

**School of Molecular and Life Sciences**

**Sustainable Latent Fingerprint Detection Protocols  
for Remote-Location and Resource-Limited Jurisdictions**

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**This thesis is presented for the Degree of  
Doctor of Philosophy  
of  
Curtin University**

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## **Declaration**

To the best of my knowledge and belief, this thesis contains no material previously published by any other person except where due acknowledgement has been made.

This thesis contains no material which has been accepted for any other degree or diploma in any university.

## **Human Ethics**

The research presented and reported in this thesis was conducted in accordance with the National Health and Medical Research Council National Statement on Ethical Conduct in Human Research (2007) – updated March 2018. The proposed study received human research ethics approval from the Curtin University Human Research Ethics Committee (EC00262), Approval Number HRE2020-0162.

Signature: .....

Date: 18/10/2023

## **Acknowledgement of Country**

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We acknowledge that Curtin University works across hundreds of traditional lands and custodial groups in Australia, and with First Nations people around the globe. We wish to pay our deepest respects to their ancestors and members of their communities, past, present, and to their emerging leaders. Our passion and commitment to work with all Australians and peoples from across the world, including our First Nations peoples are at the core of the work we do, reflective of our institutions' values and commitment to our role as leaders in the Reconciliation space in Australia.

## Abstract

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Over time, the growth of forensic science has become a worldwide practice that aims to promote justice, peace, and prosperity. In particular, latent fingerprint detection plays an important role in resource-limited Global South jurisdictions where advanced techniques like DNA profiling are not readily available. Nonetheless, most research and development in fingerprint detection has been carried out in the Global North, leading to a lack of methods specifically tailored to the challenges of the Global South. This thesis introduces the concept of frugal forensics, which presents the first systematic framework for sustainable forensic science provision, with a focus on latent fingerprint detection in resource-limited jurisdictions. Implementing sustainable methods is key to addressing the gap in forensic science services prevalent in the Global South and serves as a catalyst for international efforts to reduce inequalities between regions, in line with the United Nations Sustainable Development Goals.

The practical implementation of frugal forensics was demonstrated through its application to three different latent fingerprint techniques. As a result, two sustainable formulations tailored for the Seychelles Police Force (SPF) were developed, namely SPF 1,2-indanedione zinc (IND/Zn) and Wet SPF powder suspension. The assessments were conducted following the International Fingerprint Research Group's guidelines, which provide a framework for evaluating new or revised fingerprint detection methods. It also ensures compliance with quality assurance requirements and supports the implementation of detection techniques in operational laboratories.

A cost-effective and sustainable version of the IND/Zn formulation was developed by modifying existing formulations used by police in Australia National Centre for Forensic Studies (NCFS), Germany Bundeskriminalamt (BKA), and the UK Home Office Centre for Applied Science and Technology (CAST). The main challenge was to find a cost-effective and supply chain-friendly alternative to the expensive solvent HFE-

7100 used in the IND/Zn formulation. Based on a frugal analysis, the BKA petroleum ether-based formulation was identified as the most cost-effective and low-risk supply chain option. Solstice® Performance Fluid (PF) as an alternative carrier solvent was also evaluated. Although it was found to have potential as a substitute, some aspects of Solstice PF fall short as a carrier solvent for a sustainable reagent. Spectroscopy studies revealed that reducing excess 1,2-indanedione in the formulation enhances reagent effectiveness. The SPF formulation was modified to contain 594 mg/L of 1,2-indanedione, reducing the cost of the formulation by 40%. This formulation was assessed through laboratory and incidental fingerprint trials across five substrates, two ageing periods of one and 4 weeks, and a range of ten donors producing 1200 fingerprint halves and 100 incidental items. The findings demonstrated comparable performance between SPF and NCFS formulations. Both formulations exhibited high sensitivity and effectively developed fingerprints that were 1 week and 4 weeks old.

The recently published Wet UCIO (Unitat Central d'Inspeccions Oculars) carbon-based powder suspension was modified by replacing the surfactant solution with an in-house sodium dodecyl sulfate (SDS) surfactant solution to develop the Wet SPF powder suspension. This formulation containing 15% SDS in a 5% ethanol/water mixture was compared to the commercial Wetwop™ black powder suspension on eight different tapes with two aging periods using ten donors. The Wet SPF performed similarly to the commercial Wetwop™ with slightly better sensitivity. Using an inexpensive and easily accessible SDS salt allows for better transportation, storage, and quality control compared to commercial surfactant solutions.

Acid Nile blue as an emerging lipid-sensitive technique with promising frugal forensic attributes was also investigated. Like Oil red O, acid Nile blue was found to successfully develop charged fingerprints on wet porous substrates, with its effectiveness decreasing as the fingerprint ages. It was also observed that Nile blue targets the 'fragile fraction' of the sebaceous component and should be applied to fingerprints less than 4 weeks old. While acid Nile blue has limited sensitivity, its affordability, water-based nature, and low toxicity make it a useful tool for training and outreach programs. In addition, acid Nile blue was compared with Basic Yellow 40 (BY40) as a post-cyanoacrylate stain. The study revealed that acid Nile blue was

an effective method for staining preferentially lipid-rich fingerprints, unlike BY40, which produced no preferential development. The result suggests that acid Nile blue has a different staining mechanism than BY40, penetrating through the polymer structure to interact with the fingerprint residue. These findings can aid in developing improved post-cyanoacrylate stains by providing insight into the post-cyanoacrylate staining mechanism.

Finally, a comprehensive and objective assessment tool was developed to evaluate the sustainability of methods in the frugal forensics framework. Drawing from the fingerprint grading scale, this tool objectively evaluates all six attributes of frugal forensics, promoting transparent method selection and adhering to quality assurance requirements. The tool was successfully applied to evaluate the sustainability and implementation recommendations of the three latent fingerprint detection methods. Moreover, it can also serve as a valuable planning tool by identifying areas for improvement.

## Acknowledgements

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## Contribution to Others

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The following have contributed to the thesis, with contributions being listed in the form of CRediT (Contributor Roles Taxonomy) statements for each chapter – see <https://www.elsevier.com/authors/policies-and-guidelines/credit-author-statement>

### **Chapters 1 – 8:**

Professor Simon W. Lewis (Principal Supervisor): Project Administration, Supervision, Conceptualisation, Writing – Review & Editing.

Dr Georgina Sauzier: Supervision, Conceptualisation, Writing – review & Editing.

### **Chapter 1:**

Portions of this chapter have been published in *Forensic Science International: Synergy and Forensic Chemistry* see Appendix E for details.

Aaron J. Horrocks, Stephen M. Bleay: Conceptualization, Investigation, Writing – review & editing.

Thais Lópes, Anna L. Heavey, Jessie Parrish, Georgina Sauzier, and Simon W. Lewis: Conceptualization, Investigation, Writing – original draft, Writing – review & editing.

### **Chapter 3:**

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Aaron J. Horrocks, Amanda A. Frick: Conceptualization, Investigation, Writing – review & editing.

Georgina Sauzier, Stephen M. Bleay, Simon W. Lewis: Conceptualization, Writing – review & editing.

### **Chapter 4:**

Portions of this chapter have been published in *Forensic Science International* see Appendix E for details.

Aaron J. Horrocks, Jason Merendino, Stephen M. Bleay: Conceptualization, Investigation, Writing – review & editing.

Georgina Sauzier, Simon W. Lewis: Conceptualization, Supervision,  
Writing – review & editing.

**Chapter 7:**

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**Chapter 8:**

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Thais Lópes, Anna L. Heavey, Jessie Parrish, Georgina Sauzier, and Simon W. Lewis: Conceptualization, Investigation, Writing – original draft, Writing – review & editing.

## Publications

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This dissertation contains work which has been submitted for publication in the following peer-reviewed journals:

**Jemmy T. Bouzin**, Jason Merendino, Stephen M. Bleay, Georgina Sauzier, and Simon W. Lewis. New light on old fingermarks: The detection of historic latent fingermarks on old paper documents using 1,2-indanedione/zinc. *Forensic Science International: Reports* **2020** 2 100145.

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**Jemmy T. Bouzin**, Georgina Sauzier, and Simon W. Lewis. Forensic science in Seychelles: An example of a micro-jurisdiction forensic delivery system. *Forensic Science International: Synergy* **2021** 3 100139.

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**Jemmy T. Bouzin**, Aaron J. Horrocks, Georgina Sauzier, Stephen M. Bleay, and Simon W. Lewis. Comparison of three active 1,2-indanedione-zinc formulations for fingerprint detection in the context of limited resources and supply chain risks in Seychelles. *Forensic Chemistry* **2022** 30 100439.

DOI: <https://doi.org/10.1016/j.forc.2022.100439>

**Jemmy T. Bouzin**, Amanda A. Frick, Georgina Sauzier, and Simon W. Lewis. Preliminary evaluation of Solstice® PF as a replacement carrier solvent for Australian fingerprint detection. *Forensic Science International* **2022** 340 111465.

DOI: <https://doi.org/10.1016/j.forsciint.2022.111465>

**Jemmy T. Bouzin**, Thais Lópes, Anna L. Heavey, Jessie Parrish, Georgina Sauzier, and Simon W. Lewis. Mind the gap: The challenges of sustainable forensic science service provision. *Forensic Science International Synergy* **2023** 6 100318.

DOI: <https://doi.org/10.1016/j.fsisyn.2023.100318>

In addition, the following peer reviewed article was published during the course of this dissertation:

Kenny J. Travouillon, Christine Cooper, **Jemmy T. Bouzin**, Linette S. Umbrello, and Simon W. Lewis. All-a-glow: spectral characteristics confirm widespread fluorescence for mammals. *Royal Society Open Science* 2023 10 (10) 230325.

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## Conference Presentations

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Selected aspects of the work contained within this thesis have been presented, or have been accepted for presentation, at the following conferences:

### Oral Presentations

*Preliminary evaluation of Solstice® PF as a replacement carrier solvent for Australian fingerprint detection*, International Association of Forensic Sciences - 23rd Triennial Meeting, Sydney, September 2023 (Amanda A. Frick).

*Frugal Forensics*, International Fingerprint Research Group, Delft, Amsterdam, June 2023 (Simon W. Lewis).

*Forensics in Paradise*, Science Teachers Association of Western Australia - Future Science Conference, Perth, December 2022.

*1,2-indanedione reagent: Investigation into operational use in a small, remote jurisdiction*, Australian New Zealand Forensic Science Society - 25th International Symposium, Brisbane, September 2022.

*New light on old fingerprints: The detection of historic latent fingerprints on old paper documents using 1,2-indanedione/zinc*, Virtual 10th Annual Forensic Science Symposium - hosted by Florida International University, (online), June 2021.

*New light on old fingerprints: The detection of historic latent fingerprints on old paper documents using 1,2-indanedione/zinc*, Curtin University School of Molecular and Life Sciences Symposium, Perth, December 2020.

### ➤ Invited Presentations

*Panellist for Round Table 1 - Challenges for the future*, Forensic Document Examiners Live International Knowledge Exchange on Documents meeting, (online), March 2023.

*Forensic science in Seychelles: A micro-jurisdiction forensic delivery system*, PathWest - Forensic Biology Seminar Series, December 2021.

*Improving latent fingermarks protocols*, Inaugural Western Australian Forensic Science Forum, Perth, October 2021.

*Forensic science in Seychelles: A micro-jurisdiction forensic delivery system*, Guest Lecture to First Year Forensic Students, Curtin University, Perth 2020-23.

*A unique perspective of a small-Island-State forensic delivery system*, Australian New Zealand Forensic Science Society, Western Australia Branch meeting, Perth, August 2019.

### **Poster Presentations**

*Developing sustainable formulations enhances global access to latent fingerprint protocols*, International Association of Forensic Sciences - 23rd Triennial Meeting, Sydney, September 2023 (Simon W. Lewis).

- 1<sup>st</sup> Place Poster Prize - Fingerprint Evidence & Biometrics

*Detection of historic fingermarks on paper documents using 1,2-indanedione/zinc*, Australian New Zealand Forensic Science Society - 25th International Symposium, Brisbane, September 2022.

*Forensic Science in Seychelles: A unique perspective of a small-island-state forensic delivery system*, Australian New Zealand Forensic Science Society - 25th International Symposium, Brisbane, September 2022.

*New light on old fingermarks: The detection of historic latent fingermarks on old paper documents using 1,2-indanedione/zinc*, Royal Society of Chemistry Twitter Conference, (online), March 2021.

- 2<sup>nd</sup> Place Poster Prize

*Luminescence detection of latent fingerprints on paper surfaces after incidental handling*, Royal Society of Chemistry Twitter Conference, (online), March 2020

*Investigation into improved protocols for latent fingerprint detection technique for jurisdictions with limited resources and tropical climates*, Curtin University School of Molecular and Life Sciences Symposium, Perth, December 2019.

**Jemmy T Bouzin**, presenting author of the above-listed orals and posters, except were stated otherwise.

# Table of Contents

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<i>Declaration</i> .....	<i>i</i>
<i>Human Ethics</i> .....	<i>i</i>
<b>Acknowledgement of Country</b> .....	<b>ii</b>
<b>Abstract</b> .....	<b>iii</b>
<b>Acknowledgments</b> .....	Error! Bookmark not defined.
<b>Contribution to Others</b> .....	<b>viii</b>
<b>Publications</b> .....	<b>x</b>
<b>Conference Presentations</b> .....	<b>xii</b>
<b>Table of Contents</b> .....	<b>xv</b>
<b>List of Figures</b> .....	Error! Bookmark not defined.
<b>List of Tables</b> .....	<b>xxii</b>
<b>List of Abbreviations</b> .....	<b>xxiv</b>
<b>Chapter 1. Introduction</b> .....	<b>1</b>
1.1 <i>Global importance of fingerprint detection</i> .....	2
1.2 <i>Fingerprint composition</i> .....	5
1.2.1 Eccrine components .....	6
1.2.2 Sebaceous components.....	7
1.2.3 Other contaminants .....	8
1.3 <i>Factors affecting fingerprint composition post-deposition</i> .....	9
1.3.1 Effect of time .....	10
1.3.2 Substrates.....	12
1.3.3 Environmental conditions .....	13
1.4 <i>Fingerprint detection methods</i> .....	14
1.4.1 Porous-surface fingerprint detection methods .....	17
1.4.1.1 Amino acid sensitive reagents .....	17
1.4.1.2 Ninhydrin and DFO .....	17



1.4.1.3	1,2-indanedione .....	18
1.4.1.4	Lipid-sensitive reagents .....	19
1.4.1.5	Physical developer (PD).....	20
1.4.1.6	Oil red O (ORO) .....	20
1.4.1.7	Nile red.....	22
1.4.1.8	Nile blue .....	22
1.4.2	Non-porous-surface fingerprint detection methods .....	24
1.4.2.1	Fingerprint powder .....	24
1.4.2.2	Powder suspension .....	25
1.4.2.3	Cyanoacrylate .....	27
1.4.2.4	Post-cyanoacrylate stains .....	29
1.5	<i>Latent fingerprint detection service provision</i> .....	30
1.5.1	Seychelles latent fingerprint detection service provision.....	30
1.5.2	Challenges in operationalising latent fingerprint detection protocols .....	31
1.5.3	Sustainable forensic science provision; frugal forensics .....	33
1.6	<i>Aim and Objectives</i> .....	37
<b>Chapter 2.</b>	<b>Sample collection, experimental methods, and instrumentation .....</b>	<b>40</b>
2.1	<i>Introduction</i> .....	41
2.2	<i>Experimental considerations</i> .....	41
2.2.1	IFRG guidelines .....	41
2.2.2	Frugal forensics approach .....	42
2.2.3	COVID-19 pandemic impact .....	42
2.3	<i>Fingerprint methodology</i> .....	42
2.3.1	Donor information.....	42
2.3.2	Fingerprint collection procedures.....	43
2.3.3	Fingerprint ageing and storage conditions .....	45
2.4	<i>Substrates selection and environmental considerations</i> .....	45
2.4.1	Substrates surface selection.....	45
2.4.2	Environmental considerations.....	45
2.5	<i>Fingerprint development</i> .....	46
2.5.1	1,2-indanedione zinc .....	46
2.5.2	Powder suspension .....	48
2.5.3	Oil red O .....	49
2.5.4	Cyanoacrylate fuming.....	49
2.5.5	Acid Nile Blue .....	50

2.5.6	Basic Yellow 40 .....	51
2.6	<i>Instrumentation</i> .....	51
2.6.1	Photography of developed fingermarks .....	51
2.6.2	Fluorescence spectroscopy .....	52
2.6.3	UV-vis spectroscopy .....	54
2.6.4	ATR-FTIR spectroscopy .....	55
2.6.5	Confocal Raman microscopy .....	55
2.7	<i>Fingermark grading</i> .....	56
2.8	<i>Statistical analysis</i> .....	57
2.9	<i>Human ethics approval</i> .....	59
<b>Chapter 3.</b>	<b>A sustainable 1,2-indanedione zinc formulation.....</b>	<b>60</b>
3.1	<i>Introduction</i> .....	61
3.2	<i>Experimental</i> .....	70
3.2.1	Preliminary consideration .....	70
3.2.2	Experimental methods .....	70
3.2.2.1	Substrate.....	70
3.2.2.2	Fingermark collection .....	70
3.2.2.3	Fingermark development.....	71
3.2.2.4	Photography and grading of developed fingermarks .....	71
3.2.3	Head-to-head comparative assessment of existing IND/Zn formulations .....	71
3.2.3.1	Investigation into co-solvents.....	71
3.2.3.2	Investigation into 1,2-indanedione concentration .....	72
3.2.4	Investigation into Carrier solvent – Solstice® PF as an alternative.....	73
3.2.4.1	Overall performance of Solstice® PF.....	73
3.2.4.2	Compatibility with inks .....	73
3.2.4.3	Effect of co-solvents on ink diffusion.....	75
3.2.4.4	Reagent stability .....	75
3.2.5	Statistical analysis.....	75
3.3	<i>Results and discussion</i> .....	76
3.3.1	Head-to-head comparative assessment of existing IND/Zn formulations .....	76
3.3.1.1	Overall performance and sensitivity.....	76
3.3.1.2	Substrates .....	77
3.3.1.3	Statistical Analysis.....	80
3.3.1.4	Frugal forensics analysis of existing operational IND/Zn protocols .....	80
3.3.2	Carrier solvent – Petroleum ether.....	81

3.3.2.1	Co-solvents .....	81
3.3.2.2	1,2-indanedione concentration .....	83
3.3.3	Carrier solvent - Solstice® PF as an alternative .....	87
3.3.3.1	Overall performance of Solstice® PF.....	87
3.3.3.2	Compatibility with inks .....	90
3.3.3.3	Effect of co-solvents on ink diffusion.....	92
3.3.3.4	Reagent stability .....	94
3.3.3.5	Frugal forensics analysis of Solstice® PF as a carrier solvent for fingerprint development.....	95
3.4	<i>Conclusions</i> .....	96
<b>Chapter 4.</b>	<b>SPF 1,2-indanedione zinc comparative assessment .....</b>	<b>99</b>
4.1	<i>Introduction</i> .....	100
4.2	<i>Experimental</i> .....	102
4.2.1	Experimental consideration .....	102
4.2.2	Experimental methods .....	103
4.2.2.1	Substrate.....	103
4.2.2.2	Fingerprint collection .....	103
4.2.2.3	Fingerprint development.....	103
4.2.2.4	Photography and grading of developed fingerprints .....	103
4.2.3	Preliminary investigations .....	104
4.2.3.1	Inks compatibility.....	104
4.2.3.2	Processing conditions .....	104
4.2.3.3	Simple comparative assessment.....	104
4.2.4	Laboratory trials .....	105
4.2.4.1	Comparative assessment .....	105
4.2.4.2	Incidental fingerprints .....	105
4.2.4.3	Environmental considerations .....	106
4.2.4.4	Reagent stability .....	106
4.2.4.5	UV-visible spectroscopy .....	106
4.2.5	Statistical analysis.....	107
4.3	<i>Results and discussion</i> .....	108
4.3.1	Preliminary investigations .....	108
4.3.1.1	Inks compatibility.....	108
4.3.1.2	Ambient temperature processing conditions .....	109
4.3.1.3	Simple comparative assessment.....	109
4.3.2	Laboratory trials .....	110

4.3.2.1	Comparative assessment .....	110
4.3.2.2	Incidental fingermarks .....	115
4.3.2.3	Environmental considerations .....	117
4.3.2.4	Reagent stability .....	119
4.3.2.5	UV-vis spectroscopy .....	120
4.3.3	Statistical Analysis .....	121
4.4	<i>Conclusions</i> .....	122
<b>Chapter 5.</b>	<b>Modification of WET UCIO powder suspension for fingerprint detection on pressure sensitive adhesive tape .....</b>	<b>123</b>
5.1	<i>Introduction</i> .....	124
5.2	<i>Experimental</i> .....	130
5.2.1	Experimental methods .....	130
5.2.1.1	Powder suspension preparation .....	130
5.2.1.2	Substrates .....	130
5.2.1.3	ATR-FTIR Tape Analysis .....	131
5.2.1.4	Fingerprint collection, photography, and grading .....	131
5.2.2	Stage A – Formulation modification .....	133
5.2.3	Stage B – Supplementary studies .....	133
5.2.4	Stage C - Comparative assessment to Wetwop™ .....	134
5.2.5	Statistical analysis .....	134
5.3	<i>Results and discussion</i> .....	135
5.3.1	Tape Characterisation .....	135
5.3.1.1	PSA tape backing .....	135
5.3.1.2	PSA tape adhesive .....	139
5.3.2	Formulation modification .....	143
5.3.3	Supplementary studies .....	147
5.3.4	Comparative assessment to Wetwop™ .....	150
5.3.4.1	Overall performance .....	150
5.3.4.2	Sensitivity .....	154
5.3.5	Statistical analysis .....	155
5.4	<i>Conclusion</i> .....	155
<b>Chapter 6.</b>	<b>Investigation into acid Nile blue as an emerging lipid-sensitive and post-cyanoacrylate detection method .....</b>	<b>157</b>
6.1	<i>Introduction</i> .....	158

6.2	<i>Experimental</i> .....	161
6.2.1	Experimental methods .....	161
6.2.1.1	Reagent preparation and sample development .....	161
6.2.1.2	Substrates .....	161
6.2.1.3	Fingermark collection .....	162
6.2.1.4	Sample visualisation and evaluation.....	162
6.2.2	As a lipid-sensitive reagent on wet porous substrates.....	162
6.2.2.1	Preliminary Study.....	162
6.2.2.2	Comparison to Oil red O .....	163
6.2.3	As a post-cyanoacrylate stain.....	163
6.2.3.1	Comparison to Basic Yellow 40.....	163
6.2.3.2	Confocal Raman microscopy investigation into post-cyanoacrylate staining mechanism.....	163
6.3	<i>Results and discussion</i> .....	164
6.3.1	As a lipid-sensitive reagent on wet porous substrates.....	164
6.3.1.1	Preliminary investigations.....	164
6.3.1.2	Comparison to Oil red O .....	165
6.3.2	Suitability as a post-cyanoacrylate stain and exploring the staining mechanism. ....	169
6.3.2.1	Preliminary investigation .....	169
6.3.2.2	Comparison to Basic Yellow 40.....	171
6.3.2.3	Sequencing to enhance detection of overlapping fingermarks. ....	173
6.3.2.4	Confocal Raman microscopy investigation into post-cyanoacrylate staining mechanism.....	176
6.4	<i>Conclusion</i> .....	180
<b>Chapter 7.</b>	<b>Frugal forensics sustainability assessment tool .....</b>	<b>181</b>
7.1	<i>Introduction</i> .....	182
7.2	<i>Experimental</i> .....	184
7.3	<i>Results and Discussion</i> .....	184
7.4	<i>Conclusion</i> .....	188
<b>Chapter 8.</b>	<b>Conclusion and future work .....</b>	<b>190</b>
8.1	<i>Conclusion</i> .....	191
8.2	<i>Alternative carrier solvent for amino acid sensitive reagent</i> .....	192
8.3	<i>Powder suspension and acid Nile blue improvements</i> .....	193

8.4	<i>IFRG guidelines requirements</i>	194
8.5	<i>Sustainable forensic science service provision</i>	194
<b>Chapter 9.</b>	<b>References</b>	<b>196</b>
<b>Appendices</b>		<b>227</b>
	<i>Appendix A-1: Statistical analysis test assumptions.</i>	228
	<i>Appendix A-2: Statistical analysis - Chapter 3 R codes and results.</i>	230
	<i>Appendix A-3: Statistical analysis - Chapter 4 R codes and results.</i>	233
	<i>Appendix A-4: Statistical analysis - Chapter 5 R codes and results.</i>	237
	<i>Appendix B-1: Air-condition office datalogger temperature and relative humidity records.</i>	242
	<i>Appendix B-2: Non-air-condition Level 5 space datalogger temperature and relative humidity records.</i>	243
	<i>Appendix C-1: 3M Announcement letter (PFAS Discontinuation).</i>	244
	<i>Appendix D-1: Human ethics participant information form (English).</i>	245
	<i>Appendix D-2: Human ethics consent form (English).</i>	247
	<i>Appendix D-3: Human ethics participant information form (Creole).</i>	248
	<i>Appendix D-4: Human ethics consent form (Creole).</i>	250
	<i>Appendix E-1: Contribution statement - New light on old fingerprints paper.</i>	251
	<i>Appendix E-2: Contribution statement – Forensic science in Seychelles paper.</i>	252
	<i>Appendix E-3: Contribution statement – IND/Zn Comparative assessment paper.</i>	253
	<i>Appendix E-4: Contribution statement – Solstice® PF paper.</i>	254
	<i>Appendix E-5: Contribution statement – Mind the gap paper.</i>	255

## List of Tables

---

<b>Table 1.1:</b> The United Nations Sustainable Development Goals. <sup>9</sup> .....	4
<b>Table 1.2:</b> Summary of the main constituents of eccrine and sebaceous skin secretions. <sup>25, 28, 41</sup> .....	6
<b>Table 2.1</b> Summary of some of the chemical detection capabilities of the Seychelles Police. ....	46
<b>Table 2.2:</b> Photographic conditions optimised to each detection method.....	52
<b>Table 2.2:</b> Fingerprint grading scale and grading classification used in this study. <sup>19, 219</sup> .....	56
<b>Table 2.3:</b> UC (University of Canberra) comparative scale to assess the relative performance of two detection methods for split fingerprints.....	57
<b>Table 3.1:</b> Mass in grams and molar ratio of active material present in one litre of NCFS, BKA and CAST IND/Zn working solution. ....	62
<b>Table 3.2:</b> Comparison of physical properties of carrier solvents used in amino acid sensitive fingerprint detection reagent. <sup>238, 240, 242, 243</sup> .....	66
<b>Table 3.3:</b> Approximate cost of one litre of 1,2-indanedione zinc working solution in USD. Chemical sourced from two regular Seychelles Police suppliers based in the UK and USA. ....	69
<b>Table 3.4:</b> Details of substrates used in the head-to-head comparison of NCFS, BKA and CAST IND-Zn formulations.....	70
<b>Table 3.5:</b> Pen used in ink diffusion investigations.....	74
<b>Table 4.1:</b> Details of substrates used in the SPF IND/Zn formulation laboratory trials. ....	103
<b>Table 5.1:</b> Composition and approximate c.m.c of in-house surfactant solution used in this study. The c.m.c of SDS was taken to be 8.2 mM. <sup>297</sup> .....	130
<b>Table 5.2:</b> Details of tapes used in this study. ....	130
<b>Table 5.3:</b> Details of the number of donors, tapes and powder suspension formulations used in each experiment. ....	132
<b>Table 5.4</b> Classification of adhesive and backing composition based on IR analysis of the main polymer type.....	135

