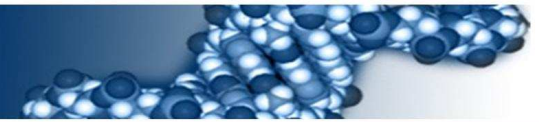


# Improving DNA Evidence Collection via Quantitative Analysis: A Systems Approach

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## Introduction

- In the collection of biological evidence from a crime scene, it is imperative to implement the most effective and robust collection method to ensure maximum DNA recovery.
- While common techniques for biological collection such as swabbing, cutting, scraping, and taping have been a mainstay in forensics, there are drawbacks of these techniques, which include, but are not limited to, the lack of surface area that may be processed, potential co-elution of PCR inhibitors, and non-optimized elution of cells from the substrate into solution.
- Due to this, an advancement, or new technique, in the area of biological evidence collection is needed in order to optimize collection from different items of interest, especially large items.
- Recent work in the field of pathogen testing suggests the use of a wet-vacuum collection system may be a valuable addition/alternative to already well-established biological collection methods (1).
- In this study, traditional biological collection methods, including the double swab method and taping, are compared to a wet-vacuum system (Microbial-Vac Systems® Inc., Bluffdale, UT) through the collection of different volumes of blood (0.075 - 75 µL) on tile, denim, and carpet.

## Methods



Figure 1. Representative image of different volumes of blood ranging from 0.075 to 75 µL on a non-porous substrate (tile).

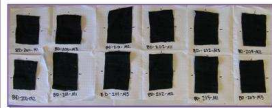


Figure 2. Representative image of different volumes of blood ranging from 0.075 to 75 µL on denim.

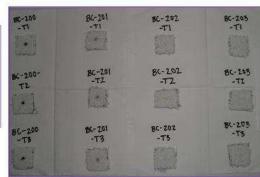


Figure 3. Representative image of different volumes of blood ranging from 0.075 to 75 µL on carpet.

75 µL of the appropriate blood dilution was spotted on each substrate, allowed to dry, and then collected using the double swab method, taping, or the M-Vac®. Therefore, the representative volume of blood tested was 75 µL, 7.5 µL, 0.75 µL, and 0.075 µL.



Whole Blood

- Extraction performed using QiAmp® Investigator extraction protocol (Qiagen, Valencia, CA).
- Quantification performed using the QuantifilerDuo® Quantification Kit (ABI, Carlsbad, CA) and the 7500 Detection System.
- Each sample was analyzed in triplicate.

## References

- Bradley B. Sadtler F. Comparisons of Meat Carcass Surface Bacterial Collection Efficiencies Utilizing a Novel Wet-Vacuum Microbial Sampler and the Sponge Method. Proceedings of 54th Annual Reciprocal Meat Conference, 2001 Indianapolis.

## Results

Table 1. Average concentrations of blood (0.075 – 75 µL) collected from tile using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
Double Swab	75 (±14)	3 (±3)	0.16 (±0.08)	0.01 (±0.01)
Tape (BVDA Instant Lifters®)	50 (±28)	1 (±1)	0.1 (±0.1)	0.02 (±0.02)
Vacuum Collection (M-Vac®)	66 (±7)	3 (±2)	0.2 (±0.1)	0.02 (±0.02)

(2 Standard Deviations)

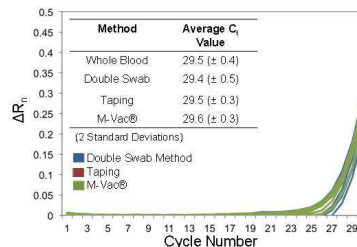


Figure 4. IPC analysis of each collection method from a non-porous substrate (tile).

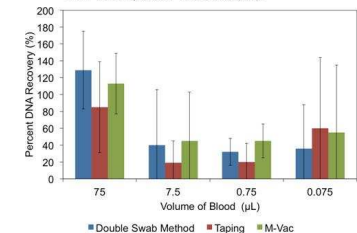


Figure 7. Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on tile with error bars representing the 2SD calculated using the theory of propagation of random error.

