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USE OF M-VAC SYSTEM FOR THE COLLECTION OF TRACE HAIRS AND FIBERS

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ALYSSA WILSON

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USE OF THE M-VAC SYSTEM FOR THE COLLECTION OF TRACE HAIRS AND FIBERS

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BY

Dr. Rhonda Williams, Committee Chairperson

Mrs. Keisha Jones, Committee Member

Mrs. Heather Schafstall, Committee Member

Dedicated to my parents, whose unwavering support, encouragement, and belief in me made this achievement possible.

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Abstract

Trace evidence is a critical component of forensic investigations and commonly include fibers and hairs. Various methods are used to recover trace materials, including taping, tweezing, scraping, combing, and vacuuming. This study evaluated the effectiveness of the M-Vac™ wet vacuum, as a collection method for recovering trace evidence. The M–Vac is routinely used for the collection of DNA evidence from porous surfaces and has made a large contribution to the forensic science community, but had yet been evaluated for its application in trace evidence recovery. Automotive carpet and bed sheets were stained with whole blood and seeded with trace fibers (polyester and cotton) or hairs (human and dog) to simulate realistic forensic conditions. Samples were collected using the M-Vac system, taping, and dry vacuuming methods. Although the M-Vac system successfully recovered some trace materials, the conventional methods consistently achieved complete recovery. There was observance of some blood transfer subsequent to collection with conventional methods. These findings indicate that the M-Vac system is less effective for trace evidence recovery but may still hold value as a multi-use collection tool in situations when DNA recovery is prioritized. Further research is warranted to determine whether the use of conventional trace evidence collection methods prior to M-Vac sampling influence DNA recovery.

Introduction

Trace evidence recovery and examinations remain an important aspect in forensic casework. Although the literature surrounding this topic covers a wide variety of content, this review will focus on types of trace evidence, fiber and hair characteristics, transference and persistence of fibers and hairs, and collection methods. As technology advances for collecting evidence at crime scenes, the preservation of trace materials should be further explored. A new method of recovering DNA evidence emerged with the creation and use of the M-Vac. The proposed research study focused on the use of the M-Vac system as a tool to recover trace evidence as an alternative collection method. This review will also focus on the M-Vac system components as well as how the system works.

Literature review

Trace Evidence Transference

The foundation for trace evidence lies in that material may be transferred or relocated between items during contact and can be recovered and examined (Palmer, 2023). This fact is based on Locard's exchange principle, which states that "every contact leaves a trace." Previous studies that have discussed transfer fibers have led the way in the understanding of the basic principles of trace material transfer and persistence. Transfers may be primary or secondary. Primary transfer is also known as direct transfer. Secondary (indirect) transfer is where fibers or hairs are transferred from an item and then transferred to another item. How fibers transfer and persist depends on a wide variety of influencing factors. These include the likelihood of a textile product to shed fibers and the nature, intensity, and duration of a contact (Deedrick, 2000; Koch, 2020). The surface quality of both the donor and recipient garments plays a crucial role in the potential shedability of fibers and the procurement of any transferred fibers (Koch, 2020). The transfer and persistence of different fiber types should be considered as well, as some fiber types may have less adherence to their recipient and potentially be easily lost.

Collection and Recovery

Depending on the circumstances of the case at hand, collection of the trace evidence may take place at the crime scene, morgue, hospital, or crime laboratory. If possible, items of evidence should be submitted for trace evidence to be collected in a controlled laboratory environment. Additionally, Koch (2020) stated, "possible source materials should be collected as reference samples at the crime scene and cuttings taken from submitted textile materials in the laboratory for comparisons to be made" (p.344). Collecting textiles and garments at an early stage in an investigation will be helpful in preserving much of the trace evidence. To prevent loss or contamination of the evidence, it is extremely important that the items be packaged and sealed immediately after seizing. If the items are not correctly packaged, fragile evidence, such as trace materials, could be easily lost or new materials could be introduced to the evidence causing cross-contamination.

Documentation

During an initial assessment, evidence items should be visually inspected, described, and processed to remove trace materials adhering to the item. Documentation of the evidence should include a general description of the item as well as information from any visible manufacturer's label, the color, the type of material, any noticeable damage, and the condition of the packaging the item was originally received in. Depending on the case circumstances, it is possible that the interior of garments may need to be examined in addition to the exterior (Powell, 2022). When examining and assessing the submitted evidence, magnification and adjustable lighting can assist in identifying and collecting the trace evidence.

Visible trace materials can be collected from the surface of items by using tweezers or forceps, taping, or vacuum collection. The location of the trace material collected from the submitted item of evidence should be documented as well. Collecting fibers or hairs from items of evidence typically involves methods such as picking, scraping, taping, and/or vacuuming the evidence. Powell (2022) provides an overview of various trace evidence collection methods. The author describes the picking process in which trace materials are detected under a low level of magnification, such as a stereomicroscope, and picked off the surface of the item. Oblique lighting or an alternate light source may aid in locating different fibers or hairs, due to variances in optical properties. Picking involves the use of a pair of sterile tweezers to collect the material in question, which is then placed in a small container, in a paper fold, on tape, or on a glass microscope slide (Koch, 2020). If no material of interest is recovered after the visual inspection, textile items such as clothing may be scraped or taped for microscopic analysis in the laboratory.

Taping. Extraneous hairs or fibers can be collected from the surface of a garment or other surface by the use of transparent adhesive tape. The tape is to be held adhesive side down, and systematically applied to the item surface of interest. The tape is then placed onto a transparent plastic sheet, which helps to preserve the recovered evidence. The taping process can be repeated several times until all the surfaces of the item have been properly processed. These completed tapings are then labeled and placed in the appropriate container. If a garment sheds fibers readily, there may be a plethora of these fibers present on the tape-lift. When tapelifting, care should be taken to help prevent overloading the tape with the "background" fibers. The "background" fibers may make it more difficult to search for trace materials of interest and they also compromise the efficiency of recovery by making the tape less adhesive. Using several pieces of tape on the item of interest can help to prevent the overloading of the background fibers.

Tapelifts can be used in two different approaches, one-to-one or zonal. A one-to-one approach is where each section of the tape is applied only once to the item or victim. This approach can help to create a “distribution map” of the transferred items by documenting the location of each taping. However, this method can be very time-consuming and also creates a large number of tapes for examination. The zonal approach is where tapings are applied to broader sections of the item. “Zonal taping provides a compromise between workload and accuracy of information as to the location of each fiber” (Powell, 2022). The importance of the location of fibers on an item comes on a case-by-case basis and may not be as crucial in some circumstances. The effectiveness of tapelifts relates back to the retentiveness of the recipient item. For example, fibers transferred to a rougher surface, such as a wool sweater, have a higher probability of becoming bonded stronger with the garment, in turn making it more difficult to recover. It has also been shown in a study conducted by Lowrie and Jackson (1991, as cited in Powell, 2022), that the use of more adhesive tape by the fiber examiner does not seem to have a significant effect on the number of fibers recovered. Once the tapelift from the donor garment has been recovered, it can then be processed, and a stereomicroscope can be used to magnify and analyze the materials present.

Scraping. The scraping technique involves suspending the evidence above a collection funnel, or a sheet of paper, and then scraping the debris from the item onto the collection surface below. The collected debris is then transferred to the appropriate receptacle, such as a Petri dish, for searching. This method is useful for items that are soiled, or where numerous types of trace evidence are present and needing to be recovered at the same time. However, concerns have been raised regarding the method’s efficiency, risk of contamination, and loss of potential for location identification and interpretation. Due to these concerns, this method is not universally used for trace evidence collection.

Vacuuming. The vacuuming collection method uses a modified vacuum cleaner with a collection filter encompassed into the hose system. The single-use sealed collection filters are specifically designed to collect trace evidence (Koch, 2020). The vacuum method is useful for items with larger surface areas. The items may include carpets or car footwells. The vacuum method has disadvantages in that the entire vacuum must be cleansed thoroughly to help prevent contamination as well as the method tends to be too efficient. It recovers substantial amounts of irrelevant debris, which makes searching for the target fibers or hairs much more difficult. The efficiency of this method relies on the capability of the machine being used as well.

Combing. The combing method uses a "seeded" comb, which is a hair comb with cotton wool that has been introduced between the teeth. This method is used to recover extraneous fibers from hair either from a victim or a perpetrator. Studies have shown that fibers transferred from garments can persist on head hair for substantial periods of time, even after washing has occurred (Salter and Cook, 1996). Care must be taken to make sure the comb and cotton wool used are free from contaminants. The comb being used should be provided in a sealed tamper-evident package and should not be used if the package appears to be compromised or contaminated.

In considering the various recovery methods of trace evidence, it is important to differentiate between a laboratory environment and the crime scene environment. A laboratory is strictly controlled with access to multiple useful instruments, such as microscopes, as well as good lighting conditions. A crime scene often introduces difficult circumstances such as unpredictability, unclean conditions, poor lighting, and limited access to specialized equipment.