<b>Table 5.5:</b> PSA tapes characteristic IR peak assignment for PSA tapes backing and adhesive. <sup>302-311</sup> .....	136
<b>Table 5.6:</b> Percentage of fingermarks graded 3&4 on each of the eight types of adhesive tape. ....	152
<b>Table 6.1:</b> Detail of substrates used in the comparative assessment to ORO. ....	161
<b>Table 6.2:</b> Details of substrates used in the post-cyanoacrylate study. ....	161
<b>Table 7.1:</b> Frugal forensics sustainability comparative score assessment tool.....	183
<b>Table 7.2:</b> Frugal forensics method recommendation score assessment tool.....	183
<b>Table 7.3:</b> Comparative score and recommendation results for SPF IND/Zn with CAST formulation, Wet SPF powder suspension with Wetwop™ formulation and post-cyanoacrylate acid Nile blue method with BY40. ....	185



## List of Figures

---

<b>Figure 1.1:</b> Schematic illustrating the influence of the “triangle of interaction” on the initial and aged composition.....	9
<b>Figure 1.2:</b> An example of a flow chart procedure for the development of latent fingerprints. <sup>16, 17</sup> .....	16
<b>Figure 1.3:</b> The reaction of 1,2-indanedione with amino acids to form Joulles’s pink. ....	18
<b>Figure 1.4:</b> Chemical structure of Oil red O. ....	21
<b>Figure 1.5:</b> Hydrolysis of Nile blue to Nile red in aqueous solution. ....	23
<b>Figure 1.6:</b> Schematic illustration of powder suspension development. Fingerprint constituents disrupt surfactant micelles exposing suspended powder particulates, causing discriminate deposition on fingerprint and not the substrate.....	26
<b>Figure 1.7:</b> Polymerisation reaction of ethyl cyanoacrylate.....	27
<b>Figure 1.8:</b> The geographical location of Seychelles.....	31
<b>Figure 1.9:</b> Illustrating the gap in operationalisation of latent fingerprint detection technique in remote and small jurisdictions.....	32
<b>Figure 1.10:</b> Number of publications on “forensic science” in a simple Bibliometric search on Scopus for the (a) Total papers 1972–2022 (b)Top 10 countries and (c) Top 10.....	34
<b>Figure 1.11:</b> The concept of sustainable forensic science provision (a) Definition (b) The three principles and six attributes that underpin it.....	35
<b>Figure 1.12:</b> Illustrating the relationship between frugal forensics and UN sustainable goals through SDG 16.....	36
<b>Figure 2.1:</b> Schematic illustrating split fingerprint and depletion series methodology for fingerprint collection and the reverse of development process A and B to minimise donor biasing on one side of the finger. ....	44
<b>Figure 2.2:</b> Schematic of photographic setup for photoluminescence fingerprints examination. ....	52

<b>Figure 2.3:</b> Experimental setup using the fibre optic probe for fluorescence measurements of amino acid spots treated with IND/Zn formulation containing different 1,2-indanedione concentrations.....	54
<b>Figure 2.4:</b> Illustrating transformation of fingerprint grades frequency results (left) into vectors (right), the basic data structure in R code to compute statistical analysis. ....	58
<b>Figure 3.1:</b> Percentage concentration v/v of carrier solvents and co-solvents in NCFS, BKA and CAST IND/Zn working solution. ....	62
<b>Figure 3.2:</b> The reaction mechanism of IND/Zn with amino acid and zinc ions as a Lewis acid catalyst. <sup>104</sup> .....	63
<b>Figure 3.3:</b> The molecular structure of HFE-7100 (C <sub>4</sub> F <sub>9</sub> OCH <sub>3</sub> ) consisting of two isomers CF <sub>3</sub> CF <sub>2</sub> CF <sub>2</sub> CF <sub>2</sub> OCH <sub>3</sub> and (CF <sub>3</sub> ) <sub>2</sub> CF <sub>2</sub> OCH <sub>3</sub> . ....	65
<b>Figure 3.4:</b> The molecular structure of Solstice <sup>®</sup> PF (trans-1-chloro-3,3,3-trifluoropropene). ....	66
<b>Figure 3.5:</b> Percentage of fingerprints graded (3&4), (1&2), and (0) out of 450 fingerprints developed in head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation.....	76
<b>Figure 3.6:</b> Comparison of percentage fingerprint grades assigned to 1 <sup>st</sup> , 5 <sup>th</sup> and 10 <sup>th</sup> depletion series in the head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation. ....	77
<b>Figure 3.7:</b> Comparison of percentage fingerprint grades assigned to fingerprints developed on five substrates in the head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation. ....	78
<b>Figure 3.8:</b> Fingerprints treated with (a) NCFS formulation, (b) BKA, (c) CAST on notepad paper showing difference in luminescence and background development. ....	79
<b>Figure 3.9:</b> Fingerprints on copy paper treated with NCFS formulation (left) and the more polar modified NCFS formulation (right) showing the marginally greater background development. ....	82
<b>Figure 3.10:</b> Fingerprints on copy paper treated with modified CAST formulation containing ethanol (left) and CAST formulation containing methanol (right).....	82

<b>Figure 3.11:</b> Difference in luminescence of fingermarks on copy paper with modified BKA formulation containing 227 mg/L of 1,2-indanedione (left) and BKA formulation containing 990 mg/L (right). .....	84
<b>Figure 3.12:</b> Amino acid spots on copy paper treated with BKA IND/Zn formulations containing different 1,2-indanedione concentrations, (a) emission spectrum excited at 505 nm illustrating the difference in luminescence intensity (b) difference in colour under visible light, (c) difference in colour under Polilight at 505 nm. ....	86
<b>Figure 3.13:</b> Amino acid spots on cardboard treated with BKA IND/Zn formulations containing different 1,2-indanedione concentrations, (a) emission spectrum excited at 505 nm illustrating the difference in luminescence intensity (b) difference in colour under visible light, (c) difference in colour under Polilight at 505 nm. ....	86
<b>Figure 3.14:</b> Latent fingermarks on copy paper treated with HFE-IND/Zn (left) and Sol-IND/Zn (right), (a) viewed under white light and (b) in luminescence mode.....	87
<b>Figure 3.15:</b> Comparison of percentage fingermark grades assigned to fingermarks treated with HFE-IND/Zn and Sol-IND/Zn on white copy paper, white envelope and cardboard.....	88
<b>Figure 3.16:</b> Fingermark halves developed on (a) white envelope treated with HFE-IND/Zn (left) and Sol-IND/Zn (right) and (b) cardboard treated with Sol-IND/Zn (left) and HFE-IND/Zn (right).....	88
<b>Figure 3.17:</b> Comparison of percentage fingermark grades assigned to a) fingermarks aged 1, 7 and 30 days b) fingermarks aged for each donor developed on white copy paper and treated with HFE-IND/Zn and Sol-IND/Zn.....	90
<b>Figure 3.18:</b> Comparison of the effect of Sol-IND/Zn and HFE-IND/Zn reagents on inks, photographed under ambient light and luminescent mode. ....	91
<b>Figure 3.19:</b> Inks spotted with neat solvent and photographed under ambient light and luminescence mode. ....	93
<b>Figure 3.20:</b> Sol-IND/Zn after 2.5 months in refrigerated storage, showing white precipitate. ....	94
<b>Figure 4.1:</b> SPF 1,2-indanedione working solution less than 1 day old (left) and more than 6 months old (right) showing difference in colour. ....	107
<b>Figure 4.2:</b> Comparison of the effect of SPF IND/Zn and NCFS IND/Zn reagents on inks, photographed under ambient light and luminescent mode. ....	108

**Figure 4.3:** Comparison of fingermarks treated with SPF IND/Zn and processed with heat press at 160 °C (left) and ambient temperature of 25 °C (right) on copy paper. ....109

**Figure 4.4:** Comparison of quartered fingermarks on blue notepad paper, developed with SPF IND/Zn (top left), NCFS IND/Zn (top right), and BKA IND/ZN (bottom left) and CAST IND/Zn (bottom right). ....110

**Figure 4.5:** Percentage of fingermarks graded (3&4), (1&2), and (0) out of 1200 fingermarks halves developed with SPF and NCFS IND/Zn formulations on five different paper substrates. ....110

**Figure 4.6:** Comparison of percentage fingermarks grades across five paper substrates, developed with SPF and NCFS IND/Zn formulation. ....112

**Figure 4.7:** Percentage of fingermarks graded (3&4), (1&2), and (0) developed for 1 week and 4 weeks samples, treated with SPF and NCFS IND/Zn formulation. ....113

**Figure 4.8:** Examples of fingermark developed with NCFS IND/Zn (left half) and SPF IND/Zn (right half) on (a) COS Premium white copy paper, (b) OfficeMax recycled white copy paper, (c) JBurrows blue notepad, (d) PPS cardboard, (e) The West Australian newspaper. ....114

**Figure 4.9:** Comparison of total number of useful incidental fingermarks detected with SPF and NCFS IND/Zn treatment on five different substrates. ....115

**Figure 4.10:** Examples of incidental fingermarks developed with NCFS IND/Zn (left) and SPF IND/Zn (right half) on (a) white plain envelope, (b) JBurrows blue notepad. Fingermarks were deposited by the same donor on each type of substrate. ....116

**Figure 4.11:** Comparison of fingermarks on white copy paper stored under non-air-conditioned and air-conditioned environments and treated with SPF IND/Zn. ....118

**Figure 4.12:** Comparison of fingermarks on cardboard stored under non-air-conditioned and air-conditioned environments and treated with SPF IND/Zn. ....118

**Figure 4.13:** Comparison of fingermark ridge details on recycled copy paper stored under an air-conditioned environment (left), non-air-conditioned environment (right) and treated with SPF IND/Zn. ....119

**Figure 4.14:** Percentage of useful fingermarks detected on white copy paper and treated with SPF IND/Zn with increased reagent age. ....120

<b>Figure 4.15:</b> Absorption spectra for SPF IND/Zn working solutions of less than 7 days old, 6, 12 and 18 months since preparation, showing the decrease in 1,2-indanedione peak absorbance. ....	121
<b>Figure 5.1:</b> The molecular structure of materials used in the adhesive of PSA tapes, a) acrylate monomer, b) natural rubber polyisoprene, c) synthetic rubber poly-cis-1,4-butadiene, d) synthetic rubber polystyrene-butadiene. ....	125
<b>Figure 5.2:</b> The molecular structure of materials used in the backing of PSA tapes, a) polypropylene, b) polyethylene, c) polyvinyl chloride.....	126
<b>Figure 5.3:</b> The molecular structure of diisononyl phthalate, a common plasticiser used in adhesive tape to impart flexibility. <sup>285</sup> .....	126
<b>Figure 5.4:</b> The products and cost of Wet UCIO and two commercial powder suspension formulations Wet Powder black Wetwop™ black. ....	128
<b>Figure 5.5:</b> The molecular structure of sodium dodecyl sulfate (SDS). ....	128
<b>Figure 5.6:</b> Schematic of latent fingerprint deposition on tape samples.....	132
<b>Figure 5.7:</b> ATR-FTIR spectra of polypropylene backing of packaging tape #1-3. Spectra have been offset for clarity.....	137
<b>Figure 5.8:</b> ATR-FTIR spectra of (a) polyethylene backing of duct tape #4 and (b) cellulose acetate backing of masking tape #5. Spectra have been offset for clarity. ....	138
<b>Figure 5.9:</b> ATR-FTIR spectra of polyvinylchloride backing of insulating tapes #6-#12. Spectra have been offset for clarity.....	138
<b>Figure 5.10:</b> ATR-FTIR spectra of acrylic adhesive packaging tape #1 and #2.....	139
<b>Figure 5.11:</b> ATR-FTIR spectra of synthetic rubber adhesive for packaging tapes #2, duct tape #4, masking tape #5 and insulating tape #6-#12. Spectra have been offset for clarity. ....	140
<b>Figure 5.12:</b> Overlay of the fingerprint region of both backing and adhesive IR spectrum of rubber adhesive tapes showing significant contribution of the backing in the adhesive spectrum of tape#6 to #12.....	141
<b>Figure 5.13:</b> Diisononyl phthalate IR spectrum <sup>312</sup> offset over rubber adhesive tape's backing and adhesive spectrum of tape #9. Showing the contribution of diisononyl phthalate in the backing and adhesive tape spectra. ....	142

**Figure 5.14:** Comparison of infrared spectra of different insulating tape synthetic rubber adhesive. The main differences are highlighted (blue shaded regions). .....143

**Figure 5.15:** Sodium dodecyl sulfate solutions (a) insoluble solution of 27 % w/v in deionised, (b) clear solution of 15 % w/v in 5 % ethanol deionised water mixture. ....144

**Figure 5.16:** Comparison of (a) the number and grades of fingermarks developed (b) the degree of powder deposition on fingermarks and background after treatment with modified WET UCIO powder suspensions prepared with 1 %, 5 %, 10 % and 27 % w/v SDS surfactant solution. ....145

**Figure 5.17:** Comparable performance and sensitivity of Wet SPF formulation (left) and 27 % SDS-modified WET UCIO formulation (right) in developing fingermarks on rubber-based adhesive tape. ....146

**Figure 5.18:** Comparison of (a) percentage of fingermarks graded (3&4), (1&2), and (0) out of 150 fingermarks (b) the degree of powder deposition on fingermarks and background after treatment with WET UCIO and Wet SPF powder suspensions on packaging tape, duct tape and insulating tape. ....147

**Figure 5.19:** Comparison of fingermarks ridge details and contrast developed on four different tapes after treatment with Wet SPF powder suspensions prepared with different additives; 5 % ethanol, 5 % PEG-400, and 10 % PEG-400 v/v.....148

**Figure 5.20:** Fingermark ridge details and contrast after treatment with Wet SPF, WET UCIO and Wet Powder on (a) Click white insulating tape #10 under white light, (b) Click black insulating tape #9 under 505 nm excitation and 550 nm filter. ....149

**Figure 5.21:** Percentage of fingermarks graded (3&4), (1&2), and (0) out of 320 fingermarks developed with Wet UCIO and Wet SPF on eight different types of tape. ....150

**Figure 5.22:** Percentage of fingermarks graded (3 &4), (1&2), and (0) out of 320 fingermarks developed with Wet UCIO and Wet SPF on eight different types of tape aged for 1 week and 4 weeks.....152

**Figure 5.23:** Examples of fingermark developed with Wet SPF (left) and Wetwop™ (right) on eight pressure sensitives tapes #2 Scotch 3M clear packaging tape, #3 Scotch 3M brown packaging tape, #4 Paint Partner silver duct tape, #5 PPS white

masking tape, #6 Nitto yellow insulating tape, #10 Click white insulating tape, #11 Delta yellow insulating tape, #12 Nitto white insulating tape.....	153
<b>Figure 5.24:</b> Percentage of fingerprints grades out of 300 fingerprint halves developed with Wet UCIO and Wet SPF on three different types of tape grouped into depletion series 1-5 and 6-10. ....	154
<b>Figure 6.1:</b> Reaction pathway proposed for the spontaneous hydrolysis of Nile Blue to Nile red in aqueous solution. <sup>146</sup> .....	158
<b>Figure 6.2:</b> The molecular structure of Basic Yellow 40 (CAS Number 29556-33-0) recommended dye for fingerprint enhancement. ....	160
<b>Figure 6.3:</b> Comparison of fingerprints developed by filtered acid Nile blue solution (left half) and unfiltered acid Nile blue solution (right half): (a) under white light with no observable yellow stains and (b) at 450 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.....	164
<b>Figure 6.4:</b> Percentage of fingerprints graded 3&4 out of 72 fingerprint halves developed with ORO or acid Nile blue under wet and dry conditions. ....	165
<b>Figure 6.5:</b> Comparison of fingerprints aged 1 day, 10 days and 30 days developed by ORO (left half) under white light and acid Nile blue (right half) at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.....	167
<b>Figure 6.6:</b> Number and grades of fingerprints developed by ORO and acid Nile blue on A4 copy paper, recycled A4 copy paper, kraft brown paper and newspaper after 1 week of submersion in tap water. ....	168
<b>Figure 6.7:</b> Comparison of fingerprints on different substrates submerged in tap water for 1 week and developed by Oil red O (left half) under white light and acid Nile blue (right half) at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter: (a) white copy paper (b) 50 % recycled copy paper (c) brown kraft paper and (d) newspaper. ....	169
<b>Figure 6.8:</b> (a) Improvised cyanoacrylate fuming chamber (b) quality of cyanoacrylate fingerprints developed after 2 hours of fuming on borosilicate microscope slides. ....	170
<b>Figure 6.9:</b> Comparison of cyanoacrylate fumed fingerprints stained by acid Nile blue for ten minutes (left half) and twenty minutes (right half), at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.....	170

<b>Figure 6.10:</b> Percentage of fingermarks grades developed out of 30 fingermark halves with acid Nile blue and Basic Yellow 40 on borosilicate glass, aluminium sheet, and polypropylene plastic lid. ....	171
<b>Figure 6.11:</b> Comparison of fingermarks developed with acid Nile blue (left half) and Basic Yellow 40 (right half) illustrating donor effect: (a) on borosilicate glass for donor 1 (b) on aluminium sheet for donor 1 (c) borosilicate glass for donor 3 (d) aluminium sheet for donor 3 (e) on polypropylene superimposed of donor 1 (top) and donor 3 (bottom). ....	172
<b>Figure 6.12:</b> Overlapping fingermark collection (a) natural over charged fingermarks (b) charged over natural fingermarks. ....	174
<b>Figure 6.13:</b> Overlapped fingermarks developed with cyanoacrylate and stained using the sequence BY40 → acid Nile blue, (top) image of results at different development stages and (bottom) assessment result using UC scale. ....	175
<b>Figure 6.14:</b> Overlapped fingermarks developed with cyanoacrylate and stained using the sequence acid Nile blue → BY40, (top) image of results at different development stages and (bottom) assessment result using UC scale. ....	175
<b>Figure 6.15:</b> Raman spectra of acid Nile blue and Basic Yellow 40 liquid smear on glass slide, acquired using WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser. ....	177
<b>Figure 6.16:</b> Raman spectra of unstained cyanoacrylate fingermark (red), cyanoacrylate fingermark stained with acid Nile blue (blue) and cyanoacrylate fingermark stained with Basic Yellow 40 (yellow) on a glass slide, acquired using WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser. ....	177
<b>Figure 6.17:</b> Visual image of the cross-section of post-cyanoacrylate stain fingermark on glass slide. The fingermark and polymer structure not apparent. The glass side was mounted edge-up. ....	178
<b>Figure 6.18:</b> Visual and Raman image overlay of the cross-section of post-cyanoacrylate stain fingermark on glass slide showing fluorescent and non-fluorescent regions. ....	179



**Figure 6.19:** Overlay of Raman spectra at two opposite ends of a post-cyanoacrylate fingerprint stained with acid Nile blue on a glass slide, acquired using a WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser. ....179

**Figure 7.1:** Comparison of frugal forensics attributes scores for (a) the SPF IND/Zn formulation with CAST as control method and (b) acid Nile blue as a post-cyanoacrylate stain with BY40 as control. All control methods were given a base score of zero. ....187

## List of Abbreviations

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ATR	Attenuated total reflection
AUD	Australian Dollars
AWRE	Atomic Weapons Research Establishment
BKA	Bundeskriminalamt
BY40	Basic Yellow 40
ca.	Circa
cmc	Critical micelle concentration
CAST	Centre for Applied Science and Technology
DFO	1,8-diazafluoren-9-one
DNA	Deoxyribonucleic acid
EU	European Union
FTIR	Fourier transform infrared
FVM	Fingermark Visualisation Manual
GC-MS	Gas-chromatography mass spectrometry
GDP	Gross Domestic Product
GWP	Global Warming Potential
HDPE	High-density polyethylene
HFE-7100	Methyl nonafluorobutyl ether
HFEs	Hydrofluoroethers

HSP	Hansen solubility parameters
IFRG	International Fingerprint Research Group
IND/Zn	1,2-indanedione zinc
IR	Infrared
JP	Joullie's pink
LDPE	Low-density polyethylene
LLDPE	Linear low-density polyethylene
MALD-MSI	Matrix-assisted laser desorption/ionisation mass spectrometry imaging
NCFS	National Centre for Forensic Studies
NHMRC	National Health and Medical Research Council
NSW	New South Wales
ORO	Oil red O
PAACSS	Performance, Accessibility, Availability, Cost, Simplicity, Safety
PD	Physical developer
PEG	Polyethylene glycol
PFAS	Per- and polyfluoroalkyl substances
PSA	Pressure sensitive adhesive
PVC	Polyvinyl chloride
RH	Relative humidity
SDGs	Sustainable Development Goals

SDS	Sodium dodecyl sulfate
SPF	Seychelles Police Force
UC	University of Canberra
UK	United Kingdom
USA	United States of America
USD	United States dollars
UV	Ultraviolet
WA	Western Australia



## Chapter 1. Introduction

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## 1.1 Global importance of fingerprint detection

Forensic science has become a global practice,<sup>1, 2</sup> serving as a means to promote justice, peace and prosperity,<sup>3-7</sup> aligning with the United Nations Sustainable Development Goals (SDGs)<sup>8</sup> - especially SDG16 (Table 1.1). These 17 goals, universally adopted by United Nations Member States in 2015<sup>9</sup> aim to protect the planet and reduce disparities between the Global North and South. The terms 'Global North' and 'Global South' are not geographic labels but relate to socio-economic development and global influence.<sup>10</sup> The Global North is usually equated with technically developed and well-resourced countries, whereas the Global South often denotes politically or culturally marginalised regions with comparatively limited income. In nations of the Global South, where resources are limited and sophisticated techniques such as DNA profiling are not readily available, latent fingerprint detection serves as a fundamental service provision<sup>11</sup> that plays an instrumental role in advancing the global practice of forensic science.

For over a century,<sup>12</sup> fingerprints have been a crucial forensic tool in criminal investigations, utilised across various jurisdictions,<sup>13</sup> regardless of their size or technological advancement. Despite their widespread and longstanding use, the detection of fingerprints remains a challenging task, particularly for developing regions in the Global South. This is attributed to the inherent fingerprint variability coupled with the Global North hegemony in the research and development of fingerprint enhancement methods.

A fingerprint is a mirror image of the ridge pattern of the finger, formed by the deposition of residue from the fingertip when in contact with a surface.<sup>14, 15</sup> Due to their highly polymorphic nature, fingerprints can be used to identify and link people, places, and things involved in criminal activity. Fingerprints found at scenes are often latent (invisible) to the naked eye, requiring chemical or physical treatment to visualise. The detection process involves exploiting the fingerprint and substrate's physical and chemical properties to produce appropriate contrast.<sup>16-18</sup> However, the detection effectiveness can be affected by the fingerprint composition, the nature of the substrate and environmental conditions. This is because fingerprint composition

is highly variable,<sup>19-23</sup> and substrates and conditions where they may be found are diverse. Consequently, various detection techniques have been developed through research, but no global optimum exists.<sup>24, 25</sup> This is underpinned by guidelines of the International Fingerprint Research Group (IFRG), which recommends evaluation of new or modified enhancement reagents using locally sourced test substrates and visualisation equipment.<sup>24</sup>

Against this backdrop, it is important to note that research to continuously develop more effective procedures for improving forensic services has primarily been carried out in countries with ample resources. This is evident from the dominance of the Global North in fingerprint detection research output,<sup>26, 27</sup> leading to a lack of techniques that have been developed and validated for jurisdictions in the Global South. Particularly, those that address the challenges such as supply chain, budget-restrictions, and limited resources. Therefore, there is a need to develop alternative fingerprint detection capabilities adapted to the vulnerabilities of the Global South so that they can be implemented effectively. This applies to fingerprint detection provision and other forensic science services alike.

This thesis presents a framework for sustainable forensic science provision, to address the gap in fingerprint service provision outlined above. To introduce the concept, this chapter begins with an overview of the chemistry of latent fingerprints and the factors that affect its composition, followed by the various fingerprint detection techniques available, and the challenges in implementation in resource-limited jurisdictions. Finally, the framework and principles of sustainable forensic science is discussed, followed by the aims and objectives. This project centres on sustainable latent fingerprint detection methods, focusing on the challenges faced by Seychelles, a small and remote jurisdiction with limited resources.



**Table 1.1: The United Nations Sustainable Development Goals.<sup>9</sup>**

<b>Goal Number</b>	<b>Goal Name</b>	<b>Goal Description</b>
SDG 1	No Poverty	End poverty in all its forms everywhere
SDG 2	Zero Hunger	End hunger, achieve food security and improved nutrition and promote sustainable agriculture
SDG 3	Good Health and Well-being	Ensure healthy lives and promote well-being for all at all ages
SDG 4	Quality Education	Ensure inclusive and equitable quality education and promote lifelong learning opportunities for all
SDG 5	Gender Equality	Achieve gender equality and empower all women and girls
SDG 6	Clean Water and Sanitation	Ensure availability and sustainable management of water and sanitation for all
SDG 7	Affordable and Clean Energy	Ensure access to affordable, reliable, sustainable and modern energy for all
SDG 8	Decent Work and Economic Growth	Promote sustained, inclusive and sustainable economic growth, full and productive employment and decent work for all
SDG 9	Industry, Innovation and Infrastructure	Build resilient infrastructure, promote inclusive and sustainable industrialisation and foster innovation
SDG 10	Reduced Inequalities	Reduce inequality within and among countries
SDG 11	Sustainable Cities and Communities	Make cities and human settlements inclusive, safe, resilient and sustainable
SDG 12	Responsible Consumption and Production	Ensure sustainable consumption and production patterns
SDG 13	Climate Action	Take urgent action to combat climate change and its impacts
SDG 14	Life Below Water	Conserve and sustainably use the oceans, seas and marine resources for sustainable development
SDG 15	Life on Land	Protect, restore and promote sustainable use of terrestrial ecosystems, sustainably manage forests, combat desertification, and halt and reverse land degradation and halt biodiversity loss
SDG 16	Peace, Justice and Strong Institutions	Promote peaceful and inclusive societies for sustainable development, provide access to justice for all and build effective, accountable and inclusive institutions at all levels
SDG 17	Partnerships for the Goals	Strengthen the means of implementation and revitalise the global partnership for sustainable development

## 1.2 Fingerprint composition

The residue left behind by a fingerprint is a complex mixture of natural secretions, contaminants, and substances present on the skin of the finger.<sup>14, 28, 29</sup> These secretions originate from three types of sweat glands: eccrine, apocrine, and sebaceous.<sup>30, 31</sup> Sweat glands are essential for regulating electrolytes and body temperature, and removal of metabolic products.<sup>31-34</sup> In the context of latent fingerprint detection, the composition originating from sweat glands is broadly classified as either eccrine or sebaceous. The eccrine secretion comprises inorganic salts and water-soluble organic compounds, while the sebaceous secretion consists of lipids and fats (summarised in Table 1.2).<sup>28, 29, 35, 36</sup> Due to the hydrophilic nature of eccrine and the hydrophobic nature of sebaceous secretions, they are respectively referred to as the water-soluble deposit and water-insoluble deposit of latent fingerprint residue.<sup>16, 17</sup>

The composition of the residue varies from person to person based on donor traits such as biological sex, age, ethnicity, psychological state, health, medication, and diet.<sup>15, 32, 37, 38</sup> Personal habits and activities, such as smoking or use of hair and cosmetic products, may introduce endogenous and exogenous contaminants.<sup>35, 36</sup> Additionally, the composition of the initial fingerprint residue deposited on a surface is influenced by deposition conditions, such as contact pressure, angle, and time duration, as well as the characteristics of the substrate.<sup>16, 36</sup> These factors can cause high variability in the composition of fingerprint residue.