Table 2. Average concentrations of blood (0.075 – 75 µL) collected from denim using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
Double Swab	9 (±1)	0.5 (±0.4)	0.01 (±0.01)	0.001 (±0.004)
Tape (BVDA Instant Lifters®)	3 (±3)	2 (±1)	0.1 (±0.2)	0.004 (±0.004)
Vacuum Collection (M-Vac®)	64 (±3)	4.8 (±0.2)	0.16 (±0.04)	0.02 (±0.04)

(2 Standard Deviations)

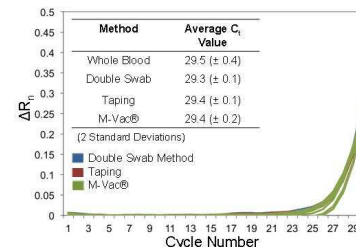


Figure 5. IPC analysis of each collection method from denim.

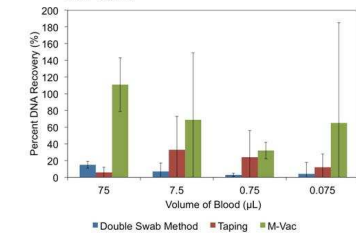


Figure 8. Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on denim with error bars representing the 2SD calculated using the theory of propagation of random error.

Table 3. Average concentrations of blood (0.075 – 75 µL) collected from carpet using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
Double Swab	27 (±9)	1 (±2)	0.01 (±0.006)	0.001 (±0.003)
Tape (BVDA Instant Lifters®)	9 (±2)	0.3 (±0.2)	0.1 (±0.2)	0.001 (±0.002)
Vacuum Collection (M-Vac®)	36 (±12)	0.6 (±0.5)	0.08 (±0.08)	0.03 (±0.02)

(2 Standard Deviations)

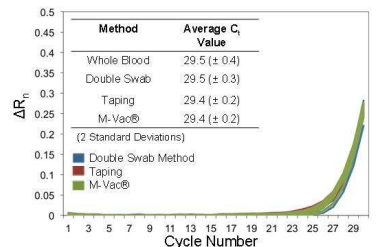


Figure 6. IPC analysis of each collection method from carpet.

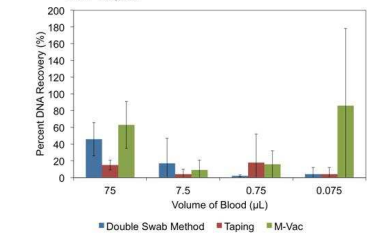


Figure 9. Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on carpet with error bars representing the 2SD calculated using the theory of propagation of random error.

