Evaluating Direct Lysis and Swabbing Techniques for Improved DNA Typing from Adhesive Surfaces

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INTRODUCTION

One of the most common types of evidence found at a crime scene is fingerprints. Over the past two decades, the forensic community has increasingly recognized the potential of fingerprints as a viable source for DNA analysis [1]. On the other hand, adhesive tapes of various kinds can be used in different criminal activities like binding or restraining victims, covering up weapons, or packaging illegal substances and are routinely submitted to crime labs as evidence. These adhesive tapes might contain latent fingerprints which in turn could be a source of touch DNA. Touch DNA, found on fingerprint residues, likely originates from various biological materials, including cell-free DNA, shed corneocytes, nucleated epithelial cells, and fragmented cells and nuclei [2,3,4,5]. Additionally, crime labs may use a variety of fingerprint lifters which too could be potential sources of touch DNA. In the past a wealth of research has been conducted on analyzing DNA from fingerprints. However, not much focus has been given on the methods of collecting DNA for efficient genetic interpretation. This is crucial since the quality of the DNA collected can significantly impact the likelihood of generating useful genetic profiles for criminal investigations. Keeping that in mind, in this project two commonly encountered adhesive tapes- Duct Tape and Electrical Tape and three types of fingerprint lifters- Hinge Lifters, Gel Lifters and Mikrosil® Casting Putty were selected as the sample types and two collection methods- Swabbing and Direct Lysis were compared to determine the best DNA collection method in terms of DNA yield and ease of implementation.

MATERIALS & METHODS

Sample Preparation:

Duct Tape and Electrical Tape:

- Fingerprints were deposited on 21 pieces of each tape type.
- 3 pieces were directly subjected to lysis while the remaining were swabbed.
- 9 pieces of Duct Tape and Electrical Tape each were set aside for swabbing with Nylon Swab and the remaining 9 pieces of each tape were set aside for swabbing with Cotton Swab.
- Swabbing for each swab type was performed using 3 different moistening agents- Water, Xylene and Lysis Buffer, in triplicates.

Hinge Lifter, Gel Lifter and Mikrosil® Casting Putty:

- 18 separate fingerprints were deposited on glass slides.
- 6 fingerprints were lifted using Hinge Lifters, Gel Lifters, and Mikrosil® Casting Putty each, with DNA free fingerprint lifting powder and DNA free brushes.
- 3 of each fingerprint lifter with the fingerprints were directly subjected to lysis.
- Remaining 3 of each fingerprint lifter with the fingerprint were swabbed using Cotton Swabs moistened with Lysis Buffer.

DNA Extraction:

DNA extraction was performed using the EZ1 DNA Investigator Kit on the EZ1 Advanced XL system (QIAGEN).

DNA Quantification and Amplification:

Samples were quantified using the Investigator Quantiplex Pro Kit (QIAGEN) on Applied Biosystems 7500 Real-Time PCR Systems (ThermoFisher Scientific). Extracted samples were amplified using the Investigator 24plex QS Kit (QIAGEN) according to the manufacturer's protocol.

Separation, Detection and Analysis:

Amplified DNA samples were separated and detected on Applied Biosystems 3500 Series Genetic Analyzer (ThermoFisher Scientific). Data was analyzed on GeneMapper ID-X Software v1.6 (ThermoFisher Scientific) with an analytic threshold of 100 RFUs.

RESULTS & DISCUSSION

Table 1: Comparison of DNA Concentrations (ng/µl) from latent fingerprints on Duct Tape and Electrical Tape samples using Swabbing and Direct Lysis methods. Direct Lysis produced the highest DNA concentrations. Cotton Swabs performed better than Nylon Swabs in terms of DNA yield. Across all sample types, Cotton Swabs in combination with Lysis Buffer consistently recovered more DNA in comparison to all other combinations of swabs and moistening agents.

Concentrations (ng/µl)	Duct Tape			Electrical Tape		
	Swab		Direct Lysis	Swab		Direct Lysis
	Nylon	Cotton		Nylon	Cotton	
Lysis Buffer	0.0037	0.0215	0.0416	0.0064	0.0258	0.045
Xylene	0.0008	0.0084		0.0096	0.0077	
H_2O	0.0007	0.0362		0.0022	0.0124	