Knowledge of the composition and influencing factors of fingerprints is key to understanding its effect on detection performance and sampling protocol in a research context. For example, fingerprint test methodology primarily uses 'natural' fingerprints collected from donors conducting their normal daily routine instead of sebaceous 'charged' fingerprints, where donors wipe their fingers across the face or forehead prior to the deposition of marks.<sup>19, 24</sup> Studies show that charged samples are lipid-rich,<sup>39, 40</sup> potentially biasing results since it may be an unrealistic representation of marks encountered in operational work. Hence, the use of charged

fingermarks is limited to the early phase trial, particularly when establishing interactions between the fingermark and the technique.<sup>19, 24</sup>

**Table 1.2:** Summary of the main constituents of eccrine and sebaceous skin secretions.<sup>25, 28, 41</sup>

Secretions	Organic constituents	Inorganic constituents
Eccrine	Amino acids	Water (>70%)
	Proteins	Chloride
	Urea	Metal ions (Na <sup>+</sup> , K <sup>+</sup> , Ca <sup>2+</sup> )
	Uric acid	Sulphate
	Lactic acid	Phosphate
	Sugars	Bicarbonate
	Creatinine	Ammonia
	Choline	
Sebaceous	Glycerides	
	Fatty acids	
	Wax esters	
	Squalene	
	Sterol	
	Sterol esters	

### 1.2.1 Eccrine components

The primary function of eccrine glands is to dissipate body heat through sweat and excrete excess water. Eccrine glands are the only type of secretory glands present on the palms and fingertips, making them a major contributor to the composition of fingermark residue. Eccrine residue primarily consists of amino acids, lactic acid, proteins, urea, uric acid, sugars, and inorganic salts.<sup>35, 36</sup>

Amino acids have been studied as sweat and fingermark constituents, as they are a primary target in fingermark detection on porous surfaces that have not been wet.<sup>31, 42-44</sup> So far, up to 20 amino acids have been quantified, with serine being the most abundant, followed by glycine and alanine.<sup>39, 45</sup> Croxton *et al.*<sup>39</sup> reported the total amino acid content to range from 20.7 ng to 345.1 ng per fingermark and did not find significant difference between charged and natural fingermarks. A recent study by Helmond *et al.*,<sup>23</sup> profiling the chemical composition of fingermarks from 463 donors using mass spectrometry, reported large inter-variability in the amino acid profile and a content range from 100 ng to 10 µg. The amount and profile of amino acids depend on several factors, including sex, age, diet, physical exercise, and general health.<sup>28, 35, 36</sup>

Inorganic compounds including salts and trace amounts of metals such as sodium, potassium, and calcium have been detected in eccrine residue.<sup>28</sup> Sodium chloride and potassium chloride salts are the most abundant. Studies suggest that the amount of chloride ions in fingermarks decreases as the donor's age increases.<sup>36, 46, 47</sup> Other than chloride ions targeted by silver nitrate, most of the relatively less reactive inorganic compounds are not subject of fingermark treatment.<sup>28</sup>

The average water content of eccrine sweat has long been reported in the literature to be 98 % by weight.<sup>31, 34, 48</sup> This has been perpetuated in a similar reported amount in fingermark residue.<sup>49</sup> According to Kent's<sup>50</sup> theoretical argument, the suggested amount is unrealistic as it fails to consider the presence of sebaceous secretions, which are primarily composed of lipids. Kent instead suggested the water content would be close to 20 % or less. Keisar *et al.*<sup>41</sup> verified this claim by utilising quartz crystal microbalance and temperature-programmed desorption-mass spectrometry to measure the complete water loss through evaporation. Their findings revealed a varying water content of 20-70 %.

### **1.2.2 Sebaceous components**

Sebaceous glands are found in varying abundance all over the body except on the palms of the hands and soles of the feet.<sup>31</sup> Sebum, a secretory product of these glands, forms a major component of fingermark composition due to contact of the fingertips with other parts of the body (e.g., hair and face).<sup>51</sup> Sebum and compounds from the epidermis contribute to the lipid composition of fingermark residue consisting of triglycerides, fatty acids, wax esters, cholesterol, and squalene.<sup>15, 39</sup>

The lipid composition has been studied both as skin surface and fingermark residue, and a good correlation between the two compositions has been found.<sup>36, 39, 52-54</sup> Free fatty acids, which form 15-25 % of sebum, are major lipid compounds identified in fingermark residue with palmitic acid identified as the most abundant.<sup>53</sup> The fatty acids found in sebum are quite diverse, from volatile short-carbon chain fatty acids (<C10) partly responsible for body odour,<sup>55</sup> to more stable long-carbon chain fatty acids (C30) targeted in the treatment of older fingermarks. Wax esters produced

exclusively by the sebaceous glands make up 20-25 % of the fingermark's residue, primarily comprising fatty acids and fatty alcohols. Squalene, a cholesterol precursor produced by all tissues, accounts for 10-15 % of fingermark residue and is usually not detected in older fingermarks due to rapid decomposition over time.<sup>52, 56</sup> Sterol and sterol esters originate mainly from the epidermis and form 1-5 %, with cholesterol and cholesterol esters being the most abundant. Triglycerides are estimated to make up 30-40 % of fingermark residue and are the least studied due to most analytical methods involving hydrolysis which break them down into glycerol and fatty acids.<sup>35</sup>

Whilst many lipid compounds are common to fingermark residue, the relative composition varies between individuals, based on donor traits and deposition characteristics, including the amount of contact of the finger with other body parts. Several studies have highlighted the inherent variability in the lipid compositions of fingermark residue.<sup>18, 35, 36</sup> Frick *et al.*<sup>57</sup> demonstrated the high intra-donor variability in the lipid components of fingermark residue of 116 donors using gas chromatography - mass spectrometry (GC-MS). Croxton *et al.*<sup>39</sup> reported significant quantitative differences in amounts of fatty acids and squalene between natural and charged fingermarks from the same donor. These studies illustrate fingermarks' inherent variability and the importance of appropriate sampling protocol in a research context.

### **1.2.3 Other contaminants**

In addition to secretory glands and epidermis, fingermark composition is also affected by contaminants.<sup>58-62</sup> There are two primary sources of contaminants: contact of the fingertip with exogenous substances and excreted metabolites on the skin surface.

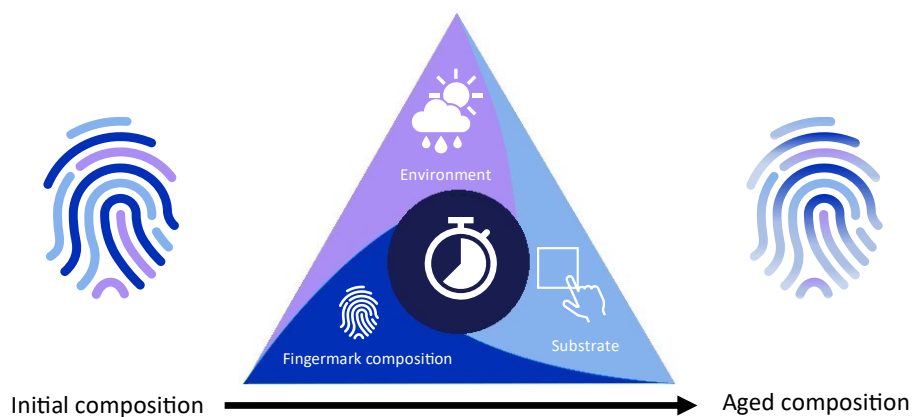
Everyday activities can transfer materials onto fingertips, mixing with the fingermark residue. Fingermark residues have been found to contain cosmetics and other consumer products like hair products, sunscreen, hand sanitisers and lotions.<sup>59, 60, 62-</sup>

<sup>64</sup> Compounds with additional forensic value, such as gunshot residue, illicit drugs and explosives, have also been identified.<sup>61, 65</sup> Other studies have successfully

identified metabolites in sweat and fingerprint residue related to smoking habits or drug consumption, which can provide additional donor information.<sup>66, 67</sup> The presence of these contaminants can affect the chemistry of the residue, potentially interfering with detection processes and affecting degradation rates. For example, handling oil-based substances such as food, lotion, and hair products can increase the lipid fraction in a fingerprint residue,<sup>63</sup> impacting the ageing and detection of latent fingerprints.<sup>68</sup>

### 1.3 Factors affecting fingerprint composition post-deposition

Following deposition, the initial composition of the latent fingerprint residue undergoes further changes, the nature of which depends on the interactions with the substrate and environmental conditions with time. This process is illustrated by the triangle of interaction<sup>19</sup> in Figure 1.1, which depicts the relationship between the three elements over time. The resulting aged composition of the fingerprint residue is a composite of the triangle of interaction from deposition to the detection time, and impacting the ability of a fingerprint to persist, and ultimately its detection.<sup>35</sup> Therefore, understanding how aged fingerprints evolve is crucial for the successful recovery of latent fingerprints. Several studies<sup>52, 69-72</sup> have explored the evolution of fingerprint composition over time to develop effective detection techniques, while others<sup>73-77</sup> have investigated ageing markers to develop methods for determining fingerprint age.



**Figure 1.1:** Schematic illustrating the influence of the “triangle of interaction” on the initial and aged composition.

### 1.3.1 Effect of time

Fingerprint residue undergoes ageing between deposition and detection through various chemical, physical, and biological processes. These processes include degradation, drying, evaporation, metabolism, migration, oxidation, and polymerisation.<sup>35, 36, 78, 79</sup> These changes are influenced by factors such as the substrate deposited on and the environmental conditions,<sup>35, 73</sup> ultimately affecting the mechanism and rate of the fingerprint ageing process.

Over time, the mass of fingerprint residue decreases mainly through evaporation of volatile components.<sup>21, 72</sup> The loss of moisture through this process has been shown to cause physical changes in the thickness of the fingerprints due to cumulation of residual organic and inorganic components.<sup>71, 80</sup> While some studies have investigated the inorganic salt compounds, such as the concentration of chloride ions over time,<sup>47</sup> most of the focus has been on the organic compounds, which are the most targeted by fingerprint detection techniques.

Amino acids have been suggested to be stable over time, particularly on paper substrates due to hydrogen bonding with cellulose.<sup>81</sup> This has been practically demonstrated with successful detection of old fingerprints using amino acid sensitive reagents on 32 and 80 year old documents.<sup>82, 83</sup> However, it has been generally reported that with increasing age of fingerprints, there is a significant decrease in the quality, indicating that amino acids binding with cellulose are not completely stable.<sup>69</sup> The progressive diffusion over time of the amino acids in the paper matrix has been suggested as a primary mechanism for the decrease in quality of developed fingerprints.<sup>35, 83</sup>

Lipids undergo degradation and decomposition over time, with unsaturated compounds such as triglycerides and squalene found to be less stable compared to saturated lipids such as wax esters, which are less affected.<sup>52, 72, 76, 84</sup> Pleik *et al.*<sup>78, 85</sup> suggested that ozonolysis is a major lipid degradation pathway in fingerprint residues in ambient air, primarily through oxidation of unsaturated triglycerides to form ozonides (1,2,4-trioxolanes), which are proposed as a possible aged marker. More

recently, Frick *et al.*<sup>86</sup> demonstrated that diunsaturated triglycerides in fingerprints undergo progressive oxidation to mono- and dihydroperoxides of fingerprints when exposed to ambient atmospheric conditions, using untargeted ultra performance liquid chromatography-ion mobility spectrometry-quadrupole time-of-flight mass spectrometry and five donors. The study found that the number of unsaturated triglycerides decreased in samples left for 7 and 28 days, while mono- and dihydroperoxides of these lipids were identified as the main components, indicating that ozonolysis is a major pathway for fingerprint degradation over time.

The physicochemical changes of latent fingerprint residue over time have been explored in relation to fingerprint age determination. Various indicators such as fingerprint quality developed with powdering techniques,<sup>74, 87</sup> changes in the ridge width thickness,<sup>71, 75</sup> variations in lipids<sup>52, 88</sup> and amino acid concentration<sup>77, 89</sup> have been studied. However, no reliable method for determining fingerprint age has been established, primarily due to the intrinsically variable nature of fingerprint composition. In a recent study, Boseley *et al.*<sup>21</sup> investigated the changes within fingerprint residue of 12 donors from deposition for the first 7-13 hours under ambient conditions using synchrotron-sourced attenuated total reflection-Fourier transform infrared (ATR-FTIR) microspectroscopy. The study revealed high variation in the initial composition and morphology of fingerprints, with substantial loss of water (14-20 µg) within the first 8 hours and relatively higher variation in lipid concentration due to degradation and redistribution.<sup>21</sup> The study highlighted the unpredictability of fingerprint age estimation, particularly when the chemistry of the initial composition is unknown.

In essence, the detection of latent fingerprints in operational cases is generally carried out days, months, or even years after deposition, hence the detectability of fingerprints depends on the stability over time of the target compounds and sensitivity of the development technique. Between deposition and processing, fingerprints undergo ageing through various processes; however, these mechanisms and the rate of the ageing processes are also influenced by the nature of the substrate and environmental conditions as discussed below.



### 1.3.2 Substrates

The nature of the substrate plays a crucial role in influencing the fingerprint deposit and in determining the appropriate enhancement method.<sup>90, 91</sup> For purposes of latent fingerprint detection, substrates are grouped into porous, non-porous and semi-porous. The porosity dictates the capacity of the substrate to absorb fingerprints and, consequently, their persistence.

Porous substrates like paper or cardboard tend to absorb fingerprint deposits to varying degrees, with different fractions of the residue penetrating to different depths.<sup>91</sup> Eccrine materials, which include amino acids, are more easily absorbed and retained due to hydrogen bonding with cellulose in the substrate matrix.<sup>81</sup> In contrast, lipid material is less likely to penetrate due to factors such as lower wettability.<sup>15, 68</sup> Almog *et al.*<sup>91</sup> demonstrated the absorption of fingerprints using amino acid sensitive reagents and fluorescence microscopy to study the depth of penetration of fingerprints cross-sections on paper. They found that the depth of penetration of fingerprints on paper correlates with the substrate's porosity, with better quality developed marks produced at depths between 40-60 microns.

When fingerprints are left on non-porous substrates like glass and plastics, they are not absorbed into the material and remain exposed on the surface.<sup>68</sup> This can cause changes to the chemical profile of the mark, such as the loss of water and other volatile substances. For instance, it was observed that the volatile compound squalene decreased more quickly on glass surfaces than on paper over a 30 day period.<sup>76</sup>

The ability of different substrates to absorb and retain fingerprint residue is crucial in determining their persistence and recoverability.<sup>16</sup> Fingerprints on non-porous substrates are more prone to physical damage, degradation, and evaporation, while porous substrates tend to minimise exposure to these factors, resulting in increased persistence. This is demonstrated by studies showing that fingerprints on paper substrate can be developed using amino acid sensitive reagents, even after several decades.<sup>82, 83</sup> In addition, the chemistry and reactivity of a substrate with fingerprint

residue can also impact recoverability. Fingermarks on metal surfaces, for instance, can cause corrosion,<sup>92, 93</sup> altering the properties of the fingermark deposit and affecting its recoverability. Within the same context, new substrates, such as biodegradable plastics, have prompted investigation<sup>94, 95</sup> of their impact on fingermark enhancement methods.

### 1.3.3 Environmental conditions

Various environmental factors, including temperature, light exposure and humidity, can impact the composition and rate of change to the fingermark residue as well as the substrate deposited on.<sup>52, 74</sup> Studies have shown that exposure to high temperatures can lead to accelerated water loss, volatile materials and chemical degradation of fingermark constituents. Richmond-Aylor *et al.*<sup>96</sup> explored the amino acid degradation products using pyrolysed GC-MS on extracted fingermark residue. Decomposition products such as 2,5-furandione from aspartic acid and 3,6-dimethylpiperazine-2,5-dione from alanine were identified. Some of these compounds were found to fluoresce, providing an additional enhancement method for fingermarks subjected to extreme temperatures.<sup>97, 98</sup> Similar results of thermal degradation have been observed with urea.<sup>98, 99</sup> Elevated temperatures also affect the lipid components, with Wolstenholme *et al.*<sup>100</sup> revealing the degradation of oleic acid at 60 °C using matrix-assisted laser desorption/ionisation mass spectrometry imaging (MALDI-MSI). In a more recent study, Kim *et al.*<sup>101</sup> explored the degradation of squalene and cholesterol over a short exposure time of 8 hours using GC-MS and reported significant loss of both compounds at 100 °C.

Light exposure has also been found to increase the oxidation rate of the lipid material. Archer *et al.*<sup>52</sup> found that squalene and oleic acid degradation were more rapid for fingermarks stored in light compared to dark conditions. Squalene was found to be undetectable after 9 days in light conditions, while it lasted for 33 days in the dark. Mountfort *et al.*<sup>56</sup> demonstrated the rapid oxidation of squalene in the presence of a photoxidiser and identified squalene monoxide and squalene epoxide as oxidation products in fingermark residue within 1 day of light exposure. Several

other studies have reported the rapid degradation of squalene under light exposure with UV radiation light having a greater impact.<sup>84, 102</sup>

Although the impact of humidity on detection methods has been studied, little research has been conducted on its effect on fingerprint composition. Paine *et al.*<sup>103</sup> found that the quality of cyanoacrylate fume prints varies with humidity. Relative humidity (RH) of 80 % was found to develop the best quality fingerprints. This was attributed to water absorption by sodium chloride salt crystals, influencing the initiation of polymerisation and the type of microstructures formed. In contrast, a previous study using silver nitrate<sup>47</sup> found that higher humidity had a detrimental effect on developed fingerprints, suggesting that chlorides diffuse faster under high RH, making silver nitrate less effective on older fingerprints. The variation between jurisdictions in the effectiveness of amino acid sensitive reagent 1,2-indanedione in its early development has also been attributed to differences in humidity, with the addition of zinc chloride to the formulation minimising this effect.<sup>104</sup> These studies show that humidity plays a crucial role in the composition of eccrine material. Water submersion has a major influence on the persistence of eccrine material, with most of the aqueous residues washed away and the lipid constituents least affected. This is supported by successfully detecting fingerprints on wetted substrates using lipid targeting detection methods such as physical developer (PD) and Oil red O (ORO).<sup>105-107</sup>

It is important to note that although environmental factors can be considered in isolation, from an operational perspective, a combination of several environmental factors are encountered, which may have a greater impact than a single factor.<sup>16</sup> Additionally, the climatic difference between geographical locations needs to be considered when developing or introducing operational detection methods.

#### **1.4 Fingerprint detection methods**

The basis of the detection process is to produce appropriate contrast between the fingerprint and the substrate to enable the discrimination required to capture and record an image. This can be achieved through optical, chemical and physical

processes that exploit certain properties of the fingerprint and substrate.<sup>16, 17</sup> Optical processes exploit the reflectivity, refractive index, emissivity, colour or fluorescence properties of fingerprints and the deposited surface when illuminated or irradiated to enhance contrast.<sup>17</sup> The physical properties of fingerprints, such as their adhesive nature or induced electrical charge, may also be exploited for visualisation. For example, fingerprint powder development is a method that takes advantage of the preferential adhesion of powder particulate to the fingerprint constituents rather than the substrate.<sup>16</sup> Chemical processes target the chemical constituents of fingerprint deposits to form coloured or fluorescent products that improve contrast with the substrate.<sup>16, 17</sup>

Optical processes are applied to all types of fingerprints and are generally non-destructive, while physical and chemical processes are more selective and target specific constituents of the residue.<sup>68</sup> Considering the array of conditions under which a fingerprint could be encountered, it is important for practitioners to understand which constituents are being targeted by each process to enhance fingerprints under various conditions effectively.

Enhancement treatments can be applied sequentially to maximise recoverability, with optical processes applied first and in between other detection methods. Physical and chemical processes are applied in order of least to most destructive, generally in the order of liquid-free, organic solvent and water-based processes.<sup>16</sup> Figure 1.2 shows an example of a flow chart practitioners use to select enhancement treatment orders. Practitioners must also consider factors like time, substrate, and environment, which can alter fingerprint composition and impact detection effectiveness.

While the most effective detection sequence may be known, resource limitations can impact practitioner's choices. These limitations are jurisdiction-specific and include restricted budget, availability and accessibility to reagents, personnel and equipment, and supply chain disruptions. This section outlines common detection methods and discusses some of the challenges associated with their operational use in resource-limited jurisdictions.

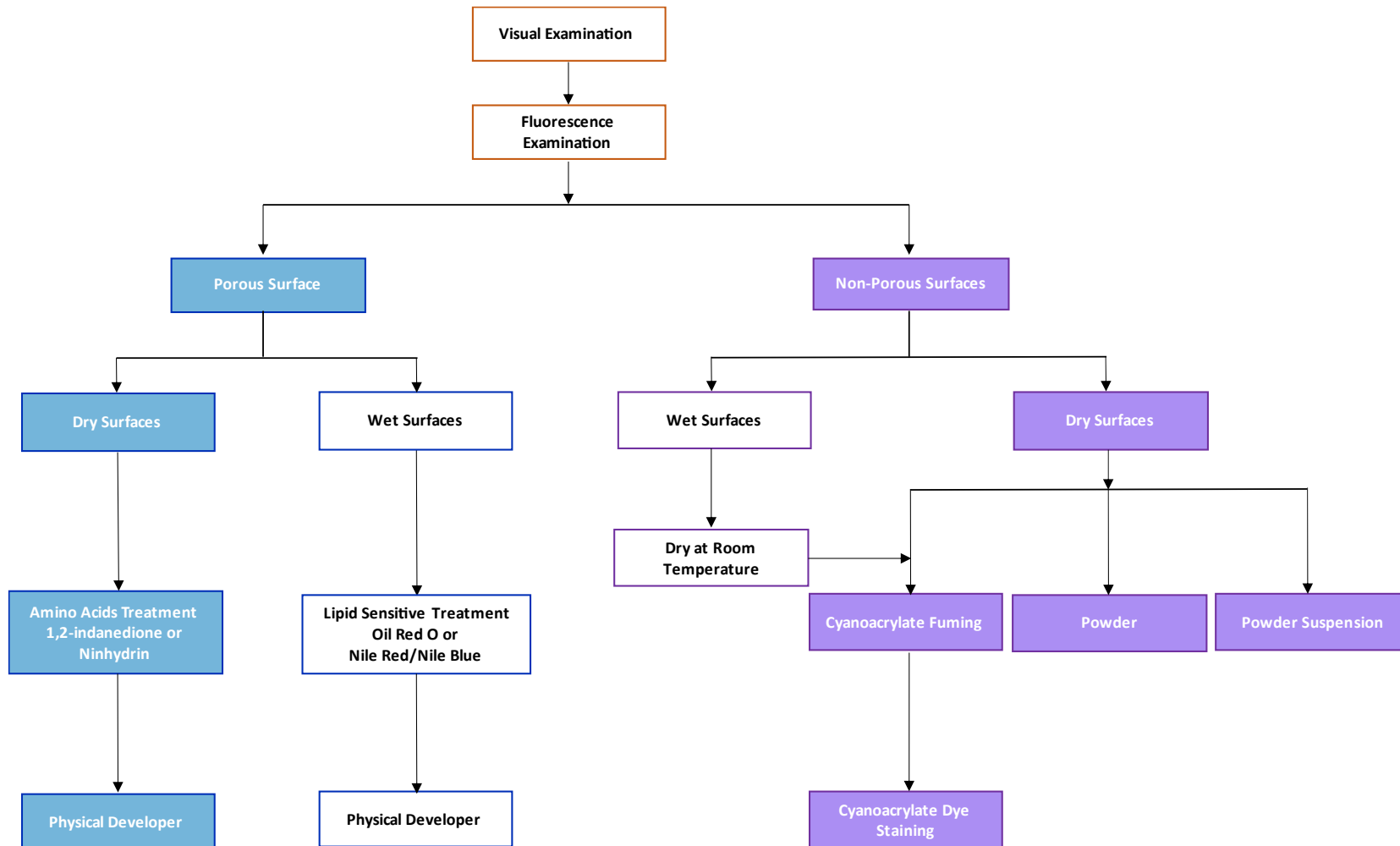


Figure 1.2: An example of a flow chart procedure for the development of latent fingerprints.<sup>16, 17</sup>

### **1.4.1 Porous-surface fingerprint detection methods**

Porous surfaces such as paper and cardboard are frequently encountered in criminal investigations. As discussed earlier, on these surfaces, water-soluble deposits like amino acids, urea, and chlorides get absorbed quickly and are contained within the substrate. In contrast, the water-insoluble deposits remain on top of the surface, with only a small amount persisting over time. This means the fingerprint pattern based on the water-soluble deposit is well preserved, and an image can be developed with amino acid sensitive reagents. However, environmental factors like water can wash away amino acids while diffusion of urea and chlorides due to humidity can affect water-soluble deposits.<sup>36</sup> In such cases, lipid-sensitive detection methods are used.

#### **1.4.1.1 Amino acid sensitive reagents**

Various amino acid sensitive reagents have been established over the years. Research in this area has mostly centred around the synthesis and optimisation of ninhydrin analogues,<sup>25, 108-110</sup> with an emphasis on increasing the sensitivity and the use of luminescence detection.<sup>14, 29</sup> Nonetheless in some instances the accessibility and cost of the development reagents limit its use in some jurisdictions.

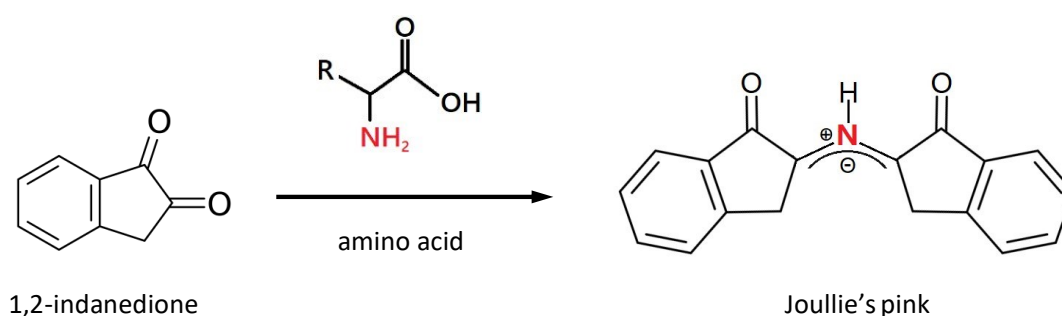
#### **1.4.1.2 Ninhydrin and 1,8-diazafluoren-9-one (DFO)**

Ninhydrin (2,2-dihydroxy-1,3-indanedione) was discovered in 1910 by Ruhemann.<sup>111</sup> It was the first amino acid sensitive reagent to be used for latent fingerprint detection. Ninhydrin reacts with amino acids to produce a purple product known as "Ruhemann's purple" through the generally accepted mechanism of Strecker degradation.<sup>25</sup> Ruhemann's purple does not have photoluminescent properties unless further treated with metal salts<sup>16</sup> which makes the method cumbersome. Photoluminescence is desirable as it offers better sensitivity and contrast, particularly on coloured or patterned surfaces. This in part led to further investigation into ninhydrin analogues<sup>112-114</sup> that can produce both coloured and photoluminescent products in reaction with amino acids. Two reagents that became of particular interest were 1,8-diazafluoren-9-one (DFO) and 1,2-indanedione.