However, the crime scene can provide the opportunity to maximize evidence recovery and aid in collecting additional information pertaining to the case. Other types of evidence and the case details should be considered to determine which method or combination of methods of recovery is appropriate. No matter which recovery method(s) is used, it is important to make sure the recovered items are preserved in such a way that prevents loss, contamination, or redistribution of trace materials prior to the examination taking place in the laboratory. Items collected must be labeled clearly and notes thoroughly recorded to ensure the evidence is recovered properly. Following these precautions can help to aid in the analyst's investigation.

Fiber Evidence

Fiber evidence is examined in crime scene investigations due to the tendency to shed and transfer during contact between different surfaces (Palmer, 2023). According to Koch (2020), fibers are encountered as trace evidence in forensic casework in many diverse forms such as shed or transfer of single fibers, threads from a damaged garment, or in the form of textiles such as clothing, carpet, or cordage. A forensic fiber examination consists of identifying the type of fiber by analyzing the properties of a fiber, the comparison of unknown fiber(s) to a known sample for reference to determine whether the fiber may have originated from the same source, and potentially observing the construction of a textile product to discover damage or if two items could have once been joined together. In order to determine information on how a textile material was manufactured, a fiber's physical, optical, and chemical elements can be useful, including the mechanical processes involved in creating and finishing a fabric, the dyes used to color the fabric, the actions taken to apply the dye(s), and any additives that may have been applied to convey a finished product's properties. A fiber examination starts with the collection of evidence at a particular crime scene. If nothing of interest is collected, or the materials with fibers present that could potentially adhere to the surface are not properly preserved for analysis in the laboratory, then no conclusions can be made (Koch, 2020).

Fiber Types

Natural fibers can be divided into three subdivisions: animal, mineral, or vegetable (Palmer, 2023). Animal fibers include protein-based fibers such as sheep wool and other similar animal sources. Cultivated silk and wild silk are considered a non-keratin-based animal fiber. The structure of hair is organized into three different layers, an outer cuticle, which surrounds the cortex (main body of the hair) and a medulla, which occupies the central core (Koch, 2020). Wool fibers are more elastic, have a reduced medulla, higher water retention, and a greater heat retention capacity than other mammalian hairs, making the fibers easier to use in textiles. The most well-known mineral fiber is asbestos. Besides being spun into yarns, asbestos has also been woven into fire-resistant fabrics and produced construction materials. Plant or vegetable fibers can be further differentiated to indicate what part of the plant the fiber is processed from: the stem, seed, leaf, or fruit. Some commonly known vegetable fibers include flax (linen), cotton, hemp, and jute. The chemical composition of vegetable fibers can vary on many levels, however the main component in all is cellulose. Cotton is a natural vegetable fiber from plant seed pods and is the most encountered natural fiber in forensic investigations. Man-made fibers made an appearance in the late 1800s in attempts to produce artificial silk that are modified natural polymers, such as rayon, and synthetic fibers such as acrylic. Even though there is a large amount of man-made fibers in forensic investigations, only a small number of generic fiber types are usually found. The most common fiber type in global production is polyester, accounting for approximately one-third of fibers produced (Textile Exchange, 2020). Cotton is also a largely produced fiber throughout the world. Other fibers commonly found globally are polypropylene, polyamide (nylon), acrylic, and wool. Natural fibers, apart from silk, are fibers with relatively short lengths ranging from 2 to 50 cm and cross sections ranging from 10 to 40 microns. Synthetic fibers and silks are produced as sustained filament yarns, which are sometimes cut to form staple yarns of specific lengths, depending on the intended use of the fiber. Fibers are often dyed to give a desired color for many fabrics. Color influences the value of identification of any particular fiber. Color is important in examination of fibers, not only because of the variety of colors used in textiles, but also the production process of the colors (Maxwell, 2016). Color-fading and any discoloration that has occurred can also help in fiber identification and association.

Fiber Examination

Examinations conducted in the laboratory include determining a fiber's optical, physical, and chemical properties to identify possible sources for comparison. Sources can be other items of evidence or known reference samples. An evaluation of whether unknown fibers are similar to a source can answer questions relevant to the case. These questions can include potential contact between people or contact between objects.

To avoid contamination in the laboratory, the fibers recovered from garments of victims and offenders need to be examined independently. The donor qualities (shedability) and recipient qualities (retention) should be checked prior to an examination if assumptions are to be made about the possible rate of transfer and persistence of fibers (Koch, 2020). A general assessment of shedability can be made by taping a garment, though a tape lift may possibly remove more fibers than those that would be typically transferred under typical circumstances. Through this tape lift, a garment can be viewed as either a low or high shedder. The shedability of a garment can also be tested by the Martindale rub test or with a simplified weighted rub test. These rub tests are designed to assess the durability of different fabrics when subjected to abrasion conditions, which could be encountered by a fabric during normal handling and wear. This test is useful for forensic purposes to count the number of fibers shed from a donor garment under predefined conditions such as weights, pressures, and a set number of intervals where the fabric is rubbed up against a neutral surface. Usually, three different pressures (light, medium, and heavy) are used in three replicated experiments. For both the Martindale rub test and the weighted rub test, a defined area of fabric is designated to search and count the number of transferred fibers (Koch, 2020). Since garments can be made of multiple different fiber types and people often wear more than one garment at a time, transfers may not be limited to a single type of fiber stemming from only one item.

Hair Evidence

Hairs recovered at a crime scene can be very valuable evidence. Examinations of hair evidence can provide information including: the type of hair, biological identifications, profiling of consumed substances, characteristics for comparisons and identifications, reconstruction of activities or incident sequencing, and also a source of forensic intelligence (Pfefferli, 2015).

Hair Types

The field of hair microscopy in forensic science is typically divided into two sub-disciplines: human and animal hair examinations. Beckert (2020) provides an overview of hair types. In humans, hairs are most commonly divided into three types: Lanugo, Vellus, and Terminal. The Lanugo subdivision includes hairs that are formed in utero and typically shed before or shortly after birth. The Vellus subdivision includes fine, short, unmedullated, and typically unpigmented hairs present on almost all skin surfaces. These hairs are typically found on areas such as the nose, forehead, and ears, but are not found on the palms of hands or the soles of feet. Terminal hairs are the typical hairs visible on children and adults. Terminal hairs can be subdivided into primary hairs and secondary hairs. Primary hairs include those found on the head, eyelash, and eyebrow. Secondary hairs, also known as secondary sex hairs, are formed during puberty in specific regions such as the pubic area, armpit, and beard. These secondary hairs are the result of vellus hairs being replaced by terminal hairs. Human hairs are typically the most encountered hair type at crime scenes and in forensic casework.

Animal hairs can be divided into three types: guard hairs, fur hairs, and vibrissa. Guard hairs, also known as overhairs, are longer, stiffer hairs in the main hair of the animal. Guard hairs are often used in microscopic identifications (Beckert, 2020). Fur hairs, also known as underhairs, are fine, often wavy, and located in the main hair of the animal. These hairs are typically uniform in diameter and devoid of any microscopical characteristics to be used in identifications. Vibrissa hairs primarily provide a tactile sensory function for the animal and are also called whiskers. Vibrissa hairs are rarely encountered in forensic casework. Cat and dog hairs are by far the most common pets in the United States, and because of this, they are likely the most encountered animal hairs in forensic cases.

Significance and Error

After meticulous collection and analysis of data, determining the significance of the derived findings and error rates need to be discussed. The significance of fiber evidence must also be considered, such as the type(s) of fibers and color(s), the number of fibers discovered and their respective locations, whether multiple different fiber types were discovered, and if cross transfer may have taken place (Maxwell, 2016). Dyed synthetic fibers or dyed natural fibers can be significant in forensic investigations, whereas common fibers such as blue denim cotton or white cotton would be less significant due to their widespread use. However, that does not mean that the presence of these fibers has no meaning at all to a case. According to Thorsen (1999), "There are a number of factors that strengthen and weaken the significance of fiber evidence including the possibility of errors" (p.15). Although there is a possibility of examiner errors, another type of error that is important is an association error. One association error described by Gaudette is where an unknown fiber is linked to a known sample when it could have another source (Gaudette, 1982). This error could be possible due to the mass production of textile fibers. Gaudette also describes two different types of errors. A type I error occurs if the examiner reports that an unknown fiber or hair did not originate from the known, when the evidence did originate from the known source (Thorsen, 1999). This type of error is also known as a false negative or an incorrect exclusion. A type II error occurs if the examiner reports an unknown fiber or hair could have originated from the known, when it did not (Thorsen,1999). This type of error is also known as a false positive or an incorrect association. Gaudette gives an exceptional summation of different factors that can either strengthen or weaken the significance of fiber evidence. Some of the strengthening factors given include: finding several materials originating from a known, finding matching fibers of several different types, finding unordinary characteristics, and cross-transference. Some of the weakening factors include finding common types and colors of fibers, finding many non-matching fibers and only a few similar fibers, and any contamination issues (Thorsen,1999). Also, when determining the significance of fiber evidence there are other factors to consider. These factors include information about the case, time elapsed before the evidence collection, the number of types of matching fibers, the quantity of matching fibers and unknown factors like the pressure of contact (Thorsen,1999). The type of physical contact between a suspect and a victim can impact the number of fibers or hairs potentially transferred and their value. For example, violent physical contact of lengthy duration will often result in many fiber or hair transfers occurring. If multiple fiber or hair type materials are found on different surfaces and materials from the crime scene, suspect, and victim, it increases the likelihood that contact occurred between the individuals and the scene (Deedrick, 2000).

