DNA transfer via blood with varying substrates, pressures, and times

Nicole L. Agre[1]* and Jamie S. Spaulding Ph.D[1]

[1]Department of Criminal Justice and Forensic Science, Hamline University, Saint Paul, MN 55104

*E-mail: nagre01@hamline.edu

Introduction

DNA is a very important piece of evidence in a criminal case due to its ability to individualize (1). Through the years of its use, DNA technology has improved to become much more sensitive. This means that smaller evidence samples are needed for a DNA profile to be generated (1). These improvements resulted in some issues with DNA. The issue of most importance is the idea of DNA transfer. DNA transfer can be broken down into two categories: direct transfer and indirect transfer (2). Direct transfer is when DNA is directly deposited by a donor onto a surface. Indirect transfer is when DNA already on a surface is transferred onto another surface.

The implications of indirect DNA transfer has on forensic science is immense. Investigators are no longer worried about if they will find DNA evidence, but how DNA evidence got there (1). One piece of evidence could generate many profiles, many not relevant to the case (3). Forensic scientists are being increasingly called upon to quantify the probability of DNA evidence being from an indirect transfer in court testimony (1, 2). Forensic scientists need to understand how DNA transfer works and what factors affect DNA transfer in order to make informed testimony to the jury. There have already been many factors that have been examined for their potential to affect DNA transfer. They include environmental factors like heat and humidity, the type of biological material, and many others (1).

The purpose of this study is to look at several factors: type of substrate, duration of contact, and pressure. These factors have been researched before, but this study aims to more accurately reflect the conditions possibly found on a crime scene. Pressures heavier than weights used in other studies will be used (3, 4). A semi porous substrate will also be investigated whereas only nonporous and porous substrates are usually looked at (3, 5).

Methodology

Transfer Scenarios

- Blood will be pipetted onto a clean glass slide and then transferred to one of the four substrates
- Each transfer will have a different pressure, substrate, and duration of contact
- Samples will be left to dry and then stored if needed

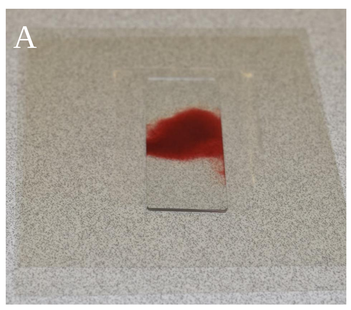

Figure 1. Photographs of the transfer scenario setup. A shows the setup without any pressure applied. The layers consist of a transparency, the glass slide, blood, the substrate, and then another transparency. B shows the setup with the addition of pressure. The appropriate weight is placed in the canister, and then the canister is placed onto the second transparency.

DNA Extraction

- DNA extractions will be performed with Qiagen's QIAamp® DNA Mini Kit using the manufacturer's protocol for dried blood spots

Quantitation

- Quantitation will be performed using a NanoDrop Microvolume Spectrophotometer (ThermoFisher Scientific)

Results

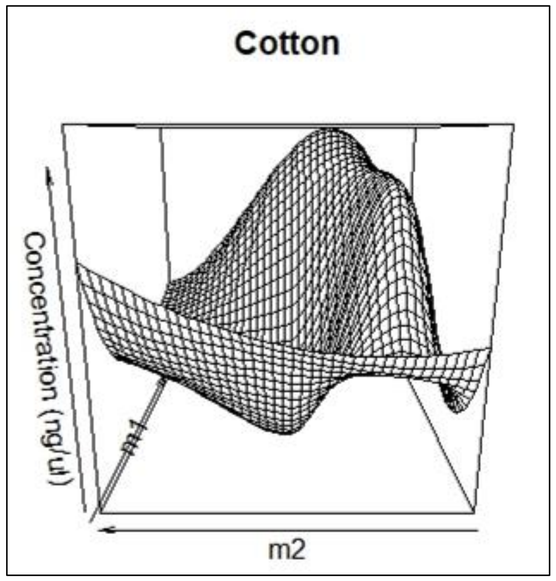

Figure 2. 3D Contour map of how variables affect DNA concentration in cotton substrate. m1 represents weight and m2 represents duration of contact.

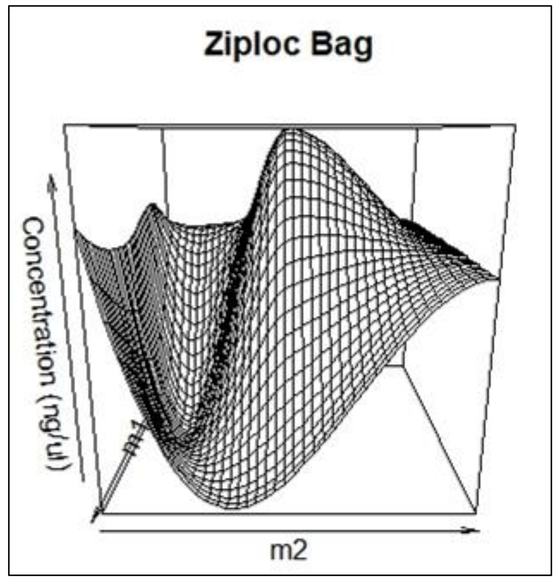

Figure 3. 3D Contour map of how variables affect DNA concentration in plastic substrate. m1 represents weight and m2 represents duration of contact.

