



Comparison of operational DNA recovery methods: Swabs versus tapelifts

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ABSTRACT

It is routine among many jurisdictions to recover DNA using tapelifts on porous substrates (e.g. clothing) and swabs on non-porous substrates (e.g. tool handles). Here, we examine this by comparing the efficiency of the NSW jurisdiction's specific swabbing and tapelift techniques on a range of porous and non-porous substrates. To test DNA recovery efficiency, 30 μ l aliquots of 1:50 and 1:100 saliva dilutions were deposited onto the substrates, left to dry overnight, recovered, extracted, quantified and a subset profiled. Tapelifts recovered more DNA and DNA profiles with more detectable alleles than swabs for both saliva dilutions on porous substrates. For non-porous substrates, similar DNA quantities and profiles were generally recovered with both methods for both saliva dilutions. These data underpin current practices to recover DNA using tapelifts for porous substrates and swabs for non-porous substrates. These data also revealed severe degradation of DNA recovered from brass, supporting the on-going need to improve DNA recovery and analysis methods for brass substrates.

1. Introduction

For DNA recovery, it is routine among many jurisdictions, particularly in Australia and the UK, for tapelifting to be used on porous substrates (such as clothing [1]) and swabbing on non-porous substrates (such as tool/weapon handles [2]). However, anecdotal evidence and emerging data [3] suggest that a fresh assessment of the collection methods for various substrates is warranted.

Here, we examine this by comparing the efficiency of the specific swabbing and tapelift techniques used in the NSW jurisdiction for sampling a range of porous and non-porous substrates. The specifics of the techniques employed are tailored to the automated DNA processing pipeline utilised within the Forensic & Analytical Science Service DNA laboratory. Briefly, a single moist-dry swab is used [4] or a tapelift of specific size to fit a 2 ml robot-ready tube when rolled.

2. Materials and methods

Saliva was donated by a consenting participant by spitting into a DNA-free 50 ml falcon tube. A preliminary experiment was conducted in which 30 μ l aliquots of four dilutions of the provided saliva (1:25, 1:50,

1:100 and 1:200) were pipetted on to two pre-cleaned substrates (cotton and tile) and were recovered by swabbing and tapelifting in duplicate per substrate and recovery method. Results from this initial experiment identified the 1:50 and 1:100 dilutions as those to be used in the main experiment. The 1:50 dilution resulted in DNA quantities corresponding to those routinely recovered from dilute body fluids, whereas the 1:100 dilution provided DNA quantities similar to those routinely recovered from trace DNA samples.

As such, 30 μ l aliquots of each of these two saliva dilutions were deposited on to pre-cleaned DNA-free substrates (5 replicates per dilution per substrate per recovery method: $n = 120$ plus controls). The substrates used were: cotton, denim and polyester for porous substrates, and tile, brass and synthetic leather for non-porous substrates. The deposited saliva was left to dry on the substrates overnight and then sampled using moist-dry rayon swabs (Medical Wire & Equipment, UK) or tapelifts (3 M tape cleaned and packaged by Lovell Surgical Solutions Pty. Ltd., Australia) in the manner employed within the NSW jurisdiction.

All samples were lysed and extracted using the PrepFiler™ Automated Forensic DNA Extraction Kit (Thermo Fisher Scientific) on the Hamilton Microlab® AutoLys STAR and Tecan Freedom EVO® robotic

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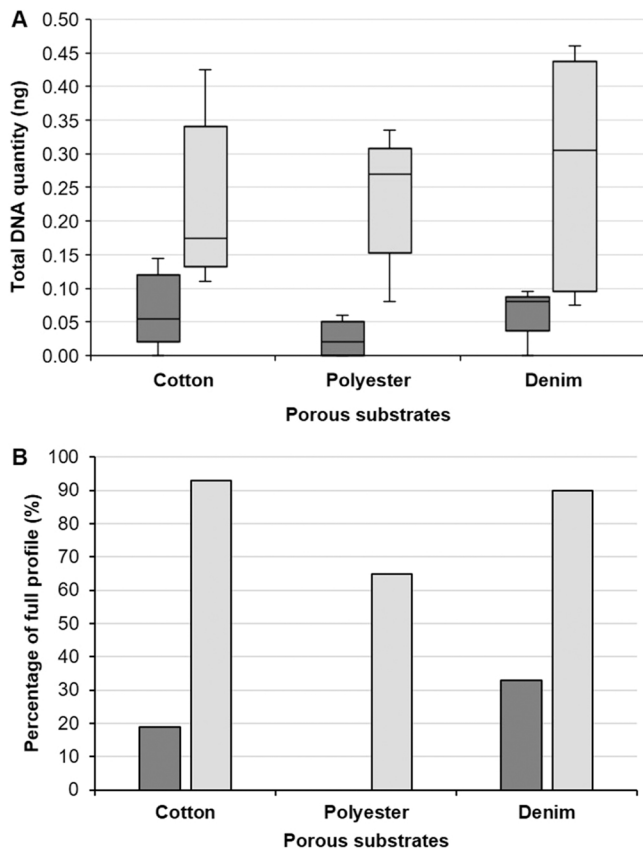


Fig. 1. DNA quantities (A) and profile percentages (B) recovered from 1:50 saliva dilution on porous substrates by swabbing (dark grey) and tapelifting (light grey).

workstations, respectively, with an elution volume of 50 μ l. Samples were quantified using the Quantifiler™ Trio DNA Quantification Kit (Thermo Fisher Scientific), and a sub-set was profiled using the PowerPlex® 21 Amplification System (Thermo Fisher Scientific) with a 0.7 ng input amount and 29 cycles.

