



Trace DNA Retrieval from Forensic Footwear Evidence

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Introduction

Footwear in forensic science is the evidence that provides important information which can be used in assisting criminal investigations in several facets, for instance, determining the type of footwear that has made the impression at the crime scene, establishing the origin of debris found on footwear and also identifying a person who has worn them. Forensic footwear evidence may come in various types at crime scenes such as sneakers, slippers, sandals, flip-flops, etc. In most cases where shoeprints were prevalent, forensic examiners would not only establish a link between the crime scene shoe impression with the specific piece of footwear by comparing the shoe impression against the shoes in question to determine whether or not the questioned shoes were at the crime scene or comparing them against the database (if available) but also prove who the wearer was through DNA analysis. According to Locard's exchange principle which concerns physical contact between two surfaces stating that every touch leaves a trace. When two objects comes into contact, there will always be an exchange of physical traces between the two surfaces [1]. For footwear, the wearer's feet definitely come into contact with the shoes. Therefore, it is anticipated that the skin cells that having deposited in the footwear definitely belong to the wearer. DNA typing from those sloughed skin cells can be used to identify the person who has worn the footwear. Up to the recent years there has been a minimal research on forensic footwear evidence. In Thailand there was no publication on DNA analysis from footwear. Sport shoes and Flip-flops are relatively popular and not infrequent to be found at crime scenes. DNA analysts often look for trace DNA besides the blood stain. The consistency of DNA profile production results from these footwear as trace DNA evidence is never seen.

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Aims and Scope

Apart from shoe impression comparison, fingerprint development and debris origin analysis on forensic footwear evidence, this article is intended to present a brief overview on factors effecting trace DNA retrieval from contact objects especially in footwear. Since there are only a few studies on footwear publicized, this article may not cover all footwear but flip-flops and other shoes.

Trace DNA

Trace DNA could be defined as the DNA amount that falls below the set up threshold and cannot be defined by an exact picogram from evidence detection through DNA profile interpretation [2]. Unlike the prevalent stain of blood or semen which is generally regarded as DNA-enriched biological materials and often producing a high rate of success. To achieve the success in DNA profiling from such a low amount, forensic practitioners should understand the basic property of trace DNA and factors that effecting its recovery as described below.

Factors Effecting Trace DNA

1. Basic property of trace DNA

The ultimate goal in DNA sample analysis is the success in producing a full profile of DNA. The goal may not always be attainable in some samples especially trace DNA evidence. In achieving such a challenging profile, forensic practitioners should never overlook but rather get to fully understand its basic property namely the transfer, the persistence and the abundance. Many of the past studies evaluated the results of trace DNA recovery from touched objects and mostly reported that three basic properties of DNA had strongly influenced upon DNA results [3-4].

1.1 Transfer process

The transfer process generally occurs in two passages, primary and secondary transfers. DNA that was transferred from a person directly to an object is regarded as a primary transfer, while the secondary transfer will concern two transfer events and through a medium, where DNA was transferred from one person to another person and subsequently to an item or a place which is possible under particular conditions [5-8]. Remarkably, the possibility of tertiary transfer of DNA to items was



reported in a past research [6]. DNA transfer from a person to an object is generally involving the shedding status of the person. A good cell shedder would transfer more cells onto the object that came into contact [9]. Additional factors effecting the transfer process were the moisture of skin cells, the anxiety state of a person that increasing his sweat resulting in more cells transferred and the surface of the substrate, like rough items would retain more cells [10]. In the issue of DNA transfer, there were a number of studies reporting the possibility of DNA transfer which could happen in several level; not only the primary and the secondary event but the tertiary was also possible [5-6]. The main argument is that false incrimination of an innocent person may happen and raise an issue in the consequence of such transfer event. Therefore, forensic practitioners must be aware whenever dealing with trace DNA.