## Conclusions

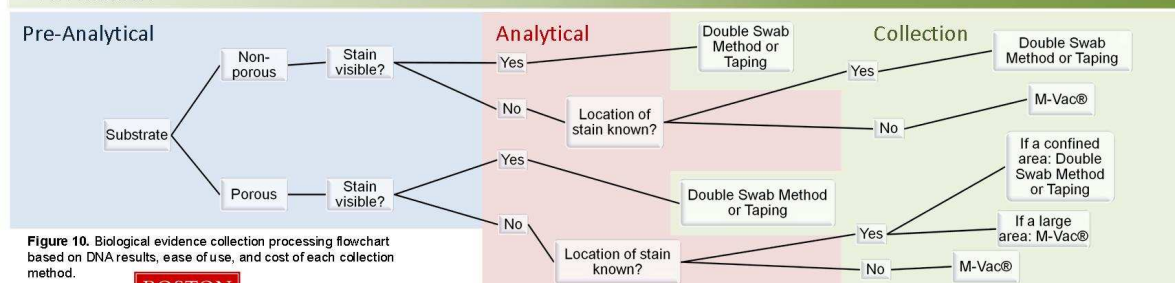


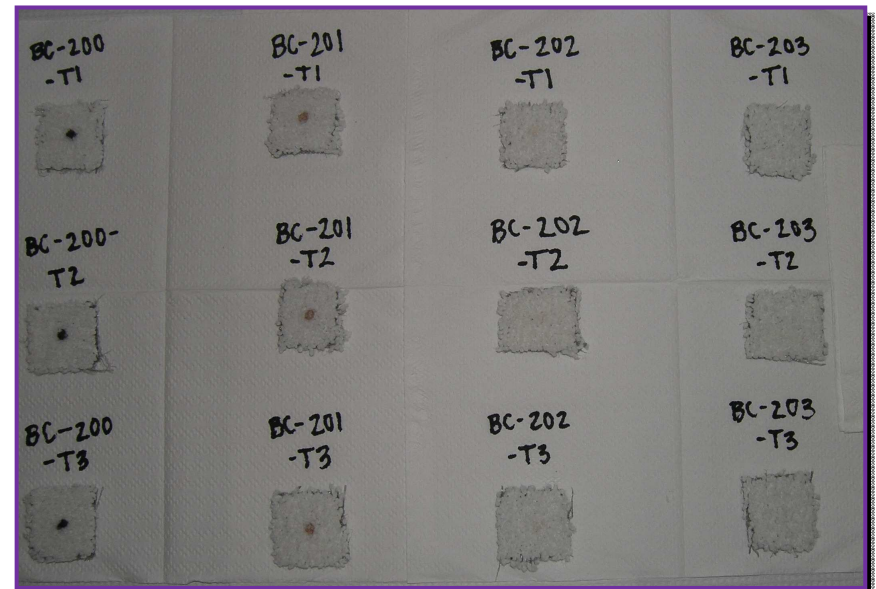
Figure 10. Biological evidence collection processing flowchart based on DNA results, ease of use, and cost of each collection method.

# Introduction

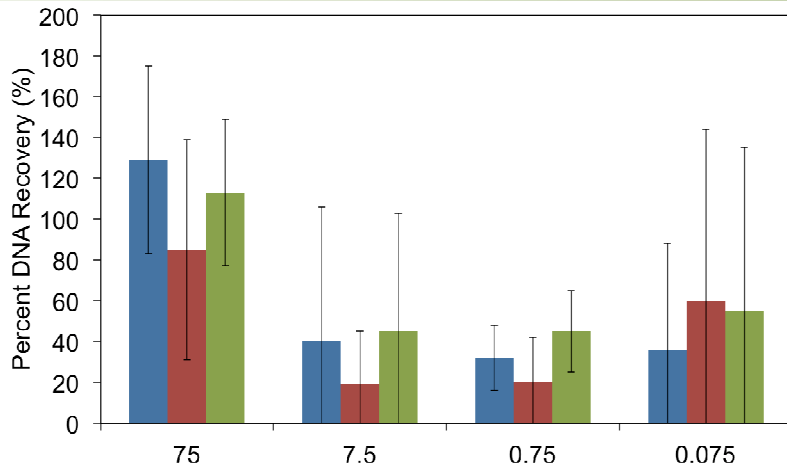
- ✧ In the collection of biological evidence from a crime scene, it is imperative to implement the most effective and robust collection method to ensure maximum DNA recovery.
- ✧ While common techniques for biological collection such as swabbing, cutting, scraping, and taping have been a mainstay in forensics, there are drawbacks of these techniques, which include, but are not limited to, the lack of surface area that may be processed, potential co-elution of PCR inhibitors, and non-optimized elution of cells from the substrate into solution.
- ✧ Due to this, an advancement, or new technique, in the area of biological evidence collection is needed in order to optimize collection from different items of interest, especially large items.
- ✧ Recent work in the field of pathogen testing suggests the use of a wet-vacuum collection system may be a valuable addition/alternative to already well-established biological collection methods (1).
- ✧ In this study, traditional biological collection methods, including the double swab method and taping, are compared to a wet-vacuum system (Microbial-Vac Systems® Inc., Bluffdale, UT) through the collection of different volumes of blood (0.075 - 75  $\mu$ L) on tile, denim, and carpet.