Statistical testing was conducted using IBM SPSS Statistics, version 28. Of the 24 combinations of recovery method, substrate and saliva dilution, five combinations resulted in DNA quantification datasets that were not normally distributed, as determined by the Kolmogorov Smirnov test ($p < 0.05$). As such, to make pairwise comparisons across all the datasets, the non-parametric Mann-Whitney U test was used to identify any significant differences in DNA quantities recovered. Effect size of significant differences was interpreted from r^2 , determined by squaring the value of r , calculated from $r = Z/\sqrt{N}$, where $N = 10$ for each comparison.

3. Results and discussion

3.1. Porous substrates

For the 1:50 saliva dilution on porous substrates, tapelifts recovered notably more DNA than swabs (Fig. 1A). This was statistically significant for cotton ($Z = -2.402, p < 0.05, r^2 = 0.58$) and polyester ($Z = -2.619, p < 0.05, r^2 = 0.69$), indicating that 58 % and 69 % of the variability is accounted for by the recovery method for cotton and polyester, respectively. The difference in DNA quantity for denim was not significant ($Z = -1.892, p = 0.056$), presumably due to the large variability in results obtained from denim for this saliva dilution (Fig. 1A). Higher DNA recovery was also observed with tapelifts than swabs for the 1:100 saliva dilution, though this was only significant for polyester and denim (data not shown). Tapelifting also resulted in DNA profiles with more

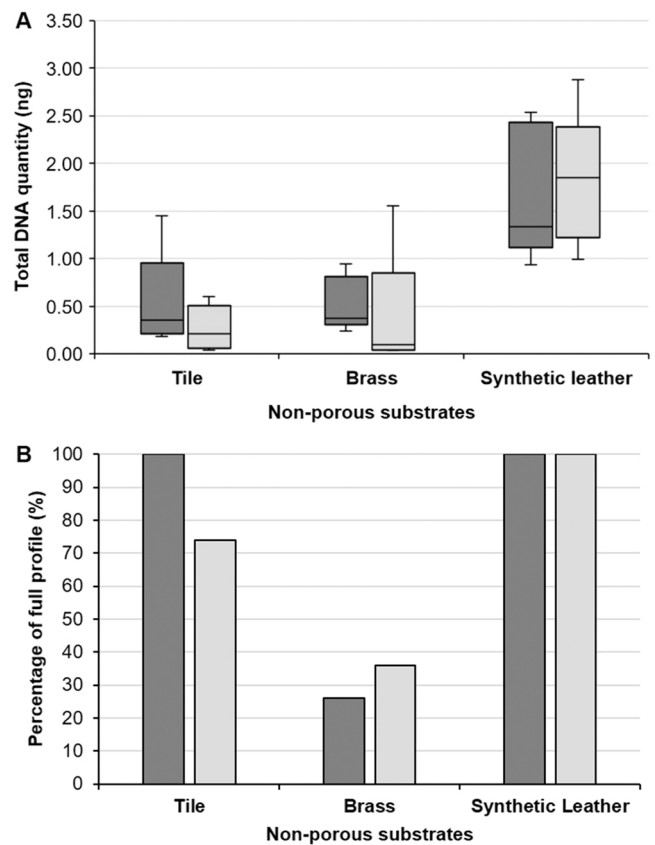


Fig. 2. DNA quantities (A) and profile percentages (B) recovered from 1:50 saliva dilution on non-porous substrates by swabbing (dark grey) and tapelifting (light grey).

detectable alleles than swabbing for the 1:50 (Fig. 1B) and 1:100 (data not shown) saliva dilutions, which was expected given the quantification data.

3.2. Non-porous substrates

For the 1:50 saliva dilution, there was no significant difference in DNA quantity recovered between tapelifting and swabbing on tile ($Z = -0.940, p = 0.347$), brass ($Z = -1.571, p = 0.116$) and synthetic leather ($Z = -0.522, p = 0.602$) (Fig. 2A). Whilst similar quantities of DNA were recovered from both tile and brass, poorer quality DNA profiles were recovered from brass than tile (Fig. 2), with ‘ski-slope’ degradation observed in the DNA profiles from brass. This supports prior findings that DNA profiling from metal substrates is challenging and may be attributed to the physicochemical properties of brass, in particular the copper within brass, causing DNA degradation [5]. For the 1:100 saliva dilution, similar DNA quantities were recovered with both methods on brass and synthetic leather, but swabbing recovered notably more DNA than tapelifting on tile (data not shown). This increase was statistically significant ($Z = -2.095, p < 0.05, r^2 = 0.44$), indicating that 44 % of the variability is accounted for by the recovery method. The DNA profile percentages obtained for the non-porous substrates reflected the DNA quantity findings for the 1:50 (Fig. 2B) and 1:100 (data not shown) saliva dilutions.

4. Conclusions

The results in this study underpin the current NSW practices to recover DNA using tapelifts for porous substrates and swabs for non-porous substrates. These data also revealed severe degradation of DNA recovered from brass, supporting the on-going need to improve DNA

recovery and analysis methods for brass and other copper-based substrates.

Conflict of interest statement

None.

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