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Touch DNA: Revolutionizing Evidentiary DNA Forensics

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Abstract

Touch DNA is an advanced technique widely employed in modern criminal justice systems in many developed countries. It aims to extract genetic information from biological substances, specifically the cells shed from the outermost layer of skin, that are left behind on touched objects. This method involves recovering trace amounts of DNA from the biological cells released during contact, even though the quantity is usually very low. The recovered DNA is further analyzed to generate a person's DNA profile. Since dead cells are not really visible to the naked eye, successfully locating and recovering them can be challenging. Performing DNA profiling from the samples that are just touched is quite difficult, hence, requires a highly sensitive approach to its proper recovery, extraction, and amplification of the segment. The methods which are used for the collection, sampling procedure, preservation, removal of contaminants, quantification of DNA, the amplifying of the genetic material, and the subsequent analysis and interpretation of the findings all play a role in how well touch DNA analysis works. Various techniques have been created over time to gather touch DNA. Reliable DNA profiles are produced thanks to the use of sophisticated kits, tools, and well-equipped forensic laboratories, which benefit the criminal justice system.

Keywords: Touch DNA; DNA Profile; RT-PCR; Forensic Science; Quantification

Abbreviations: NGS: Next-Generation Sequencing; ANDE: Accelerated Nuclear DNA Equipment.

Introduction

Touch DNA particularly refers to the availability or the presence of DNA that has been transferred through direct contact between an object or surface and a person's skin cells. It is an essential concept in forensic science as it helps and enables the investigator to collect and analyze trace or minute amounts of DNA that are left behind by individuals who might have come in contact, touched, or handled an item.

On the basis of very foundational Locard's Principle, when a person comes in any contact with or touches an object, they leave behind microscopic skin cells, which include the epithelial cells present along the outermost layer of the skin. These cells contain DNA that can be recovered and analyzed to provide evidentiary identification of the individual who might have left the DNA sample. This novel technique of analysis involves the extraction, amplification, and further analysis of DNA from these skin cells [1].

The importance of touch DNA in forensic science lies in its potential to help link individuals to crime scenes or any such items of evidence. Even small amounts of DNA, such as those left behind by a single touch, can be sufficient

for analysis using modern forensic techniques. This is particularly valuable in cases where other biological shreds of evidence, such as blood or semen, may be absent or present in insufficient quantities for analysis.

Touch DNA is possible to be found in various areas, including clothing, weapons, tools, door handles, and personal belongings. By analyzing touch DNA, forensic scientists can help establish the legal evidentiary linkage between suspect and crime scene, provide additional evidence for identification purposes, or exclude innocent individuals from suspicion [2].

Therefore, it is highly important to note that touch DNA analysis has its own limitations. Depending on the quality and quantity, touch DNA can vary such as duration and intensity of contact, surface type, type of contact, and environmental conditions. Touch DNA may also be easily contaminated by other DNA sources, making proper collection and preservation techniques crucial to ensure accurate results.

Touch DNA has gained increased interest in many infamous cases, such as the acquittal of Timothy Masters and the JonBenét Ramsey homicide investigation. The first ever documented report on the capacity to extract DNA samples from these epithelial cells was made in 1997 by Van Oorschot and Jones.

This breakthrough has proven to be valuable in various cases, including those involving murder, rape, and sexual assault [3].

Humans shed tons of these skin cells each day, and these cells can be transferred onto objects that come into contact with our skin. This transfer of skin cells, if able to deposit a sufficient amount or number of cells at any given crime scene, provides potential evidence that can link the perpetrator or depositor to the crime. For example, useful DNA profiles can be analyzed and obtained from clothing worn by the perpetrator, which may contain transferred skin cells. The forensic evidence's evidentiary value is further increased by the ability to identify the wearer of the shoe using DNA profiles found on the inside of the shoes.

A touch DNA study conducted by Castella and Mangin analyzed 1,739 case samples and found that 26% of those samples successfully yielded suitable DNA profiles. Similarly, Raymond et al. executed a touch DNA study on 252 case samples and encountered that 44% of them produced suitable DNA Profiles [4-14].

Forensic science has witnessed the development of sensitive DNA typing kits aimed at obtaining DNA profiles from trace amounts of cells found on various objects, including postage stamps, documents, bullets, knives, and door handles. The progress made in this field has facilitated the effective retrieval of touch DNA from a wide range of objects, including bottles, letters, personal possessions, imprints on the forehead, utensils, food, cans, handled objects, envelopes, and postage stamps. Additionally, investigations have explored the possibility of obtaining touch DNA from items such as socks, shoes, cigarette butts, hats, upper garments, chewed gum, gloves, and underwear.

