UNIVERSITI SAINS MALAYSIA

UNIVERSITI SAINS MALAYSIA

TOUCH DNA PROFILING FROM PROCESSED FINGERPRINT

Dissertation submitted in partial fulfillment of the requirements for the degree of Bachelor Sciences in Forensic Science

HAFIDZA BT BAHARUM

School of Health sciences
Universiti Sains Malaysia
Health Campus
16150, Kubang Kerian, Kelantan
Malaysia

April 2009

CERTIFICATE

This is to certify that the dissertation entitled

"Touch DNA Profiling from fingerprint"

is the bona fide record of research work done by

Ms. Hafidza Bt. Baharum

during the period of December 2008 to may 2009 under my supervision.

IN THE NAME OF ALLAH

THE MOST BENEFICIENT

THE MOST MERCIFUL

AKNOWLEDGEMENTS

In the name of Allah, the most gracious, the most merciful, shalawat and remembrance for the Holy Prophet, Muhammad S.A.W for the blessing in giving me strength, health, and patient along this final semester to complete the research paper.

I would like to address my sincere thanks and deepest gratitude to my supervisor Dr. Zafarina Bt. Zainuddin, for her great assistance, consultation, proper guidance and faith during the period of preparation this precious dissertation. Without her expert advice and gracious comment, this research could not have been possible.

I would like also to forward my utmost gratitude to Ms. Tengku Puteri Nadiah bt. Tengku Baharudin Shah a master's student and Mrs. Nur Hazlindawati bt. Abd. Rashid a PhD's student in Forensic Laboratory for their constant support and assistance during my hard days conducting this research. Without their contribution, this research would not become a reality.

I'm in debt for the rest of my life to my parents, Mr. Baharum b. Zainal and Mrs. Rusnani Bt. Amiruddin to whom I dedicate this research:

Everything I have ever accomplished and ever will accomplish goes to their credits. Without their love, support and pray, none of this would have been worthwhile.

My appreciation to, Ms. NurAlifah Bt. Jali for her time spent and cooperation. My research partner, Ms. Zuliana Bt. Zakariya, and all my friends who had generously contributed their idea and friendly support for me to finish this project.

APRIL 2009

HAFIDZA BT BAHARUM

CONTENTS

LIST OF ABBREVIATIONS

µL	=	Microliter
bp	=	base pair
DNA	=	Deoxyribonucleic acid
PCR	=	Polymerase chain reaction
RT-PCR	=	Real- Time polymerase chain reaction
STR	=	Short tandem repeat
ρg	=	Picogram

LIST OF TABLES

LIST OF FIGURES

ABSTRACT

Forensics identification using fingerprint analysis is only possible if there is a complete pattern of ridges obtained from the crime scene. In this study we have attempted to perform DNA analysis on processed smudged fingerprints. A subject was requested to deposit complete and portioning fingerprints on glass surface. The deposited fingerprints were exposed to 18 °C, 28 °C, 34 °C, and 40 °C for an hour, 3 days, and 7 days. The fingerprints were then visualized using black powder, were measured and one of sample was recovered using tape lifting method. A total of 2µl of the extracted DNA was quantitated using Real Time PCR (Quantifiler kit). 2 samples with DNA concentration above 1.0 ng was used for amplification using Amp FℓSTR Identifiler kit and another 2 samples concentration above 0.2 ng was used for amplification using AmpFℓSTR Minifiler kit. Both kits failed to amplify with DNA and therefore optimization is required for the collection and extraction methods used in this study.

1. INTRODUCTION

1.1 Forensic DNA analysis

Forensic DNA analysis began in 1985 with an introduction of DNA fingerprinting by an English scientist, Sir Alec Jeffreys (Jeffreys, *et al.*, 1985) and was submitted as evidence into court by 1986. Forensic DNA profiling became important in many fields such as anthropology and evolution (Cann, *et al.,* 1987; Vigilant, *et al.*, 1989), since DNA could be obtained from any source of biological material such as blood, semen, skin and saliva (Wiegand *et al.*, 1997).

The Association of Chief Police Officers and Crown Prosecution Service, UK, had initially suspended the use of forensic DNA analysis as evidence in court, but had quickly accepted after one true case of Omagh bombing in 1998, which has been solved by DNA key evidence. A year after, Forensic Science Service in UK has successfully applied DNA profiling to the high profile cases in United Kingdom (Graham, 2008). From there on, many criminal and civil cases were solved through this method and it becomes admissible as evidence in the court of law.

1.2 Touch DNA or low copy number (LCN) DNA

The term touch DNA is actually referred to low copy number (LCN) DNA (van Oorschot *et al.*, 1997). In the last few years the progress in DNA profiling methods together with the increased sensitivity of the DNA marker system, has led to the possibility of detecting very low level of DNA. Low copy number (LCN) DNA profiling method enable the successful investigation of sample which has simply been touched (Whitaker *et al.*, 2001; Gill, 2002; Rutty, 2002; Rutty *et al.*, 2003).