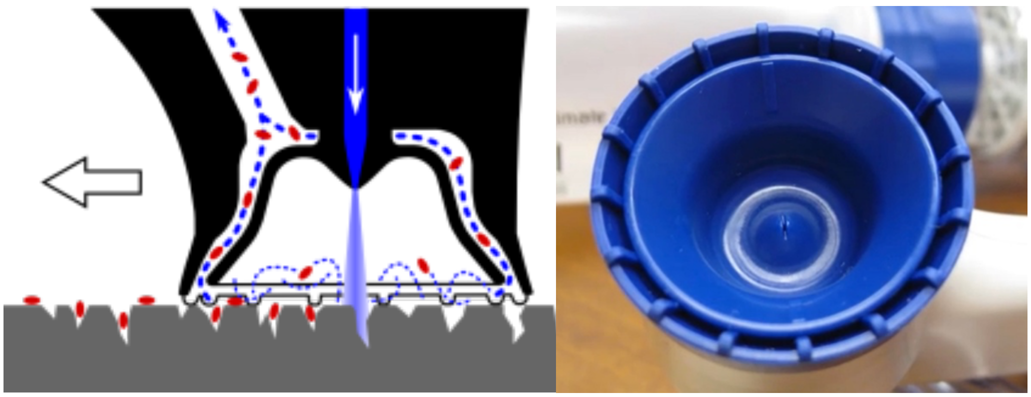
M-Vac Background

The Microbial Vacuum (M-Vac®) Wet-Vacuum System (CSI Equipment Ltd., UK) was initially developed for use in the food industry as a device to sample food for potential pathogens (Ryan, 2016). However, the developers discovered in the early 1990s that with a couple minor modifications the system could pose a huge benefit in the collection of deoxyribonucleic acid (DNA) evidence in the field of forensic science. There are other techniques that are commonly used, such as swabbing to collect DNA evidence; however, there are often issues recovering adequate quantities of DNA when these methods are used on rough, porous surfaces (Glater, 2022). Since this adaptation was made, the M-Vac has been used to collect DNA evidence targeting large and porous surfaces (Vickar, 2018), which are defined as a surface that has small or minute spaces or holes through which liquid can pass through. Some examples of porous surfaces are untreated wood, carpet, and some stones or rocks. In a study conducted by the FBI in 2020, it was found that the M-Vac yielded more total DNA than wet-swabbing on 18 different porous substrates (McLamb, 2020).

Components and Use

The M-Vac can be considered simply as a hand-held wet vacuum cleaner (Ryan, 2016). Although the M-Vac does not spray cleaner, it sprays nuclease-free, DNA-free, sterile buffers onto the item of interest (Ryan, 2016). The M-Vac collection head sprays the buffer solution directly onto the surface through a hole in the center of the head. While the M-Vac is spraying the buffer onto the surface, it simultaneously vacuums up the liquid along with any cells and DNA present on the surface into a sterile container (Vickar, 2018). The sample is collected through the slots on the outside of the collection head. An example of the collection head design and operation is illustrated in Figure 1.

Figure 1. M-Vac Collection Head



The solution is filtered through either an 0.45 µm or 0.20 µm polyethersulfone (PES) vacuum filter. The biological material is retained on this filter. Once the filter is dried, the filter can be removed from the M-Vac. The filter can then be processed for DNA analysis following the same standard procedures used for cuttings or swabs. In cases where a larger surface area must be sampled, the collection container can be removed from the vacuum, the contents filtered, and then replaced to continue sampling the area (Glater, 2022). In some samples, differing amounts of debris can be collected along with the sample, which can cause difficulty when attempting to analyze the sample. In efforts to prevent this potential problem from occurring, the M-Vac system offers an optional pre-filter system. The pre-filter can be used to remove any debris larger than 40 µm, however it also poses a risk of losing some of the target cells. This filtration helps to remove any contaminants or dirt present that can influence the ability to process the collected DNA. All parts of the vacuum that make contact with the evidence, as well as the recollected buffer, are made to be single use, which helps to eliminate the possibility of cross-contamination. The system is very straightforward to operate and use without the need for meticulous training. An informative demonstration video is available on the M-Vac® Systems website to aid in the training and understanding of using the M-Vac system for collection of DNA material. The user interface contains a pressure gauge, a flow gauge, and three different switches: power, solution, and vacuum (Vickar, 2018). Once the tubing is connected and the solution pressure has reached the desired level, the system is ready for use. When attaching the collection head, care must be taken to ensure sterility is maintained by avoiding any external contact with the connection points, since these points come into contact with the collected sample (Vickar, 2018). After the collection, the M-Vac process requires additional time due to the filtration process. Even though the M-Vac collection is more complex and time consuming than the double swabbing technique, which consists of using a wet swab followed by a dry swab rubbed across the substrate, the system is almost straightforward, requiring minimal training. Several studies have shown that the M-Vac performs better than the other collection methods for the recovery of DNA in bloodstains as well as dried saliva on different types of materials. In one study, it was determined that the M-Vac could successfully collect DNA material from tile, denim, carpet, and brick surface materials in sufficient quantities for analysis (Gunn, 2013).

Summary

The literature supports the value of trace evidence and types likely to be encountered in forensic casework. Textile fiber and hair evidence are likely the most commonly found transfer and trace evidence in forensic science. There are several different recovery methods used for the collection of trace evidence; however, the research suggests that taping is the most practiced method. Vacuuming is a method that is beneficial to use on larger surface areas but is not commonly practiced due to the large number of insignificant debris that can be collected as a result. Technological advances in the recovery of DNA have expanded the possibilities for trace evidence recovery. The M-Vac could be used as an alternative recovery method to collect trace evidence, similar to the vacuuming method. Exploring this application of M-Vac could support its usage as a viable multi-use tool for the collection of both DNA and trace evidence.

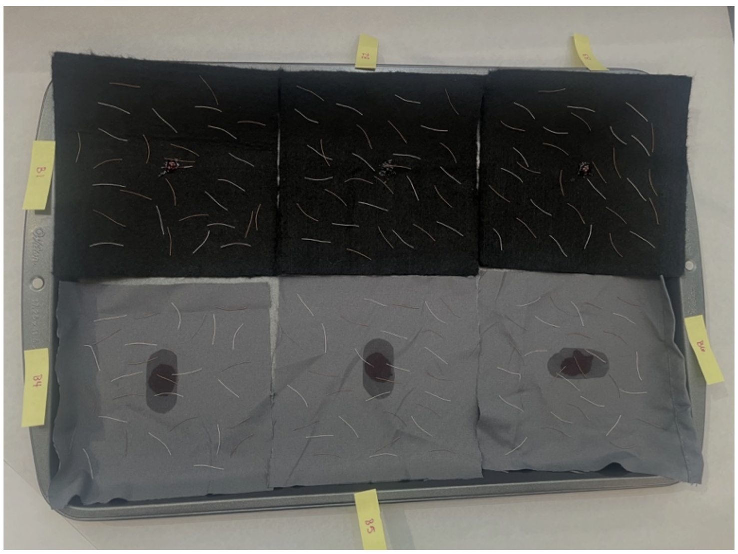
Materials and Methods

Medium and Sample Preparation

A roll of black, low pile, polyester automotive carpet (78” x 40”) (Halero) and a gray, polyester queen-sized flat bed sheet (102” x 90”) (Utopia, 400 thread count) were purchased from Amazon and cut into equal pieces of 6 in x 6 in. There were a total of 27 automotive carpet squares and 27 bed sheet squares. The automotive carpet squares and sheet squares were used for the mediums in this study. Automotive carpet and bed sheets were chosen for trace evidence collection due to the porous nature of the surfaces as well as being commonly encountered in forensic casework. Two different fiber types were chosen for this study, polyester and cotton. Polyester and cotton fibers were chosen based on their commonality in production. All-purpose thread, pink polyester (Superior Threads, 50 wt.), and brown cotton (Coats, 35 wt.), were purchased from Amazon. The thread was cut into segments of approximately 2-2.5 centimeters for ease in sample preparation. Two different hair types were chosen in this study, human hair and dog hair. Human hair was chosen based on its significance to forensic casework and dog hair due to canines being a popular pet. Blond human hair extensions (Nodcay brand, 14”) were purchased from Amazon and cut into 2-2.5 cm segments. Samples of dog hair (Siberian Husky) were brushed from the dog and donated. Hairs consisted of varying colors from white to gray and ranged from 3 to 5 cm in length.