This particular article tries to shed light upon both conventional and the latest sampling methods for touch DNA, outlining their benefits, evidential value, and limitations. It highlights that the successful analysis requires trained laboratory officers, Investigation Officers, or Law officers to properly lift, handle, and transport exhibits from the crime scene to Forensic Science Laboratory. The quality of the generated STR profiles obtained can be influenced by several factors, including the time gap, pressure, area, time, and most importantly state of mind of the personnel involved. Obtaining a high-quality STR profile can further be enhanced by employing increased touch pressure, expanding the touch area, and prolonging the touch time.

Furthermore, it has been observed rough substrates are more likely to retain touch DNA due to their texture, unevenness, and presence of valleys and creases. Over time, various collection methods have been developed for touch DNA, and well-equipped forensic laboratories with sophisticated kits and instruments play a crucial role in providing reliable DNA profiles, thereby assisting the Criminal Justice System [4].

Modes of Lifting Touch DNA Samples

Swabbing Techniques: Traditionally preference has been given to dry sterile cotton swabs used for collecting touch DNA samples at crime scenes. However, wet sterile cotton swabbing has now replaced dry swabbing due to its improved effectiveness. Despite this, swabbing techniques have limitations in recovering sufficient DNA, leading to the development of tape-lifting techniques.

While cotton swabs are still used for collecting touch DNA from smooth surfaces, they result in significant DNA loss from rough and porous substrates. The Indianapolis Police Department started using prepackaged touch DNA swab kits (TriggerPro) in 2008–09 to gather touch DNA samples from confiscated firearms. These kits contain moistened swabs with antimicrobial fluid. Furthermore, the utilization of non-polar surfactants in lieu of sterile distilled water as wetting agents on cotton swabs has been implemented to improve the retrieval of touch DNA from suspected items found at crime scenes [5].

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Tape Lifting: Many jurisdictions routinely employ tape lifting for collecting touch DNA from fabrics. Water-soluble tape lifts, such as SceneSafe Fast™ mini tapes, are now available and can increase the amount of touch DNA collected.

Cutting/Scraping: While cutting/scraping the suspected touch area provides the best method for obtaining touch DNA, it is a destructive technique. Therefore, non-destructive techniques such as swabbing and tape lifting are typically employed for lifting touch DNA from crime scenes.

FTA Card: For direct PCR amplification, specialized equipment like FTA Cards and MicroFLOQ swabs have been created, lowering the possibility of contact DNA loss during the extraction process and measurement.

Touch DNA Extraction

Organic Extraction: Bright and Petricevic [15-24] conducted a study using organic extraction methods (phenol: CHCl3) and Chelex 100 resin to recover DNA from collected swabs of the hands and feet of the volunteers. The study successfully demonstrated that the organic extraction procedure resulted in a greater yield of DNA.

Automated DNA Extraction: Silica-based magnetic systems, such as the EZ1/XL BioRobot by Qiagen and the Maxwell® FSC by Promega- Automated DNA extraction system, offer the advantages of producing high-quality genomic DNA and minimizing the risks of contamination that may arise from manual handling. The Low DNA Content BioChipSet (LDC BCS) integrated into the fully automated Accelerated Nuclear DNA Equipment (ANDE) is specifically engineered for efficient DNA purification, microfluidic ultrafiltration, as well as subsequent amplification, electrophoretic separation, and detection of amplified fragments. Additionally, a 96-well centrifugal filtration plate has been developed for automated DNA extraction from touched objects. This plate enables the processing of 92 samples on a robotic workstation within a timeframe of 90 minutes [12].

Quantification

This step is crucial in determining the subsequent processing of the sample. DNA quantification helps in reducing the number of (off-, over-, or under-) amplified samples and provides normalized profiles for reliable data and its interpretation.

Different methods for DNA quantification include UV-spectrophotometry, Micro/Nanodrop (which relies on UV-spectrophotometry), and Real-Time PCR. Real-Time PCR is considered more advantageous than spectrophotometry because it accurately determines the initial concentration of DNA being amplified, while absorbance measurements can fluctuate due to contaminants [7].