DFO was first synthesised by Druey and Schmidt in 1950<sup>25</sup> and introduced as a fluorescent detection technique for latent fingerprints by Pounds and Grigg in 1990.<sup>112</sup> Although DFO is not a direct analogue of ninhydrin, it follows a similar reaction mechanism to produce a red product, which is fluorescent under excitation in the green region of the spectrum (430 to 580 nm).<sup>68</sup> Unlike ninhydrin, the reaction does not require secondary treatment with metal salts. The advantage of a one-step reaction and the noticeable increase in sensitivity compared to ninhydrin, lead to DFO gaining widespread use in the UK.<sup>16</sup>

#### 1.4.1.3 1,2-indanedione

1,2-indanedione is a more recent amino acid sensitive detection method for porous surfaces. It was first proposed as a fingerprint reagent in 1997 after initial observation in the 1990s at the University of Pennsylvania.<sup>113, 114</sup> The compound was shown to be sensitive to amino acids and produce a highly fluorescent, bright pink complex known as Joullie's pink (JP) (Figure 1.3).<sup>108</sup> The reaction mechanism has been characterised to involve Strecker degradation<sup>115</sup> similar to ninhydrin and DFO. Joullie's pink has a maximum absorption at 550 nm and a luminescence emission at 555 nm when excited with a green light source (500–530 nm).



**Figure 1.3:** The reaction of 1,2-indanedione with amino acids to form Joullie's pink.

Following the promising initial study of 1,2-indanedione as an alternative to ninhydrin and DFO, researchers worldwide conducted comparative assessment. Variable results were obtained, with research groups in Australia<sup>116</sup> and Israel<sup>117</sup> reporting superiority of 1,2-indanedione over DFO, whilst researchers in Canada<sup>118</sup> and the UK<sup>119, 120</sup> found the reverse. The contradicting results were highlighted in a

2004 survey conducted by Wallace-Kunkel *et al.*<sup>121</sup> in Australia, New Zealand, USA, UK, and Europe on detecting and enhancing latent fingermarks on porous surfaces. The authors reported a lack of widespread use of 1,2-indanedione, with only 28 % of respondents having used it in casework. This is likely due to various formulations and processing conditions adopted by different laboratories worldwide, as well as differences in the chemistry of the local substrate or environmental conditions.

A modified 1,2-indanedione formulation was developed for Australian conditions containing premixed zinc chloride, and was found to be more resilient to humidity fluctuations.<sup>115, 122</sup> Spindler *et al.*<sup>104</sup> characterised the role of zinc(II) in the 1,2-indanedione zinc (IND/Zn) formulation as a Lewis acid catalyst, stabilising a key intermediate during a rate-limiting hydrolysis step in forming JP. IND/Zn is recognised as the most sensitive amino acid fingerprint detection method on porous surfaces following several comparative studies<sup>122-125</sup> to ninhydrin and DFO. This has led to its widespread use in various jurisdictions.<sup>124, 126, 127</sup> More recently, pseudo-operational trials<sup>125, 127, 128</sup> conducted in the UK demonstrated the superiority of IND/Zn to DFO, and it is expected to be fully implemented in casework by the UK Home Office Centre for Applied Science and Technology (CAST) Fingerprint Visualisation Manual. However, implementation in micro-jurisdictions such as Seychelles remains a concern due to cost and supply chain disruption of the standard carrier solvent HFE-7100 (methyl nonafluorobutyl ether) used in the formulation.

#### **1.4.1.4 Lipid-sensitive reagents**

When items have been wetted, commonly used amino acid sensitive reagents are ineffective since amino acids dissolve in water. In such cases, lipid-sensitive reagents such as PD, ORO or Nile red are used to target the water-insoluble components. The water-insoluble deposit can be grouped into 'fragile' and 'robust' fractions.<sup>105, 107, 129, 130</sup> The robust fraction consists of proteins and lipo-proteins that can form hydrogen bonds with the cellulose content of paper, allowing them to remain on the surface for longer periods of time. The fragile fraction includes unsaturated fatty acids, lipids, and triglycerides, which quickly undergo chemical changes when exposed to air.<sup>78</sup> It is suggested that the lipid reagents target different fractions of the water-insoluble



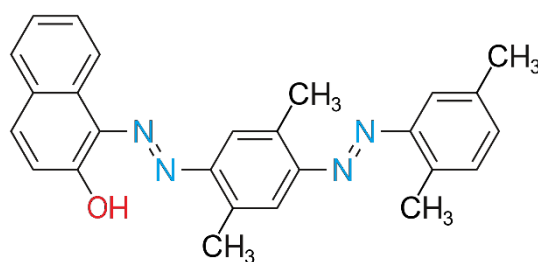
deposit, given variation in the quality of developed marks between fresh and older fingermarks. However, some of these methods are complex and use toxic reagents, limiting their use in some jurisdictions due to cost and access to trained personnel and equipment. A prime example is the use of PD.

#### **1.4.1.5 Physical developer (PD)**

The PD technique was developed as a fingermark detection method in early 1970 at the Atomic Weapons Research Establishment (AWRE) in the UK.<sup>68</sup> Its development was based on an adaptation of the silver nitrate solution development process that was commonly used in the photographic industry. The PD solution contains silver ions, ferrous and ferric salt, citric acid and a surfactant in a delicate formulation.<sup>17</sup> It works by selectively depositing silver particles on the ridges of a fingermark, resulting in a light grey image. Due to alkaline binders and fillers present in some paper substrates, an acid prewash with maleic acid forms part of the treatment process to avoid background development.<sup>16, 106</sup> PD has been reported to be an effective method in developing fingermarks exposed to extreme conditions such as, immersed in water,<sup>131</sup> temperatures excess of 100 °C<sup>132</sup> and up to 90 year old fingermarks<sup>82</sup> have been successfully developed. Nonetheless, the complexity of the method (e.g. requiring several different stock solutions and baths, along with sensitivity to light and contamination) and its high cost limit its use in some jurisdictions.<sup>133, 134</sup>

#### **1.4.1.6 Oil red O (ORO)**

Oil red O (1-([4-(xylylazo)xylyl]azo)-2-naphthol) is another alternative method for detecting fingermarks on wet porous surfaces. It is a lysochrome used as a biological stain for lipids since the 1920s.<sup>135</sup> In the early 2000s, ORO was used as a reagent to visualise lip marks<sup>136</sup> and in 2004 Beaudoin *et al.*<sup>129</sup> introduced it as a latent fingermark detection technique. ORO is a diazo dye (R–N=N–R') with non-ionising structural conformation shown in Figure 1.4. Hence it can dissolve in the lipid components of fingermark residue, staining the ridges red and the background pink.



**Figure 1.4:** Chemical structure of Oil red O.

The ORO process developed by Beaudoin *et al.*<sup>129</sup> consists of three steps (staining, neutralisation and drying) and two stock solutions; the stain solution dissolved in methanol and a buffer solution. The reagent and treatment processes are more economical and less tedious than PD. However, the ORO process was found to be more time-consuming taking up to 90 minutes to develop fingermarks.<sup>68</sup> Frick *et al.*<sup>137</sup> proposed a non-toxic modified ORO formulation by dissolving the stain in propylene glycol instead of methanol. The amount of ORO in the formulation was reduced, making it more straightforward and economical to prepare. The development process was also modified, involving a 15-minute immersion in the reagent, resulting in an efficient process in contrast to the original method. The authors reported comparative performance between the original and modified ORO detection techniques on various paper substrates, including under wet conditions.

Several studies have conducted comparative assessments of ORO and PD. Studies using lipid-rich fingermark deposits have reported the superiority of ORO,<sup>105, 138, 139</sup> whereas PD was found to be more effective with natural fingermarks.<sup>107, 140</sup> In essence, ORO is generally effective in developing a charged fingermark that is less than a few weeks old, although the detection of a 20 year old fingermark has been reported.<sup>141</sup> PD is a superior method for detecting older and uncharged fingermarks, that have been exposed to water. The variation in effectiveness has been attributed to ORO targeting the fragile fraction, which is more susceptible to environmental factors. Whereas PD targets the stable robust fraction, hence the ability to detect older fingermarks and on items under prolonged exposure to water. Nonetheless, ORO (particularly the modified formulation) offers an alternative for jurisdictions where PD may not be feasible, subject to recognition of its limitations.

#### 1.4.1.7 Nile red

Nile Red (9-diethylamino-5H-benzo[a]-phenoxazine-5-one) is a benzophenoxazine lipophilic stain that can stain fingermarks for visualisation.<sup>142</sup> It has photoluminescent properties and was initially introduced as a fluorescent stain for intracellular lipid staining in the 1980s.<sup>143</sup> More recently, Braasch *et al.*<sup>144</sup> proposed it for detecting latent fingermarks on wetted paper. Nile red preferentially dissolves in neutral lipids of the fingermarks and can be visualised under luminescent mode. Due to poor solubility and quenching of fluorescence in aqueous environment, methanol is used to dissolve Nile red.<sup>142</sup>

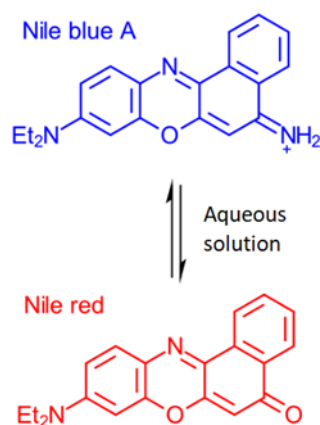
A recent study<sup>144</sup> compared Nile red to PD and ORO on wetted papers in a pseudo-operational study. The study found that Nile red was effective on fingermarks less than 1 month old, but like ORO, it was outperformed by PD on older marks. However, Nile red has the advantage of a simple and easy preparation process and develops luminescent fingermarks with better contrast on dark and patterned surfaces. Unfortunately, the high cost of Nile red (ca. \$760 USD per gram)<sup>145</sup> and the use of toxic reagent methanol limits its operational use. Frick *et al.*<sup>146</sup> developed an alternative method to overcome these issues using the basic phenoxazine dye Nile blue in aqueous solution.

#### 1.4.1.8 Nile blue

Nile blue A or Nile blue (benzo[a]phenoxazin-7-ium,5-amino-9-(diethylamino)-, sulfate) is another benzophenoxazine dye that is commonly used as a histological stain.<sup>147-149</sup> It was initially trialled as a fluorescent latent fingermark detection technique in the 1980s.<sup>150</sup> Other tested forensic applications include a post-cyanoacrylate stain<sup>151</sup> and lip print detection,<sup>152</sup> where it is dissolved in an alcoholic solution.

Frick *et al.*<sup>57, 146</sup> developed an aqueous-based formulation to eliminate the use of organic solvents. Nile blue is a cationic dye, readily dissolving in aqueous solution but exhibiting low levels of luminescence compared to Nile red.<sup>142</sup> In aqueous solution, a small amount of Nile blue undergoes spontaneous hydrolysis to Nile red (Figure 1.5).

The presence of the two stains results in a dual-purpose fingerprint detection reagent. Nile red dissolves preferentially in neutral lipids, producing photoluminescence, while Nile blue interacts with the acidic components, resulting in visible dark blue development and suppression of background fluorescence.



**Figure 1.5:** Hydrolysis of Nile blue to Nile red in aqueous solution.

Studies by Frick *et al.*<sup>146</sup> demonstrated aqueous Nile blue's effectiveness in developing lipid-rich fingerprints on a range of wet porous and non-porous surfaces, including some adhesive surfaces. The advantage of this formulation is that it is water-based, making it a safe and organic solvent-free procedure. Moreover, Nile blue dye is inexpensive (ca. \$3.20 USD per gram)<sup>153</sup> compared to Nile red. Its low toxicity, cost, and ease of use make it a promising option for jurisdictions with limited resources or for non-scientifically trained personnel. It also underlines an avenue to adapt to challenges encountered in resource-limited jurisdictions through the use of alternatives chemicals and modified reagents.

Further studies by Crocker<sup>154</sup> have shown that modifying the aqueous Nile blue formulation with sulfuric acid can increase the technique's sensitivity. This is because the presence of acid increases the hydrolysis of Nile blue into Nile red, resulting in increased Nile red concentration as demonstrated using luminescence spectroscopy. Similarly, the preliminary study reported that acidified Nile blue produces fingerprints with greater luminescence than aqueous Nile blue on wet porous

substrates. Other studies have also investigated the potential of Nile blue as a post-cyanoacrylate stain, which is discussed in section 1.4.2.4.

## **1.4.2 Non-porous-surface fingerprint detection methods**

When a fingerprint is deposited on non-porous substrates like glass or plastics, it remains on the surface and is vulnerable to damage from environmental factors.<sup>90</sup> Therefore, solvent-based detection methods are not generally preferred as they are likely to wash away the fingerprint residue. Instead, it is recommended to use dry powders or cyanoacrylate fuming methods.<sup>16, 17</sup> However, on challenging surfaces and where the fingerprint is partially fixed, such as on adhesive tape, using a liquid powder suspension is more effective.

### **1.4.2.1 Fingerprint powder**

The powdering technique is one of the oldest reported latent fingerprint detection methods on smooth non-porous surfaces, and it remains a common method used at crime scenes.<sup>15</sup> Various commercial powders, such as magnetic, granular (carbon black and titanium dioxide white), metal flake (aluminium and brass alloy), and fluorescent powders, are available for different applications. However, some of these powders were developed in the 1920s<sup>155</sup> and appeared not to have been developed within a standardised approach, underlining the need for a revisit under the more current assessment framework.<sup>24</sup> This is especially important when considering the method's sustainability.

The development of latent fingerprints with powder occurs based on the preferential adhesion to the fingerprint constituents in contrast to the substrate background.<sup>15</sup> Several mechanisms contribute to the overall adherence, including particle shape, size, surface chemistry and electrostatic charge.<sup>156</sup> The presence of grease and liquid in fingerprint deposits has also been identified to increase adhesion through increased contact surface area and the liquid surface tension capillary action.<sup>68</sup> This explains why the powdering technique is not suitable for old, dried fingerprints on porous substrates. Additionally, surfaces with high adhesion to fingerprint powders,

such as adhesive tape, result in poor contrast between the fingerprint and substrate, presenting a challenge.

#### **1.4.2.2 Powder suspension**

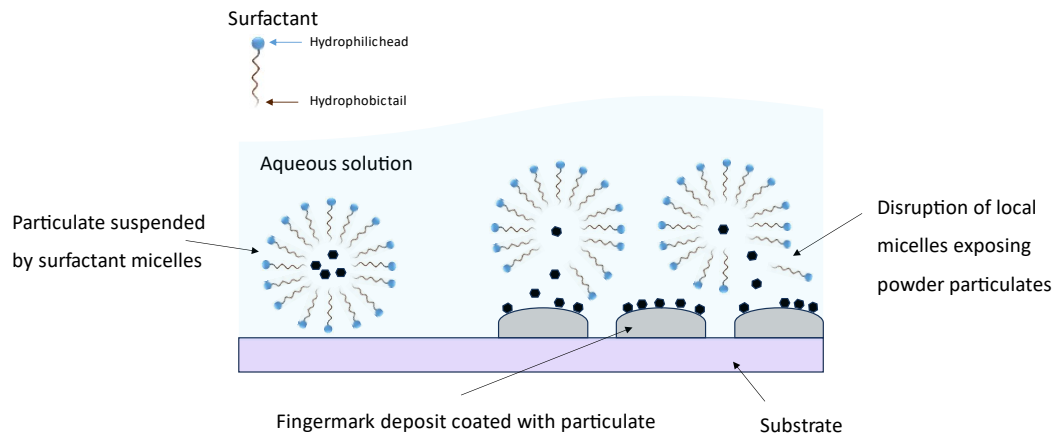
Powder suspension is an effective and recommended method for developing latent fingerprints on the sticky side of non-porous tapes.<sup>16</sup> It consists of a powder suspended by a surfactant in water to form a paste that is then lightly painted over the adhesive tape using a brush, followed by washing with tap water. An early process was adopted from Japan and commercialised in the 1990s as “Sticky-side Powder” by the Lightning Powder Company.<sup>157</sup> This formulation, containing grey and black powder in Kodak Photoflo surfactant water mixture, was compared to gentian violet, the primary method used on adhesive tapes in 1996.<sup>158</sup> The sticky-side powder method was found to outperform gentian violet and led to investigations into alternative formulations; using black,<sup>159</sup> grey<sup>160, 161</sup> and white<sup>162, 163</sup> commercial fingerprint powders combined with different surfactants.

In the UK, several powder suspension formulations were compared against Sticky-Side Powder to determine their effectiveness on various surfaces. Results showed that a black formulation containing precipitated magnetic iron oxide was more effective in treating light-coloured non-adhesive surfaces.<sup>68</sup> Further research revealed that its effectiveness is highly influenced by the type of iron (II/III) oxide powder used.<sup>164-166</sup> On dark non-porous adhesive surfaces, a white powder suspension formulation based on titanium dioxide was found to be more effective,<sup>68</sup> which was supported by other independent studies.<sup>163, 167</sup>

Meanwhile surfactants such as Liquinox<sup>162</sup>, Kodak Photoflo<sup>160, 161</sup> and Triton™ X-100<sup>16</sup> were found to be the most effective for the various formulations. However, some surfactants have faced supply chain issues, such as Kodak Photoflo being discontinued due to commercial viability and Triton X-100 being subjected to environmental regulations.<sup>168</sup>

The exact mechanism of powder suspension is unknown, but it has been suggested that surfactant micelles encapsulate the powder particles, and certain components

of the fingerprint residue disrupt the micelles, exposing the powder and causing selective deposition on the fingerprint,<sup>166</sup> illustrated in Figure 1.6. Based on the observation that powder suspension develops fingerprints on wetted surfaces<sup>169, 170</sup> and does not appear to be selective towards sebaceous fingerprints, it has been suggested that the eccrine component encapsulated by the water-insoluble constituents is responsible for the destabilisation of the surfactant micelles.<sup>16</sup>



**Figure 1.6:** Schematic illustration of powder suspension development. Fingerprint constituents disrupt surfactant micelles exposing suspended powder particulates, causing discriminate deposition on fingerprint and not the substrate.

Various pre-mixed commercial proprietary black and white powder suspension formulations have become available in recent years,<sup>171-174</sup> with carbon-based powder suspension generally recommended as the most effective on non-porous light-coloured adhesive surfaces. However, their operational use may be limited in small laboratories with restricted budgets due to the high cost of commercial powder suspension. For example, the black powder Wetwop™ costs \$340 USD per litre.<sup>171</sup> Another consideration when using a pre-mixed product is the general lack of control over formulation quality, and shelf-life<sup>168</sup>. This has led to interest in developing cheaper in-house alternatives.

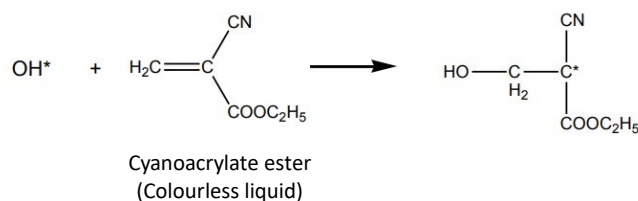
A recent example is the development of WET UCIO powder suspension formulation,<sup>175</sup> consisting of carbon black Sirchie powder mixed with a commercial Gran Velada surfactant solution. This formulation has been reported to be more effective than the commercial Wetwop™ on natural fingerprints deposited on eight

types of adhesive tapes.<sup>175</sup> The WET UCIO powder is estimated to cost c.a \$64 USD per litre, which is at least five times cheaper than Wetwop™. The development of more economical reagents increases the accessibility of the method to the Global South.

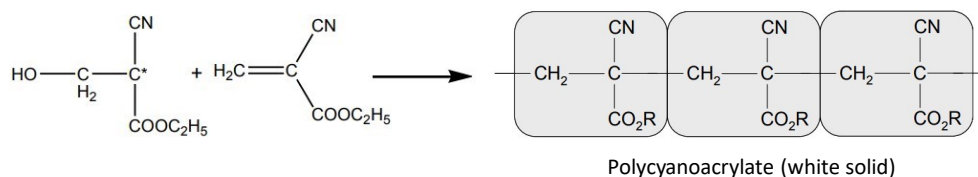
### 1.4.2.3 Cyanoacrylate

Cyanoacrylate fuming also known as superglue fuming, is one of the most common methods used on non-porous surfaces such as glass and plastics.<sup>15</sup> This technique was discovered independently in Japan, America and the UK in the late 1970s.<sup>176, 177</sup> The glue is based on colourless cyanoacrylate esters, specifically methyl-2-cyanoacrylate or ethyl-2-cyanoacrylate, which are monomeric liquids that polymerise into a white solid in the presence of weak bases. Although the exact mechanism for polymerisation growth on fingerprints is not well understood, it has been suggested that anionic polymerisation can be initiated by water and other weak bases in the residue.<sup>178</sup> Polymerisation occurs due to the strong inductive effects of the cyano group (–CN) and the ester group (–COOR), which makes the alkyl cyanoacrylate susceptible to nucleophilic attack. Once nucleophile initiation occurs, it will react with another alkyl cyanoacrylate monomer and the propagation of the reaction results in a long polycyanoacrylate polymer chain. An example of ethyl cyanoacrylate polymerisation reaction is shown in Figure 1.7.

#### Initiation:



#### Polymerisation:



**Figure 1.7:** Polymerisation reaction of ethyl cyanoacrylate.



Polycyanocrylate has been observed to occur preferentially on greasy and moist fingerprint deposits. Lewis *et al.*<sup>179</sup> found that moist fingerprints prior to development produce high quality cyanoacrylate fingerprints. However, sebaceous fingerprints produce better quality fingerprints than eccrine when allowed to age. This was suggested to be due to sebaceous material retaining moisture rather than its constituents contributing to the polymerisation reaction. Differences in the morphology of the polymer structure have been observed under scanning electron microscopy.<sup>68</sup> Sebaceous fingerprints exhibit clumps of noodle-like polymer structure compared to longer microstructure with eccrine marks. Further studies have shown that high relative humidity increases polymerisation, which is thought to be due to rehydration by sodium chloride salts in the fingerprint deposit facilitating the initiation process. Paine *et al.*<sup>103</sup> reported that the highest quality developed fingerprints were obtained at 80 % RH, which aligns with the theory that water is an initiator given that sodium chloride is saturated at 75 % RH,<sup>180</sup> the point at which it draws in water from immediate surroundings. The morphology of polymer structure above 70 % RH is noodle-like compared to flat film-like microstructure at 60 % RH.<sup>103</sup> Farrugia *et al.*<sup>181</sup> suggested the difference in morphology can be attributed to the abundance of water at high RH leading to a soft anion initiation causing one directional polymer growth.<sup>182</sup> At lower RH, initiation by a hard anion results in many active centres of polymer growth.<sup>183</sup> The noodle-like morphology is favoured as it provides better visualisation due to increasing light scattering interactions with staining dyes.

Initially, the processing method of cyanoacrylate involved a simple improvised fuming cabinet comprising a sealed chamber and drops of cyanoacrylate at the bottom.<sup>184</sup> In an attempt to decrease the processing time and enhance the quality of developed fingerprints, the use of heat<sup>185</sup> to increase the rate of evaporation of the glue and other accelerating agents such as sodium hydroxide<sup>184</sup> and sodium carbonate<sup>186</sup> were investigated. As the literature shed light on the polymerisation mechanism and optimum conditions for fingerprint development, dedicated controlled humidity superglue cabinets were developed. However, due to the high cost of commercial cabinets, many police agencies still use improvised fuming

chambers, such as an aquarium and a hotplate as a heat source.<sup>68</sup> Other adaptations include using a small dish of water to increase humidity or small fans for even development in large chambers.<sup>17</sup> These adaptations demonstrate how jurisdictions with limited resources can access latent fingerprint detection methods. However, safety precautions must be taken to avoid health risks, associated with the suggested carcinogenic hazards with heating cyanoacrylate above 200 °C. This underscores the importance of safety whilst adapting fingerprint enhancement techniques.

#### **1.4.2.4 Post-cyanoacrylate stains**

Developed cyanoacrylate fingerprint is a white deposit, and photography can be challenging on light-coloured or multicoloured surfaces. Post-treatment methods such as powdering and staining are employed to enhance visualisation.

Enhancement using staining techniques can be achieved using coloured and fluorescent dyes, with the latter recommended due to higher sensitivity.<sup>17</sup> Rhodamine 6G and Basic Yellow 40 (BY40) are two of the most commonly used stains. Rhodamine 6G was suggested by Menzel *et al.*<sup>150</sup> in the 1980s, and it is one of the most widely used post-cyanoacrylate stains. However, the most effective formulations use methanol, which is toxic, and there is also an ongoing debate on the carcinogenic effect of rhodamine 6G.<sup>68</sup> As an alternative BY40 was proposed,<sup>68</sup> which has lower toxicity since it uses ethanol. Nonetheless, the use of alcohol is detrimental to surfaces such as varnishes and some ink-printed surfaces. A water-based formulation of BY40 has been explored but exhibited lower effectiveness. This has led to the investigation of other dyes that can be used with a water-based formulation, such as basic red 14 and Nile blue. Nile blue has additional benefits being an inexpensive dye that is water soluble. Although preliminary study by Chesher *et al.*<sup>151</sup> reported weak development with Nile blue as a post-treatment stain for cyanoacrylate, there has been limited research on its potential in the literature. More recently a series of preliminary studies<sup>154, 187, 188</sup> at Curtin University have shown promise for Nile blue as a safe and cost-effective post-cyanoacrylate enhancement technique.

Furthermore, the mechanism of post-cyanoacrylate staining is not well understood. One of the few suggestions is that the cyanide (CN<sup>-</sup>) anions of polycyanoacrylate may form weak van der Waals interactions with basic dye cations.<sup>68</sup> A fundamental understanding of the staining process can direct the use of new dyes and sequencing processes.

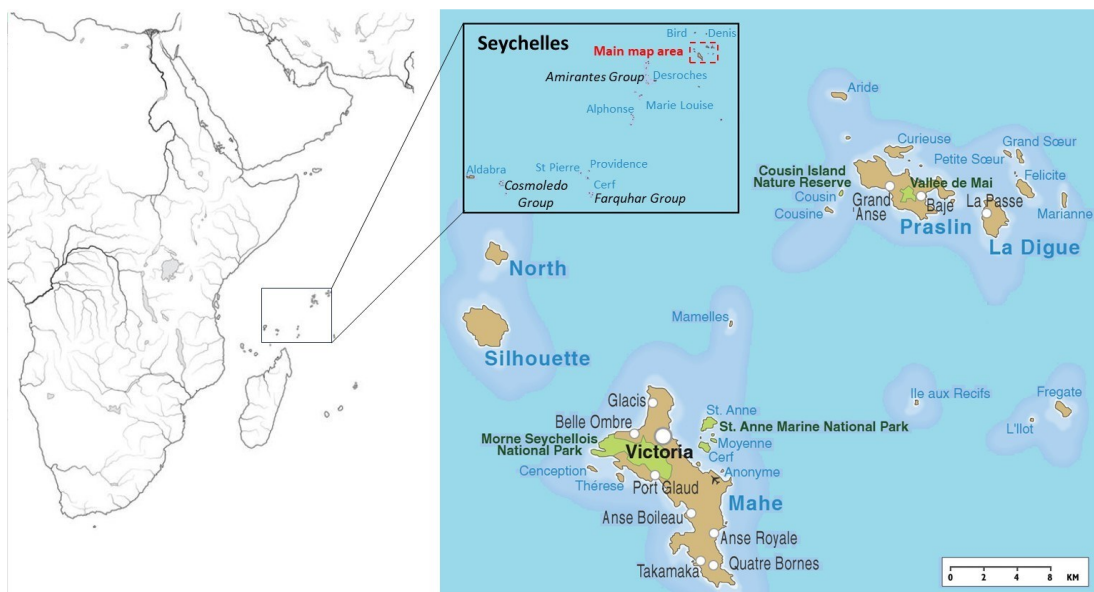
## **1.5 Latent fingerprint detection service provision**

The detection of latent fingerprints is an important tool in both humanitarian and criminal investigations worldwide<sup>26, 189, 190</sup> Therefore, it is essential for jurisdictions to have access to effective latent fingerprint detection techniques and the ability to ensure the continuity of service. The previous section highlighted some of the obstacles that resource-limited jurisdictions face in implementing fingerprint enhancement methods, as well as potential solutions. The next section examines the specific case of Seychelles, a remote, small jurisdiction, before expanding the focus to a global perspective. Amid the discussion, the concept of sustainable forensic science service provision is presented as a viable solution.