# Methods

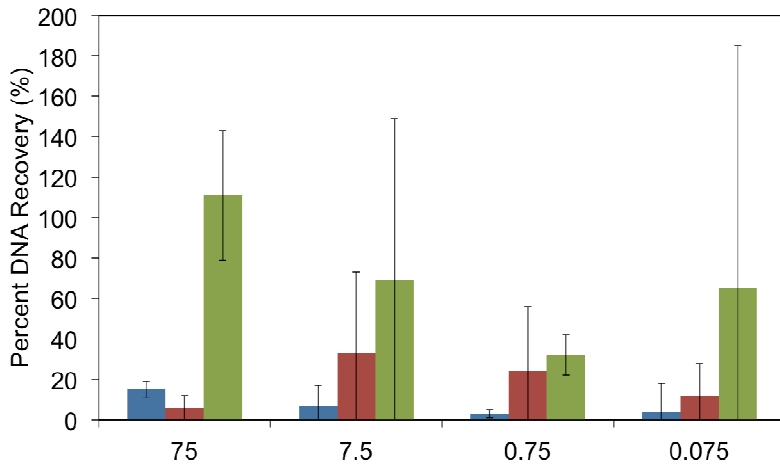
- ✧ 75  $\mu$ L of the appropriate blood dilution was spotted on each substrate (tile, denim & carpet), allowed to dry, and then collected using the double swab method, taping, or the M-Vac®. Therefore, the representative volume of blood tested was 75  $\mu$ L, 7.5  $\mu$ L, 0.75  $\mu$ L, and 0.075  $\mu$ L.
- ✧ Extraction performed using QiAmp® Investigator extraction protocol (Qiagen, Valencia, CA).
- ✧ Quantification performed using the QuantifilerDuo® Quantification Kit (ABI, Carlsbad, CA) and the 7500 Detection System.
- ✧ Each sample was analyzed in triplicate.



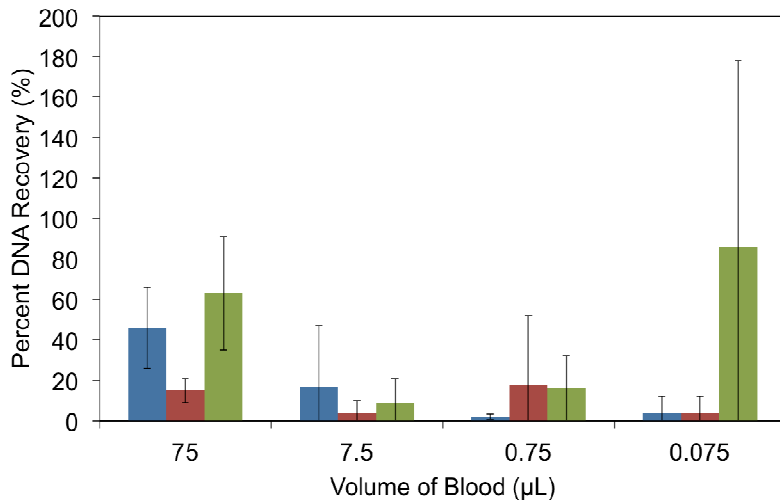
# Results



**Figure 7.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on tile with error bars representing the 2SD calculated using the theory of propagation of random error.