Amplification and Detection

The Amplification of extracted DNA followed by Amplicons plays a crucial role in the DNA profiling process. The entire profiling is set up on the rapid amplification of specific loci, which are then analyzed. Multiplexing, a technique used for STR profiling, involves the use of sophisticated kits provided by various manufacturers. These kits include the AmpFlSTR SGM PlusTM, AmpFlSTR Identifiler Plus, GlobalfilerTM PCR Amplification Kit system (Applied Biosystems), Investigator 24Plex QS kit (Qiagen), PowerPlex® 21/18D System (Promega), powerplex fusion 6c, VersaPlex® 27PY System and others. Following recommended protocols, DNA amplification is performed using these kits.

When only partial profiles are acquired, utilizing Mini STR kits is another method of creating a full profile. By increasing the number of PCR cycles, successful STR profiling can be achieved even with low initial sample concentrations [8].

Capillary Electrophoresis

After amplification, DNA fragments are separated using capillary electrophoresis, typically employing Genetic Analyzer instruments such as 3100, 3130, 3500, or 3500XL. The data obtained from the electrophoresis are analyzed using Gene Mapper ID-X software.

Technique	Sensitivity	Accuracy
Quantitative PCR (qPCR)	High	High
Digital PCR	Very high	High
Microfluidic-based PCR	High	High
Capillary Electrophoresis	Moderate to high	Moderate to high
Next-generation sequencing (NGS)	Very high	High
Droplet Digital PCR (ddPCR)	Very high	High

Table 1: Comparison of Various Quantification, Detection and Profiling Techniques [1,5,6,26].

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Cutting-edge technology like the Accelerated Nuclear DNA Equipment (ANDE) and RapidHIT ID System for Identification allows samples to be processed immediately after collection. This automated procedure involves steps like cell lysis, amplification, capillary electrophoresis, and the creation of highly specific STR profiles in under 2 hours. The streamlined process is achieved through self-contained sample cartridges, which consolidate the entire procedure into a single device and a single task initiated by the user.

Next-Generation Sequencing (NGS)

Next-generation sequencing (NGS) has made significant advancements in producing a vast number of sequence reads quickly and generating large DNA sequences in variable-sized fragments. This technology allows the creation of libraries from entire genomes or specific regions of interest without prior knowledge of their sequences. In forensic science, limited sample availability and DNA degradation pose significant challenges. However, the decreasing DNA input requirement for NGS library preparation opens up the possibility of sequencing nearly any sample, enabling maximum information retrieval from biological remains. Moreover, the application of NGS in microbiome profiling can provide valuable insights for crime scene characterization [8].

NGS technologies will play a pivotal role in DNA human typing in scenarios such as mass disasters or incidents where forensic specimens and samples are compromised or degraded. By utilizing NGS, simultaneous analysis of standard autosomal DNA markers (such as short tandem repeats [STRs] and single nucleotide polymorphisms [SNPs]), mitochondrial DNA, and markers specific to the X and Y chromosomes becomes achievable. This comprehensive approach enhances the accuracy and efficiency of forensic analysis, aiding in the identification and characterization of individuals in forensic investigations [9].

Results

Interpretation of the results thus obtained from DNA samples involves classifying profiles as

- Full DNA profiles,
- partial DNA profiles,
- mixed DNA profiles (containing DNA from multiple individuals), or
- No result,

This totally depends on the number of alleles that have been detected. After STR profiling, touch DNA interpretation can be performed. A match between two DNA profiles is considered an “inclusion,” while profiles that do not match are considered an “exclusion.”

Author	Sampling Method	Important Findings
Comment, et al. [10]	A comparison was conducted over a duration of five months to evaluate the effectiveness of five distinct ISO 18385-certified forensic swabs in collecting DNA samples. The assessment encompassed 1094 samples of “touch” DNA.	The swabs exhibited significant variations in DNA profiling success rates and DNA extraction efficiency. Collaborators’ evaluations of the swabs’ handling also differed significantly. The study highlights the importance of considering a holistic approach in selecting swabs for DNA sampling, as convenient handling can reduce contamination risk, improve sampling efficiency, and increase staff satisfaction. The findings suggest that controlled laboratory conditions alone may not be sufficient for choosing the best swab, and additional factors should be taken into account during the selection process.
Thornton [2]	Utilization of ATR-FTIR to investigate the interactions between DNA and metals. Employ three distinct collection techniques, namely the wet-dry double-swabbing method, tape-lifting, and the M-Vac® wet-vacuum DNA collection system. Employ the centrifugal separation method to extract the samples.	ATR-FTIR is used to evaluate the interactions happening between the DNA and Metal at the surface level. Three collection methods were assessed for ‘touch’ DNA recovery from metal substrates. Centrifugal separation method tested for efficient extraction of cellular and cfDNA.

