# A novel approach to latent fingermark detection using aptamer-based reagents

by

Michael Wood

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# Certificate of authorship and originality

I certify that the work in this thesis has not previously been submitted for a degree nor has it been submitted as part of the requirements for a degree except as fully acknowledged within the text.

I also certify that the thesis has been written by me. Any help that I have received in my research work and the preparation of the thesis itself has been acknowledged. In addition, I certify that all the information sources and literature used are indicated in the thesis.

Michael Wood

DATE

## **Acknowledgements**

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### **Abbreviations**

ACE-V Analysis, Comparison, Evaluation - Verification

ADP Adenosine diphosphate

AIDS Acquired immune deficiency syndrome

ALISA Aptamer-linked immobilised sorbent assay

AMD Age-related macular degeneration

AMP Adenosine monophosphate

AMPs Antimicrobial peptides

ASPV Apple stem pitting virus

ATP Adenosine triphosphate

AuNPs Gold nanoparticles

BSA Bovine serum albumin

CE Capillary electrophoresis

CEDIA Cloned enzyme donor immunoassay

CE-SELEX Capillary electrophoresis SELEX

DAB Diaminobenzidine

DCM Dichloromethane

DFO 1,8-diazafluoren-9-one

DMAC Dimethylaminocinnamaldehyde

DNA Deoxyribose nucleic acid

dsDNA Double-stranded DNA

ECL Electrochemiluminescence

EDDP 2-ethylidene-1,5-dimethyl-3,3-diphenylpyrrolidine

EDTA Ethylenediaminetetraacetic acid

ELISA Enzyme-linked immunosorbent assay
ELONA Enzyme-linked oligonucleotide assay

EMIT Enzyme-multiplied immunoassay technique

FAM 5(6)-Carboxyfluorescein

FBI Federal Bureau of Investigation

FRET Fluorescence resonance energy transfer

FTIR Fourier transform infrared

HCl Hydrochloric acid

HEX Hexachloro-6-carboxyfluorescein

HFEs Hydrofluoroethers

HIV Human immunodeficiency virus

HMDS Hexamethyldisilazane

HPLC High performance liquid chromatography

IND 1,2-Indanedione

IND-Zn 1,2-Indanedione-zinc

ISA Individual autoantibody profile

KCl Potassium Chloride

K<sub>d</sub> Dissociation constant

LIF Laser-induced fluorescence

Matrix Laser Assisted Desorption/Ionisation - Mass

MALDI-MS/P

Spectrometry/Profiling

MCAR Mixed cell agglutination reaction

MMD Multi-metal deposition

MW Molecular weight
NaCl Sodium chloride

Maci Souldin Chloride

NAD<sup>+</sup> Nicotinamide adenine dinucleotide

NBT Nitro blue tetrazolium

NCFS National Centre for Forensic Studies

NIR Near infrared

ORO Oil red O

PAGE Polyacrylamide gel electrophoresis

PCR Polymerase chain reaction

PD Physical developer

PEG Poly(ethylene glycol)

PSMA Prostate-specific membrane antigen

PVC Polyvinyl chloride

PVDF Polyvinylidene fluoride

QD's Quantum dots

RCMP Royal Canadian Mounted Police

RDT Rapid diagnostic tests

RNA Ribonucleic acid

RP Ruhemann's purple

RP-Cd Ruhemann's purple-cadmium

RP-Zn Ruhemann's purple-zinc

RT-PCR Reverse transcription PCR

RTX Ruthenium tetroxide

SDS Sodium dodecyl sulfate

SELEX Systematic evolution of ligands by exponential enrichment

siRNA Small interfering RNAs

SMD Single-metal deposition

SND Single-metal nanoparticle deposition

SPR Surface Plasmon Resonance

ssDNA Single-stranded deoxyribonucleic acid

TAR Trans-activation response

TBS Tris-buffered saline

THC  $\Delta^9$ -Tetrahydrocannabinol

THF Tetrahydrofuran

TiO<sub>2</sub> Titanium dioxide

TLC Thin layer chromatography

TMCS Trimethylchlorosilane

TNT Trinitrotoluene

TTBS Tween 20 and tris-buffered saline

UK United Kingdom

UTP Uridine triphosphate

UV Ultra Violet

VEGF Vascular endothelial growth factor

VMD Vacuum metal deposition

VSC Video spectral comparator

### **Abstract**

Research into latent fingermark detection and visualisation has taken many paths over the years as researchers and practitioners explore numerous methods to improve existing reagents. The majority of past research has resulted in providing small, incremental improvements to existing techniques. Currently, some researchers have opted to seek more transformational improvements in detection sensitivity, selectivity and visualisation. One such area being investigated is utilising immunology to target proteins, amino acids and drug metabolites in the latent fingermark deposit. Research to date has indicated that antibodies have great potential in providing these transformational improvements due to their ability to bind to certain fingermark components with high sensitivity and selectivity.

Following on from the antibody research, aptamers have been highlighted as the next potential immunogenic technique for several reasons, including reduced health and safety issues, lower cost, greater sensitivity and selectivity, and ease of design and versatility. Aptamers are specifically selected oligonucleotides comprised of either ribonucleic acid (RNA) or single-stranded deoxyribonucleic acid (ssDNA). Due to the selection strategies employed, aptamers can be designed to target most molecules and bind to them with detection limits in the sub-micromolar to nanomolar ranges. Although aptamers have been successfully used in a variety of highly sensitive and selective detection devices, they have not been investigated for use in the detection and visualisation of latent fingermarks prior to this project.

Initially, this project focussed on aptamers targeting amino acids as a means of visualising latent fingermarks. However, it was found that strong, non-specific interactions occurred with both the aptamer and the fluorescent tag, resulting in a lack of success with this approach.

In order to address these issues, aptamers selected to the protein lysozyme were used on fingermarks placed on both PVDF and plain white copier paper. Lysozyme was selected as it was found to be a component in human sweat, while aptamers selected to lysozyme, with binding affinities in the nanomolar range were available. It was found that the aptamer-based reagents possessed high levels of sensitivity with the clear detection of lysozyme at very low concentrations (1 ng). Latent fingermarks from various donors were able to be detected on both substrates, with primary and secondary level detail being clearly visible. Results,

however, were very inconsistent, with marks older than a couple of days being difficult to detect. This was found to be due to the degradation of lysozyme in the latent fingermark. Unfortunately, aptamers to other, possibly more suitable, fingermark components that would circumvent this problem were not available for this project. Despite the difficulties encountered, this project has, for the first time, demonstrated the potential of detecting and visualising latent fingermarks with an aptamer-based reagent. The study has laid the groundwork for future successful investigations that exploit the benefits of aptamers while overcoming the limitations identified in this project.