1.2 Persistence

A few studies concerning the persistence of DNA were conducted. One of these were to evaluate time if having an effect on the amount of DNA. A known DNA concentration of saliva was used as a DNA input and being left in a controlled and undisturbed condition. Different periods of time were applied (1-Day, 1-Week, 2-Week and 4-Week time). The findings indicated that the amount of DNA output was observed but comparatively decreased over time [11]. The other two studies were also intended to assess the length of time. The experiments which were designed to simulate real cases revealed a substantial decrease in DNA yield from outdoor window frames over time and no DNA was found after two weeks. Interestingly, if the samples were left in a dark, cool and undisturbed environment, it was possible to observe DNA profiles after six weeks [3-4]. In the issue of DNA persistence, the findings from these studies insinuated that the sooner DNA collected the greater chance DNA detected.

1.3 Abundance

There has been a limited research on this area not like the transfer process. The studies on this part was solely to measure a baseline level of trace DNA retrieved on the entry points of residential burglary like the frames of door and window. The results disclosed a comparative low in the baseline level of DNA [4]. This could assume that trace DNA by its nature is of transient and fragile.



2. Cell Shedding Ability

Apart from the three basic properties mentioned above there's still another factor that has an affect on trace DNA. There have been a large number of researchers conducting studies on the tendency of individuals to shed their skin cells onto handled items and similarly indicated that there was a huge variation [5,9, 12-17]. There were several factors affecting the shedding ability namely the effect of hand dominance and the length of time between the contact and the hand washing. Such finding suggested that categorization of shedding ability of a person was not possible. Another previous studies conducted on DNA retrieval from flip-flops indicated that a good shedder transfers more cells on the flip-flops than a bad shedder did. Moreover, the ability to shed cells was not consistent within a person. A person could be both a good shedder and a bad [9,11]. In the issue of cell shedding status of a person, Lowe et al. had reported that there was a variation among individuals in their tendency to deposit their cellular materials onto an object that they touched. The cause of shedding variation among persons and within a person was yet unknown [5]. A very interesting research were conducted, assessing the shedding ability between human limbs, in particular, hands and feet. Volunteers' hands and feet were swabbed for DNA recovery. This research applied the double swab technique to the hands and feet of six volunteers to test the shedding ability and reported that lower DNA amounts were obtained from the feet compared to the hands [18]. The finding could relatively imply that there would be less chance to recover DNA from forensic footwear evidence.

3. Collecting and Recovering Methods

When happening to deal with the challenging trace evidence like trace DNA, a crime scene examiner should develop a systematic approach. Beginning with a sample collection; a proper selection of moistening agent, a high quality of swab item and a swabbing technique to jointly use in the DNA collection step. Sample collection is very important and typically regarded as the starting point of DNA profiling. Conducting a good start is half the work. At this point, the higher yield of collected DNA would imply the higher chance of success in DNA profiling. Even though the advances in DNA technology in recent years are available for forensic practitioners to generate a profile from a low amount of DNA, the ability to collect invisible cellular materials from a contact object is even more important.



3.1 Moistening Solutions and Swab Items

In general, sterile water and DNA-free water have been used to moisten the swab to collect biological evidence such as blood, semen and even trace evidence at crime scenes. The subsequent quality and quantity sometimes come along with a doubt especially from trace DNA. There have been several studies described below disclosed various results of moistening solutions evaluation to use in trace DNA retrieval. A swab moistened with ethanol was also one of those swabbing techniques which reported as the best collection technique for firm and not wet surfaces [19]. Sterile distilled water and 95% Ethanol were comparatively tested in their efficiency in trace DNA recovery in a study of DNA retrieval from flip-flops. The result indicated that 95% Ethanol recovered higher DNA yields than sterile water [11]. Sarah et al. had studied the effect of swabbing solution on DNA retrieval from touch samples. In the experiment, water and six detergent-based solutions were compared. The report revealed that detergent-based swabbing solutions gave better results than water while sodium dodecyl sulfate (SDS) and Triton X-100 increased DNA yield significantly [20]. Another interesting studies on the evaluation of solutions and swabs. The experiment was designed to evaluate the effect of swab brands and moistening agents used in touch DNA collection. Four brands of swab were used; Bode SecurSwab, Puritan DNA Free Cotton Tipped Applicator, HI-VAN Lab swab and EO swab and six moistening agents were used; DNA-free water, phosphate-buffered saline, ethanol, sodium dodecyl sulfate, isopropanol and IQ lysis buffer. The findings demonstrated that both solutions and cotton swabs jointly influence the DNA collection process. Not specific brand nor agent alone worked best, nevertheless, isopropanol together with the EO swab produced the highest yield of DNA [21]. A wise selection of moistening solutions and appropriate swabbing items will enhance the ability to collect cells from the contact surface.

