, Department of Human Genetics, K.U. Leuven, Campus Gasthuisberg O&N, Herestraat 49-bus 602, B-3000 Leuven, Belgium
^b Laboratory of Technical and Scientific Police, Leuven, Belgium

Abstract. This study describes the quantitative and qualitative results of the evaluation of different methods (cellotape, swab or gauze) for recovering fingerprints from objects, the isolation of 'Low Copy Number' DNA (LCN-DNA) with silica or magnetic beads, the influence of time on the conservation of DNA on swabs used for sampling fingerprints and the ability to analyze DNA from fingerprints after dactyloscopic enhancement. © 2005 Published by Elsevier B.V.

Keywords: Latent fingerprints; Low copy number DNA; Quantifiler; STRs; SNPs

1. Introduction

Several studies have demonstrated the feasibility of using latent fingerprints for forensic DNA analysis [1–4]. However, the analysis of these LCN-DNA samples is not trivial and leads frequently to no results, partial results or the recovery of mixed DNA profiles. As this kind of evidence material is increasingly being submitted by the police for DNA analysis, we wanted to evaluate if current methodologies of sampling and DNA extraction in the laboratory are optimal for DNA analysis of latent fingerprints.

2. Materials and methods

Fingerprints were applied by pressure for 10 s onto clean microscope glass slides by 6 different donors. The fingerprints were recovered by using cellotape, cotton swabs with

* Corresponding author. Tel.: +32 16 345860; fax: +32 16 345997. *E-mail address:* ronny.decorte@med.kuleuven.be (R. Decorte).

^{0531-5131/ © 2005} Published by Elsevier B.V. doi:10.1016/j.ics.2005.09.079

physiologic water, cotton swabs with lyses buffer (ATL; Oiagen) or cotton gauze with physiologic water. The 'double-swab' method was applied when swabs were used [3]. Four different methods were evaluated for DNA extraction according to the procedures described by the manufacturers: the QIAamp DNA Mini Kit (Qiagen), the QIAquick PCR Purification Kit (Oiagen), a combination of both kits by using the flow-through, after binding of the DNA on the QIAamp DNA Mini Kit columns, on the QIAquick PCR Purification Kit columns, and the ChargeSwitch® Forensic DNA Purification Kit ('CST'; Invitrogen). The effect of storage was investigated by sampling fingerprints with swabs (physiologic water or ATL-buffer) and keeping at room temperature for 1, 2, 4, 6 and 8 weeks in closed paper envelopes before DNA extraction. The effect of fingerprint enhancement methods was studied by using either white powder, black powder, cyanoacrylate fuming or enhancement of cyanoacrylate with basic yellow. The LCN-DNA extracts were evaluated quantitatively and qualitatively, respectively, with the Quantifiler[™] Human DNA Quantification kit, and by the amplification of Short Tandem Repeats (STR; AmpFISTR® SGM Plus® and Profiler[®], Applied Biosystems, and PowerPlex[®] Y, Promega) or Single Nucleotide Polymorphisms (SNP) on the Y chromosome (De Maesschalck et al., in preparation).

3. Results and discussion

3.1. Quantitative results

The two-step procedure of using the flow-through of the 'DNA Mini' columns onto the 'PCR Purification' columns was based on the assumption that DNA from fingerprints might show some degradation and be better isolated with the 'PCR Purification' columns. The results of these experiments confirmed this hypothesis: the positive control (blood on FTA card) showed a 55 times lower amount of DNA in the flow-through, while the DNA from the fingerprints was almost equally divided between the two methods (average ratio of 1:1.2). The total amount of DNA recovered with the two-step procedure (average of 5.63 ng) was in line with the 'PCR Purification' method (7.65 ng). The 'PCR Purification' kit showed also a 2-fold higher amount of DNA than the 'DNA Mini' kit (3.45 ng) or the 'CST' kit (3.19 ng). This difference was probably due to the presence of only one 'Good Shedder' (GS) among the donors and 5 'Poor Shedders' (PS), and to differences in the amount of epithelial cells present on the glass slides. The amount of DNA obtained from the GS was 13.8-fold higher than from the PS. Further experiments where the swabs were stored for several weeks at room temperature did not confirm the observed higher DNA recovery with the 'PCR Purification' kit.

Current practice of sampling fingerprints on objects relies on the use of cotton swabs. Use of alternative methods such as cellotape and cotton gauze showed in this study that the swab method is preferable as it resulted in a higher amount of DNA recovered: average of 4.76 ng versus 0.5 ng with the cellotape and 1.56 ng with gauze. Therefore, the swab method was used in the remaining study. Similarly, use of a lyses buffer (ATL) instead of physiologic water for recovering the cells from fingerprint deposits did not increase the amount of DNA, even when swabs were stored at room temperature for several weeks. These experiments showed also no trend (in-or decrease) in the amount of DNA recovered from the swabs that were stored for different time periods indicating that loss of DNA on swabs from fingerprints is not a rapid process.

3.2. Qualitative results

The obtained DNA from the different methodologies was used for the amplification of STR loci. The DNA profiles were compared to the profiles of the donors and evaluated for the

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number of loci amplified (success rate), the presence of mixed profiles, allele-drop-out and discordant genotypes. No difference was observed in the ability to amplify DNA between the different sampling/extraction methods except for the 'PCR Purification' kit where no positive result was obtained. However, the loci could be amplified after a 2-fold dilution of the DNA extract indicating the presence of an inhibitor, which was not identified in the Quantifiler® assay. There was no trend in the success rate from DNA on swabs stored up to 8 weeks before DNA extraction indicating that DNA degradation is a slow process after sampling and storage at room temperature. The average success rate for the 21 loci amplified ranged between 50% and 99%, while artifacts such as mixed genotypes, allele-drop-out and discordant loci were observed respectively with a frequency of 8-45%, 1-23% and 0-6%. These artifacts were mainly seen for the extracts from the PS. The majority (>70%) of the profiles from the GS were complete profiles without artifacts, which was in contrast to the profiles from the PS (1-3%). The donor of the fingerprint could be identified in more than 82% of the GS profiles and 28% of the PS profiles when this was extended to profiles with at least 10 STRs and the presence of the donor profile in a mixed profile. The presence of mixed DNA profiles indicates the presence of additional DNA on the glass slides. The origin of this DNA could not be determined but secondary transfer cannot be excluded [4]. Finally, the DNA extracts from the GS (male) were used for the analysis of STRs and SNPs on the Y chromosome. A 100% success rate was obtained with the Y-SNPs while this was 82% for the Y-STRs.

3.3. DNA analysis after dactyloscopic enhancement of fingerprints

Fingerprint samples from 3 individuals (2 PS and 1 GS) were processed for dactyloscopic enhancement as described in Section 2 and extracted with the 'DNA Mini' kit. Quantification of the DNA showed that the amount of DNA recovered for the GS was in line with the results before enhancement, in contrast to the PS's showing a lower amount of DNA. DNA typing of the extracts showed similar results as before enhancement indicating that the fingerprint enhancement methods did not resulted in inhibition of the amplification process. A complete profile for the GS was obtained while the PS's showed a mixed profile. The origin of the supplementary DNA profile remains unknown and could be the result of secondary transfer. Further experiments are necessary in order to confirm these obtained results and to identify the origin of the additional DNA profiles.

Acknowledgements

Jean-Jacques Cassiman is a holder of the 'Arthur Bax and Anna Vanluffelen chair of Human Genetics'.

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