### **1.5.1 Seychelles latent fingerprint detection service provision**

Seychelles is an archipelago of 115 islands in the West Indian Ocean covering a total land mass of 444 square kilometres, spread over a vast sea area of 1.4 million square kilometres (Figure 1.8). Because it is located near the equator, Seychelles has a tropical climate with temperatures ranging from 21 °C to 23 °C and relative humidity between 70% and 80%.<sup>191</sup> It is the smallest African state with a population of approximately 100,000 and a fragile economy primarily based on tourism, contributing 70 % of the gross domestic product (GDP).<sup>192</sup> Relative to population size, the Seychelles Forensic Section has modest resources and trained local staff within a unified forensic science service, which operates on a hybrid of local service provision and overseas outsourcing under the auspices of the Seychelles Police. The potential socio-economic impact of crime on a small island community and fragile tourism

economy has led the Government to invest in strengthening forensic science service provision,<sup>193</sup> including latent fingerprint detection and identification capabilities.



**Figure 1.8:** *The geographical location of Seychelles.*

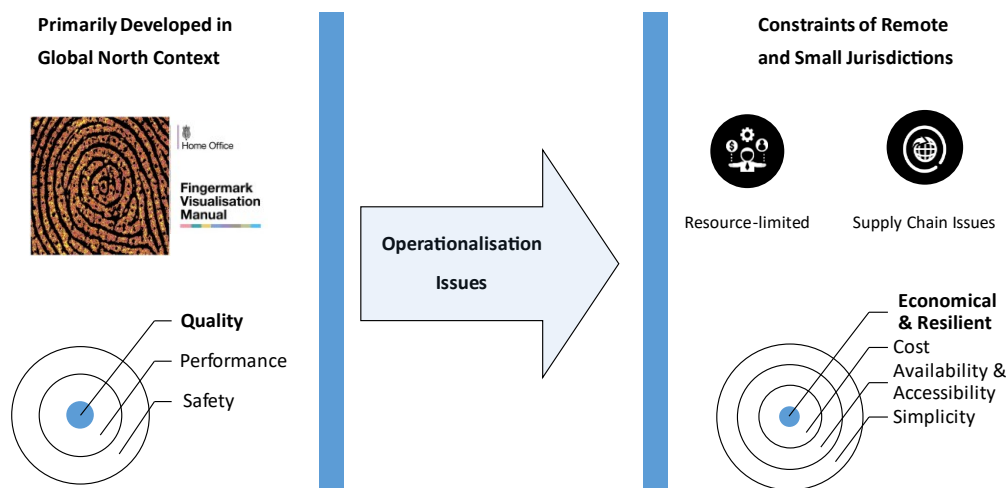
Established in 1968, the Seychelles latent fingerprint section was the first local forensic science service provision.<sup>194</sup> Its administration and delivery system are primarily based on the UK model, as Seychelles was a former British colony. This is evident with most of the personnel trained in the UK and using the CAST Fingerprint Visualisation Manual<sup>16</sup> as the basis for applying fingerprint detection methods despite major differences in climatic conditions. In 2013, the Seychelles Police Force (SPF) enhanced its fingerprint identification capabilities by acquiring an automated fingerprint identification system under the framework of the Interpol capacity building for Eastern African countries funded by the European Union (EU).<sup>195</sup> However, as a prerequisite, the system's effectiveness depends on an enhanced latent fingerprint detection capability.

### **1.5.2 Challenges in operationalising latent fingerprint detection protocols**

When implementing new or modified latent fingerprint detection protocols in Seychelles, there are two primary considerations due to its locality: i) the limited budgetary and technical resources and ii) supply chain interruptions. As a small jurisdiction, Seychelles has limited resources, making it essential to adopt an

economical, safe, and simple latent fingerprint detection method to optimise resource management. Secondly, Seychelles is geographically remote as shown in Figure 1.8, and sits at the end of the commercial supply chain. As a result this increases the risk of supply chain disruption.<sup>196</sup> Therefore ensuring a readily available and easily accessible detection method is crucial to service continuity.

Over the last two decades, a significant amount of research has been into developing novel and improving existing latent fingerprint detection methods.<sup>26, 125, 197, 198</sup> Overwhelmingly, this research has been carried out in the northern hemisphere and highly developed nations with abundant resources and non-tropical climates. The primary focus of these studies has been to improve performance of detecting quality and quantity of fingerprints, but not necessarily to address challenges faced by jurisdictions with limited resources and supply chain issues. Therefore, the operationalisation of these methods can be problematic in a small jurisdiction as illustrated in Figure 1.9. To overcome these challenges and provide sustainable fingerprint service provision, smaller jurisdictions require cost-effective and resilient detection methods.



**Figure 1.9:** Illustrating the gap in operationalisation of latent fingerprint detection technique in remote and small jurisdictions.

As an example, the carrier solvent for the amino acid sensitive reagents has been standardised over the last two decades to HFE-7100 (methyl nonafluorobutyl ether).<sup>120, 199</sup> Despite its ideal properties as a carrier solvent, HFE-7100 is expensive

(ca. \$240 USD per litre) and prone to restricted supply<sup>168, 200</sup> due to environmental regulations. The supply risk is exacerbated as it is a single-source proprietary solvent from 3M™ Novec™<sup>201</sup> and under demand from multiple industries. Manufacturing plant issues and the COVID-19 pandemic have increased the lead time for this product in large jurisdictions like Australia and the United States by up to 9 months over the last 2 years,<sup>202, 203</sup> underscoring the global extent of supply chain disruption. It follows from the perspective of a restricted-budget laboratory in a remote location that the reliance on HFE-7100 poses a substantial risk to service continuity.

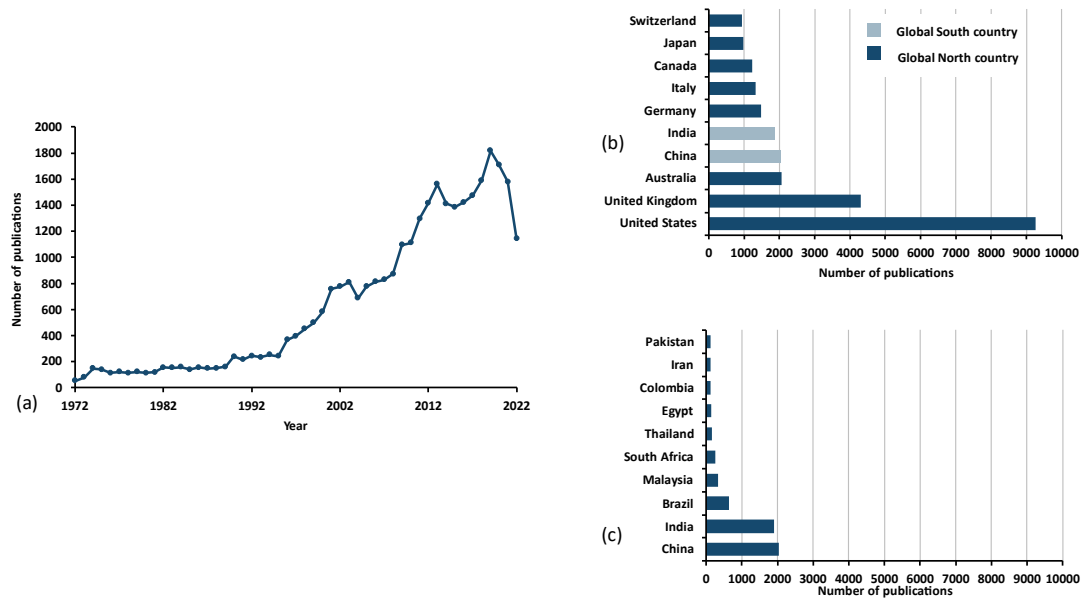
This signifies a gap in developing and validating latent fingerprint detection techniques for operational use. The question that arises is how small jurisdictions such as Seychelles can adapt to access and develop fit-for-purpose fingerprint detection capabilities and maintain continuity of service. In other words, how to establish a sustainable provision for latent fingerprint detection.

### **1.5.3 Sustainable forensic science provision; frugal forensics**

The challenges that Seychelles faces in the field of latent fingerprint detection reflect the broader issue of limited forensic science resources in Global South jurisdictions. A Scopus bibliometric search of articles using “forensic science” as a search term reveals that the space is dominated by literature emanating from the Global North (Figure 1.10). A similar conclusion may be drawn from a more detailed recent study<sup>27</sup> examining research trends in forensic fingerprint detection and interpretation.

The research dominance, resourcing, and advanced capabilities of the Global North may convey the view that forensic science provision is globally well-developed. However, the stark reality is that most Global South nations face substantial obstacles in forensic science service provision compared to the Global North. For example, the literature reveals that challenges of the Global North revolve around high-level issues such as forensic backlogs,<sup>204, 205</sup> human bias or error influencing decision-making,<sup>206-208</sup> and the need for evaluative and activity-level reporting.<sup>209, 210</sup> By contrast, the Global South faces fundamental resourcing issues such as a lack of fingerprint or DNA databases,<sup>211, 212</sup> inadequate training,<sup>213, 214</sup> and supply chain

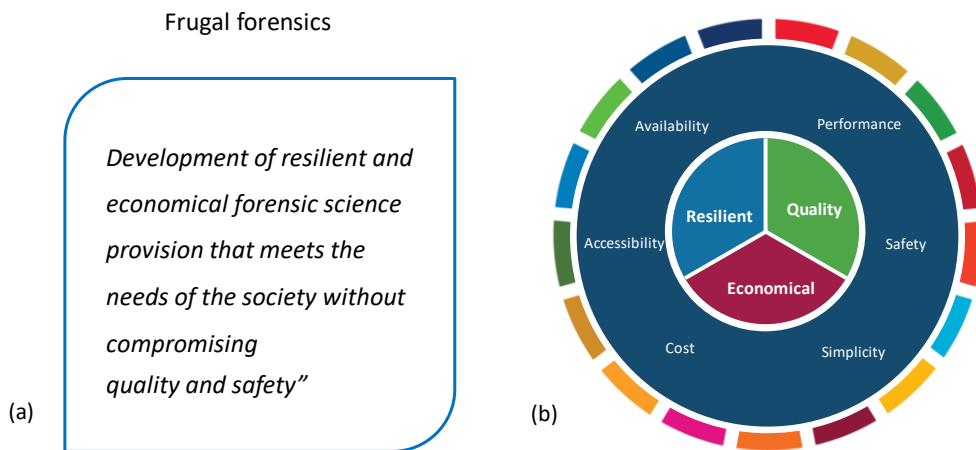
problems.<sup>196, 215</sup> Hence it is not surprising that the limited forensic research literature from the Global South primarily focuses on the challenges around developing and strengthening fundamental forensic science capability and service provision. Against this backdrop, it is important to note that latent fingerprint service provision is common across all jurisdictions and is generally a primary forensic science service in the Global South.<sup>11</sup>



**Figure 1.10:** Number of publications on “forensic science” in a simple Bibliometric search on Scopus for the (a) Total papers 1972–2022 (b) Top 10 countries and (c) Top 10.

This thesis proposes an approach to how Seychelles and, to some extent, broader Global South jurisdictions can adapt to the challenges of their locality and develop sustainable fingerprint detection provisions. This can be achieved by recognising vulnerabilities within the process and building resilient capacity to deliver innovative, simple, and economical services meeting fit-for-purpose quality benchmarks. For example, in the case of Seychelles investigating the use of more economical chemical analogues of fingerprint reagents, or sourcing of more accessible or locally available alternatives would provide a more resilient fingerprint detection method. The investigation should be carried out in a standardised framework to ensure the quality of the results.

This approach follows a concept referred to in this thesis as frugal forensics, defined as the development of resilient and economical forensic science provision that meets the needs of society without compromising quality and safety. This concept is derived from a combination of frugal innovation<sup>216</sup> and the Brundtland Commission’s definition of sustainable development.<sup>217</sup> Frugal forensics is based on three core principles; Resilience, Economics and Quality, and six attributes; Performance, Accessibility, Availability, Cost, Simplicity and Safety (PAACSS) (Figure 1.11), which underpin the sustainable provision of forensic science services. At the heart of this approach is shifting the focus from ‘pure’ performance to a holistic consideration of jurisdictional vulnerabilities while ensuring that risks or limitations are recognised and documented to ensure transparent, high-quality service provision.



**Figure 1.11:** The concept of sustainable forensic science provision (a) Definition (b) The three principles and six attributes that underpin it.

Although this approach focuses on fingerprint detection, the considerations raised in relation to economics, resilience and quality also apply to the sustainability of the broader forensic science service provision. While it may not have been explicitly defined previously, sustainable forensic science provision is not a new concept, as by nature and necessity, it is embraced in Global South countries due to their unique needs. However, its development thus far has been piecemeal and ad hoc. The challenge is for it to be systematically integrated and aligned to international standards as far as can be practically achieved. For latent fingerprint detection, the IFRG guidelines<sup>24</sup> provide an objective evaluation and reporting framework for



performance assessment of new or modified methods. The guidelines specifically recommend testing methods in the environment where they are to be operationally used, aligning with the concept of frugal forensics as ‘meeting the needs’ of the locality. The recommendation for local validation extends this approach a step further by evaluating not only the method's performance but also its sustainability in the light of local challenges.

Embracing this concept has the potential to contribute towards the United Nations Sustainable Development Goals (SDGs).<sup>9</sup> These goals consisting of 17 intertwined targets, aiming to build a set of shared global targets for a just and equitable world. The ‘SDG 16 –Peace and Justice’ goal implies that forensic science service provision is essential in achieving sustainable development. Developing a sustainable latent fingerprint detection protocol under the banner of frugal forensics (Figure 1.12) increases accessibility to forensic science service provision in jurisdictions where such provision would have been limited or otherwise absent. At the core of the SDGs is the drive to reduce inequalities between nations and to transfer resources, knowledge, technology, and capability to the Global South. The collaboration of Curtin University and the Seychelles Police Forensic Section through this thesis is a prime example.



**Figure 1.12:** Illustrating the relationship between frugal forensics and UN sustainable goals through SDG 16.<sup>218</sup>

## 1.6 Aim and Objectives

This study focuses on enhancing the accessibility of latent fingerprint detection techniques and enabling their use in remote and resource-limited jurisdictions. The research is conducted within the frugal forensics framework, which recognises the constraints of these jurisdictions to develop adaptable and fit-for-purpose detection techniques. This concept can be used to develop sustainable latent fingerprint protocols, increasing access to fingerprint detection capabilities in jurisdictions where they were previously unavailable or restricted.

The study aims to achieve four main objectives:

- Develop a sustainable IND/Zn formulation to facilitate its use in the Global South jurisdictions as a recognised most sensitive amino acid sensitive reagent for detection of fingerprints on paper substrates (Chapters 3 and 4).
- Modify a recently published carbon-based powder suspension formulation for non-porous surfaces to address the accessibility issue of commercial surfactant solutions (Chapter 5).
- Investigate Nile blue as an emerging, low-cost, and easily accessible method that can be used as a lipid-sensitive reagent and post-cyanoacrylate-fuming stain (Chapter 6).
- Develop a comprehensive evaluation tool to assess the sustainability of latent fingerprint detection methods (Chapter 7).

This thesis adheres to the IFRG guidelines, which provide best practice guidelines for evaluating new or modified fingerprint detection methods. These guidelines are essential due to the intrinsic high variation of fingerprints, and it's important to account for multiple donors, ageing conditions, and result assessment. Additionally, it fulfils quality assurance requirements to facilitate validation and its adoption of detection methods in operational laboratories. Chapter 2 outlines the experimental considerations in line with the IFRG guidelines, followed by an overview of the experimental methods and instrumentation used throughout this study.

Chapters 3 and 4 investigated how to enhance the accessibility of existing latent fingerprint detection methods for the Global South jurisdictions. Chapter 3 begins with a preliminary head-to-head comparison of three active IND/Zn formulations to identify the most suitable formulation that can achieve sustainability with minimum modifications. Subsequently the different chemical components of the formulation were assessed in the context of a frugal forensics approach. This provided direction in terms of methodology and modification of the formulation to come to a final formulation that was assessed through laboratory trials in Chapter 4, including the use of incidental fingerprints to provide a more realistic casework sample. This will provide a formulation that is more accessible to other jurisdictions and enhance the recoverability of fingerprints.

Chapter 5 investigates the use of sodium dodecyl sulfate salts to prepare an in-house surfactant solution to replace the commercial Gran Velada solution used in a recently published carbon-based WET UCIO powder suspension formulation. The study is divided into two parts; the first involves characterising pressure-sensitive tapes using ATR-FTIR analysis. The second part consists of modifying the WET UCIO formulation and assessing it in comparison to the original and commercial powder suspension formulations. This will improve the accessibility of the low-cost WET UCIO formulation to other jurisdictions.

Chapter 6 investigates the benzophenoxazine dye Nile blue as an emerging lipid-sensitive latent fingerprint detection technique. Firstly, its effectiveness as a lipid-sensitive method was compared to ORO on wet porous surfaces. Secondly, its potential as a post-cyanoacrylate stain was investigated by comparing it with BY40. Additionally, the staining mechanism was explored using confocal Raman microscopy. This gives an insight into the use of acid Nile blue as a frugal forensics method and a fundamental understanding of its staining mechanisms that can assist in developing post-cyanoacrylate dyes.

Chapter 7 addresses how the methods investigated in Chapters 3-6 can be objectively evaluated within the frugal forensics framework. This involves developing an assessment tool for a holistic evaluation of the frugal forensics attributes in

consideration of the jurisdiction-specific challenges. This tool can assist with decision-making and identify areas that need modification to achieve sustainability. Moreover, it provides a transparent and objective evaluation for end-users of forensic information and aligns with ensuring quality assurance in fingerprint service provision.

## **Chapter 2. Sample collection, experimental methods, and instrumentation**

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## **2.1 Introduction**

This chapter outlines the experimental methods used in the study. It begins with an overview of the methodology and experimental considerations, followed by discussion of the fingerprint collection procedures and an outline of the fingerprint development methods. Finally, it provides details of the instrumentation, fingerprint grading system and statistical analysis used in this study.

## **2.2 Experimental considerations**

### **2.2.1 IFRG guidelines**

Fingerprints are intrinsically highly variable, and the methods used to enhance them vary greatly, making it necessary to have a standardised approach to compare new or modified enhancement techniques. A standardised approach was highlighted by Kent in 2010<sup>219</sup> and Sears *et al.* in 2012<sup>19</sup> due to inconsistencies in the methodology and reporting of fingerprint detection research. In response, the International Fingerprint Research Group (IFRG) published guidelines<sup>24</sup> in 2014, providing a standardised research framework for assessing detection techniques from the initial concept to the final casework implementation. This thesis adheres to IFRG guidelines with deviations clearly stated and the reason for the deviation.

The framework divides the research process into four phases: initial proof-of-concept studies (phase 1), optimisation and comparative assessment with an established method (phase 2), validation using large numbers and more realistic samples (phase 3) and operational evaluation (phase 4). As a method progresses through different phases, the research methodology changes to manage the number of samples while obtaining meaningful results. For example, phase 1 uses a smaller sample size, while phases 3 and 4 have more rigorous evaluations using realistic samples. The different experiment in this study covers phase 1 to phase 3 and broadly consist of two strands. Firstly, method development and preliminary comparative study to improve relevant parameters in line with the frugal forensics approach. Secondly, if the modified method is deemed viable, it was then compared to existing techniques through

laboratory trials on incidental fingermarks or simulated casework substrates to assess its operational suitability.

### **2.2.2 Frugal forensics approach**

The method progression also depends on the resources at the disposal of researchers and the operational laboratories.<sup>24</sup> The aim of this thesis is to address the challenges posed by the limited availability and accessibility of resources in some jurisdictions, which hinder the use of effective latent fingerprint detection methods. Therefore, using minimal equipment for operational simplicity, cost-effectiveness, and health and safety are underlying considerations.

### **2.2.3 COVID-19 pandemic impact**

Due to part of the project coinciding with the COVID-19 pandemic, consideration had to be taken given the limited availability and access to donors. The number of donors was kept at a minimum but within the recommendation of the IFRG guidelines. Additionally, laboratory trials intended to be conducted in Seychelles were scaled back to Western Australia (WA) due to travel restrictions in Western Australia and the Seychelles that lasted until mid-2022.

## **2.3 Fingerprint methodology**

The standardised methodology limits fingerprint variability by addressing key variables such as the number of donors, collection and storage procedures, substrates and environmental conditions, development and visualisation conditions, and analysis and reporting. As a result, experiments are designed to reduce the number of uncontrollable variables as well as account for the intrinsic variability of fingerprints.<sup>19, 24, 219</sup>

### **2.3.1 Donor information**

To ensure accurate results fingerprint reagent testing is carried out on a range of donors across both biological sexes and covering a wide range of ages. A total of seventeen donors participated in this study, nine females and eight males aged 22-

60 years. The IFRG recommends using 5-15 donors for phase 2 and  $\geq 20$  donors for phase 3. Due to COVID-19, the study used five donors for the initial study and ten for laboratory trials. However, the number of samples for the laboratory trials was increased, as studies have shown that meaningful results can be obtained either in the form of a large donor pool or a large number of marks per donor.<sup>220</sup>

### **2.3.2 Fingermark collection procedures**

Fingermark donors were instructed to lightly rub their hands together and provide natural fingermarks by lightly pressing the finger on the substrate surface for  $\sim 5$  seconds. Care was taken to ensure that the donor had not washed their hands, eaten or come into contact with chemicals in the 30 minutes prior to sample collection to reduce contamination and ensure a natural build-up of surface secretions. Multiple samples collected from the same fingermark donor were collected at least with 30 minutes intervals between depositions of each sample to allow secretions to replenish on donors' fingertips.

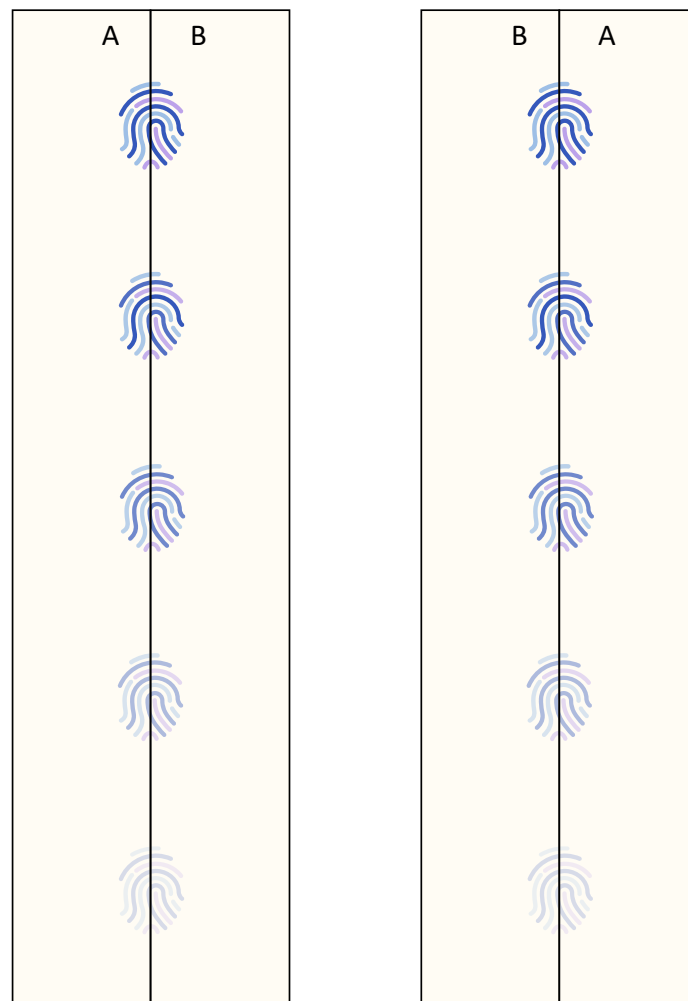
Natural fingermarks, which have not been intentionally groomed for increased material, were used throughout this study, except in Chapter 6, where charged fingermarks were used to investigate the selectivity and donor effect of the lipid-sensitive acid Nile blue method. For charged fingermarks, donors were instructed to wipe their fingers across the face or forehead prior to the deposition of marks.

In casework, fingermarks are not deliberately deposited; incidental fingermarks provide a more realistic sample in assessing fingermark detection method in line with phase 3 of the IFRG guidelines. Incidental fingermarks deposited through the process of inspecting the contents of an envelope, simulating activity in cases of theft, were used in Chapter 4. When charged or incidental fingermarks are used, it is clearly outlined in the relevant chapters.

Split depletion series fingermark collection method was used in this study unless stated otherwise. When comparing the effectiveness of two fingermark development techniques, the split depletion method is preferred for fingermark collection. It involves depositing a series of fingermarks, dividing it in half, and processing each



half with a different technique before recombining for assessment. This allows for a direct comparison of the two techniques within a single fingermark, which reduces variability in the amount and composition of the deposited fingermark material.<sup>19, 24</sup> The use of depletion series allows for testing for sensitivity, with progressively less material available for detection with successive deposition (Figure 2.1). To minimise donor biasing on one side of the finger, a second depletion series was collected from the same donor in order to reverse the development process on the opposite side.



**Figure 2.1:** Schematic illustrating split fingermark and depletion series methodology for fingermark collection and the reverse of development process A and B to minimise donor biasing on one side of the finger.

An additional technique that can be employed to quickly evaluate a formulation or make small adjustments is the quartered fingermark approach.<sup>19</sup> This method entails

placing a single natural fingerprint at the centre of a substrate that is sectioned into four equal parts. Subsequently, the fingerprint is divided into four parts, with each quarter treated with different formulations or the same formulation that varies by only one component, such as the processing time. Quartered fingerprints methodology was used in Chapters 4 and 6.

### **2.3.3 Fingerprint ageing and storage conditions**

Fingerprints samples were stored under ambient conditions in an office cupboard (ca. 20 °C, 56 %RH) for at least 24 hours prior to development unless stated otherwise. To study the effects of sample age, samples were stored under the same conditions for 7 and 30 days. These periods were chosen because fingerprint composition undergoes the most changes within the first 24 hours<sup>21</sup>, and the typical time frame casework samples are likely to be processed. Other storage conditions including samples stored under non-airconditioned environment to simulate the condition in Seychelles and samples submerged in water to simulate wet surfaces, was used in Chapter 4 and 6 respectively. The details of each condition are provided under the relevant chapters.

