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Contents lists available at ScienceDirect

Forensic Science International: Genetics

journal homepage: www.elsevier.com/locate/fsig



Probability of detection of DNA deposited by habitual wearer and/or the second individual who touched the garment



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ARTICLE INFO

Article history:
Received 25 February 2015
Received in revised form 5 September 2015
Accepted 2 October 2015
Available online 17 October 2015

Keywords: DNA Touch DNA Wearer Indirect transfer Background DNA Likelihood ratio

ABSTRACT

Available literature on the detection of transferred DNA does not address the interpretation issues in relation to who wore rather than touched the garment. To acquire a greater knowledge of the rate of detectable wearer, toucher and background DNA, 63 males wore their own underpants for 12 h. The inside-waistband was handled by one of 11 female volunteers for 15 s. The waist-band was mini-taped and subjected to DNA profiling with the AMPF ℓ STR® NGM SElectTM kit.

The findings show that on worn garments the probability of observing reportable DNA profiles is 61.9%. The wearer was detected as a single profile or part of a mixed profile in 50.8% of samples. When the wearer was present in a mixture, he was always observed as the major contributor. The toucher was detected on 11.1% of underpants. Reportable background DNA (non-wearer and non-toucher) occurs in 14.3% of samples and may affect the assessment of who wore the garment.

Greater knowledge of the frequency of detection of reportable wearer DNA and/or toucher allows scientists to evaluate the likelihood of observing a matching profile if an individual wore a garment rather than touched it in disputed case scenarios.

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1. Introduction

Forensic scientists often examine clothing that was discarded at or near to a scene [1,2] for the determination of who the wearer was. However, with the onset of 'touch DNA profiling' [2–4] and the possibility of indirect transfer [5–9] the true wearer of the garment can be called into question.

Some research on trace DNA suggests the last user/toucher results as the major contributor to mixed DNA profiles. This has been observed on clothing [10], steering wheels [2] and wallets [11]. On the other hand, a recent study by Van Oorschot et al. [12] found that the primary user's DNA profile was retained on soft porous items even after use by a second person [12]. Meakin and Jamieson's review of current data concludes that the quality of a DNA profile cannot infer the mode or duration of deposition of this DNA or determine the last user of an item or wearer of a garment [4]. This available literature does not address the interpretation issues in relation to who wore the garment.

To help the scientist to address the issue of whether the observed DNA profile on clothing is from the wearer or another source, the Authors sought to establish data on the frequency of detection of DNA from wearer, toucher or others when individuals wore and handled worn garments under normal circumstances. Using an evaluative approach [13] the data can be used to estimate the likelihood of the detection of the DNA profile if the garment was worn or touched.

2. Materials and methods

2.1. Collection of sample underwear

A total of sixty three male volunteers aged between 18 and 35 years of age wore their own underpants for a minimum of 12 h under normal circumstances. Each underpants was returned in a self-seal plastic bag. The waistband of the underwear was used as a proxy for cuffs and collars of worn clothing which would have direct contact with the wearer's skin. The samples were stored in a secure environment at room temperature.

All volunteers provided buccal DNA samples using sterile sealed plain cotton swabs (Copan, Brescia, Italy). All samples were

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collected according to the guidelines approved by the Limerick Institute of Technology Ethics Committee.

2.2. 'Toucher' scenario experiment

A total of eleven female volunteers washed their hands 1 h prior to the experiment. Each female removed an item of worn underpants from a sterile bag and then held the worn underpants by the inside waistband for 15 s with their bare hands [Fig. 1]. Afterwards, they refolded the garment back into the bag and returned it for analysis. The females then rewashed their hands and went about their daily schedule for 1 h where they repeated the procedure on another underpants. This procedure was repeated by the female volunteers until all 63 male underwear samples were touched by a single individual. A single mini-tape (WA Products, Birmingham), was used to lift DNA from around the inside of the whole waistband. The mini-tape was cut-up with sterile scissors and added to nucleic acid optimiser (NAOTM) baskets situated in 2 mL microcentrifuge tubes (Copan, Brescia, Italy) and stored at $-20\,^{\circ}$ C until analysis.

Each female 'toucher' gave a reference sample on FTA Micro cards (Whatmann, UK) to compare with any DNA profiles observed on the underpants.

2.3. Persistence study

As there was an interval of two years between the wearing of the underpants and the subsequent touching and minitaping of the waistbands, a study was undertaken to assess the persistence of wearer DNA during storage. A total of ten male volunteers were given a new pair of cotton underpants distributed in self-seal bags. Cotton underwear was provided as it was the fabric type that had the highest representation among the original 63 male volunteers. The volunteers were requested to wear the underwear for approximately 12 h under normal circumstances and return the underwear for analysis. The inside of the waistbands were separated into multiple equal sized sections (2 cm in length) labelled A, B or C (A, B, C, A, B, C...) right around the circumference of the waistband [Fig. 2]. This approach was taken in case there had been uneven distribution of the wearer in parts of the waistband. All of the sections labeled A were mini-taped with a single mini-tape upon receipt of sample (t-0), all sections B after 6 weeks and sections C after 12 weeks. All sections were mini-taped with multiple touches with the tape-lift. The underwear was stored in a secure, no traffic area during the time intervals.



Fig. 1. Photograph of female 'toucher' touching the inside waistband of underwear worn by a male volunteer under normal circumstances as part of toucher scenario.



Fig. 2. Photograph of the inside waistband of male underwear, which was separated into equal sized sections for mini-taping at a given timeline: sections A were minitaped initially, sections B after 6 weeks and sections C post 12 weeks. The letters A, B and C are for illustration purposes.

Each male volunteer gave a reference sample on FTA Micro cards (Whatmann, UK) to compare with any DNA profiles observed on the underpants.

2.4. DNA extraction from samples

Mini-tapes were added to a lysep column seated in a 2 mL microcentrifuge tube (Copan, Brescia, Italy) where 280 µL prewarmed ATL buffer and 20 µL of Proteinase K was added to the sample. The sample was vortexed and incubated on a shaker for 15 min at 56 °C with 1400 rpm rotation, the sample was subsequently centrifuged at 9600 rpm for 2 min to filter the lysate through lysep column. DNA from lysate was extracted via the use of a Hamilton Star extraction robot with the addition of 900 µL binding buffer, 100 μ L uniform magnetic bead solution and 300 μ L of sample lysate to the deep well plate (DWP). The DWP was transferred to the heated shaker for 600 s (1000 rpm) at 80 °C and subsequently to the magnet for 60 s. The lysate was removed to a waste plate. The DWP was placed back on heated shaker, where 500 µL of wash 1 buffer was added and the DWP shaken for 30 s at 1000 rpm. The plate was moved to the magnet for 60 s and the wash 1 buffer removed. This process was repeated with the addition of 500 µL of wash 2 buffer. This procedure was repeated for further wash steps. The plate was air dried at room temperature for 600s and 100 µL elution buffer was added. The plate was moved to the heated shaker for 300s at 70°C at 1000 rpm. The elution buffer containing the DNA was subsequently removed for DNA quantification.

DNA from reference buccal swabs was extracted with use of a silica based method using Omega Bio-tek E.Z.N.A.® Forensic DNA kit (VWR, Ireland) following manufacturers guidelines on buccal swab DNA extraction [14].

FTA Micro cards were processed by addition of one punch to a pre-wet (nuclease-free dH_2O) 96 well plate by use of the BSD punching unit and TECAN EVO 200 robot.

Blank tape-lifts and blank swabs were used as negative controls to ensure procedure was contamination free. Positive controls

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