To simulate a situation in which the M-Vac would be utilized for the recovery of both DNA and trace evidence, blood was incorporated in this study. On each carpet square and sheet square, 25 μl of whole human blood was transferred to the center of each square using a pipette. After the blood deposition, a sample of 30 fibers or hairs, 15 of each fiber type or 15 of each hair type combined, were randomly distributed over the entire surface area of each carpet or sheet square. An example of this distribution is illustrated in Figure 2.

Figure 2. Set-up of Samples



Note. The image seen above shows the set-up for samples B1-B6. 25 μl of whole blood was transferred to the center of each medium square, followed by the random distribution of fibers.

The squares were stored in a sterile environment to dry overnight. The collection process was completed the following day. The carpet and sheet samples were divided into the following categories for collection of trace evidence: taping, dry vacuum, and wet vacuum. Three replicate samples were performed for each collection. Control samples without trace evidence were collected to assess background interference of the surface medium. All samples are listed in Table 1.

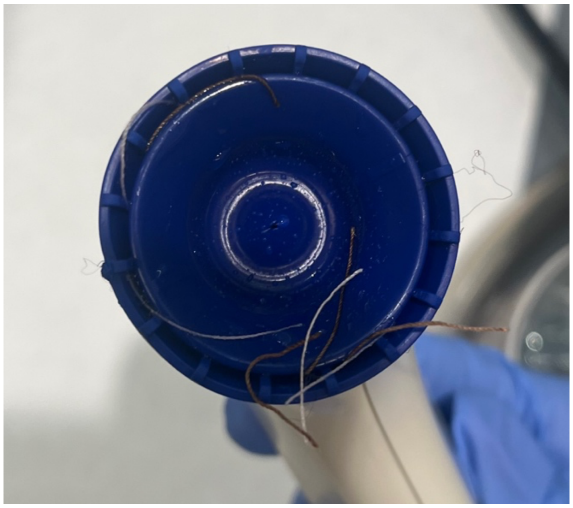
Table 1. Numbering of Samples

		Control (Blood Only)	Fibers Combined	Hairs Combined
M-Vac	Auto Carpet	A1-A3	B1-B3	C1-C3
	Bed Sheet	A4-A6	B4-B6	C4-C6
Taping	Auto Carpet	D1-D3	E1-E3	F1-F3
	Bed Sheet	D4-D6	E4-E6	F4-F6
Dry Vacuum	Auto Carpet	G1-G3	H1-H3	I1-I3
	Bed Sheet	G4-G6	H4-H6	I4-I6

Wet-Vacuum Method

The M-Vac® (M-Vac® Systems, Inc.), consisting of a Support Equipment Case (SEC) 100, a sampling device with a collection bottle, a buffer solution, and plastic tubing, was used for wet-vacuum collection. Collection was performed using the hand-held collection device and dispersing buffer solution under vacuum force. The collection device was passed over the entire surface area of the medium until 75 mL of buffer solution had accumulated in the collection bottle. Per manufacturer recommendations, the sampling device was moved in a backward motion with a slight overlap of passes. The sample collected was then vacuum filtered by pouring the entire sample into the filter apparatus while applying vacuum suction to the apparatus. The concentration filters were removed from the filter apparatus with a sterile scalpel and allowed to air-dry. A sample was collected on a square with no trace evidence present to assess the background interference of the surface medium. A total of 3 replicates of each sample type were conducted in the same fashion with 30 hairs or fibers present on the medium. For each sample, the target fibers or hairs on each filter were counted as well as those left on the carpet or sheet surface to account for any trace evidence not collected during the process. Any fibers or hairs that were collected on the collection head of the M-Vac were collected by either hand picking the trace evidence off the head using sterile plastic tweezers or by spraying the sterile buffer solution into a sterile petri dish and swirling the head around in the solution to collect any trace materials off the head. An example of fibers being collected on the collection head can be seen in Figure 3. The concentration filters of all samples were stored in a sterile environment.[i]

Figure 3. Fibers Collected on Collection Head

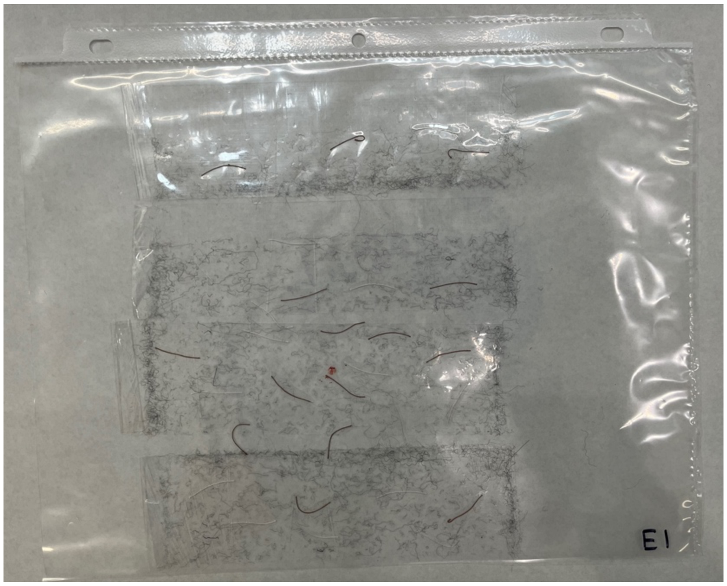


Note. The image seen above shows the fibers collected on the collection head for sample B3. All samples differed in amount of trace materials collected on the collection head.

Taping Method

A one-to-one taping method, several pieces of tape are used to cover the entire surface, was used in this study to ensure the entire surface of the medium was completely covered and for maximum recovery. Although a one-to-one taping method is mostly utilized when the trace evidence needs to be pinpointed to a certain location on the background surface, this study was not focusing on this aspect. For each sample, the entire surface was covered with clear packing tape, each piece measuring 2 inches wide with slight overlap between neighboring pieces to ensure the coverage was complete. For each collection, the tape pieces were removed in reverse order to their placement. Once each tape piece was removed from the medium surface, it was placed on a separate clear acetate sheet, with only one acetate sheet for each replicate. These sheets were then labeled for which type of trace evidence was being collected for later evaluation. An example of the clear acetate sheet used for recovery can be seen in Figure 4.

Figure 4. Clear Acetate Sheet with Recovered Tapings



Note. The image seen above shows the fibers collected from sample E1. The distribution of trace evidence had some variation among samples.

For each sample, the target fibers or hairs on each tape were counted and those left on the surface were also counted to account for any trace evidence not collected during the process. A control sample without application of trace fibers or hairs was collected with the taping method to assess background influences of the surface medium. There were 3 replicates performed for each material selected with 30 fibers or hairs present. After each replicate and control test was completed with the taping method, the M-Vac was used to collect the blood present on each square using the wet-vacuum method described previously. After all collections with the M- Vac were completed, each concentration filter was labeled and stored in a sterile environment.

Vacuum Method

A regular shop vacuum device (shop-vac brand, model L250) with a filter trap was used to collect the trace materials from the medium surface. This method utilized one-time use chambers (Arrowhead Forensics) that fit over one end of the vacuum cleaner, and the other end was the nozzle. Once the entire area was vacuumed, the filter cloth, encased by two plastic ends, containing the collected material, was removed and packaged with labels for later evaluation. After each collection the medium surface was examined to determine if any of the trace material added was still present on the surface to account for any trace evidence not collected during the process. This was done for each replicate of both trace evidence types. Once all replicates were completed, each packaged filter was opened and evaluated to determine the amount of material collected. An example of what the dry vacuum filters looked like after collection can be seen in Figure 5.

Figure 5. Dry Vacuum Filter with Collected Trace Material

Note. The image above shows the dry vacuum filter after collection for sample H1.

The filters with trace hairs present were scraped with a sterile metal spatula to evaluate how many hairs were present due to the inability to see the dog hairs clearly on the filter. There were 3 replicates performed for each material selected with 30 fibers or hairs present. A control test was used for the vacuum collection method as well to assess the background on the filters between the two mediums. After each replicate and control test was completed with the vacuum method, the M-Vac was used to collect the blood present on each square using the wet-vacuum method described previously. After all collections with the M-Vac were completed, each concentration filter was labeled and stored in a sterile environment.