## **2.4 Substrate selection and environmental considerations**

### **2.4.1 Substrate surface selection**

The IFRG guidelines<sup>24</sup> recommends testing under local conditions and substrates. Since the experiments were conducted in WA, every effort was made to source representative samples encountered in casework in Seychelles. This includes cardboard, a common surface encountered by the Seychelles Police. However, most substrates encountered in Seychelles are imported from multiple overseas sources, creating a diverse and dynamic situation. The substrates used in this study are specified in the experimental section in Chapters 3-6.

### **2.4.2 Environmental considerations**

As part of this study, it was intended that the final laboratory trials be conducted in Seychelles; however, due to COVID-19 travel restrictions the experiments had to be

conducted in WA. Considerations were made regarding the difference in temperature and humidity between the two locations, which may affect IND/Zn efficacy as outlined in Chapter 4. A preliminary experiment under elevated humidity and temperature to mimic the tropical conditions of Seychelles was conducted to evaluate the suitability of IND/Zn formulation under such conditions.

## 2.5 Fingerprint development

In this study, several fingerprint detection methods in line with some of the current capabilities (Table 2.1) of Seychelles Police were investigated. Chapters 3 and 4 investigated IND/Zn formulations, Chapter 5, carbon-based powder suspension formulations and Chapter 6, acid Nile blue, ORO, cyanoacrylate and BY40. The following section outlines the details of each development procedure, including reagent information, solution preparation and development methodology.

**Table 2.1** Summary of some of the chemical detection capabilities of the Seychelles Police.

Detection method	Reagents	Equipment
Amino acid	Ninhydrin	Treatment: Spray bottle and extractor Development: Overnight using ambient temperature
Powder suspension	Wetwop black and Wetwop white	Treatment: Brush, wash bottle, glass tray
Superglue fuming	Cyanoacrylate	Treatment: Cyanoacrylate fuming chamber
Superglue fluorescent dye staining	Basic yellow	Treatment: Wash bottles and glass trays Visualisation: Polilight, Nikon DSLR camera

### 2.5.1 1,2-Indanedione zinc

1,2-Indanedione is an amino acid sensitive fingerprint reagent commonly utilised to develop fingerprints on porous surfaces such as paper. In this study, three operationally active IND-Zn formulations were used, published by i) the National Centre for Forensic Studies (NCFS),<sup>221</sup> in use in Australia, ii) Bundeskriminalamt

(BKA),<sup>222</sup> in use in Germany, and iii) the Centre of Applied Science and Technology (CAST),<sup>127</sup> in use in the UK.

### **Materials**

1,2-Indanedione (Reddy Chemtech), ethyl acetate ( $\geq 99.7\%$ , UNIVAR - APS), glacial acetic acid ( $\geq 99.7\%$ , Lab-Scan), methanol ( $\geq 99.7\%$ , Honeywell – Burdick & Jackson), ethanol ( $\geq 99.5\%$ , UNIVAR - Ajax Finechem), zinc chloride ( $\geq 98\%$  Sigma Aldrich), HFE-7100 (3M Novec), petroleum ether 40-60 °C (VWR Chemicals), Solstice® PF (Honeywell). Reagents were used as received without further purification.

### **Sample preparation**

NCFS formulation was prepared as outlined by Stoilovic *et al.*,<sup>221</sup> 0.598 g of 1,2-indanedione dissolved in 124.8 mL of ethyl acetate and 5.2 mL of glacial acetic acid. 870 mL of carrier solvent (HFE-7100 or Solstice® PF) was added, followed by 4 mL of zinc chloride stock solution (8 g zinc chloride in 200 mL of ethanol).

BKA formulation was prepared as outlined by Becker *et al.*,<sup>222</sup> 1.0 g of 1,2-indanedione dissolved in 45 mL of ethyl acetate and 10 mL of glacial acetic acid. 900 mL of carrier solvent (petroleum ether 40-60°C) was added, followed by 10 mL of zinc chloride stock solution (200 mg zinc chloride in 30 mL of ethanol).

CAST formulation was prepared as outlined by Nicolasora *et al.*,<sup>127</sup> 0.250 g of 1,2-indanedione dissolved in 45 mL of ethyl acetate, 10 mL of glacial acetic acid and 45 mL of methanol. 1000 mL of carrier solvent (HFE-7100 or Solstice® PF) was added, followed by 1 mL of zinc chloride stock solution (100 mg zinc chloride in 4 mL of ethyl acetate and 1 mL acetic acid).

The developed SPF IND/Zn formulation was prepared using 0.60 g of 1,2-indanedione dissolved in 45 mL of ethyl acetate and 10 mL of glacial acetic acid. 900 mL of carrier solvent (petroleum ether 40-60 °C) was added, followed by 10 mL of zinc chloride stock solution (200 mg zinc chloride in 30 mL of ethanol).

All formulations of 1,2-indanedione zinc were stored in an amber glass bottle away from sunlight.

## **Sample development**

The samples were developed by submerging them in IND/Zn working solution (~150) in a glass tray for 5-10 seconds, then air dried in a fume hood. Fresh working solution was used after every ten samples treatment or when the solution turned cloudy. All treated dry samples were processed in a heat press at 160 °C for 10 s in line with the NCFS and BKA methods. This processing condition was chosen as a simple and cost-effective method that aligns with frugal forensic approach compared to oven processing at 100 °C, which CAST recommends. The SPF method follows the same sample development as the BKA and NCFS methods. All treated samples were photographed and stored in a laboratory cabinet to limit light exposure.

### **2.5.2 Powder suspension**

Powder suspension is a method that can be used to develop latent fingerprints on adhesive surfaces such as the sticky side of pressure sensitive adhesive (PSA) tapes. In this study, various powder suspension formulations were used; two commercial pre-mixed formulations Wetwop™ black and Wet Powder black, and a recently published<sup>175</sup> WET UCIO formulation including modified versions of it outlined in Chapter 5.

#### **Materials and sample preparation**

Wet UCIO formulation was prepared as outlined by Claveria,<sup>175</sup> 1.5 g of carbon black powder (Sirchie) was mixed with 10 mL of 27 % w/v sodium dodecyl sulfate solution (Gran Velada) to make a paste similar in texture to thin paint.

Modified WET UCIO powder suspension formulations were prepared by substituting the Gran Velda solution with in-house surfactant solutions of various SDS concentrations and additives. SDS salt (laboratory grade, Chem Supply), absolute ethanol (analytical grade, Scharlab), polyethylene-400 (laboratory grade, Chem Supply) and deionised water were used to prepare in-house SDS surfactant solutions using volumetric glassware. The composition of the various modified Wet UCIO powder suspension formulations and the in-house surfactant solutions utilised in this study are specified in Chapter 5.

Commercial powder suspension products Wetwop™ Black (Lightning Powder Company) and Wet Powder Black (Kjell Carlsson Innovation) were manually agitated for 10 seconds before use. A working solution of approximately 10 mL was poured into a clean beaker.

### **Sample development**

The application of all powder suspensions followed the same methodology. A separate clean squirrel brush was used for each powder suspension formulation to paint the paste onto the sticky side of adhesive tapes and left for 10-15 seconds. The tapes were rinsed with cold tap water, air dried, and photographed.

### **2.5.3 Oil red O**

ORO is a lipid-sensitive reagent for development of fingermarks on porous surfaces. In this study, a modified ORO formulation reported by Frick *et al.*,<sup>137</sup> was used.

### **Materials and sample preparation**

ORO was prepared as outlined by Frick *et al.*,<sup>137</sup> 0.50 g of ORO (Agros Organic, ChemSupply Australia) was dissolved in 1000 mL of propylene glycol (Laboratory grade, ChemSupply Australia) and heated to 95 °C with constant stirring until fully dissolved. The solution was cooled to room temperature and stored in an amber glass reagent bottle in laboratory cabinet.

### **Sample development**

The samples were developed by submerging in ORO solution (~150 mL) in a glass tray for 15 minutes with manual agitation for 30 seconds at the beginning of the treatment. The samples were rinsed with deionised water, air dried on paper towels, and photographed.

### **2.5.4 Cyanoacrylate fuming**

Cyanoacrylate fuming of latent fingermarks was conducted using an improvised non-temperature and humidity controlled chamber used in previous work by McGann *et al.*<sup>187</sup> and Boseley *et al.*<sup>188</sup> at Curtin University. The available laboratory conditions

have demonstrated appropriate development on non-porous surfaces following the procedure outlined below.

### **Materials, sample preparation and development**

Cyanoacrylate fumed print was prepared by placing samples in an improvised chamber (Ikea Socker Greenhouse, 35cm x 45 cm x 22 cm). Three drops (~1.5 g) of Loctite 401 instant adhesive ( $\geq 99.5$  % w/w ethyl-2-cyanoacrylate, Henkel Australia; Blackwoods) contained in small piece of aluminium foil (Capri Heavy Duty Catering Foil, China) was placed in the middle of the chamber. The chamber was sealed with tape and the samples were allowed to develop for 2 hours (no fan). Following development, the samples were removed and allowed to cure overnight inside a laboratory cabinet.

### **2.5.5 Acid Nile Blue**

In this study, acid Nile Blue was used as a lipid-sensitive reagent for the development of fingermarks on wet porous substrates as well as a post-cyanoacrylate stain. The treatment of porous surfaces and post-cyanoacrylate fumed prints follow the similar procedure as outlined below.

### **Materials and sample preparation**

Acid Nile was prepared as outlined by Crocker *et al.*,<sup>154</sup> 0.05 g (6.8  $\mu\text{mol}$ ) of Nile blue (analytical grade, Sigma-Aldrich, USA) was dissolved in 1000 mL of 0.3 M sulfuric acid (analytical grade, Ajax Finechem) and heated to 95 °C for 1 hour with constant stirring. The solution was cooled to room temperature and stored in an aluminium-wrapped glass reagent bottle in laboratory cabinet to avoid photo degradation.

### **Sample development**

The samples were developed by submerging in acid Nile blue solution (~150 mL) in a glass tray for 20 minutes with agitation of the solution across the samples. The samples were rinsed with deionised water, air dried on paper towels, and photographed. All treated samples were stored in a laboratory cabinet to limit light exposure.

### **2.5.6 Basic Yellow 40**

Basic Yellow 40 is a post-cyanoacrylate stain and was used in this study for comparative assessment with acid Nile blue.

#### **Materials and sample preparation**

BY40 was prepared as outlined by Bandey *et al.*,<sup>16</sup> 1.0 g of BY40 (ChemSupply Australia) was dissolved in 500 mL ethanol ( $\geq 99.5\%$ , UNIVAR - Ajax Finechem). The solution stored in an aluminium-wrapped glass reagent bottle in laboratory cabinet.

#### **Sample development**

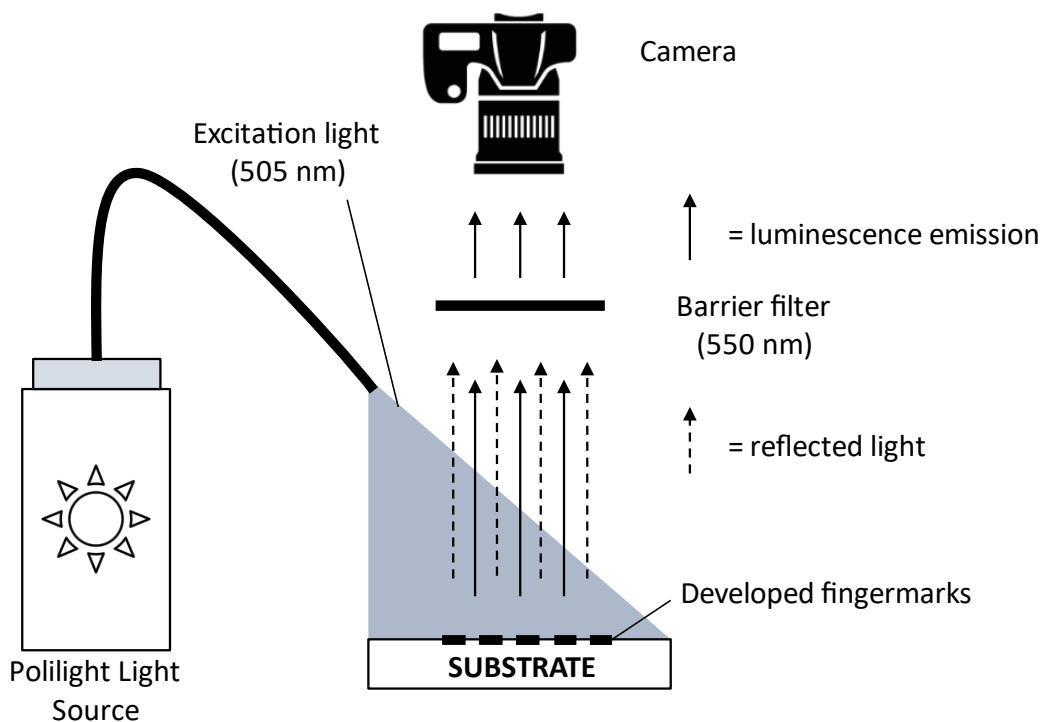
The samples were developed by submerging in BY40 solution (~100 mL) in a glass tray for 10-15 seconds. The samples were rinsed with deionised water, air dried on paper towels, and photographed.

## **2.6 Instrumentation**

### **2.6.1 Photography of developed fingermarks**

Developed fingermarks were photographed using a Nikon D300 camera on manual exposure mode, mounted on a Firenze Mini Repro stand at 35 cm above the samples and fitted with a 60 mm lens. Samples were illuminated using a Polilight PL500 (Rofin Australia Pty. Ltd., Australia) or dual incandescent light globes (Mirabella). For luminescence examination a long-pass barrier filter with 1% transmittance was fitted on the camera lens to block the reflected light and allow the emitted longer wavelength to pass through as illustrated in Figure 2.2. The camera settings, illumination and barrier filter specification optimised for each development method are shown in Table 2.2. Camera aperture f/11 and ISO200 were consistent across all methods. All digital images were recorded using Nikon Camera Control Pro (version 2.31.0).





**Figure 2.2:** Schematic of photographic setup for photoluminescence fingerprints examination.

**Table 2.2:** Photographic conditions optimised to each detection method.

Detection Treatment	Mode	Illumination	Camera filter	Shutter speed
1,2-indanedione zinc	Photoluminescence	Polilight 505 nm	550 nm	1/2 sec
Powder suspension	Absorbance	2 incandescent globes	None	1/10 sec
Cyanoacrylate fuming	Diffused reflectance	2 incandescent globes	None	1/10 sec
Acid Nile Blue	Photoluminescence	Polilight 505 nm	550 nm	1/2 sec
Basic yellow 40	Photoluminescence	Polilight 450 nm	495 nm	1/2 sec

### 2.6.2 Fluorescence spectroscopy

Fluorescence spectroscopy was used in Chapter 3 to investigate the observable variation in luminescence intensity with IND/Zn due to variation in 1,2-indanedione concentration. The measurements were conducted on amino acid spots containing

alanine, leucine and phenylalanine. The amino acids phenylalanine and leucine were used as representative for aromatic and aliphatic amino acids, and alanine is commonly used as a model for amino acids due to its faster reaction rate and fewer side reactions compared to glycine.<sup>104, 223</sup>

Amino acid solution was prepared by dissolving 200 mg of L-alanine (Sigma Aldrich, 98% w/w) in 20 mL of MilliQ water, and the solution was mixed thoroughly. The procedure was repeated with L-phenylalanine and L-leucine (Sigma Aldrich, 98% w/w), and then 5 mL from each solution was combined to produce a mixture of amino acid solution. From the mixture of 10 mg/mL amino acid concentrations, series dilution was used to produce solutions of 5 mg/mL, 1 mg/mL and 0.5 mg/mL.

The substrate samples were prepared in strips, each with five spots in the form of circles (5 mm radius) spaced 20 mm apart, drawn with a pencil. Each spot was impregnated with 1  $\mu$ L aliquots of the amino acid solution using a micropipette. The samples were stored and treated similarly to other fingerprint samples. Each strip was treated with BKA IND/Zn formulation containing different concentration of 1,2-indanedione. One strip was also treated with a formulation of IND/ZN without 1,2-indanedione zinc. This was used as control samples to verify for contamination that may cause photoluminescence.

Fluorescence spectra were obtained using a Cary Eclipse Fluorescence Spectrophotometer (Agilent) combined with a fibre optic probe for *in situ* measurements. The extended probe tip was placed on the substrate ensuring a consistent distance between the probe to the amino acid spot during measurements (Figure 2.3). The excitation wavelength was set at 505 nm, and emission spectra were collected between 540 and 650 nm with a slit width of 5 nm. A background spectrum of each untreated substrate was collected prior to sample analysis. The data acquisition was carried out using the Agilent Cary Eclipse Scan Application (Version: 1.2(147)) and exported as C.S.V file for analysis in Excel.



**Figure 2.3:** *Experimental setup using the fibre optic probe for fluorescence measurements of amino acid spots treated with IND/Zn formulation containing different 1,2-indanedione concentrations.*

### **2.6.3 UV-vis spectroscopy**

Visible spectroscopy was used in Chapter 4 to assess the stability of the SPF IND/Zn reagent by monitoring the 1,2-indanedione peak absorbance over time. Spectral scans of the SPF IND/Zn working solutions were performed using an Agilent Cary 60 UV-Vis Spectrophotometer. An Agilent rectangular quartz cuvette (10 mm pathlength) was used, and sample was scanned in the visible wavelength region (400–700 nm) with a scan interval of 0.5 nm and scan speed of 300 nm/s. A corresponding reference blank contained solvents as the sample but did not contain 1,2-indandione, was run as a blank-corrected baseline. The data acquisition was carried out using the Agilent Cary WinUV Scan Application (Version 2.00) and exported as C.S.V file for analysis in Excel.

#### **2.6.4 ATR-FTIR spectroscopy**

Attenuated total reflection - Fourier transform infrared (ATR-FTIR) spectroscopy was used to analyse the composition of the backing and adhesive of PSA tapes in Chapter 5. A Thermo Scientific Nicolet iS50 Fourier Transform Infrared (FTIR) spectrophotometer with a single-bounce attenuated total reflectance (ATR) diamond crystal was used to obtain  $4\text{ cm}^{-1}$  resolution spectra in absorbance mode over a range of  $4000 - 400\text{ cm}^{-1}$ , with 64 accumulated scans. For each tape, a ca. 5 cm strip was cut and placed over the crystal, and spectral analysis was performed using the pressure arm to maintain a consistent contact pressure. The backing and the adhesive side were analysed using separate strips with measurements taken at three locations for better representative composition. The crystal was cleaned with ethanol between different tape sample measurements, followed by a background scan. ATR correction was applied to each spectrum using the in-built software (Omnic 9, Thermo Fisher Scientific), with the replicate spectra then averaged and exported as C.S.V files for analysis.

#### **2.6.5 Confocal Raman microscopy**




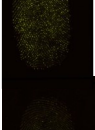
To investigate the post-cyanoacrylate staining mechanism in Chapter 6, Confocal Raman microscopy was used to investigate the position of the stain in relation to the cyanoacrylate polymer. All samples were prepared on 1 mm borosilicate microscope slides (VWR International, Leuven). Dye samples were prepared by placing one drop of each dye onto a microscope slide. For the fingerprint samples, charged split fingerprints were deposited on two microscope slides secured side by side with cellulose tape. Three types of fingerprint samples were prepared: unstained cyanoacrylate fumed, acid Nile blue post-cyanoacrylate stain and BY40 post-cyanoacrylate stain fingerprints. All fingerprints were developed according to the methods outlined in sections 2.5.4 to 2.5.6. Care was taken to ensure the fingerprints were intact along the physical boundary between the two slides during fingerprint development. After treatment the slides were carefully separated to examine the cross-section of the fingerprints using Confocal Raman Microscopy.

Confocal Raman microscopy studies were conducted using a confocal Raman microscope (alpha 300R, WITec, Ulm, Germany) equipped with a 532 nm frequency doubled Nd:YAG laser and a thermoelectrically cooled charge-coupled device (CCD) detector. The laser light was coupled into the microscope using a single-mode fibre and brought on to the sample using a dichroic mirror and a 100× microscope objective (NA 0.9). The spatial resolution is about 300 nm with a spectral resolution of 0.02 cm<sup>-1</sup>. The sample was mounted onto a piezo driven scanning stage with a position accuracy x and y directions. The WITec Control FOUR software was used for data acquisition and WITec Project FOUR 4.1 for spectral processing. Raman images were acquired with a scan speed of 18.6 s/line and 0.23 s integration time. A stress-free sample silicon wafer was used to calibrate the reference peak position.

## 2.7 Fingerprint grading

To assess development performance, photographs of fingerprints were graded on ridge detail and contrast using a modified 5-point system from the UK Home Office CAST.<sup>19</sup> A photographic representation of the CAST system 1,2-indanedione is shown in Table 2.3. Fingerprint grades were further categorised into groups, particularly grades 3 or 4, which were considered 'useful' for identification.<sup>220</sup> The UC (University of Canberra) scale shown in Table 2.4 was also used in Chapters 3 and 6 as a comparative scale to assess the relative performance between two treatments based on the difference in ridge details and contrast.<sup>224</sup> However, the CAST scale was found to be more effective for providing additional information on the general trends across the different techniques. It is also compatible with categorizing grades and making comparisons with other studies. For this reason, all results presented are based on the CAST assessment scale unless stated otherwise. The fingerprints were graded by two evaluators with experience with fingerprint grading system and each grading a different set of experiments. The use of a grading system is subjective to human bias; however, studies show that independent fingerprint graders produce reliable data for assessing fingerprint quality.<sup>225</sup>

**Table 2.3:** *Fingerprint grading scale and grading classification used in this study.*<sup>19, 220</sup>

Grade	Friction ridge detail developed	Contrast of ridge detail	Photographic examples	Classification
4	Very strong development: full ridge details	Very good contrast		Useful for comparison
3	Strong development more than 2/3 of fingerprint continuous ridges	Good contrast		Useful for comparison
2	Limited development, ridge details present but not likely to be used for identification purposes	Moderate contrast		Detected, but not suitable for comparison
1	Weak development; evidence of contact but no ridge detail	Poor contrast		Detected, but not suitable for comparison
0	No fingerprints detected	No contrast		No fingerprints detected

**Table 2.4:** UC (University of Canberra) comparative scale to assess the relative performance of two detection methods for split fingerprints

Score	Definition
2	Half-print treated with method A exhibits far greater ridge detail and or contrast than the corresponding half-print treated with method B
1	Half-print treated with method A exhibits slightly greater ridge detail and or contrast than the corresponding half-print treated with method B
0	No significant difference between the corresponding half-print
-1	Half-print treated with method B exhibits slightly greater ridge detail and or contrast than the corresponding half-print treated with method A
-2	Half-print treated with method B exhibits far greater ridge detail and or contrast than the corresponding half-print treated with method A

## 2.8 Statistical analysis

When performing statistical analysis on fingerprint grades results, it is important to recognise that CAST grades are ordinal data hence a categorical scale. Therefore, most parametric tests and descriptive statistical analysis, such as means and averages are not appropriate.<sup>226</sup> Instead, the non-parametric counterparts are

recommended. In this study, the non-parametric tests Mann-Whitney U test and Kruskal-Wallis test were used to assess whether the grades are higher or lower on average between two or more treatments. A chi-square test was used to assess whether the treatments have different distributions in fingerprint grade frequencies. Additionally, the only accepted parametric test, the difference of proportions test, was performed to assess whether the proportion of grade 3s and 4s (useful fingerprints) of one treatment differs from the other.

Statistical analysis software R version 4.3.0 was used to assess whether there was significant difference in the fingerprint grades between different IND/Zn treatments in Chapters 3 and 4, and between different powder suspension formulations in Chapter 5. The statistical analysis was performed by transforming the data into vectors in Excel. Vectors are the basic data structure in R and are created by using the `c()` function to combine variables of the same type, separated by commas. Repeated vectors or frequency of variables can be represented using the `rep()` function. Figure 2.4 shows an example of a fingerprint grade frequency table transformed into vectors in Excel. The vectors were input as R codes to compute the statistical analysis in R. All statistical tests were conducted at 95% confidence level ( $p < 0.05$ ). All statistical data analysis including fingerprint grade frequency table, R codes and results are provided in Appendix A.

Grade frequency table			Grade frequency as vectors		
Grades	Treatment A	Treatment B	Vector type	Treatment A	Treatment B
Grade 0	136	130	As repeated vectors to compute Mann-Whitney U test and Kruskal-Wallis test $\Rightarrow$	<code>c(rep(0,136), rep(1,67), rep(2,93), rep(3,202), rep(4,102))</code>	<code>c(rep(0,130), rep(1,69), rep(2,94), rep(3,192), rep(4,115))</code>
Grade 1	67	69			
Grade 2	93	94			
Grade 3	202	192	As frequency distribution to compute chi-squared test	<code>c(136,67,93,202,102)</code>	<code>c(130,69,94,192,115)</code>
Grade 4	102	115			

**Figure 2.4:** Illustrating transformation of fingerprint grades frequency results (left) into vectors (right), the basic data structure in R code to compute statistical analysis.

## **2.9 Human ethics approval**

This study required low-risk ethics approval to collect samples from human participants, in accordance with the National Health and Medical Research Council's (NHMRC) National Statement on Ethical Conduct in Human Research (2007). Ethics approval HRE2020-0162 was granted by the Curtin University Human Research Ethics Committee on the 6th of April 2020 prior to sample collection. As part of the requirements of approval, each participant received an information sheet and consent form, which provided information about; i) the study and its purpose, ii) privacy and confidentiality of their information, iii) the risk from participating iv) the voluntary participation in this study, and v) their rights to withdraw from the study at any time. The ethics approval, the participant information sheet and consent forms are provided in Appendix D.



## Chapter 3. A sustainable 1,2-indanedione zinc formulation

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Portions of this chapter have been published in the following articles:

Jemmy T. Bouzin, Aaron J. Horrocks, Georgina Sauzier, Stephen M. Bleay, and Simon W. Lewis. Comparison of three active 1,2-indanedione-zinc formulations for fingerprint detection in the context of limited resources and supply chain risks in Seychelles. *Forensic Chemistry* **2022** 30 100439.

DOI: <https://doi.org/10.1016/j.forc.2022.100439>

Jemmy T. Bouzin, Amanda A. Frick, Georgina Sauzier, and Simon W. Lewis. Preliminary evaluation of Solstice® PF as a replacement carrier solvent for Australian fingerprint detection. *Forensic Science International* **2022** 340 111465.

DOI: <https://doi.org/10.1016/j.forsciint.2022.111465>

### 3.1 Introduction

1,2-indanedione zinc (IND/Zn) has been widely recognised as one of the most effective amino acid treatments for the detection of latent fingerprints on porous substrates.<sup>82, 124, 125</sup> Many jurisdictions have adopted it as a primary technique to increase the recoverability of latent fingerprints.<sup>122-126, 227</sup> However, the Seychelles Police have not yet implemented the use of IND/Zn due to validation studies in the UK having only recently been carried out in 2018,<sup>125, 127, 128</sup> noting that Seychelles Police generally adopts fingerprint detection technique from the UK Fingerprint Visualisation Manual (FVM). Secondly, and more importantly, there are concerns regarding the cost and accessibility of the carrier solvent HFE-7100 (methyl nonafluorobutyl ether), as discussed in Chapter 1. This raises questions about the operational sustainability of the method in a small and remote jurisdiction like Seychelles. This chapter aims to identify and develop a sustainable formulation of IND/Zn that can be used in Seychelles.