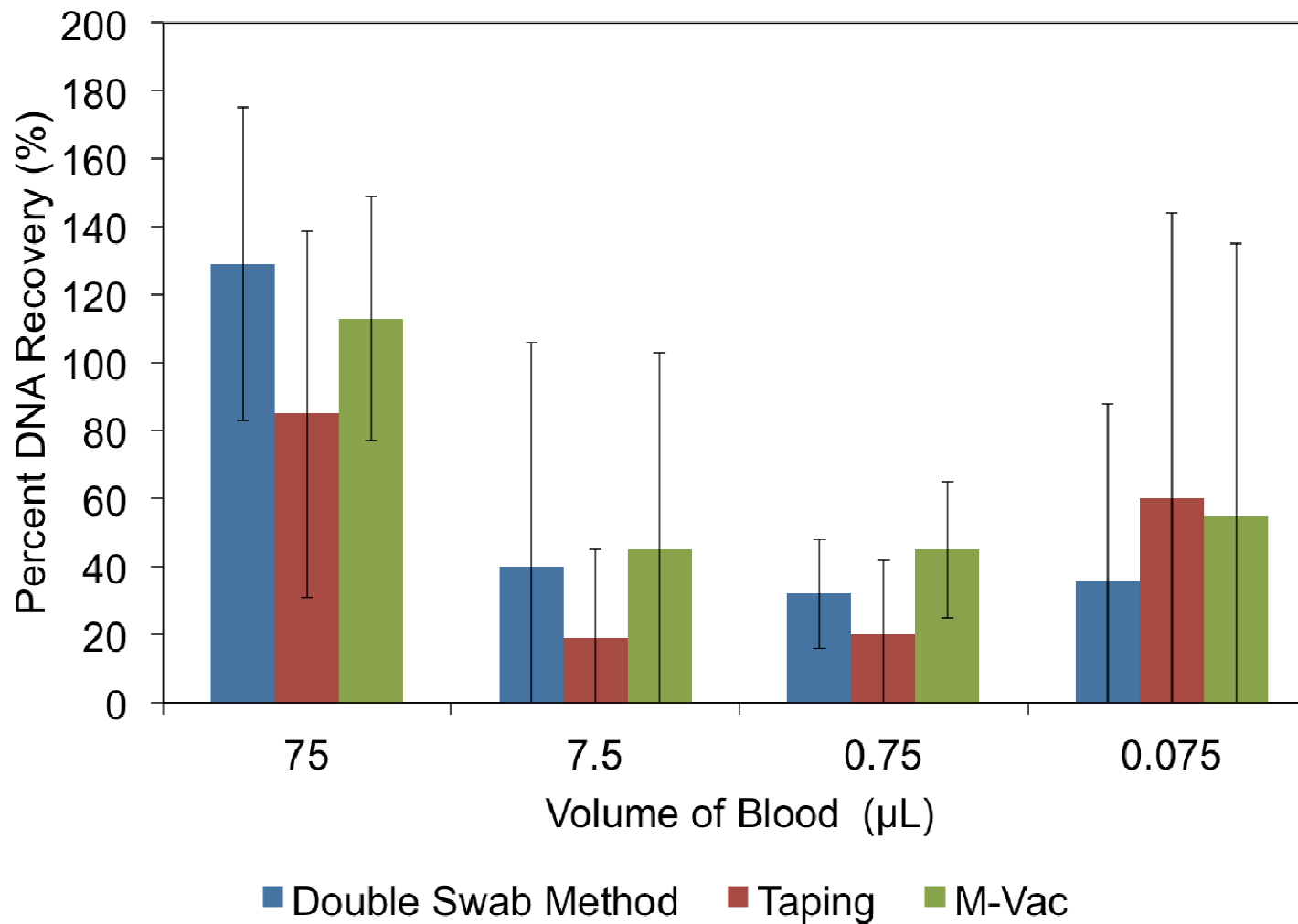


**Figure 8.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on denim with error bars representing the 2SD calculated using the theory of propagation of random error.