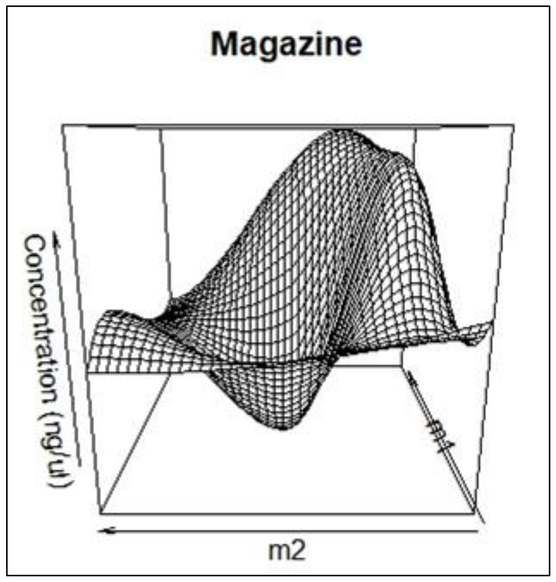

Figure 4. 3D Contour map of how variables affect DNA concentration in magazine substrate. m1 represents weight and m2 represents duration of contact.

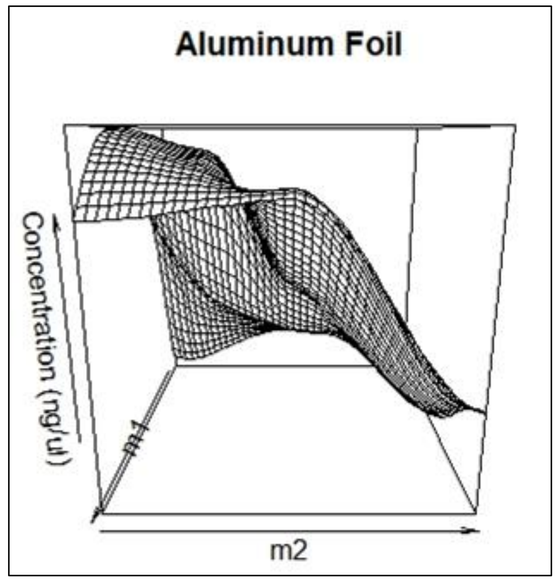

Figure 5. 3D Contour map of how variables affect DNA concentration in aluminum foil substrate. m1 represents weight and m2 represents duration of contact.

Kruskal - Wallis Test

	Kruskal - Wallis Chi Squared	P-value
Weight	3.3721	0.3377
Substrate	64.842	$5.422e^{-14}$
Duration of Contact	7.7844	0.05068

Table 1. A summary of the Kruskal - Wallis tests that were performed on the non-parametric data. Only the substrate variable was significant.

Dunn Test

	P-value
Aluminum Foil-cotton	$1.2416e^{-2}$
Aluminum Foil - Magazine	$2.1872e^{-8}$
Cotton - Magazine	$7.1258e^{-3}$
Aluminum Foil - Ziploc	1.000
Cotton - Ziploc	$6.3956e^{-5}$
Magazine - Ziploc	$7.5094e^{-13}$

Table 2. A summary of the Dunn test that was performed on the substrate data to see which pairs were significant. All comparisons except for the aluminum foil and ziploc comparison were significant.

Conclusions

- The level of DNA concentration was affected by the substrate that the blood was transferred to
  - Table 1
- Substrates differed in how weight and duration of contact affected DNA transfer through the blood
  - Table 2, Figures 2-5
  - Figure 2 and Figure 4 are similar looking despite being different types of substrates (porous and semi-porous respectively) but not significantly different according to Table 2
- Systematic loss of DNA occurs at each stage of the transfer process (not shown)
  - Depositing the sample, the contact with the primary substrate, the transfer itself, extraction, and quantitation

Deliverables

- Quantitative measurement and characterization of bloodstain transfer
- Images of bloodstain transfer
- Statistical analysis of all data and R code for reproducibility
- Recommendations for implementation of the developed methodology within forensic crime labs

Future Directions

- Further investigation into magazine data, as a negative control of just magazine showed a DNA concentration comparable to bloodstained cotton
  - Possibility of dyes interfering with the spectrophotometer
- Looking at how much DNA is lost at each step in the process of a transfer
- Possibly compare magazine to other porous substrates

References

1. van Oorschot, R.A.H., Meakin, G.E., Kokshoorn, B., Goray, M., Szkuta, B. "DNA Transfer in Forensic Science: Recent Progress towards Meeting Challenges." Genes (2021), 12, 1766. https://doi.org/ 10.3390/genes12111766

2. Meakin, G., Jamieson, A., "DNA transfer: Review and implications for casework", Forensic Sci. Int. Genet. (2013), http://dx.doi.org/10.1016/j.fsigen.2013.03.013

3. Goray, M., Eken, E., Mitchell, R.J., van Oorschot R.A.H., "Secondary DNA transfer of biological substances under varying test conditions", Forensic Sci. Int. Genet. (2009), doi:10.1016/j.fsigen.2009.05.001

4. Hefetz, I., Einot, N., Faerman, M., Horowitz, M., Almog, J., "Touch DNA: The effect of the deposition pressure on the quality of latent fingerprints and STR profiles, " Forensic Sci. Int. Genet. (2019), 38, 105-112, https://doi.org/10.1016/j.fsigen.2018.10.016

5. Verdon, T.J., Mitchell, R.J., van Oorschot R.A.H., "The influence of substrate on DNA transfer and extraction efficiency" (2012), https://doi.org/10.1016/j.fsigen.2012.09.004

Acknowledgements

- Dr. Jamie Spaulding, Department of Criminal Justice and Forensic Science at Hamline University, for research support
- SCUR Program and Professor Sharon Preves and Professor Leif Hembre for funding and research support
- The Department of Biology for allowing the usage of their NanoDrop and vortex
- Professor Betsy Martinez-Vaz for offering instruction on the NanoDrop and answering questions that were DNA focused