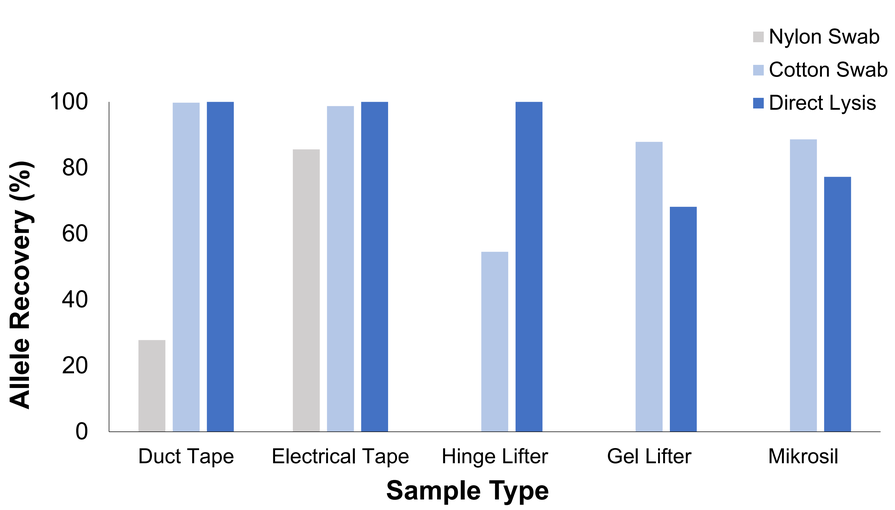

Figure 1: Comparison of average allele recovery obtained from Swabbing and Direct Lysis of all adhesive substrates. Overall, Nylon Swabs did not recover as many alleles as Cotton Swabs. Direct Lysis resulted in 100% allele recovery for Duct Tape, Electrical Tape and Hinge Lifters. In contrast, Gel Lifters and Mikrosil® Casting Putty recovered a greater percentage of alleles when swabbed with Cotton Swabs moistened with Lysis Buffer.

CONCLUSIONS

- Crime lab analysts should consider examining adhesives with latent fingerprints using not only fingerprint examination but also DNA analysis to increases the probative value of evidence.
- Among Swabbing methods, Cotton Swabs with Lysis Buffer was most effective, particularly for Gel Lifters and Mikrosil® Casting Putty.
- Overall, Direct Lysis produced the highest DNA yields for Duct Tape, Electrical Tape, and Hinge Lifters.
- The findings of this project suggest that the collection method should be optimized based on the sample type.
- In the future, a subset of samples will be sequenced using the ForenSeq Kintelligence Kit to evaluate their potential as probative samples for FGG purposes.

Table 2: Comparison of DNA Concentrations (ng/µl) from latent fingerprints lifted with Hinge Lifters, Gel Lifters, and Mikrosil® Casting Putty using Swabbing and Direct Lysis methods. Direct Lysis recovered higher average DNA concentrations for Hinge Lifters while Swabbing resulted in higher DNA recovery for Gel Lifters and Mikrosil® Casting Putty.

Concentrations (ng/µl)	Hinge Lifters	Gel Lifters	Mikrosil® Casting Putty
Cotton Swab/Lysis Buffer	0.0025	0.0084	0.0046
Direct Lysis	0.0549	0.0019	0.0014

- When comparing moistening agents, Lysis Buffer was able to recover at least 30 pg/µL of DNA when used to moisten both Nylon and Cotton Swabs for both Tape types (Table 1). In contrast, H_2O and Xylene did not effectively recover DNA when moistening Nylon Swabs used to collect DNA from Duct Tape (**Table 1**).
- Nylon Swabs were not as effective in recovering DNA or a greater percentage of alleles from Duct Tape as they were with Electrical Tape (**Table 1; Fig. 1**). Additionally, Nylon Swabs recovered less DNA and alleles from Tape samples than did Cotton Swabs (**Table 1; Fig. 1**). Hence, when comparing swabbing of latent fingerprint lifters and Direct Lysis, only Cotton Swabs moistened with Lysis Buffer were examined.
- Overall, results indicated that the Direct Lysis method yielded higher concentrations of DNA from Duct Tape (**Table 1**), Electrical Tape (**Table 1**), and Hinge Lifters (**Table 2**), while Swabbing recovered more DNA from Gel Lifters and Mikrosil® Casting Putty (**Table 2**).
- As expected, when comparing average allele recovery, complete DNA profiles were obtained using Direct Lysis of Duct Tape, Electrical Tape, and Hinge Lifters (**Fig. 1**).
- The combination of Cotton Swabs with Lysis Buffer achieved >98% allele recovery for both Duct Tape and Electrical Tape (**Fig. 1**). In case of Gel Lifters and Mikrosil® Casting Putty 87.88% and 88.64% allele recovery was observed, respectively, which was higher than that recovered using Direct Lysis. Swabbing Hinge Lifters recovered substantially less DNA than subjecting the substrate to Direct Lysis.
- Overall, Swabbing techniques yielded lower allele recovery compared to Direct Lysis (**Fig. 1**).

ACKNOWLEDGEMENTS

We extend our gratitude to QIAGEN for providing the reagents essential to this project. We also thank the Department of Forensic Science and the Graduate and Professional School at Sam Houston State University for their partial funding and the opportunity to conduct this research.

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