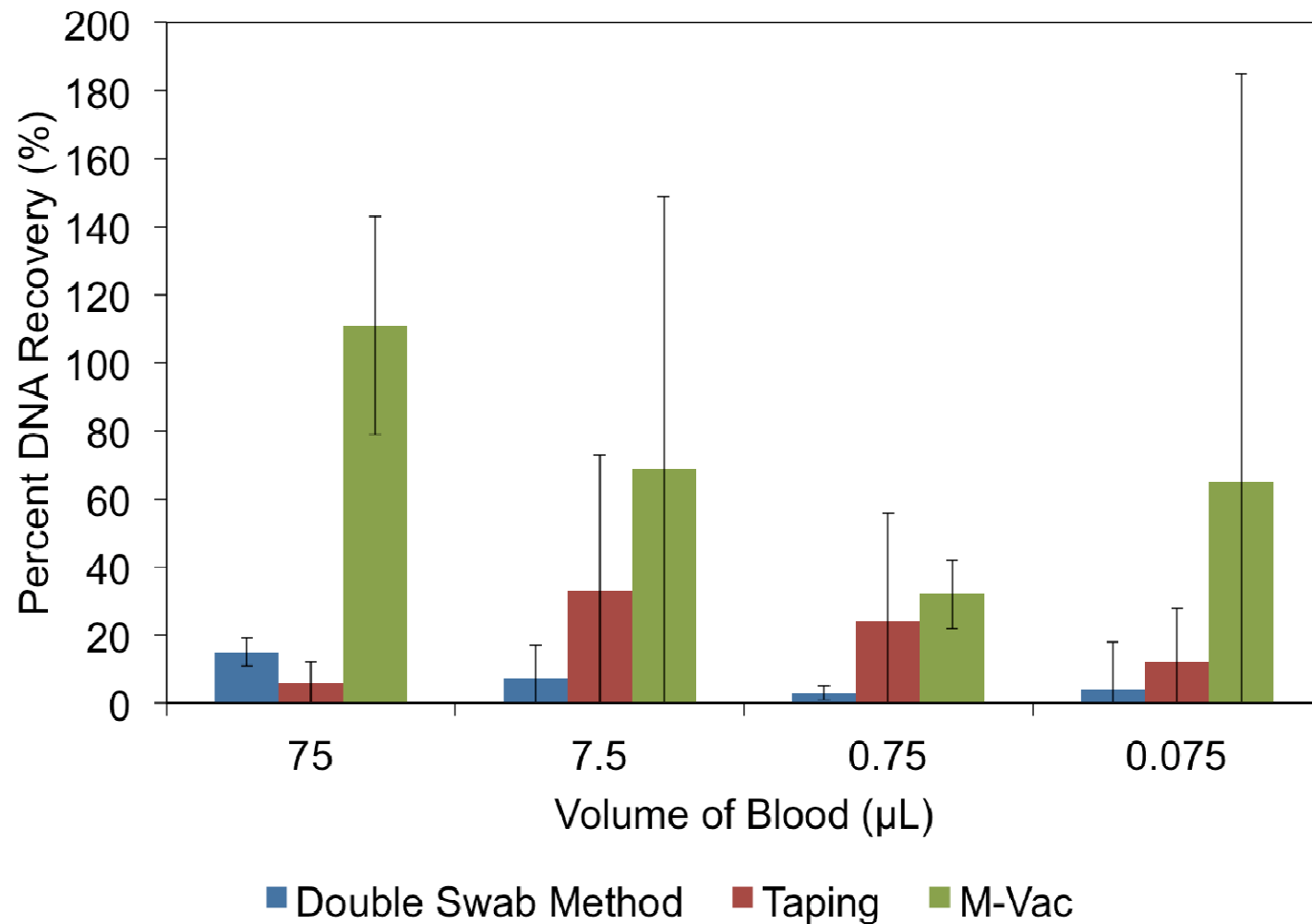


**Figure 9.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on carpet with error bars representing the 2SD calculated using the theory of propagation of random error.