This term (touch DNA) was coined in publication by Dr Peter Gill of the Forensic Sciences Services in 2000 (Gill *et al.*, 2000). Touch DNA or low copy number (LCN) DNA profiling involved starting material (DNA) less than 100pg. Touch evidence such as latent fingerprints is often submitted to forensic laboratories for DNA analysis (Pang, *et al.*, 2007). Several studies have demonstrated the feasibility of using latent fingerprints for forensic DNA analysis (van Ooschot *et al.*, 1997; van Hoofstat *et a*l., 1999; van Rentengham *et al.*, 2000).

1.3 DNA from latent fingerprints

Fingerprint is an impression of the friction ridges of the fingers (Sewell *et al.*, 2008). Fingerprint evidence is one of the most positive investigative means of identifying people (Swanson, 2005). Every fingerprint is unique and formed on a person before birth and remains unchanged until the bodies decomposed after death.

In modern usage for forensic sciences the term latent fingerprints means any chance or accident impression left by the friction ridge skin on a surface, regardless of whether it is visible or invisible at the time of deposition (Swanson, 2005).

The visibility of latent prints depend on the physical condition of the person who left the print on the surface of the object (Sewell *et al.*, 2008) and on the angle of reflection of the light by which they are viewed (Swanson, 2005). The visibility of prints also depends on the time that has passed since they were placed, and the amount of heat to which they have been exposed (Balogh *et al.*, 2003). The amount of time they stay on an object is affected by atmospheric conditions, air currents, and humidity (Balogh *et al.*, 2003). But even when the object has been exposed to adverse conditions, it may be possible to obtain prints (Findlay, *et al.*, 1997; Hellmann, *et al.*, 2001; Hochmeister, *et al.*, 1998; Fridez and Coquoz, 1996).

Black powder, super glue technique, ninhydrin, and white powder are the processing techniques used to visualize the invisible latent fingerprint found at the crime scene. In most crime scenes, it is difficult to recover clear pattern of fingerprints. Commonly, only smudge and distorted pattern of fingerprints were collected from crime scene and submitted to Forensic laboratory for DNA analysis (Pang, *et al.*, 2007). Several studies have shown that it is possible to obtain and type DNA from single fingerprints (Van Oorschot *et al.*, 1997; Van Hoofstat *et al.*, 1999; Van Renterghem *et al.*, 2000; Zamir *et al.*, 2000).

It was found that, epithelial cells from latent fingerprints contained nuclei-free corneocytes, with minimal incidence of nucleated cells (Balogh, *et al.*, 20003). Enough nucleated cell could be found and hence there is a possibility that forensically typed DNA could be extracted from latent fingerprints.

Former studies have shown that even a single skin contact, documented by a latent fingerprint, can transfer enough DNA for genetic analysis (Van Renterghem, *et al.*, 2000). This result was confirmed by Van Oorschot and Jones, (1997), where single cell left on the surface can transfer enough DNA for successful STR typing. Therefore fingerprints are possible DNA source for forensic DNA investigation.

1.4 Polymerase Chain Reaction (PCR)

Most forensic cases, deal with very limited DNA sample and sometimes the sample are highly degraded. During 1986, Kerry Mullis, had overcome the entire problem in forensic field by introducing and establishing PCR technique. The function of PCR is to amplify the samples to certain quantity so that enables further analysis to be done.

Three steps which are important in PCR application include denaturing, annealing, and extension. In the first cycle, the target DNA is separated into two strands by heating at 94°C. In order to allow the primer to anneal, the temperature is then reduced before the melting temperature. The annealing temperature is depending on the primer lengths and sequences. After annealing, the temperature is increased to 72°C for optimal elongation. If PCR is 100% efficient, one target molecule would become amplified after n cycles.

With modern PCR based technology, advanced Real Time PCR was created to detect and quantify the specific sequence in a DNA sample (Nailis, *et al.*, 2006). The method of quantification is 5' nuclease assay, which is very exact, human specific and sensitive where it can detect low level DNA in sample.

TagMan ® probe consist of 18-22 base pair oligonucleotide probe which is labeled with a reporter fluorophore at the 5' end and a quencher fluorophore at the 3' end (Nolan, *et al.*,2006). A fluorogenic probe, complementary to the target sequence is added to the PCR reaction mixture. Till the time the probe is not hydrolyzed, the quencher and the fluorophore remain in proximity to each other, separated only by the length of the probe. This proximity however does not completely quench the fluorescent of the reporter dye and background fluorescence is observed.