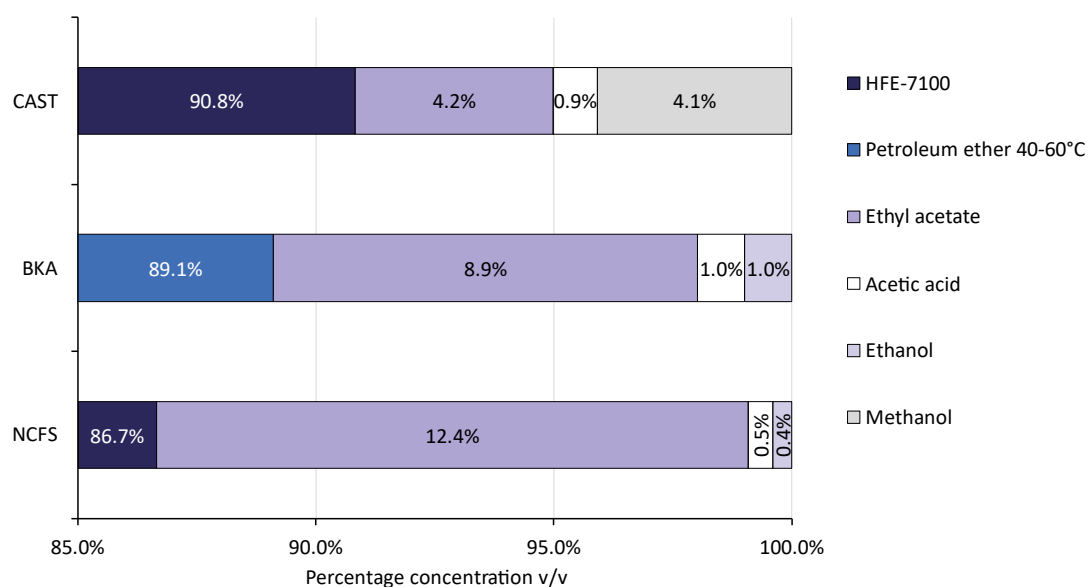
There are several published IND/Zn formulations, and as a starting point, three active formulations used by police agencies worldwide were identified. These were a formulation recommended by the Australian National Centre for Forensic Studies (NCFS)<sup>17</sup> and used by the Australian Federal Police; a formulation developed by the Bundeskriminalamt (BKA)<sup>222</sup> in use by the German Federal Police, and a formulation developed by the Centre of Applied Science and Technology (CAST)<sup>127</sup>, presently Defence Science and Technology Laboratory, in use in the UK. The CAST formulation was chosen as the default formulation for adoption by the Seychelles Police, given their usual implementation from the UK FVM. The NCFS formulation was used as a benchmark for testing under Australian conditions, while the BKA formulation was chosen for its use of an alternative carrier solvent to HFE-7100.

The composition of IND/Zn formulations used in different countries can vary significantly for reasons ranging from reagent performance under local conditions to safety. These differences can be grouped into three categories: (i) the active ingredients consisting of 1,2-indanedione and zinc chloride; (ii) the co-solvents, which help dissolve the active chemical and provide the appropriate pH conditions

for reaction with amino acids; and (iii) the carrier solvent, which acts as a medium to transport the active ingredients to the amino acid sites of the fingerprint deposit in the substrate and evaporate quickly. Table 3.1 and Figure 3.1 illustrate the differences in composition between the NCFS, BKA and CAST IND/Zn formulations.

**Table 3.1:** Mass in grams and molar ratio of active material present in one litre of NCFS, BKA and CAST IND/Zn working solution.

	Mass in grams per litre of working solution		
	NCFS	BKA	CAST
1,2-indanedione	0.596	0.990	0.227
Zinc chloride	0.159	0.066	0.018
Molar ratio IND:Zinc	3.5:1	14:1	12:1

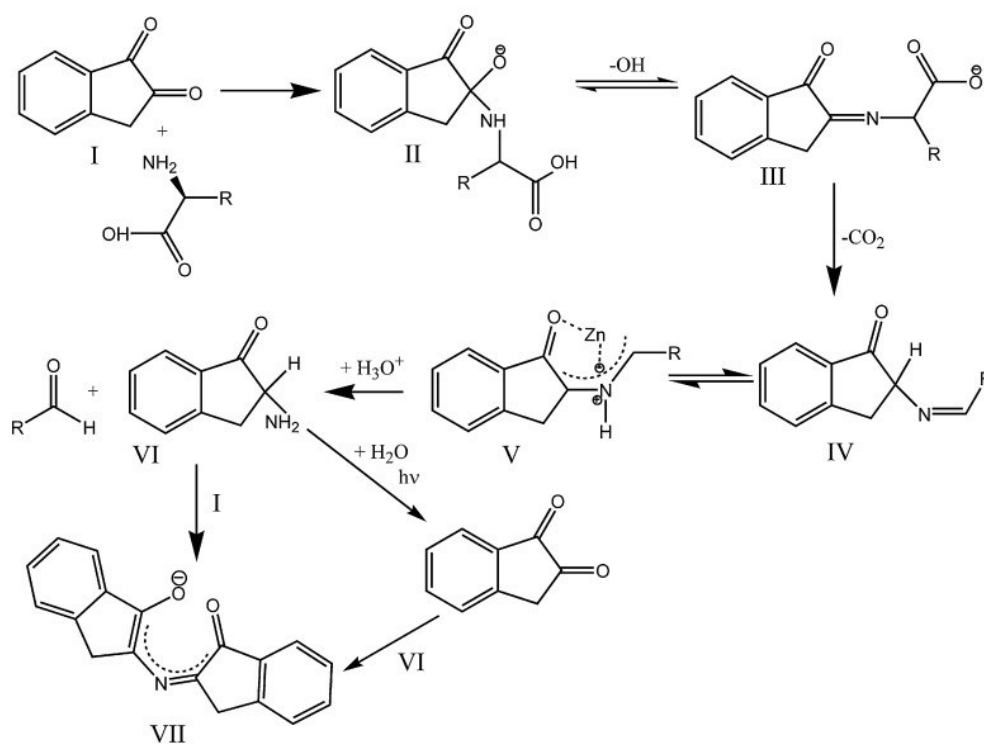


**Figure 3.1:** Percentage concentration v/v of carrier solvents and co-solvents in NCFS, BKA and CAST IND/Zn working solution.

### Active chemicals

Initial studies into 1,2-indanedione reagents showed that the interaction between 1,2-indanedione and amino acids is influenced by several factors, including the ambient humidity, the type of solvent used, the chemistry of the paper substrate and the presence of heat.<sup>110, 115, 117, 119-122, 223, 227-229</sup> Compared to earlier formulations, the addition of zinc chloride by Australian researchers was found to increase

luminescence intensity and produce more reproducible results regardless of local temperature and humidity conditions.<sup>115, 122</sup> Consequently, most 1,2-indanedione formulations contain zinc chloride in the working solution, usually in the order of 1:4 to 1:25 to indanedione (Table 3.1). Spindler *et al.*<sup>104</sup> demonstrated that zinc (II) plays a crucial role as a Lewis acid catalyst in stabilising the intermediate V in the Strecker degradation reaction of 1,2-indanedione with amino acids (Figure 3.2). The interactions of zinc (II) with the 1,3-dipole of intermediate V, accelerate the hydrolysis process to 2-amino-1-indanone (VI), which is otherwise a rate-limiting step. This was confirmed with intermediate V not detected when the reaction occurs in the presence of zinc (II) ions, indicating swift conversion of V to VI.



**Figure 3.2:** The reaction mechanism of IND/Zn with amino acid and zinc ions as a Lewis acid catalyst.<sup>104</sup>

The study<sup>104</sup> also found that using a 10:1 excess of 1,2-indanedione, which imitates standard fingerprint development, resulted in a slightly higher relative abundance of Joullie's Pink (JP) compared to stoichiometric reactions. However, this also resulted in an increased presence of yellow-brown coloured oligomer by-products. Furthermore, analysis of the products using both visible absorption and luminescence spectroscopy revealed an inverse correlation between their visible

absorbance and luminescence. Samples with strong chromophores exhibited low luminescence, and those with weak chromophores demonstrated high luminescence. This may be due to the non-fluorescent oligomeric side products masking the luminescence of JP.

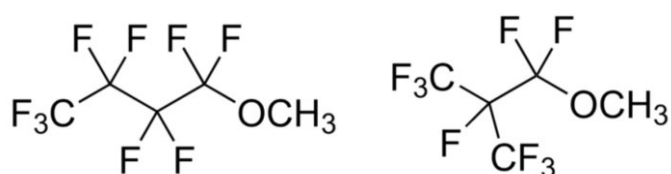
### **Co-solvents**

To maintain the solubility of 1,2-indanedione in non-polar carrier solvent, a small amount of polar solvents such as ethyl acetate, ethanol or methanol are included in all formulations. Early spectroscopic studies revealed that the presence of ethanol in the 1,2-indanedione formulation without zinc salts led to the formation of a hemiketal (2,2-diethoxy-1-indanone),<sup>228</sup> resulting in reduced luminescence and reagent stability.<sup>117</sup> However, recent studies by Nicolasora *et al.*<sup>127</sup> under UK conditions demonstrated that the inclusion of methanol in the CAST formulation led to stronger luminescence. This was attributed to the addition of zinc ions in the formulation, although the mechanism behind it is still not fully understood. It was also observed that excessive methanol can cause ink and ridge detail diffusion, which reduces the quality of developed fingerprints. Additionally, most formulations contain a small amount of acetic acid to counteract the alkaline nature of paper stocks and enhance the development of latent fingerprints. One exception is a formulation by Wiesner *et al.*,<sup>117</sup> where acetic acid was found to cause diffused fingerprints on local paper stocks.

### **Carrier solvents**

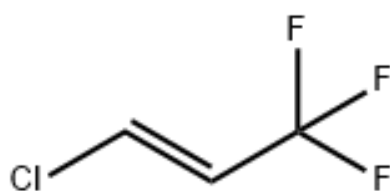
The carrier solvent makes up the bulk of the working solutions, with most formulations using HFE-7100.<sup>16, 17, 122, 230</sup> This is because the previous standard solvent, CFC-113 (1,1,2-trichloro-1,2,2-trifluoroethane) was restricted in the 1990s due to its contribution to ozone depletion.<sup>231, 232</sup> HFE-7100 has been generally preferred as it produces superior ridge detail and luminescence<sup>116</sup> and is also non-flammable, low in toxicity and minimises diffusion of printed and handwritten inks.<sup>231-233</sup> However, concerns have been raised recently over a potential EU ban<sup>234</sup> due to its contribution to fluorinated greenhouse gases and climate change as an organofluorine chemical. HFE-7100 is a hydrofluoroether (HFE) consisting of two isomers (Figure 3.3) with zero ozone depletion potential and low Global Warming

Potential (GWP).<sup>201</sup> Consequently, it falls under Annex II of Regulation (EU) No 517/2014 and reporting on its production, import, and export quantities is obligatory.<sup>200</sup> More importantly, HFE-7100 belongs to the "forever chemicals" class of per- and polyfluoroalkyl substances (PFAS). PFAS are a class of synthetic organofluorine compounds that have gained attention from environmental regulators and the public due to their widespread environmental presence, resistance to degradation and potential adverse health effects.<sup>235, 236</sup> Potential restrictions in the production and use of HFEs would result in limited supply of HFE-7100 globally. Consequently, there has been renewed interest in researching potential alternatives to HFE-7100 as a carrier solvent.<sup>233, 237, 238</sup>



**Figure 3.3:** The molecular structure of HFE-7100 ( $C_4F_9OCH_3$ ) consisting of two isomers  $CF_3CF_2CF_2CF_2OCH_3$  and  $(CF_3)_2CFCF_2OCH_3$ .

Solstice<sup>®</sup> PF (trans-1-chloro-3,3,3-trifluoropropene) depicted in Figure 3.4, is marketed as an alternative to HFE-7100 in the context of an industrial degreaser.<sup>239</sup> It has a lower GWP, as shown in Table 3.2, and is priced at half the cost of HFE-7100.<sup>233, 239</sup> Recent trials conducted by Olszowska *et al.*<sup>233</sup> in the UK, have indicated that Solstice<sup>®</sup> PF can serve as a substitute carrier solvent for HFE-7100 in amino acid sensitive fingerprint detection reagents. Studies conducted using CAST IND/Zn revealed that formulations prepared with Solstice<sup>®</sup> PF performed comparably to the standard HFE-7100-based formulations, with no significant differences in fingerprint appearance or ink diffusion.<sup>233, 238</sup> On some substrates, such as brown paper, Solstice<sup>®</sup> PF-based IND/Zn was found to outperform HFE-7100, demonstrating improved sensitivity and ridge detail.<sup>238</sup>



**Figure 3.4:** The molecular structure of Solstice® PF (*trans*-1-chloro-3,3,3-trifluoropropene).

Petroleum ether has also been trialled, however, HFE-7100 has generally favoured,<sup>116, 117, 127</sup> despite early studies by Bicknell *et al.*<sup>227</sup> and Serrano *et al.*<sup>240</sup> reporting both solvents to be equally effective. Petroleum ether 40-60 °C is a mixture of low boiling point aliphatic hydrocarbons primarily composed of pentanes and hexanes.<sup>241</sup> It is an inexpensive and readily accessible laboratory reagent<sup>124, 242</sup> with the caveat of being a flammable solvent. However, its use in a properly fitted chemistry laboratory with a fume hood and safety procedures in place, particularly regarding the use of open flame and storage conditions, should significantly reduce the risk.

**Table 3.2:** Comparison of physical properties of carrier solvents used in amino acid sensitive fingerprint detection reagent.<sup>239, 241, 243, 244</sup>

	HFE-7100	Solstice® PF	Petroleum ether 40 -60°C
Appearance	Colourless	Colourless	Colourless
Boiling Point ( °C)	61	19	40-60
Heat of Vaporisation at Boiling Point (kJ/kg)	112	194	No data available
Freezing point ( °C)	-135	-107	No data available
Flash Point ( °C)	No data available	No data available	-35
Vapour pressure at 25 ( °C) (kPa)	28	126	28
Liquid density at 25 ( °C) (g/mL)	1.52	1.26	No data available
Solubility of water in solvent at 25°C (ppmw)	95	460	No data available
GWP (100 Year)	320	1	No data available
PFAS	Yes	No	No
HSP Value (δD, δP, δH)	13.7,2.2,1.0	15.5,4.8,2.2	14.9,0,0 (Hexane)

Table 3.2 also displays the Hansen solubility parameters (HSP) for each carrier solvent. Hansen's 3-parameter solubility model is commonly used as a practical guide when selecting solvents for coating systems or optimising solvents for cleaning applications.<sup>245, 246</sup> HSP model predicts solubility based on three parameters: dispersion forces ( $\delta D$ ), polarity ( $\delta P$ ), and hydrogen bonding ( $\delta H$ ). These parameters describe the solubility compatibility between two systems by evaluating the HSP distance ( $R_a$ ) using the equation below. The HSP distance ( $R_a$ ) between two molecules determines how compatible they are in terms of solubility. The smaller the  $R_a$  value between two molecules, the more likely they will be soluble with each other.

$$R_a^2 = 4(\delta D^2 - \delta D1)^2 + (\delta P^2 - \delta P1)^2 + (\delta H2 - \delta H1)^2$$

Hansen parameter tables are available and can also be determined experimentally.<sup>243-246</sup> HSP can serve as an initial tool to identify potential solvent replacement, such as finding an alternative carrier solvent for amino acid reagent. For instance, if HFE-7100 is the target replacement solvent, Solstice® PF and petroleum ether's  $R_a$  values are 6.8 and 5.4, respectively, indicating that petroleum ether is a more compatible replacement.

### **Frugal forensics approach**

Due to the variations in composition, different formulations of IND/Zn can vary markedly in properties such as polarity, pH and cost-effectiveness. These differences can affect the performance of the reagent and its viability for use in specific jurisdictions. As discussed in Chapter 1, techniques employed in Seychelles should ideally have minimal environmental impact, be cost-effective and have minimal supply chain disruption due to limited resources and remote location. Table 3.3 provides a cost analysis of each formulation and shows the cost for one litre of the working solution based on the prices of the reagents from at least two regular Seychelles Police suppliers in the UK and USA. Overall, the petroleum-based BKA formulation is at least two times more cost-effective than the HFE-based formulations and has a shorter lead time for reagent supply. As previously mentioned, due to environmental regulations, HFE-7100 is prone to supply chain disruption. Moreover, and rarely discussed in the open literature, is that HFE-7100 is



a proprietary fluid of 3M Novec™, making it a single source chemical. This can have a direct impact on supply chain, as was seen in 2022 with a global supply chain concern for HFE-7100 due to manufacturing issues at the 3M production facility and exacerbated by the COVID-19 pandemic.<sup>202, 203</sup> In contrast, petroleum ether is a readily available solvent from multiple suppliers, making it a more likely candidate for frugal forensics application in Seychelles.

Similar arguments in terms of the cost and availability of 1,2-indanedione can be raised. However, 1,2-indanedione is present in small amounts in the working solution, ranging from 0.227 g/L in the CAST formulation to 0.990 g/L in the BKA formulation. This results in a meagre cost factor, except in the BKA formulation where the relatively high concentration coupled with the low cost of the petroleum ether makes it a major cost factor (56 %), as shown in Table 3.3. Reducing the 1,2-indanedione concentration would lead to additional savings, provided that it does not significantly affect its performance. As for supply, a remote jurisdiction can buy multiple years' worth of 1,2-indanedione crystal stock, which is reportedly highly stable.<sup>247</sup> Likewise, no immediate issues are expected with zinc chloride salt and co-solvents; both commonly available reagents with trivial differences in cost. Ensuring an effective concentration ratio of the active chemicals<sup>104</sup> would be more important. However, some formulations may tolerate a given modification differently from others, with potential impact on reagent effectiveness and stability. Hence any modification should be evaluated in line with the IFRG guidelines before operationalisation.

**Table 3.3:** Approximate cost of one litre of 1,2-indanedione zinc working solution in USD. Chemical sourced from two regular Seychelles Police suppliers based in the UK and USA.

Category	Chemicals	NCFS		BKA		CAST		Price Source/Lead Time
		Chemical average cost	Category % cost	Chemical average cost	Category % cost	Chemical average cost	Category % cost	
Active ingredients	1,2-indanedione	\$43.00	15%	\$71.49	58%	\$16.25	6%	Arrowhead Forensics, Fisher Scientific Fisher Scientific, Sigma -Aldrich
	Zinc chloride	\$0.04		\$0.02		\$0.01		
Co-solvents	Ethyl acetate	\$7.56	3%	\$5.42	5%	\$2.51	2%	Fisher Scientific, Sigma -Aldrich Fisher Scientific, Sigma -Aldrich Fisher Scientific, Sigma -Aldrich Fisher Scientific, Sigma -Aldrich
	Acetic acid	\$0.22		\$0.42		\$0.77		
	Methanol	-		-		\$1.05		
	Ethanol	\$0.09		\$0.23		-		
Carrier solvents	HFE-7100	\$237.29	82%	-	37%	\$246.48	92%	Arrowhead Forensics (unavailable), Sigma-Aldrich (45 days) Fisher Scientific, Sigma -Aldrich (2 days)
	Petroleum ether bp 40-60 °C	-		\$44.69		-		
Total cost		\$288.20		\$122.26		\$267.06		

\*Cost as of December 2022

## 3.2 Experimental

### 3.2.1 Preliminary consideration

The three active IND/Zn formulations require different processing conditions: it is recommended that items treated with CAST IND/Zn be processed in an oven at 100 °C, <sup>248</sup> while the NCFS and BKA method stipulates the use of a heat press set to 160 °C. <sup>17, 222</sup> In line with the frugal forensics approach, it was decided to use the heat press process which does not require major equipment and is more cost-effective.

### 3.2.2 Experimental methods

#### 3.2.2.1 Substrate

Nine paper- and cardboard-based substrates commonly encountered by the Seychelles Police were chosen and summarised in Table 3.4. All substrates were sourced from Officeworks Australia as representative of substrates encountered in Seychelles.

**Table 3.4:** Details of substrates used in the head-to-head comparison of NCFS, BKA and CAST IND-Zn formulations.

Substrate	Description	Colour	Weight (gsm)/Thickness
1	Winc Copy paper A4	White	80
2	PPS Envelope, 100% Recycled	White	80
3	Jburrows Notepad A4, 8mm ruled	Blue	70
4	PPS Cardboard	Brown	3.5 mm
5	PPS Heavy duty Cardboard (Fibreboard)	Brown	6.5 mm

#### 3.2.2.2 Fingerprint collection

Natural, split depletion fingerprints were collected from five donors (3 males and 2 females) between 22 and 51 years old in line with an IFRG phase 1/2 study. Each donor deposited 1<sup>st</sup>, 5<sup>th</sup> and 10<sup>th</sup> depletion series on each substrate. The remaining untreated depletions were deposited on a separate piece of the same type of substrate. This allowed an assessment of the sensitivity as well as the overall performance across multiple substrates whilst managing the number of samples. The

collection procedure was repeated to reduce donor biasing on one side of the finger, as discussed in section 2.3.2.

Samples were stored in a cupboard under office conditions (~20 °C) for at least 24 hours and cut into halves prior to IND-Zn treatment unless stated otherwise.

### **3.2.2.3 Fingerprint development**

The IND-Zn working solution and sample development for NCFS, BKA, and CAST were prepared as outlined by Stoilovic *et al.*,<sup>221</sup> Becker *et al.*,<sup>222</sup> and Nicolasora *et al.*<sup>127</sup> respectively and described in section 2.5.1. A number of modified formulations were also prepared and are specified under the relevant sections. All modified formulations followed the same preparation and development process as the parent formulation unless stated otherwise.

### **3.2.2.4 Photography and grading of developed fingerprints**

Processed samples were re-combined and photographed as outlined in section 2.6.1 and graded using the CAST assessment and UC scale as outlined in section 2.7.

## **3.2.3 Head-to-head comparative assessment of existing IND/Zn formulations**

The three active IND/Zn formulations were compared directly with each other on five substrates: white copy paper (Winc), white envelope (PPS), blue notepad (JBurrows), cardboard (PPS) and fibreboard (PPS). In order for each treatment to be compared head-to-head, three sets of fingerprints were collected on each substrate from each donor (1. NCFS|CAST, 2. BKA|NCFS, 3. CAST|BKA).

### **3.2.3.1 Investigation into co-solvents**

To further investigate the role of polar solvents, the NCFS formulation, which exhibited the least luminescence in the head-to-head comparison, was compared to a modified NCFS formulation containing a higher proportion of polar solvents. The modified formulation was prepared by increasing the ethanol concentration to 4.1 % v/v, similar to the proportion of alcohol in the CAST working solution, via a 1:1 volumetric reduction in the amount of ethyl acetate. The comparison was carried out

on two substrates which showed the least (white copy paper) and the most (blue notepad) background development in the head-to-head comparison.

### **Methanol versus ethanol**

The benefit of methanol in the CAST IND/Zn formulation reported by Nicolasora *et al.*<sup>127</sup> was also investigated by comparing the CAST formulation to a modified version in which methanol was directly substituted with ethanol in a 1:1 volumetric ratio. The same substrates as in section 3.2.3.1 were used.

### **3.2.3.2 Investigation into 1,2-indanedione concentration**

To investigate the impact of reducing the 1,2-indanedione concentration, the BKA formulation containing 990 mg/L of 1,2-indanedione was compared to a modified version containing 227 mg/L of 1,2-indanedione (similar concentration as in the CAST IND/Zn). This was investigated on two substrates which IND/Zn was the most (white copy paper) and the least (cardboard) effective in developing fingermarks in the head-to-head comparison. To assess the subtle differences of the developed fingermarks between the two formulations, the UC scale was utilised with BKA 990 mg/L as the control.

### **Fluorescence spectroscopy studies**

Interestingly, reducing the 1,2-indanedione concentration was observed to produce fingermarks with stronger luminescence. To verify this observation, fluorescence spectroscopy was carried out on amino acid spots treated with three BKA formulations containing 990 mg/L, 594 mg/L (approximately equivalent to the concentration in the NCFS formulation) and 227 mg/L of 1,2-indanedione respectively. A fourth BKA formulation without 1,2-indanedione was prepared and used as a control. The same two substrates as above (section 3.2.3.2) were used, and four separate strips of each substrate were impregnated with five 1 µL aliquots of 10 mg/L amino acid solution (alanine, leucine and phenylalanine). All strips were allowed to dry for 24 hours, mimicking the standard fingerprint methodology. Each of the four BKA formulations was used to develop one strip of each substrate, following the same standard procedure for developing fingermarks with 1,2-

indanedione. The amino acid spots were then analysed using a Cary Eclipse Fluorescence Spectrophotometer (Agilent) combined with a fibre optic probe for *in situ* measurements, as outlined in section 2.6.2. Luminescence examination was also carried out using a Polilight (PL 500) for all strips at three stages: prior to impregnation with amino acids, prior to development with IND/Zn and after development to investigate for possible luminescence contamination.

### **3.2.4 Investigation into Carrier solvent – Solstice® PF as an alternative**

Solstice® PF was investigated as an alternative carrier solvent using the NCFS IND/Zn. The NCFS formulation provides a direct comparison with the investigation<sup>233</sup> carried out on the CAST formulation by substituting HFE-7100 with Solstice® PF in a 1:1 volumetric ratio. For the purposes of this study, the terms HFE-IND/Zn and Sol-IND/Zn are used to refer to IND/Zn reagents prepared using HFE-7100 and Solstice® PF, respectively and pertain specifically to the NCFS formulation unless stated otherwise. The performance of Sol-IND/Zn and HFE-IND/Zn were directly compared across a range of paper-based substrates and aged fingermarks. Additionally, the compatibility of the Solstice® PF based reagent with pen inks and its long-term stability was also investigated.

#### **3.2.4.1 Overall performance of Solstice® PF**

To assess the overall performance and sensitivity, fingermark samples were collected on three substrates: white copy paper (COS Premium), white envelopes (PPS) and cardboard (PPS, 100% FSC recycled). For this part of the experiment each donor deposited one set of three fingermarks on each substrate. For investigations into sample age, three sets of fingermarks from each donor were collected on copy paper (COS Premium) and allowed to age 1, 7 and 30 days, respectively.

#### **3.2.4.2 Compatibility with inks**

To investigate effects of the carrier solvent on ink diffusion, a collection of eighteen different pens (Table 3.5) was used. A printed template on A4 copy paper (COS Premium) was filled in with each of the pens and left to dry for at least 15 minutes

prior to immersion. The template was then divided in half, and each half was treated with HFE-IND/Zn or Sol-IND/Zn as described above, omitting the heat treatment. Samples were handled such that any diffused ink would run horizontally across the page as much as possible, rather than vertically across the other inks. The samples were left to air dry on paper towels before examination under both ambient and luminescence conditions using a Polilight (PL 500).