■ Double Swab Method ■ Taping ■ M-Vac

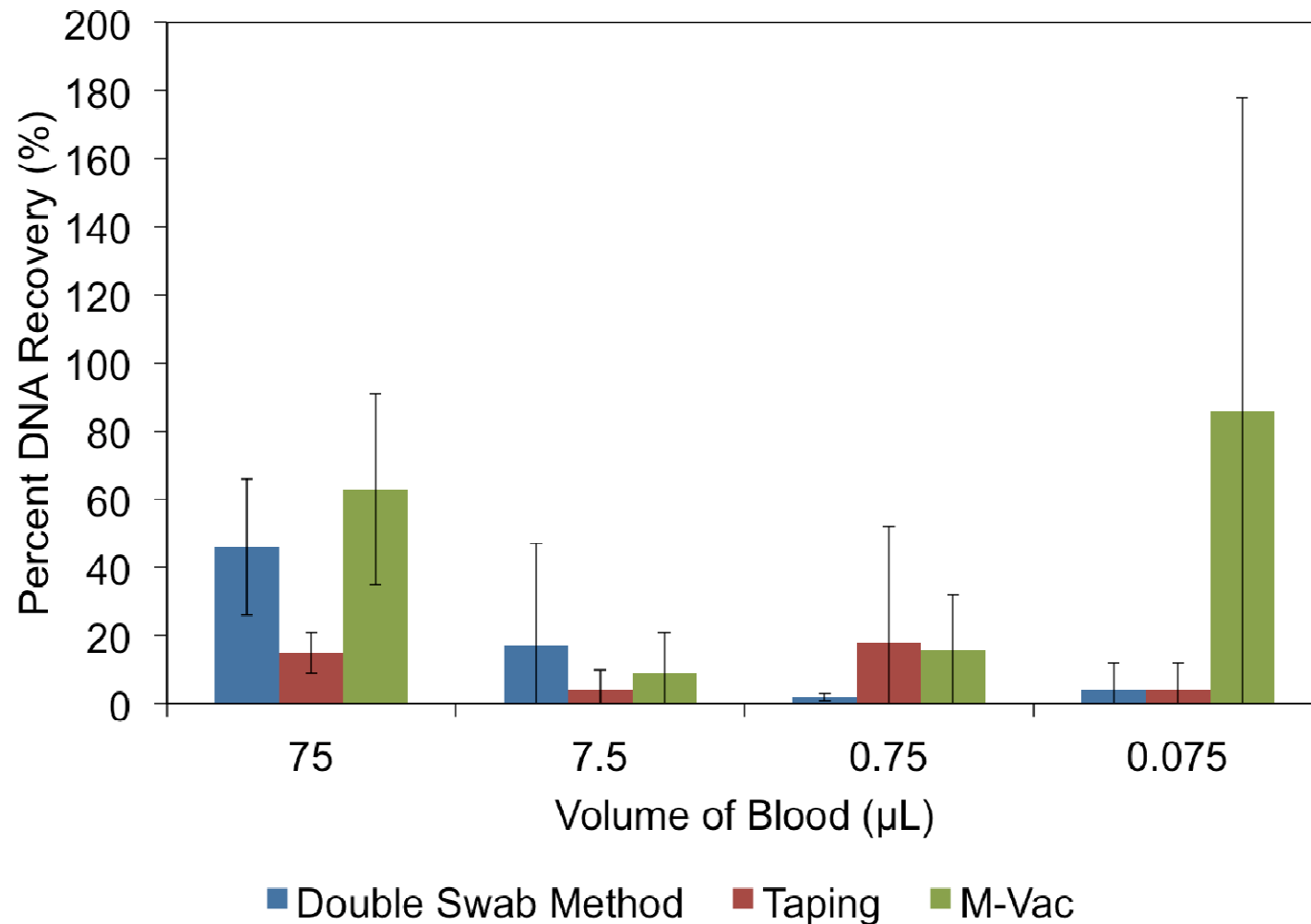


**Figure 7.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on tile with error bars representing the 2SD calculated using the theory of propagation of random error.