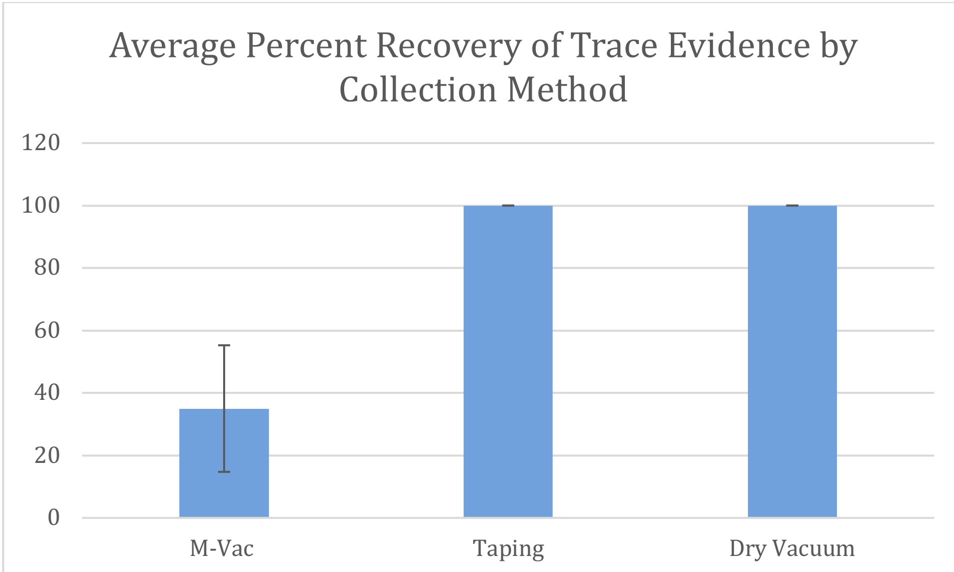
Results

Trace Evidence Recovery Efficiency

The taping and dry vacuum methods resulted in one-hundred percent recovery rates for all samples. For the taping method samples, all trace materials were collected from the medium surface using the one-to-one taping approach. After collection, all tape sheets were examined to validate that all trace materials were collected. After evaluation, all 30 trace fibers or hairs were collected using the taping method for all replicates. For the dry-vacuum samples, all trace materials were collected from the medium surface using the vacuum method. After collection, all trace vacuum filters were examined to validate that all trace materials were collected. After evaluation, all 30 trace fibers or hairs were collected using the vacuum method for all replicates. Samples used as control samples did not produce any data pertaining to trace materials, however it was observed that the carpet medium produced more background interference compared to the bed sheet medium. Overall, the wet vacuum method recovered a range of trace evidence from 1 to 20 with no samples resulting in one hundred percent recovery. The recovery totals for each type of trace evidence are listed in Appendix A. The average percent recovery for each type of trace evidence can be found in Appendix C.

The average percent recovery of combined trace evidence of all three collection methods is illustrated in Figure 6.

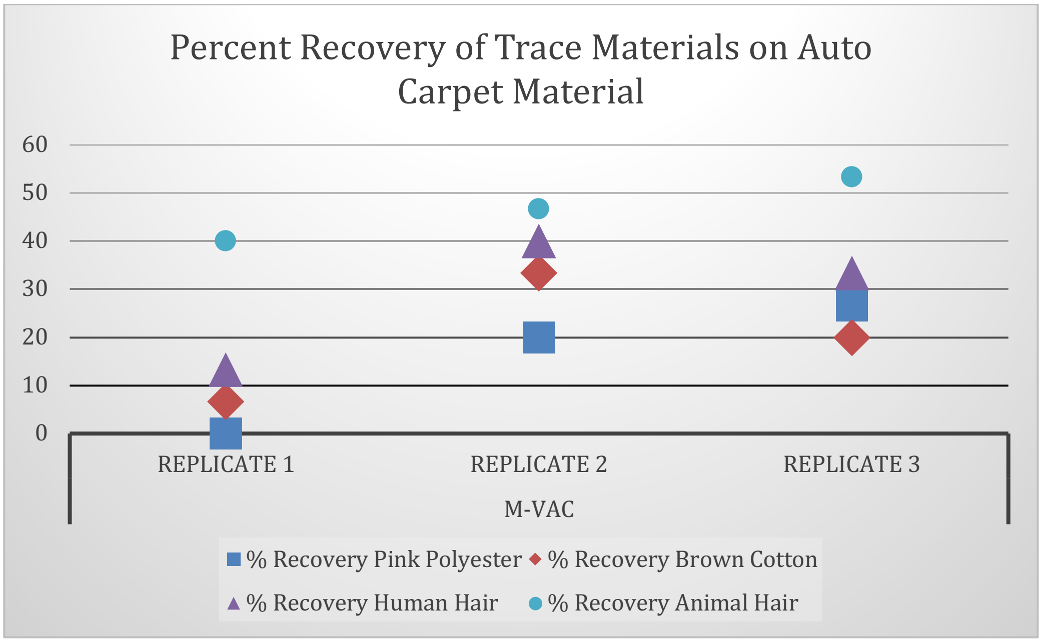
Figure 6. Percent Recovery of Trace Evidence by Collection Method

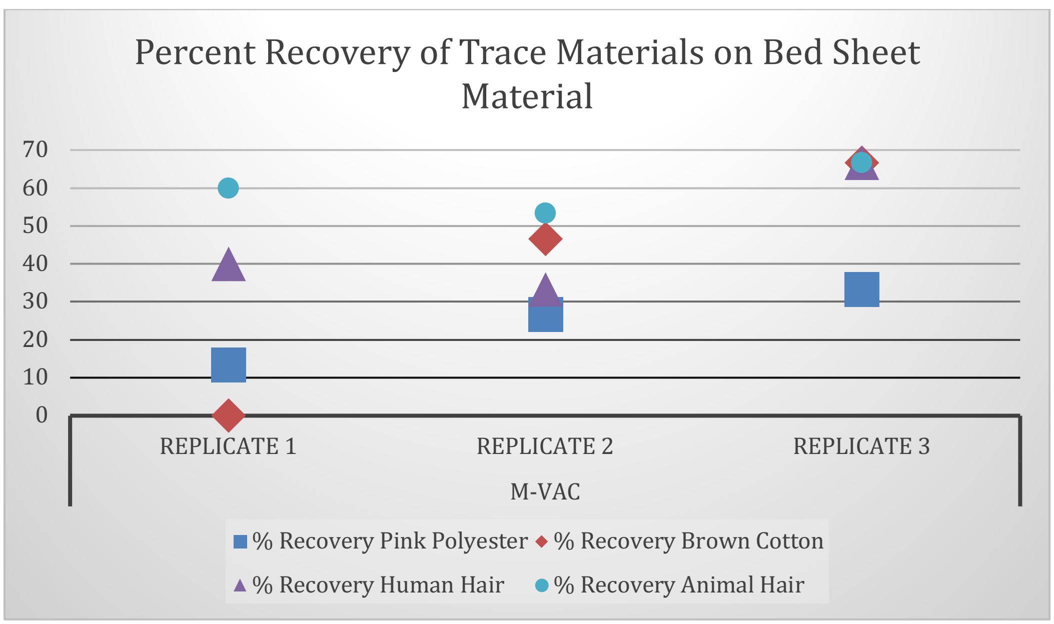


M-Vac Recovery

Figure 7 shows the effect of the surface type on trace evidence recovery for all replicates of wet vacuum collection. Overall, the trace recovered from the bed sheet seemed to result in a higher recovery rate than the carpet with the wet vacuum. A two-sample t-test was performed to compare the effectiveness of trace material collection between the automotive and bed sheet substrates. There was not a significant difference in trace materials collected between the automotive carpet ($M = 4.167$, $SD = 2.44$) and the bed sheet ($M = 6.33$, $SD = 3.28$); $t(20) = -1.834$, $p = 0.081$.

Figure 7. Effect of Surface Type on Trace Evidence Recovery with Wet Vacuum Method