During PCR, the probe anneals specifically between the forward and reverse primer to an internal region of the PCR product. The polymerase then carries out the extension of the primer and replicates the template to which the TaqMan® is bound. The 5' exonuclease activity of the polymerase cleaves the probe, releasing the reporter molecule away from the close vicinity of the quencher. The fluorescence intensity of the reporter dye, as a result increases. This process repeats in every cycle and does not interfere with the accumulation of PCR product.

The advantages of using this probe is providing specification to the detection of PCR product and cleaved probe provide a permanent record of amplification. Besides that by using this probe the fluorescent signal is directly proportional to the number of amplicon generated and it will produce low background of noise.

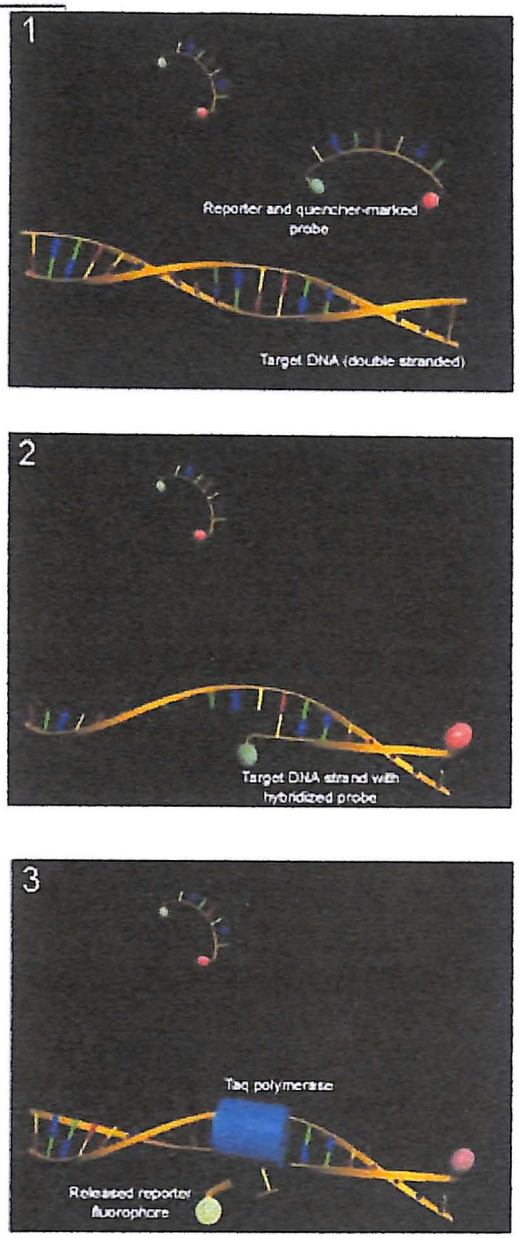

Figure 1: Basic concept of amplification and quantification by RT-PCR.

1) In intact probes, reporter fluorescence is quenched. (2) Probes and the complementary DNA strand are hybridized and reporter fluorescence is still quenched. (3) During PCR, the probe is degraded by the Taq polymerase and the fluorescent reporter released.

1.5 STR typing

Tandem repeated DNA sequences are widespread throughout the human genome and show sufficient variability among individuals in a population. They have become important in several fields including genetic mapping, linkage analysis, and human identity testing (Butler, 2005). These tandem repeated regions of DNA are typically classified into several groups depending on the size of the repeat region. Minisatellites (variable number of tandem repeats, VNTRs) have core repeats with 9-80 bp, while microsatellites (short tandem repeats, STRs) contain 2-5 bp repeats.

The forensic DNA community has moved primarily towards tetranucleotide repeats, which may be amplified using the polymerase chain reaction (PCR) with greater advantage than dinucleotide repeats (Carolyn, *et al.*, 2007). The variety of alleles present in a population is such that a high degree of discrimination among individuals in the population may be obtained when multiple STR loci are examined.

The AmpFℓSTR MiniFiler polymerase chain reaction amplification kit developed by Applied Biosystems enables size reduction on eight of the larger STR loci amplified in the Identifiler kit, which will aid recovery of information from highly degraded or inhibited DNA samples. The MiniFiler Kit amplifies CSF1PO, FGA, D2S1338, D7S820, D13S317, D16S539, D18S51, and D21S11 as well as the sex-typing locus amelogenin.

1.6 The importance of DNA typing from fingerprint

In United Kingdom, forensic scientist had been performing DNA typing from fingerprint for several years. Analyzing DNA from fingerprint proves valuable in investigation where there is a lack of body fluid, such as blood (Gill, 2001). This actually helpful when there was a forced entry and perpetrator may have touched the door handle. In the case of vehicle crime, a steering wheel usually will yield DNA from the driver. An exhibit that is often received for examination in robbery cases or terrorist activity is processed fingerprint. This type of exhibit can often, but not always, be successfully processed for fingerprint used for comparison.