**Figure 8.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on denim with error bars representing the 2SD calculated using the theory of propagation of random error.

## Results - Carpet



**Figure 9.** Percent DNA recovery of blood (0.075 – 75 µL) using various collection methods on carpet with error bars representing the 2SD calculated using the theory of propagation of random error.

# Results - Data

**Table 1.** Average concentrations of blood (0.075 – 75 µL) collected from tile using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
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Tape (BVDA Instant Lifters®)	50 (±28)	1 (±1)	0.1 (±0.1)	0.02 (±0.02)
Vacuum Collection (M-Vac®)	66 (±7)	3 (±2)	0.2 (±0.1)	0.02 (±0.02)

(2 Standard Deviations)

**Table 2.** Average concentrations of blood (0.075 – 75 µL) collected from denim using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
Double Swab	9 (±1)	0.5 (±0.4)	0.01 (±0.01)	0.001 (±0.004)
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(2 Standard Deviations)

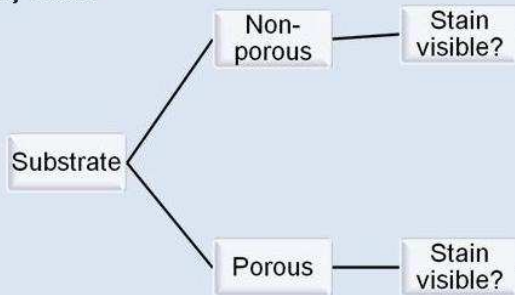
**Table 3.** Average concentrations of blood (0.075 – 75 µL) collected from carpet using various collection methods (in ng/µL).

	75 µL Blood	7.5 µL Blood	0.75 µL Blood	0.075 µL Blood
Whole Blood	58 (±17)	7 (±8)	0.51 (±0.08)	0.03 (±0.03)
Double Swab	27 (±9)	1 (±2)	0.01 (±0.006)	0.001 (±0.003)
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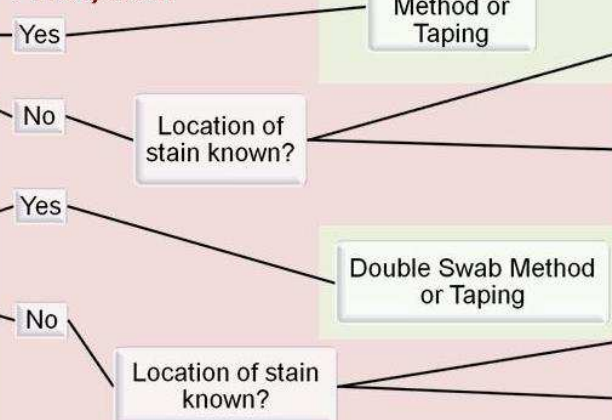
(2 Standard Deviations)

# Conclusions

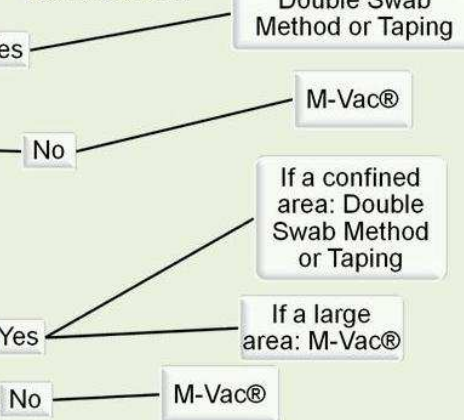
## Pre-Analytical



## Analytical



## Collection



**Figure 10.** Biological evidence collection processing flowchart based on DNA results, ease of use, and cost of each collection method.