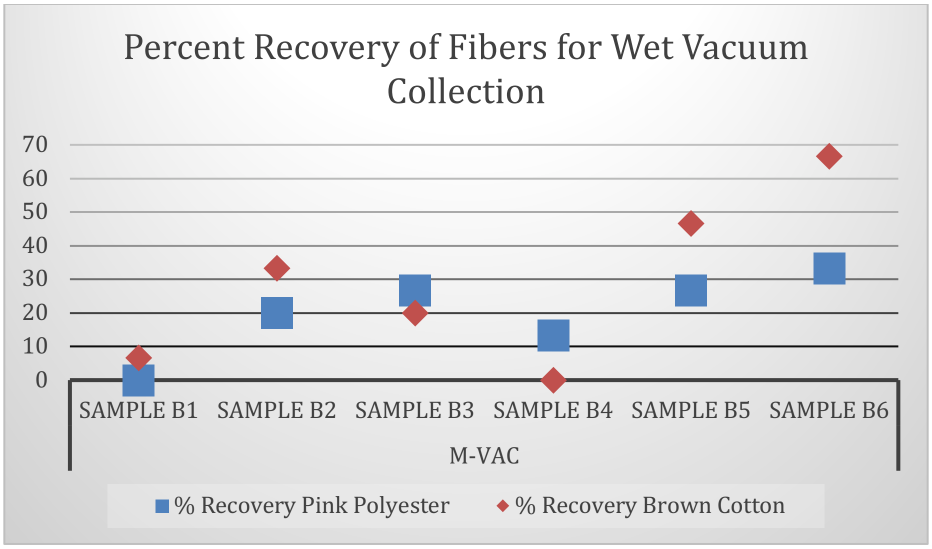


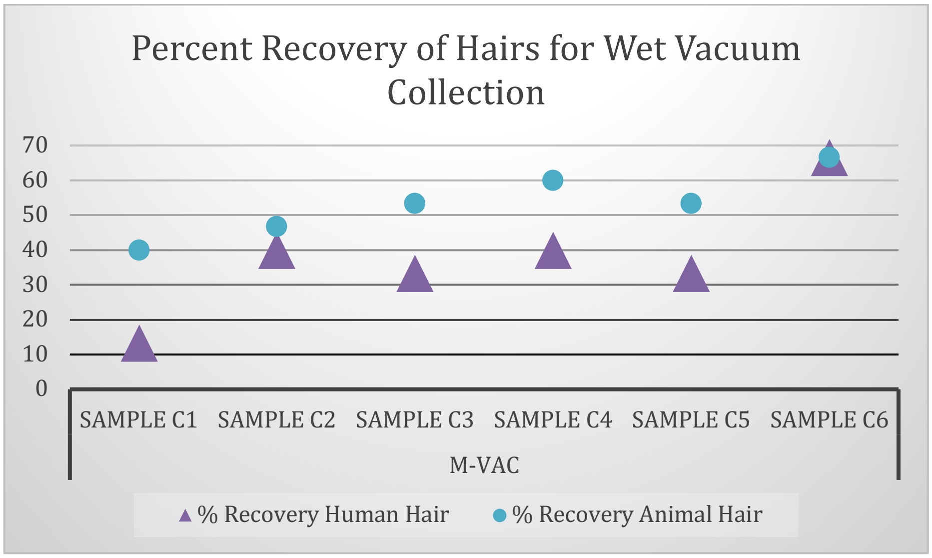


For samples containing trace fibers, the amount of combined collected fibers ranged from a total of 1 to 15. The amount of polyester fibers collected ranged from 0-5, while the amount of cotton fibers collected ranged from 1 to 10 among replicates samples. For samples containing trace hairs, the amount of combined collected hairs ranged from 8 to 20. The amount of human hairs collected ranged from 2 to 10, while the amount of animal hairs collected ranged from 6 to 10. Overall, the trace hairs had a better recovery rate than the fibers for the wet vacuum method.

Figure 8 shows the percent recovery of trace fibers and hairs for the wet vacuum method over all replicates.

Figure 8. Comparison of Hair and Fiber Recovery for Wet Vacuum Collection Method



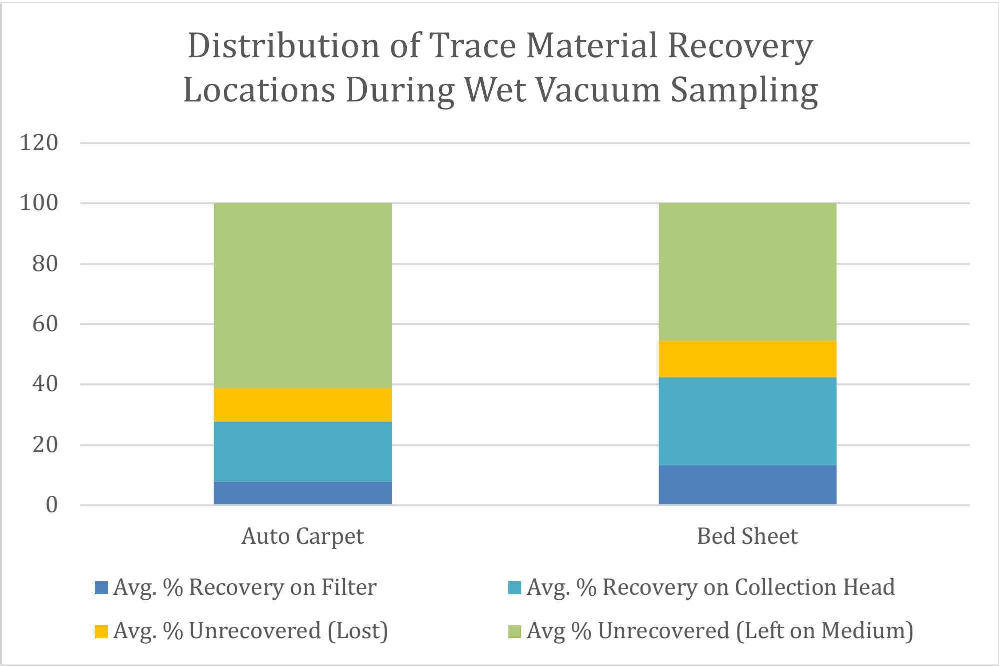


A two sample t-test was performed to compare the effectiveness of trace material collection with the wet vacuum between the trace fibers and trace hairs. There was a significant difference in the effectiveness of trace material collection between trace fibers (M = 3.67, SD =

2.902) and trace hairs (M = 6.83, SD = 2.329); t(21) = -2.947, p = 0.007. These results indicate that trace hairs were collected more efficiently than trace fibers with the M-Vac.

Figure 9 shows the distribution of trace material recovery locations of all wet vacuum sampling replicates.

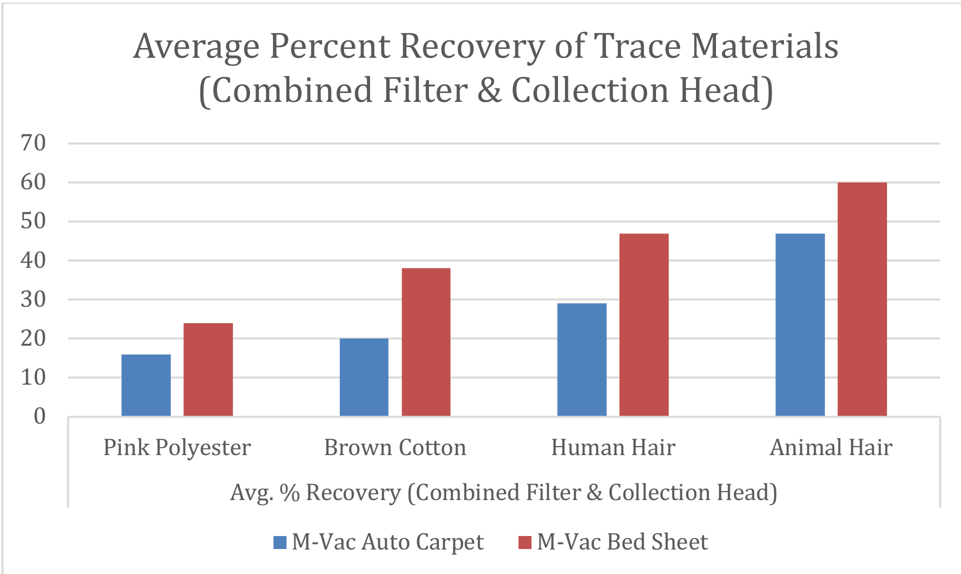
Figure 9. Distribution of Trace Material Recovery Locations During Wet Vacuum Sampling



There were more trace materials recovered from the M-Vac collection head compared to the concentration filter. This could be due to the limited amount of space available for the solution and materials to pass through on the collection head. The bed sheet material recovered the most trace material overall, both on the filter and collection head, but also had slightly more trace material that was unrecovered. The auto carpet substrate retained the most fibers, resulting in less overall recovery. This is likely due to the trace materials getting caught in the thicker medium. The collection of hairs resulted in a loss of trace material recovered, which can likely be attributed to the hairs being embedded into the grooves of the M-Vac collection head, thereby limiting visibility.

The average percent recovery of all four trace materials using the wet vacuum method is shown in Figure 10.

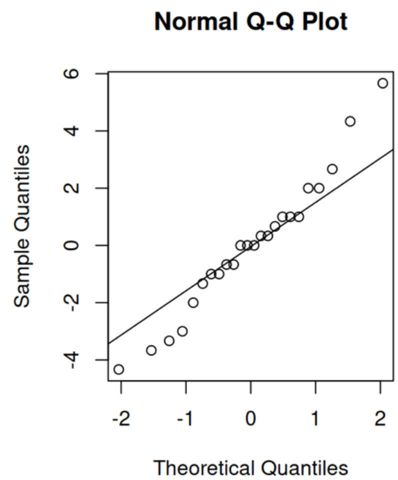
Figure 10. Average Percent Recovery of Trace Materials Using Wet Vacuum Method



Overall, animal hair had the highest recovery rates, while the pink polyester fiber had the lowest. A chi-squared test of goodness-of-fit was performed to determine whether the four trace materials were equally collected. Collection of the four materials was not equally distributed in the population, $X^2(3, N = 3) = 15.587$, $p = 0.001$.

A one-way ANOVA was also performed to compare the effect of the four different trace materials on collection efficiency with the wet vacuum. A one-way ANOVA revealed that there was a statistically significant difference in mean material collection between at least two trace materials ($F (3,20) = [4.175]$, $p = 0.019$).