While fingerprint requires an interpretable ridge pattern to be useful, DNA analysis does not. Balogh *et al.* (2003) emphasized that the main reason that taking a DNA swab from a fingerprint would be beneficial is if the fingerprint examiner had no sufficient ridge detail to make a fingerprint match. Another reason for taking a DNA swab, when fingerprint is smudged and has no real visible detail but still can develop chemically, and subsequently provide a DNA profile. When DNA profile was obtained from a fingerprint of no identification value, the probative value of the DNA information can be similar to that of fingerprint identification (Sewell *et al.*, 2008).

Touch DNA application is exclusively used in many country, as a last resort screening tools in the investigation stage of a criminal case to narrow down the universe of the suspect and/or eliminate the wrongfully accused. Ideally with time the technology to obtain touch DNA will become more discriminating and its use as an investigation tool will help bring justice in those cases where more traditional profiling technique have been exhausted.

2. LITERATURE REVIEW

Besides conventional biological material such as blood and sperm, epithelial cells from latent fingerprint are targeted in forensic science. Low copy number (LCN) DNA profiling method enable the successful investigation of samples which have been touched (Whitaker *et al.*, 2001; Gill, 2000; Rutty, 2002; Rutty *et al.,* 2003). The studies using latent fingerprints applied to surface of glasses (Renterghem, *et al.,* 2002), T-shirt left on crime scene (Schulz and Reichert, 2000), non porous surface (van Oorshot, and Jones, 1997), and paper (Balogh, *et al.*, 2003) reported the detection of STR profiles. In contrast to the relatively high number of epithelial cells from saliva or from excessively pressured fingerprint during strangulation (Wiegand, *et al.*, 1997), the experiment with latent fingerprint are expected to generate only a very small number of epithelial cells (Balogh *et al.*,2003).

Previous study had shown that, it is clearly possible to obtain DNA from fingerprints left by simple skin contact on object and to determine the genetic DNA profile (Renterghem *et al.*, 2002; Schulz *et al.*, 2000 and van Oorshot *et al.*, 19997). The different with this study is the method of collection and extraction applied to yield maximum number of DNA in epithelia cells from latent fingerprints.

One study had shown that, in order to achieve the best performance of extraction in term of sensitivity, QIAquick PCR Purification kit is suggested because DNA recovered from latent fingerprint is 38.4%, while using QIAamp DNA Mini kit (17.3%), combination between QIAquick PCR Purification kit and QIAamp DNA Mini kit (28.2%) and combination of QIAquick PCR Purification kit and QIAamp DNA Mini kit with Charge Switch Forensic DNA Purification kit yield 16% (Leemans, *et al.*, 2006). In contrast, the study done by others, show that Invisorb Forensic kit is more sensitive because DNA obtain is 80% compared with phenol/chloroform and Nucleo Spin C+ T kit from Machery-Nagel which only yield 10% of initial DNA from latent fingerprints (Balogh *et al.*, 2003).

Double swab technique was originally advocated by Sweet (1997), for recovery of saliva from skin. The use of the double swab technique improved the recovery of cells compared to the use of the classical stain recovery technique (Sweet, *et al.*, 1997). Furthermore, the double swab technique was also used to retrieve trace level of DNA in the study of the primary and secondary DNA transfer (Bright, and Petricevic, 2004).

The fact is supported by Leeman *et al.* (2006), whereby swab method is preferable as it resulted in a higher amount of DNA recovered (4.76ng) compare to cello tape (0.5ng) and gauze (1.56ng) technique. DNA recovered in some of the second dry swabs contained sufficient amount of DNA to yield a DNA profile.

It was shown that the double swab technique improved the quality of the resulting DNA profiles (Pang, *et al.,* 2007), so that the double swab technique for recovering touched evidence at crime scene is recommended.

The high sensitivity and effectiveness of the method may not only be a result of the improved extraction technique but can also be attributed to the low copy number (LCN) PCR technique applied. The sensitivity of the PCR has been increased by subsequently raising the number of cycles to 28, 32 and 38 (Balogh *et al.,* 2003).

The successful use of the LCN method with an increased number of PCR amplification cycle has been described by other authors. For example, analyzed fingerprints are effective with 28-40 cycles (Van Hoofstat, *et al.,* 1998). One study shows that PCR with 38 cycles proved to be the most effective in contrast to 32 and 28 cycles From his study (Balogh *et al.,* 2003), increasing the number of amplification cycles to 38 did not raise the incidence of artifact and stutters, so that the result of this experiment is compromised. The benefit of increased sensitivity derived from raising the number of amplification cycle has to be balanced against a reduction of profile quality (Barbaro *et al.,* 2000).