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Bini C, et al. [11]	120 fingerprints deposited on glass surfaces by 20 volunteers before and after using an alcohol-based hand sanitizer. The influence of the hand sanitizer on the deposition, transference, and recovery of touch DNA was assessed. Real-time quantitative PCR (qPCR) is used to quantify the DNA samples. 76 samples with DNA concentrations above 15 pg/µl were typed for 21 autosomal STRs using the GlobalFiler® PCR Amplification Kit.	After using the sanitizer, the samples showed descended quantities of the recovered-transferred DNA, particularly in the with salivary DNA. However, all 76 amplified samples (63.3% of the total) exhibited at least 10 typed loci. Furthermore, 83-100% of the DNA profiles were consistent with the reference samples based on a likelihood ratio (LR) value of at least 10^6.
Jansson, et al. [12]	27 pairs of hands, n = 29 for facial DNA analysis, n = 14 for sebum secretion analysis. Active hands: hands used “as usual” Inactive hands: hands not allowed to touch anything Comparison of DNA deposition from active and inactive hands	The study compared the amounts of DNA deposited from active and inactive hands. It analyzed the association between deposited DNA and accumulated facial DNA, sebum secretion levels, and time since hand or face wash. Statistical analysis, including paired t-tests and correlation calculations (Pearson’s and Spearman’s), was conducted to examine the relationships between variables. The sample size varied depending on the specific analysis conducted. The amount of DNA deposited from active hands (2.1 ± 2.7 ng) was higher than that from inactive hands (0.83 ± 1.1 ng) (paired t-test, $p = 0.014$). Individual levels of deposited DNA were highly associated with DNA accumulation on the skin of the face (Pearson’s correlation: $r = 0.90$, $p < 0.00001$; Spearman’s ranked correlation: $rs = 0.56$, $p = 0.0016$). No correlation was found between DNA levels on hands or forehead and time since hand or face wash. No correlation was observed between DNA levels and individual differences in sebum secretion levels (Pearson’s correlation: $r = -0.13$, $p = 0.66$).
Alketbi SK [13]	Cotton swab in combination with microFLOQ® swab (CS+MF). - 100% allele recovery rate for blood and saliva samples. - 84% allele recovery rate for trace samples. - Significant difference in average signal (RFU) between evidence types ($p < 0.001$).	The study explored the use of direct amplification techniques with CS+MF to preserve collected samples for re-analysis or additional testing.
Haase, et al [14]	68 samples of Nylon vs CottonNylon swabs collected shed cells more efficiently. Visualized with Diamond™ Nucleic Acid Dye (DD) DNA yield higher with Puritan® swabs DNA quantification using Quantifiler Trio kit. OneTouch™ processed samples had highest yield	16 out of 41 gun shell casing samples had no STR profilesStochastic effects observed in most samples Full profiles (17 STRs) obtained with OneTouch™ Direct PCR, chelex extraction, and mechanical rupture similar performance Full mtDNA profiles obtained for 65% of the samples Partial mtDNA profiles obtained for 15% of the samples
Pfeifer CM, et al. [5]	234 Samples of STR analyses	When second users handled tools barehanded in a simulated burglary, the first user’s DNA was rarely detected on their handles (1/40 cases).When second users used gloves while breaking up the burglary setup, the first user’s DNA matched the handle profile in 37% of the casesTouch DNA in burglary-related cases with two consecutive users

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Lim, et al. [15]	The study involved experiments to determine the success of DNA recovery from the surface of cable sheaths. Three different substances were deposited on the sheaths: sweat, extracted DNA, and fingermarks. The study also compared the double swab technique and mini-taping as options for DNA recovery.	DNA recovery: The study found that there was generally no significant difference between using swabs or mini-tapes to recover DNA from the non-porous cable sheaths. Comparison of treatments: Cyanoacrylate fuming (CNA fuming) performed better than wet powder suspensions (WPS) in terms of subsequent DNA recovery and profilingAfter CNA fuming, there was an average increase in DNA recovered via swabbing and taping (more than 4x and 8x, respectively) compared to no treatment.. DNA profiling: 50% of the DNA recovered after CNA fuming generated full DNA profiles.
Kallupurackal, et al. [16]	Total of 80 Samples using: Single-swabbing, double-swabbing, and adhesive tapes	Results indicate COPAN FLOQTM, double-swab technique, and regular swabbing techniques with cotton swabs performed equally well across all tested methods.
Verdon, et al. [17]	Two types of tapes with different adhesive strengths (Scotch1 MagicTM tape and Scenesafe FASTTM mini tapes) were compared for tape lifting from touch deposits on four different fabrics. Wet/dry swabbing with cotton swabs was also used as a comparison.	Scenesafe FASTTM tape extracted significantly more DNA and detected a higher proportion of alleles compared to Scotch1 MagicTM tape. The amount of DNA and number of donor alleles generally increased when the tape was reapplied to the surface, but there was a threshold of collection for both tape types. For two out of four substrates, Scenesafe FASTTM tape collected more DNA and generated a greater median number of donor alleles compared to swabbing. There was no significant difference in the number of alleles between swabbing and taping from flannelette fabric.A tape with stronger adhesion (such as Scenesafe FASTTM tape-lifters) is recommended. Multiple applications of tape are suggested, although excessive sampling can diminish collection efficiency Tapelifting is advantageous over swabbing for fabrics, except in the case of flannelette with easily removable loose fibers