Figure 11. Q-Q Plot for ANOVA Test

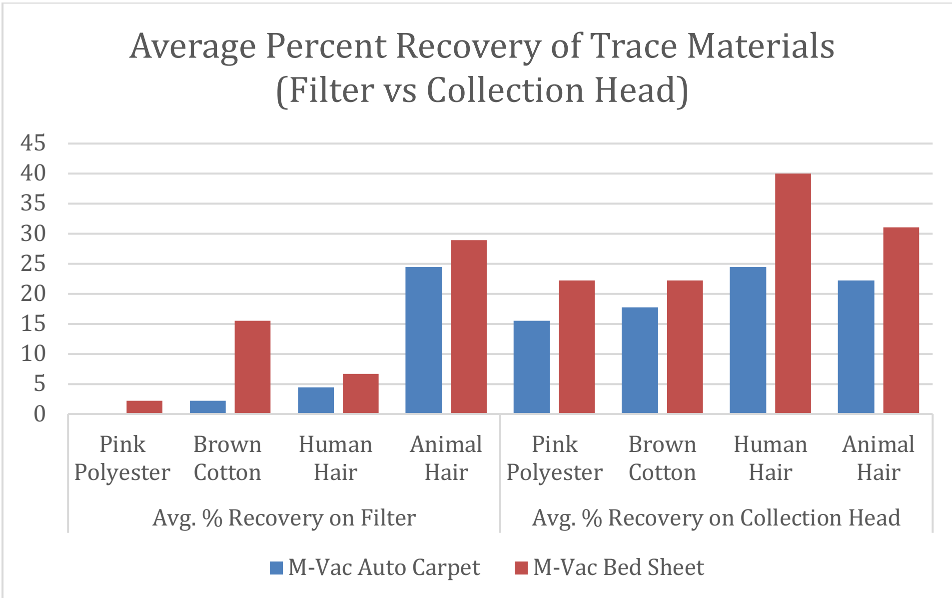


Visual inspection of the normal Q-Q plot of residuals showed minor deviation at the edges, suggesting minor departures from normality, but likely acceptable for ANOVA. A Shapiro-Wilk test indicated that the data was approximately normally distributed $W(20) = 0.972$, $p = 0.721$.

Tukey's HSD Test for multiple comparisons found that the mean value of material collection was significantly different between white dog hair and pink polyester fiber ($p = 0.014$, 95% C.I. = [0.869, 9.13]). There was no significant difference between white dog hair and brown cotton fiber ($p = 0.093$), white dog hair and human hair ($p = 0.411$), human hair and pink polyester fiber ($p = 0.299$), brown cotton fiber and pink polyester fiber ($p = 0.803$), or brown cotton fiber and human hair ($p = 0.803$).

Figure 12 shows the average percent recovery of all trace materials using the wet vacuum method separated by location.

Figure 12. Average Percent Recovery of Trace Materials Using Wet Vacuum Method Based on Location



Overall, most of the trace materials collected were collected on the collection head instead of the concentration filter. A two-sample t-test was performed to compare M-Vac collection efficiency between the two collection locations. There was a significant difference in the collection efficiency between the concentration filter (M = 1.583, SD = 1.816) and the collection head (M = 3.667, SD = 1.857); $t(46) = -3.9294$, p = 0.0002. These results indicate that the collection head yielded higher trace material recovery than the concentration filter. The amount of trace materials collected with the M-Vac separated by the location the material was found after collection, either on the concentration filter or on the M-Vac collection head, can be found in Appendix B. The average percent recovery of trace materials separated by location, either the concentration filter or M-Vac collection head, can be found in Appendix D.

Discussion

The M-Vac collections revealed that it is possible to collect trace materials with this tool. However, when compared to traditional trace evidence collection methods, the M-Vac was less efficient at recovering trace hairs and fibers on surface mediums than the other methods explored. Trace evidence collection using traditional methods of taping and dry vacuuming resulted in recovery of all hairs and fibers. In comparison, M-Vac had an average percent recovery rate of 35%. These results support traditional methods would be preferential if trace evidence is the primary focus. However, there could be some case scenarios in which the maximum collection of biological material is vital. As some biological evidence can be lost during traditional trace evidence collection methods, M-Vac may be the most appropriate method for collection of biological evidence while simultaneously collecting some trace materials. Therefore, the research suggests that M-Vac is not an ideal collection method when trace material recovery is important to a case.

Overall, the animal hairs had the highest average percent recovery rate compared to all the other trace materials for this study. The animal hairs were easily caught on the collection head, as well as they seemed to be the thinnest in diameter, so the thickness of trace materials could be a factor in how well the M-Vac can collect materials. However, some of the trace hairs were lost during collection, so if the materials are thin, it is easier to get caught in the head and not be visible.

There was also loss of the trace human hairs during collection however, there was fewer human hairs lost when compared to the amount of animal hairs lost. This could also be due to the thickness of the hairs. No fiber materials were lost during collection, possibly because the threads were thick enough to not get caught down in the notches of the collection head. The tubing of the M-Vac was also assessed to determine if any hairs were trapped after collection; however, no hairs were seen in the tubing following collections.

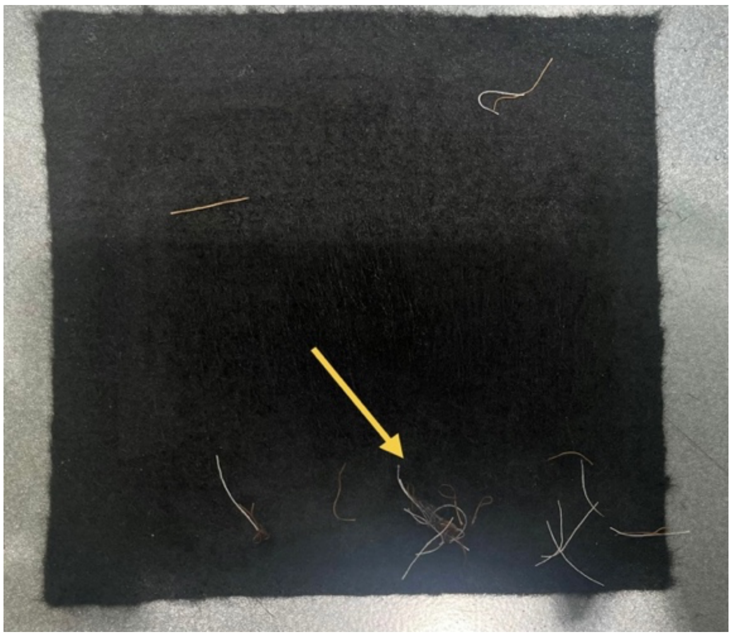
This study used hairs and fibers in lengths ranging from 2-5 cm, which the M-Vac was able to collect through the collection head. However, further studies are needed to determine if the M-Vac would be able to collect hairs or fibers longer than the lengths used. It is believed that longer trace materials would be more difficult for the M-Vac to collect due to the size and design of the collection head.

Thread was used as the fibers for this study instead of using a raw fiber material. This was for ease in sample preparation as well as ease of visualization after collection. Further studies would be needed to determine if raw fiber materials could be collected by the M-Vac. Since most raw fibers are thin in nature it could be possible to be collected; however, visualization would be difficult as most raw fiber is light in color.

The color of the trace materials being collected should also be a consideration when determining if the M-Vac would be a useful collection tool for trace evidence. In this study, the animal hair used was a white color, which made it difficult to visualize on both the M-Vac concentration filter as well as the dry vacuum collection filter. Due to the material of the M-Vac concentration filter, using a high magnification microscope, the animal hairs were able to be seen. However, the material of the dry vacuum filter made it difficult to look at under a microscope and determine the location of the animal hairs, due to the similar nature of the filter fibers, which is why the filters needed to be scraped with a metal spatula to determine the amount of animal hairs collected. After completion of this study, it is suggested that since the M-Vac utilizes a white concentration filter, trace materials of darker colors would be better for collection.

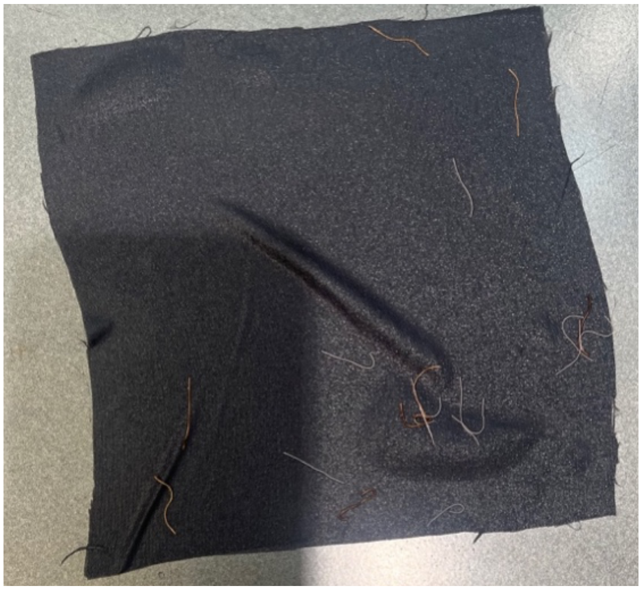
Medium material is also another factor of consideration when using the M-Vac. It was observed that the trace materials could be easily tangled in the automotive carpet fibers during collection due to the way the collection head is used creating a resistance on the surface. An example of this can be seen in Figure 13. The bed sheet material was easier to collect from based on observations; however, once the material became saturated from the buffer solution, the trace materials stuck to the material more, making it more difficult for the M-Vac to collect the material. An image of the bed sheet material after collection can be seen in Figure 14.