3.2 Swabbing technique

When dealing with trace DNA forensic practitioners need efficient sample collection methods. A single use of moistened swab for collecting cells from surfaces may not cover all in a sense but following by a second, dry swab would even be practical. The use of this technique is generally known as a double swab technique. There was a huge number of publications regarding the DNA recovery enhancement reporting that the double swabbing technique was most widely used and similarly recommended the



double swab technique [4,9,16,18,22-26]. One interesting research applied the double swab technique to the hands and feet of six volunteers and reported that lower DNA amounts were obtained from the feet compared to the hands [18]. The ability to cover entire cells deposited on an object depends on the methods used for trace DNA collection. Choosing an appropriate collection method will promise a favorable result.

DNA Recovering from Forensic Footwear Evidence

There has been a limited publication in trace DNA retrieval from footwear. In a past research Bright et al. had conducted a research on trace DNA profiling from shoe insoles and reported a success in recovering DNA profile from the wearer. The interesting results were that the hand could give a higher DNA yield than that of the foot through the double swab method, while the more swabs used, the more DNA recovered. Surprisingly, the instep of the foot had been found to be the better area for swabbing than the sole because higher DNA yields could be retrieved from instep area. Therefore, the suitable area for sampling should be the area that came in contact with the instep of the foot. The explanation to this incident was that microbial decomposition was one of the factors that lowered the DNA yields [18]. However, bacterial DNA often interfered with the specificity at the DNA quantification process through the NanoDrop instrument [11], but had no effect on the Quantifiler™ which is specific to human DNA and produced reliable results [27-28]. A study on DNA-related flip-flops was conducted to investigate trace DNA retrieval from worn flip-flops. Volunteers used flip-flops with designated duration of time (1-Day, 7-Day, 14-Day and 30-Day time) in a controlled environment. The findings revealed that a 14-Day wearing gave the highest yields, while the amount of DNAs were increasing over time. But a sharp decline of DNA yields was observed on the 30-Day session. The decrease of DNA could be caused by microbial decomposition. The study also suggested that 95% ethanol was more effective than sterile distilled water and suitable to use for trace DNA retrieval [29]. Another two studies concerning the recovery of DNA from shoes. Studied on twenty-one shoes and forty-three respectively with a focus on three areas of those shoes; the heel, the lace/tongue and the insole. Of the three areas the lace/tongue gave the most information of the wearer than the rest because the lace/tongue were the major area that came into contact with the wearer's hand. The report concluded that the shedding status of the wearer and the environmental condition influenced the determination of the result



[30].Moreover, the presence of chemical powder used in fingerprint development was also reported to have an influence on trace DNA retrieval [10,31].The ability to collect all cells from footwear is very crucial. A wise selection of moistening solution with high-quality swabbing items should be introduced.

Conclusion

In legal proceedings, forensic footwear evidence is applicable in assisting criminal investigations from certain aspects as described in the introduction section. The summary of findings and comments pertinent to trace DNA recovery from past publications have remarked the forensic practitioners to realize that there are several things to take into consideration. Whether they were the studies of DNAs from footwear or other contact objects, yields of trace DNA depended heavily on a wide range of factors including the collection methods, the comprehension of three basic properties, the nature of donor in cell shedding, the substrate for DNA to transfer to and the environmental condition the DNA exposed to, as well as the duration of time between the contact and the detection of transferred DNA.

In the author's view, for forensic DNA analysis, the two most difficult and important parts could be the mixture and the degradation of the trace DNA. The mixture issue might not be controllable and need a careful interpretation. Trace DNA is prone to degradation due to its nature. The sooner it is processed, the greater chance it will be retrieved. The success in trace DNA profiling is a combination of many elements. Even though we have implemented the state of the art technology and methods together with a dream team of knowledgeable and high-profiled DNA experts as well as a sophisticated laboratory, we won't attain a single profile if we fail to implement a good start. Sample collection at crime scene is very a crucial starting point. "A good start is half the work."



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