Table 2: Comparing Studies to Understand the Efficacy, Sensitivity, Reliability, and Accuracy of Touch DNA.

Based on Table 2, it can be definitively deduced that Touch-DNA as a regular technique and its scope in Forensic Laboratories has so much potential owing to its sensitivity (ability to work with minimal and minute quantities), accuracy to generate the Profiles, and providing reliable, reproducible, specific results pertaining to the Touch DNA evidence. This technique is continuously undergoing research and development with improved results and definite application in Forensic Evidentiary value [18].

Challenges

Several challenges can arise during DNA analysis. Sampling and handling problems may occur due to various methods employed for the collection of touch DNA, such as swabbing, tape lifting, cutting, or scraping. Special care should be taken to minimize the contamination via personnel collecting the DNA and other sources at the crime scene. Contamination can also arise from improper handling, sampling, preservation, or environmental effects. The risk of contamination should be mitigated through the use of proper protective equipment and strict protocols [19].

Preservation and transportation of biological samples also pose challenges. Proper drying, refrigeration, or freezing techniques are used to preserve biological samples, and transportation requires careful handling to prevent degradation or decomposition. Efforts are being made to develop storage devices that can maintain sample integrity at the preferred room Temp. , reducing the need for freezing or refrigeration [20-25].

Benefits

- **Specificity:** Even a small quantity of DNA from a few cells from the crime scene can provide a complete DNA profile.
- **Speed:** Fast and efficient.
- **Versatility:** Applicable to a variety of surfaces.
- **Small Sample Requirement**: Easily performed with a very small size of the sample.
- **Technological Advancements**: The development of sensitive kits and sophisticated equipment has improved touch DNA technology.

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Limitations

- **Variable Deposition**: The amount of touch DNA deposited can vary depending on factors such as surface texture.
- **Individual Differences**: Different individuals may leave varying amounts of touch DNA, and factors like stress or sweating can affect the quantity.
- **Degradation**: Touch DNA is highly prone to biological and environmental degradation, including any contamination by fungi and bacteria.
- **Sample Destruction**: Touch DNA samples are small and easily destroyed, making recreation impossible.
- **Contamination**: DNA aerosols may cross-contaminate the lab environment, making other DNA unsuitable for further profiling.
- **Dependent on Trained Personnel**: Touch DNA analysis requires well-trained personnel and errors in processing techniques can lead to incorrect or incomplete DNA profiles.
- **Risk of False Interpretation**: False or erroneous interpretation of touch DNA results can lead to misleading conclusions and potentially wrongful outcomes in a case (Nunn 2013).

Conclusions

The primary goal of forensic science is to enhance the assessment of evidence. Various sources and reports provided by operational scientists have indicated that during the collection and analysis process, as much as 90% of the DNA in a sample can be lost. However, other studies suggest that this loss averages around 39%. Gaining a deeper understanding of the challenges at each step of the DNA testing workflow is crucial for potential improvements in the efficiency of all types of biological samples, such as semen, blood, and saliva. By enhancing the efficiency of sample collection from different substrates and improving the subsequent extraction process, we can increase the number of samples that yield successful profiles, particularly when the initial DNA quantities are low or environmentally compromised, as is often the case with touch samples. By gaining a better understanding of DNA yields and the factors contributing to loss, targeted process improvements will enable touch DNA samples to become more commonly utilized with standardized and enhanced methods.

However, obtaining all the available DNA remains a challenge, depending on the type and porosity of the substrate [26-30].

The success of touch DNA as a forensic tool depends on optimizing techniques from sample collection to profile generation. Advances in methods, kits, instruments, and well-equipped laboratories have improved the process. However, careful planning and consideration of the limitations are essential for individual cases to ensure accurate interpretation of results.

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