Figure 13. Trace Materials Caught in Medium Material Fibers



Note. The image seen above shows trace fibers tangled in the automotive carpet medium fibers from sample B3. Trace materials being caught in the medium material fibers varied between samples. Samples using the bed sheet material did not have this issue.

Figure 14. Bed Sheet Material after Wet Vacuum Collection



Note. The image above shows the bed sheet material and leftover trace fibers from sample B5. It is of significance that the same amount of buffer solution was used for all replicates; however, the bed sheet material became more saturated than the automotive carpet, resulting in the fibers being more adhered to the sheet material.

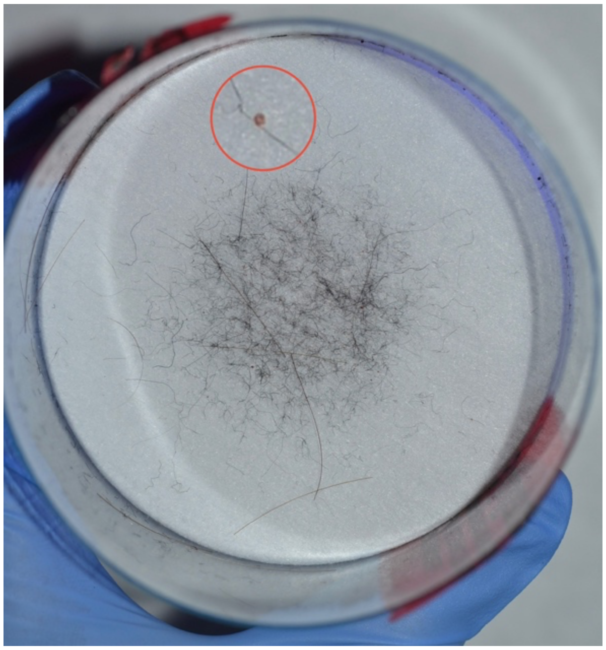
The control sample collections with each collection method revealed that the automotive carpet material was more likely to 'shed' fibers to be transferred onto either the collection filter or tape sheet. The bed sheet material seemed to only transfer fibers from the edges that had been cut during sample preparation. The dry vacuum, in comparison to the wet-vacuum method, showed to have more background material transferred based on observations. This could possibly be due to the difference in the amount of suction between the two vacuums. The dry vacuum seemed to generate more suction during the collection process.

Due to the M-Vac using a buffer solution during collection, it was noticed that once the trace materials were dampened, they seemed to be more difficult to collect due to the surface tension. Collection of trace materials could possibly be improved if a 'dry pass' was used over the surface to collect materials on top of the surface, then turning the buffer solution on. This would almost be similar to using the dry vacuum method to collect the trace materials before using the M-Vac for DNA collection. This method may be more efficient for DNA analysis, depending on if the other traditional methods deplete the amount of DNA recovered, which could be found in the future study. It was visualized that when using the traditional recovery method for trace evidence, that amounts of blood were recovered on the tape strips as well as the dry vacuum filter (Figures 15 & 16). This visualization could mean that the amount of DNA recovered by the M-Vac could be affected by the use of traditional trace evidence recovery methods before DNA collection. The future study using the preserved M-Vac samples collected from this study should be able to determine if traditional trace evidence collection methods influence the amount of DNA recovered.

Figure 15. Visualization of Blood on Taping Collection

Note. The image seen above shows the blood visualization on the taping collection of sample E2. Visualization of blood varied between samples, not all samples contained visualization of blood.

Figure 16. Visualization of Blood on Dry Vacuum Filter



Note. The image above shows the dry vacuum filter from sample I1. The red circled area is magnified for better visualization of the blood transferred from the medium. Not all filters showed blood transference.

This study used a limited number of replicates for each combination of trace materials. The use of only three replicates may limit the statistical power overall confidence in the observed trends, suggesting that future research should employ a larger number of replicates to support more robust conclusions. This also resulted in the confidence intervals having a wider range.

While statistical analyses indicate that the M-Vac method performs more effectively in the recovery of trace hairs compared to trace fibers, it is recommended that future studies assess each trace material type independently to clarify differences in collection efficiency.

Conclusion

The goal of this project was to investigate the effectiveness of the M-Vac system for collecting trace evidence from two different materials, automotive carpet and bed sheets, using four types of trace materials: pink polyester fibers, brown cotton fibers, human hair, and animal hair. The results demonstrated that, while the M-Vac system was effective in recovering trace materials, its recovery efficiency was substantially lower than that of the traditional methods. Both taping and dry vacuuming consistently achieved complete recovery across all replicates.

The findings of this study indicate that the M-Vac system is capable of collecting trace hairs and fibers. When used on both medium materials, the M-Vac demonstrated greater effectiveness in collecting trace hairs than trace fibers. However, most of the trace materials collected were collected from the collection head, rather than the concentration filter. Compared to a traditional dry vacuum for trace materials, based on observations the M-Vac generates less suction , which may account for its reduced effectiveness in collecting trace materials. Although animal hairs were the easiest trace material for the M-Vac to collect, some loss occurred during collection. This loss may be attributable to the design of the collection head, which could allow hairs to become trapped in its grooves.

Future research should investigate whether the M-Vac system can function effectively as a multi-use tool for both trace evidence and DNA collection, specifically assessing whether its use results in higher DNA yields compared to protocols in which traditional trace evidence collection methods are performed prior to DNA sampling. Although blood transfer was observed in some samples following traditional trace evidence collection methods, evaluation of this finding was beyond the scope of the present study. A follow-up study utilizing the preserved M-Vac concentration filters from this project could further evaluate whether prior use of conventional trace collection techniques influences the quantity and quality of DNA recovered during subsequent M-Vac sampling.

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Appendix A: Amount of Trace Materials Collected with M-Vac Method

	Amount of Trace Material Collected (Combined Filter & Collection Head)	
Sample	Pink Polyester	Brown Cotton
B1	0	1
B2	3	5
B3	4	3
B4	2	0
B5	4	7
B6	5	10
Sample	Brown Human Hair	White Dog Hair
C1	2	6
C2	6	7
C3	5	8
C4	6	9
C5	5	8
C6	10	10

Appendix B: Amount of Trace Materials Collected with M-Vac Method Based on Location

	Amount of Trace Material Collected			
	Trace Material on Filter		Trace Material on Collection Head	
Sample	Pink Polyester	Brown Cotton	Pink Polyester	Brown Cotton
B1	0	0	0	1
B2	0	1	3	4
B3	0	0	4	3
B4	0	0	2	0
B5	0	2	4	5
B6	1	5	4	5
Sample	Human Hair	Animal Hair	Human Hair	Animal Hair
C1	0	4	2	2
C2	1	3	5	4
C3	1	4	4	4
C4	0	5	6	4
C5	1	4	4	4
C6	2	4	8	6

Appendix C: Average Percent Recovery of Tracc Materials with Wet Vacuum Method

Trace Material	Surface Type	Mean Percent Recovery (90% CI)	SD	Lower Bound	Upper Bound
Pink Polyester	Auto Carpet	16	13.8	0	0.499
	Bed Sheet	24	10.2	0	0.652
Brown Cotton	Auto Carpet	20	13.3	0	0.579
	Bed Sheet	38	34.2	0	0.838
Human Hair	Auto Carpet	29	13.8	0	0.719
	Bed Sheet	47	17.6	0	0.940
Animal Hair	Auto Carpet	47	6.67	0	0.940
	Bed Sheet	60	6.67	0.134	1

Appendix D: Average Percent Recovery of Trave Materials with Wet Vacuum Method Based on Location

Trace Material	Surface Type	Location	Mean Percent Recovery (90% CI)	SD	Lower Bound	Upper Bound
Pink Polyester	Auto Carpet	Filter	0	0	0	0
		Collection Head	15.56	13.8	0	0.499
	Bed Sheet	Filter	2.22	3.85	0	0.162
		Collection Head	22.22	7.69	0	0.617
Brown Cotton	Auto Carpet	Filter	2.22	3.85	0	0.162
		Collection Head	17.78	10.18	0	0.541
	Bed Sheet	Filter	15.56	16.78	0	0.499
		Collection Head	22.22	19.24	0	0.617
Human Hair	Auto Carpet	Filter	4.44	3.85	0	0.240
		Collection Head	24.44	10.18	0	0.653
	Bed Sheet	Filter	6.67	6.67	0	0.304
		Collection Head	40	13.33	0	0.865
Animal Hair	Auto Carpet	Filter	24.44	3.85	0	0.652
		Collection Head	22.22	7.69	0	0.617
	Bed Sheet	Filter	28.89	3.85	0	0.719
		Collection Head	31.11	7.69	0	0.751

[i] All M-Vac concentration filters were stored in sterile environment after collections for potential use in a future study.