**Table 3.5:** *Pen used in ink diffusion investigations.*

<b>Pen type</b>	<b>Brand/model</b>	<b>Colour</b>	<b>Sample number</b>
Whiteboard marker	Artline 577	Black	1
	Anko	Blue	2
	Staples Remarx	Green	3
Permanent marker	Sharpie Fine Point	Black	4
	Sharpie Fine Point	Blue	5
	Sharpie Fine Point	Pink	6
Gel pen	BIC Gel-ocity	Orange	7
	BIC Gel-ocity	Purple	8
	BIC Gel-ocity	Red	9
Felt tip pen	Artline 200	Black	10
	COS Fineliner	Blue	11
	Texta Point 188	Red	12
Ballpoint pen	Artline Smoove	Red	13
	Artline Smoove	Blue	14
	Staedtler 430 M	Black	15
	BIC Economy	Black	16
	BIC Cristal M	Blue	17
	BIC Cristal M	Red	18

### **3.2.4.3 Effect of co-solvents on ink diffusion**

Based on the notably greater diffusion of samples immersed in Sol-IND/Zn, additional investigations were carried out to determine the potential contribution of each co-solvent. These investigations utilised pens (6, 8 and 14) that had shown differing degrees of visible and/or luminescent diffusion. Swatches from each pen were deposited on white copy paper and left to dry for at least 15 minutes prior to spotting with solvents using a capillary tube. Ink replicates were spotted with neat solvents as well as single and double 'knockout' mixtures based on Sol-IND/Zn. The 'knockout' mixtures were prepared according to the NCFS IND/Zn formulation without 1,2-indanedione or zinc chloride, omitting one (single) or two (double) of the co-solvent's ethyl acetate, acetic acid and ethanol. Omitted solvents were replaced by an equivalent volume of Solstice® PF. The samples were left to air dry on paper towels before being examined under ambient and luminescence conditions and then compared to control swatches of each ink. To investigate the effect of the drying time of inks, this test was repeated on ink samples left to dry for 5 days.

### **3.2.4.4 Reagent stability**

A 500 mL batch of Sol-IND/Zn was prepared and immediately stored at 4 °C in a sealed Schott bottle. This reagent was used to treat latent fingerprints on copy paper (as described above) at 1.5, 2.5 and 4 months after preparation, and was not handled or used at any other time. Fingerprints were collected at each time interval of reagent testing. Sample halves treated with the aged reagent were compared to those treated with 'fresh' Sol-IND/Zn (less than 7 days old). Visual inspection of the solution was also conducted at these times.

### **3.2.5 Statistical analysis**

Statistical analysis software R (version 4.3.0) was utilised to assess whether there was significant difference in the fingerprint grades between the IND/Zn treatments. The non-parametric Kruskal-Wallis test was used for the head-to-head comparison ( $n=450$ ) of NCFS, BKA and CAST, as more than two treatments were being tested. The data was subjected to a chi-squared test to assess whether the treatments have



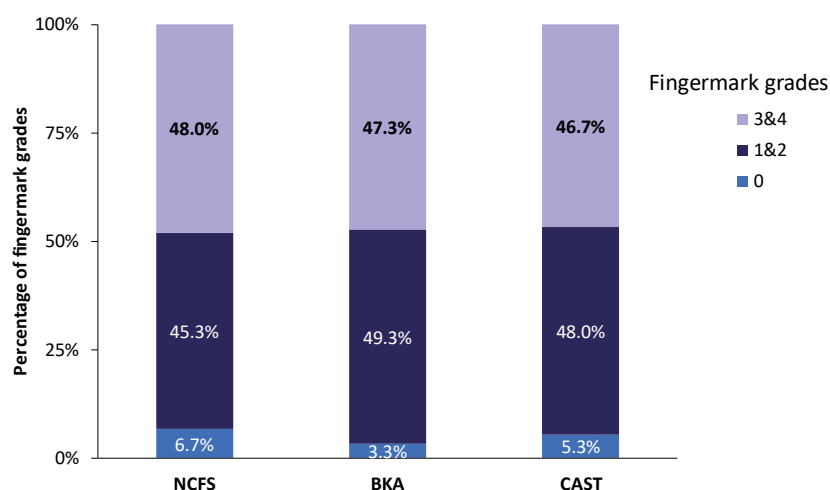
different distributions in fingerprint grade frequencies. All statistical tests were conducted at 95% confidence ( $p < 0.05$ ).

### 3.3 Results and discussion

#### 3.3.1 Head-to-head comparative assessment of existing IND/Zn formulations

##### 3.3.1.1 Overall performance and sensitivity

The head-to-head comparative assessment generated a total of 450 fingerprint halves, and the results are summarised in Figure 3.5 for overall performance and Figure 3.6 for sensitivity. All three formulations showed comparable performance, with NCFS formulation developing 48.0 % of useful fingerprint marks, BKA 47.3 % and CAST 46.4 %.

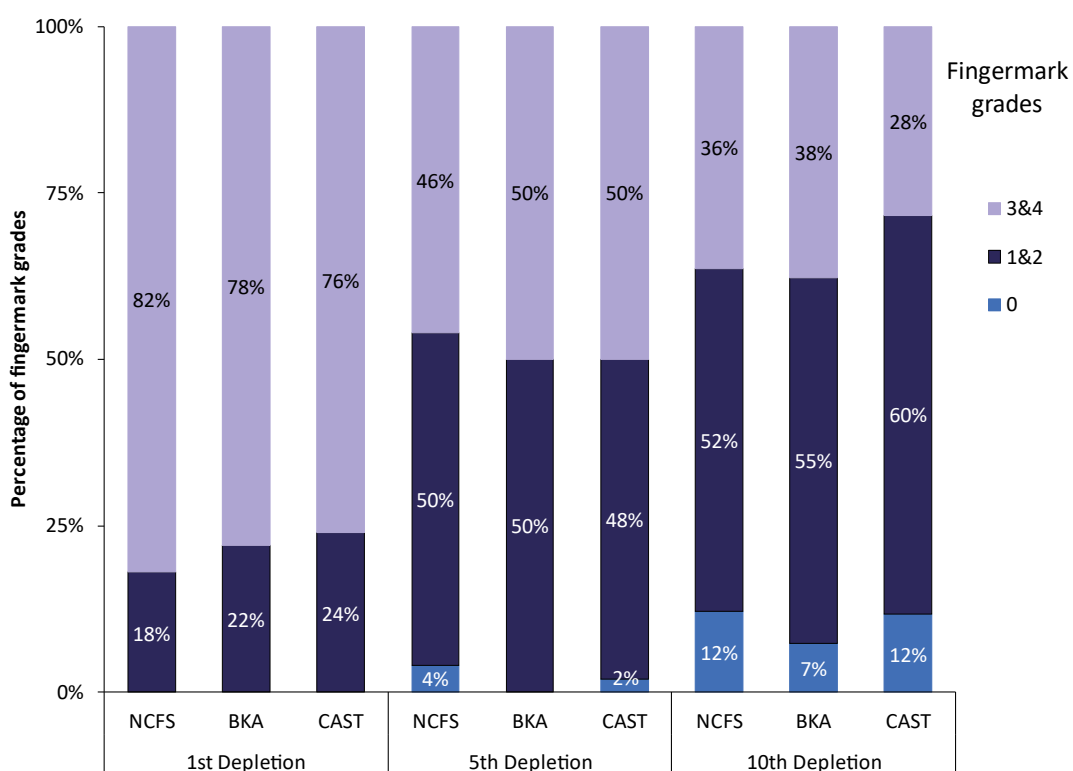


**Figure 3.5:** Percentage of fingerprints graded (3&4), (1&2), and (0) out of 450 fingerprints developed in head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation.

Similar trends of comparable performance were also observed when assessing the sensitivity of the three formulations. As expected, there was a gradual decrease in the number of fingerprints detected down the depletion series. All three formulations were highly sensitive with at least 28 % of the 10<sup>th</sup> depletion series was graded as useful fingerprints (Figure 3.6).

The result reveals that despite the differences in composition, the three IND/Zn formulations have similar effectiveness in developing latent fingerprints on

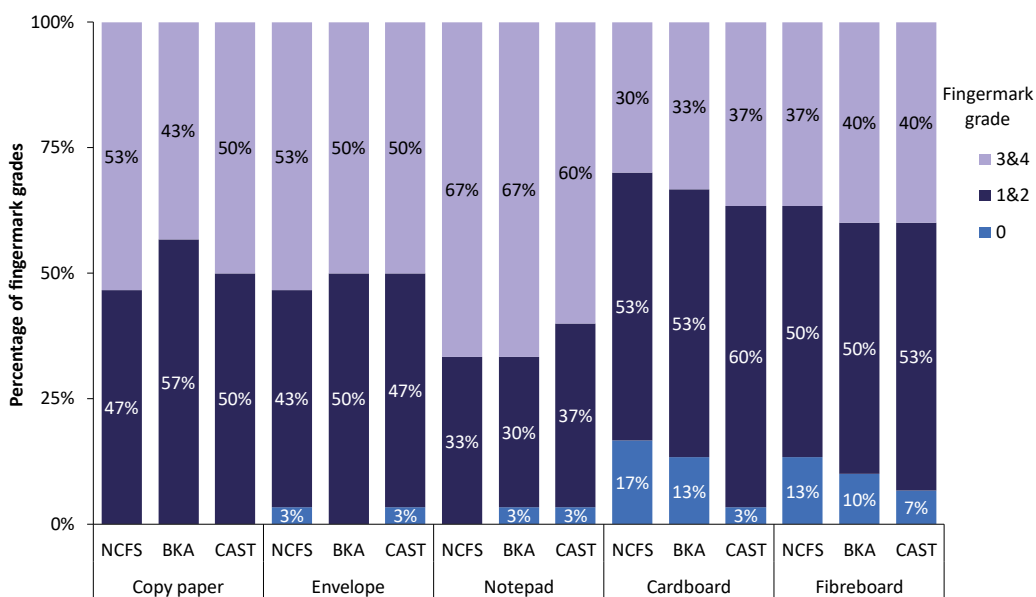
substrates used in this study. The high reproducibility is likely due to the addition of zinc salt, making IND/Zn reagents less susceptible to humidity.<sup>115, 122, 227</sup> Spindler *et al.*<sup>104</sup> demonstrated that incorporating zinc ions into a 1,2-indanedione solution helps stabilise a crucial intermediate during the rate-limiting hydrolysis process involved in producing JP. This is an improvement from earlier non-zinc salt containing 1,2-indanedione formulations, as it makes it easier to adopt formulations and modify them. This is particularly important within the frugal forensics context, where the emphasis is on cost and supply chain rather than optimum performance.



**Figure 3.6:** Comparison of percentage fingermark grades assigned to 1<sup>st</sup>, 5<sup>th</sup> and 10<sup>th</sup> depletion series in the head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation.

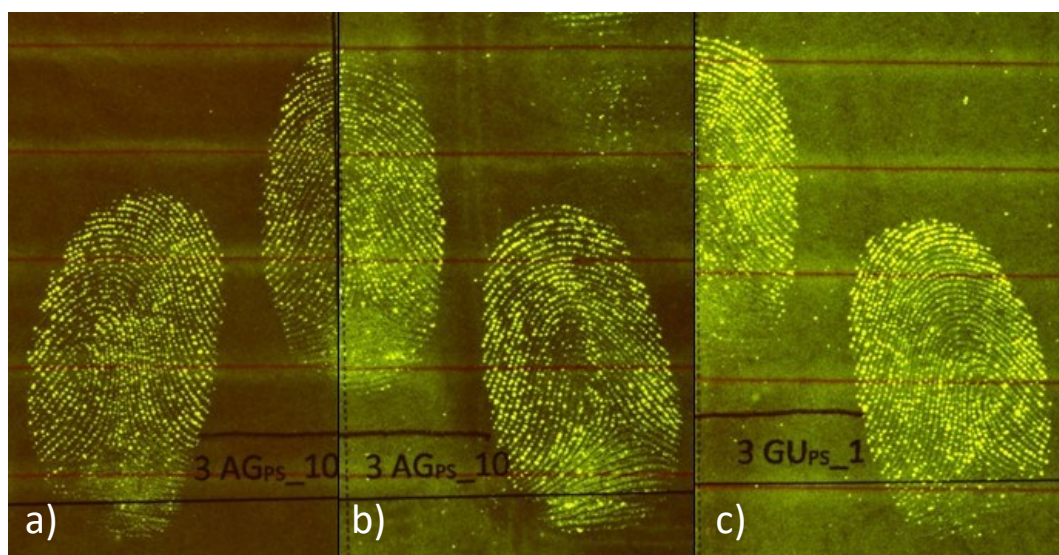
### 3.3.1.2 Substrates

The NCFS, BKA and CAST formulations were all successful in developing fingermarks on each of the five substrates, as shown in Figure 3.7. Across the various substrates, the formulations exhibited similar trends, with the most useful fingermarks developed on notepad paper and the least number on cardboard surfaces.



**Figure 3.7:** Comparison of percentage fingermark grades assigned to fingermarks developed on five substrates in the head-to-head comparison between NCFS, BKA and CAST 1,2-indanedione zinc formulation.

However, it was observed that fingermarks developed with CAST and BKA formulations exhibited brighter luminescence than those developed with NCFS formulation. Conversely, NCFS produced fingermarks with slightly better contrast and less background development. This was particularly evident on the coloured notepad with printed lines, where the NCFS formulation exhibited a clean background and lower luminescence fingermark ridge details. In contrast, the CAST and BKA formulations produced a yellowish background, as shown in Figure 3.8, but this did not significantly impact the fingermark grades.



**Figure 3.8:** Fingermarks treated with (a) NCFS formulation, (b) BKA, (c) CAST on notepad paper showing difference in luminescence and background development.

As can be seen in Figure 3.8, the CAST formulation exhibited marginally stronger luminescence and background development. This is likely due to its higher percentage of polar solvents. Methanol and acetic acid make up the highest proportion of highly polar solvents in the CAST formulation (5.0% v/v), while the NCFS formulation contains the least amount in the form of ethanol and acetic acid (0.9% v/v). The observation is consistent with the findings of Nicolasora *et al.*<sup>127</sup>, which showed that the combination of methanol and zinc chloride resulted in increased luminescence intensity, but excess methanol caused more background development. Previous studies have also reported that the carrier solvent can affect formulation properties.<sup>116, 122, 227</sup> However, the NCFS formulation which also uses HFE-7100 but with a much lower proportion of highly polar solvents (alcohol and acetic acid), exhibited the least luminescence and background development compared to the CAST formulation. This suggests that although the carrier solvent may influence the luminescence intensity and background development, the co-solvents likely have a greater impact. Further investigation into the benefits of polar solvents was therefore conducted and discussed in section 3.3.2.

The decreased effectiveness on cardboard is consistent with other studies,<sup>125, 126</sup> due to low quality paper and a less porous surface. The chipboard and fibreboard used in

this study are made from recycled paper, which has a lower cellulose content and shorter fibres compared to standard paper.<sup>249, 250</sup> The cellulose matrix in paper has been suggested to play an important role in the reaction between the IND/Zn reagent and amino acids in latent fingerprints by directing JP's fluorophoric conformation via hydrogen bonding.<sup>104</sup> Furthermore, cardboard boxes are generally made with a layer of high grammage Kraft paper to reduce porosity and increase durability.<sup>250, 251</sup> The level of porosity is particularly relevant in the development of fingerprints on paper and cardboard surfaces, since less porous surfaces tend to absorb fingerprint deposits to a lesser extent,<sup>91</sup> resulting in weaker development. This is likely because the fingerprint deposits close to the surface are less protected causing a decrease in the target material. It was observed that BKA and CAST formulations were slightly more effective on cardboard than NCFs formulations. The marginally stronger luminescence exhibited by the CAST and BKA formulations may be beneficial in developing fingerprints on cardboard, as weak development is generally seen on less porous substrates.

### **3.3.1.3 Statistical Analysis**

A Kruskal-Wallis test performed on the head-to-head comparative data showed that there is no significant difference in the fingerprint grades between the three treatments ( $p = 0.93$ ). Similarly, the chi-squared test showed that there is no significant difference in the distributions of fingerprint grade frequencies between the different treatments ( $p = 0.73$ ). The statistical results reinforce the comparative performance observed between the three active IND/Zn formulations (NCFs, BKA and CAST). The R code and results of the statistical analysis are provided in Appendix A-2.

### **3.3.1.4 Frugal forensics analysis of existing operational IND/Zn protocols**

Although the formulations have similar performance, their composition can have significant implications when applied in a frugal forensics context. This is especially true when it comes to the carrier solvents, which present challenges related to cost and supply chain risks when implementing an IND/Zn formulation for the Seychelles

Police. The use of HFE-7100 in the NCFS and CAST formulations is expensive and subject to supply chain disruption, jeopardising service continuity. In contrast, the BKA formulation's use of petroleum ether 40-60 °C is more cost-effective and readily available from multiple suppliers, allowing for better budget and supply chain risk management. Therefore, the BKA formulation was identified as the more sustainable method that the Seychelles Police Force can adopt with minimal modifications.

The publication of the head-to-head comparative findings in 2022 prompted the New South Wales (NSW) Police to conduct trials on the NCFS formulation.<sup>202, 252</sup> Their study revealed that substituting HFE-7100 with petroleum ether resulted in comparable performance and an estimated annual savings of \$40,000 AUD.<sup>253</sup> Additionally, in late 2022, 3M announced the discontinuation of PFAS production by the end of 2025 (see Appendix C). This has increased the urgency to find an alternative carrier solvent. Altogether, it highlights the thesis's timeliness and the potential impact of the frugal forensics approach to both the Global North and South.

### **3.3.2 Carrier solvent – Petroleum ether**

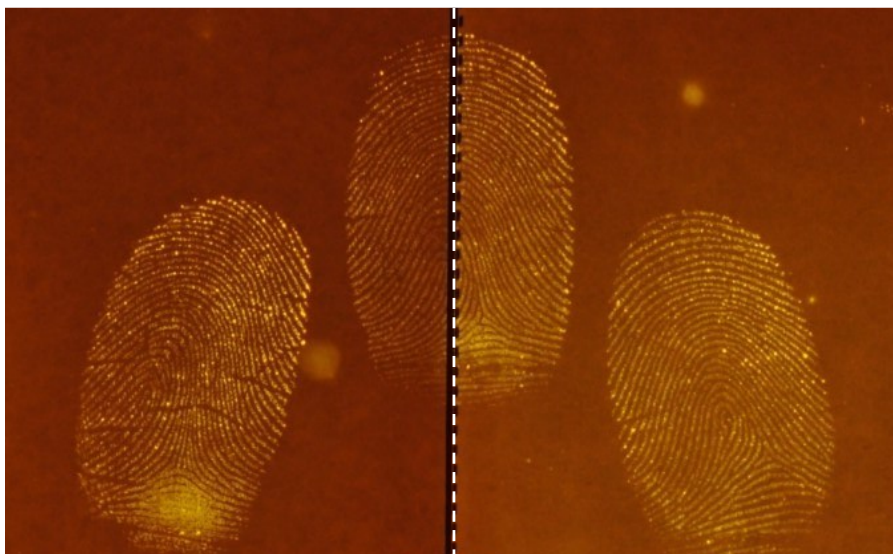
The sustainability of the BKA formulation was established largely based on the carrier solvent - petroleum ether. Hence, further investigation was conducted to examine whether the co-solvents and the amount of 1,2-indanedione could be optimised to enhance the sustainability of the formulation.

#### **3.3.2.1 Co-solvents**

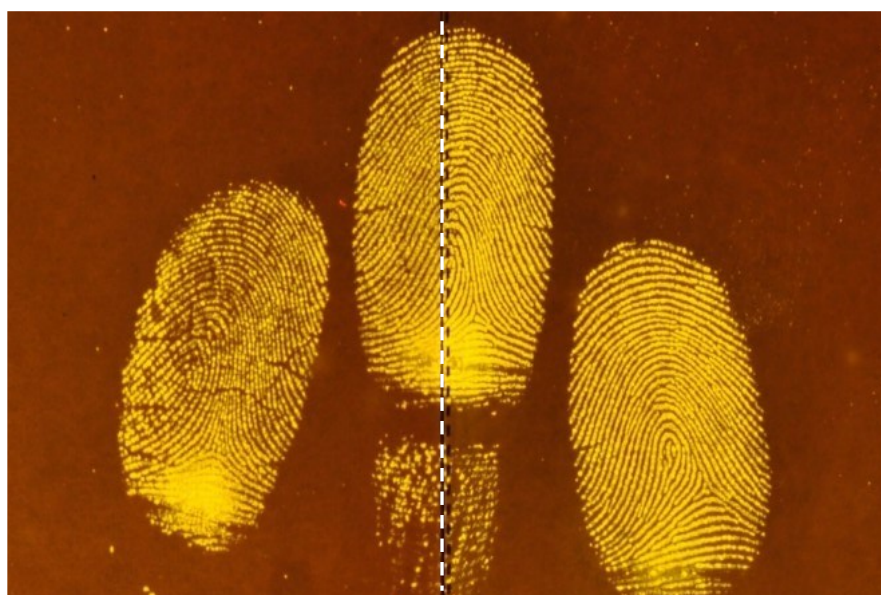
Investigation into the role of polar solvents revealed that the more polar modified NCFS formulation displayed marginally stronger luminescence on both substrates (white copy and notepad paper), similar to the CAST formulation as shown for the copy paper in Figure 3.9. The findings support the suggestion that increased polarity of the co-solvent enhances the luminescence of developed fingermarks, possibly due to greater solubility of 1,2-indanedione.<sup>127</sup> However, as mentioned previously, this also leads to increased background development and ridge diffusion. Therefore, there is a need to balance sensitivity and quality of the developed fingermarks when modifying the polar solvents in the IND/Zn formulation.

### **Methanol versus ethanol**

Assessment of the developed marks indicated that the use of methanol or ethanol in the formulation resulted in similar effectiveness. However, marginally stronger luminescence was observed with the methanol-based formulation, as shown in Figure 3.10. This was attributed to the relatively higher polarity of methanol compared to ethanol.



**Figure 3.9:** Fingermarks on copy paper treated with NCFS formulation (left) and the more polar modified NCFS formulation (right) showing the marginally greater background development.



**Figure 3.10:** Fingermarks on copy paper treated with modified CAST formulation containing ethanol (left) and CAST formulation containing methanol (right).

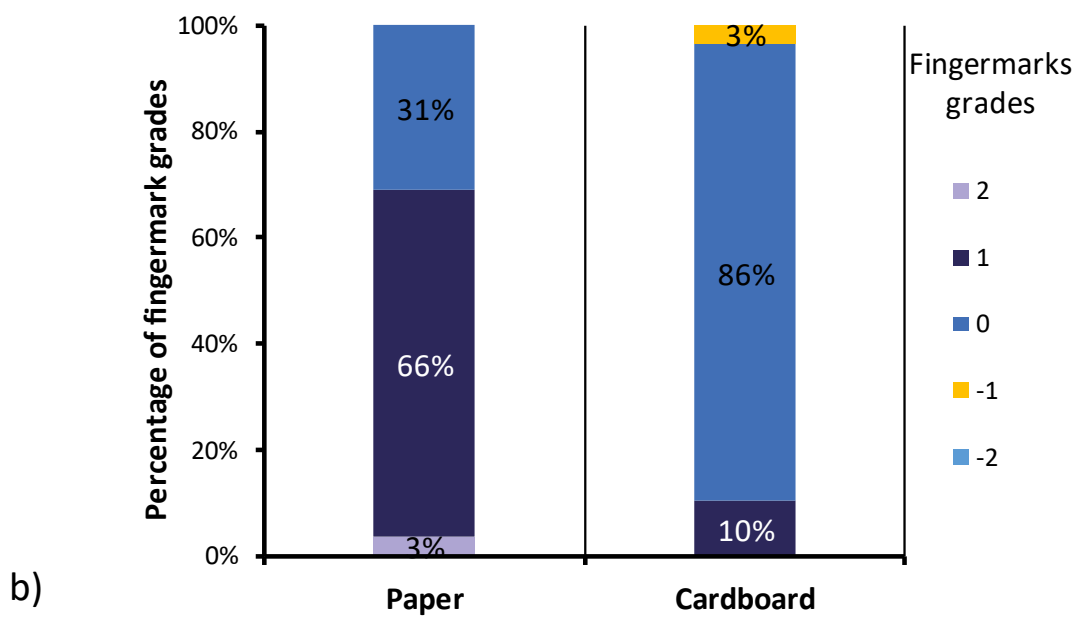
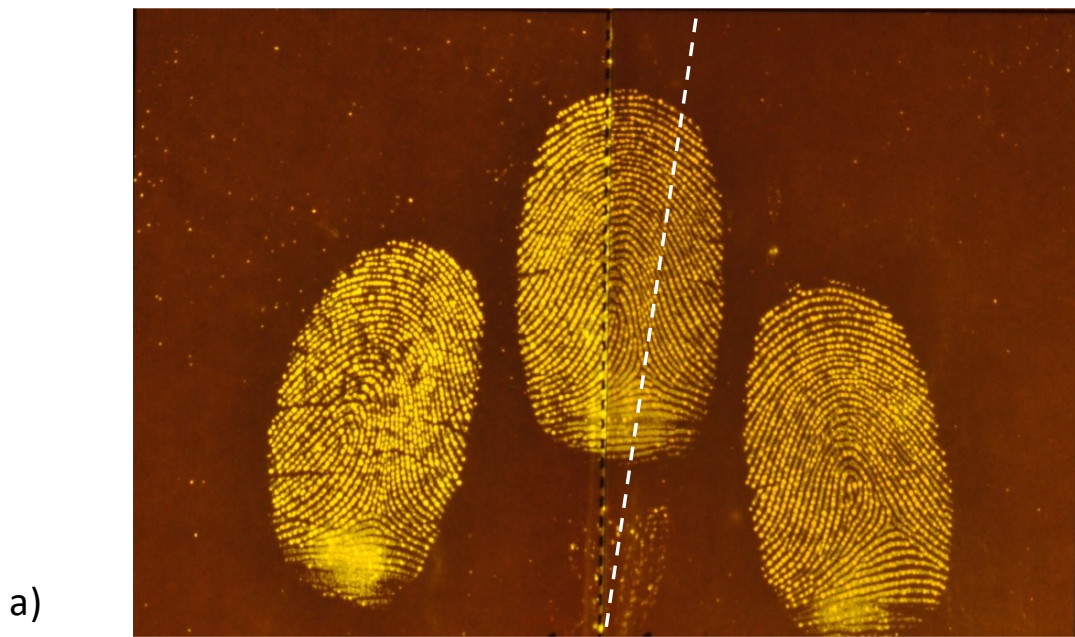
### **Frugal forensics analysis of co-solvents in BKA formulation**

With BKA identified as a more sustainable formulation, it was decided not to modify the polar solvents since it already strikes the right balance with similar luminescence intensity to CAST formulation and only minor background development compared to NCFS. With regards to the use of alcohol, ethanol was preferred over methanol due to the toxic nature of the latter. Nonetheless, the considerable cost of 1,2-indanedione in the formulation (~ 58 %) prompted further investigation into the impact on effectiveness in reducing its concentration.

#### **3.3.2.2 1,2-indanedione concentration**

Interestingly, reducing the 1,2-indanedione concentration was found to improve the luminescence of developed fingermarks. The BKA 227 mg/L formulation exhibits better contrast compared to the BKA 990 mg/L formulation, as shown in Figure 3.11. This was more pronounced on white copy paper with a 69% of UC scale scores of 1 or 2 compared to 10 % on cardboard. The result was attributed to excess 1,2-indanedione in the BKA 990 mg/L formulation, forming non-fluorescent oligomeric side products masking the luminescence of JP, which is consistent with Spindler *et al.*'s<sup>104</sup> study investigating the reaction mechanism of 1,2-indanedione with amino acids. The lower paper quality and less porous surface of cardboard may explain why this observation is less pronounced on cardboard.





**Figure 3.11:** Difference in luminescence of fingerprints on copy paper with modified BKA formulation containing 227 mg/L of 1,2-indanedione (left) and BKA formulation containing 990 mg/L (right)(a). Difference in UC Scale percentage scores between paper and cardboard (b).

### Spectroscopy studies

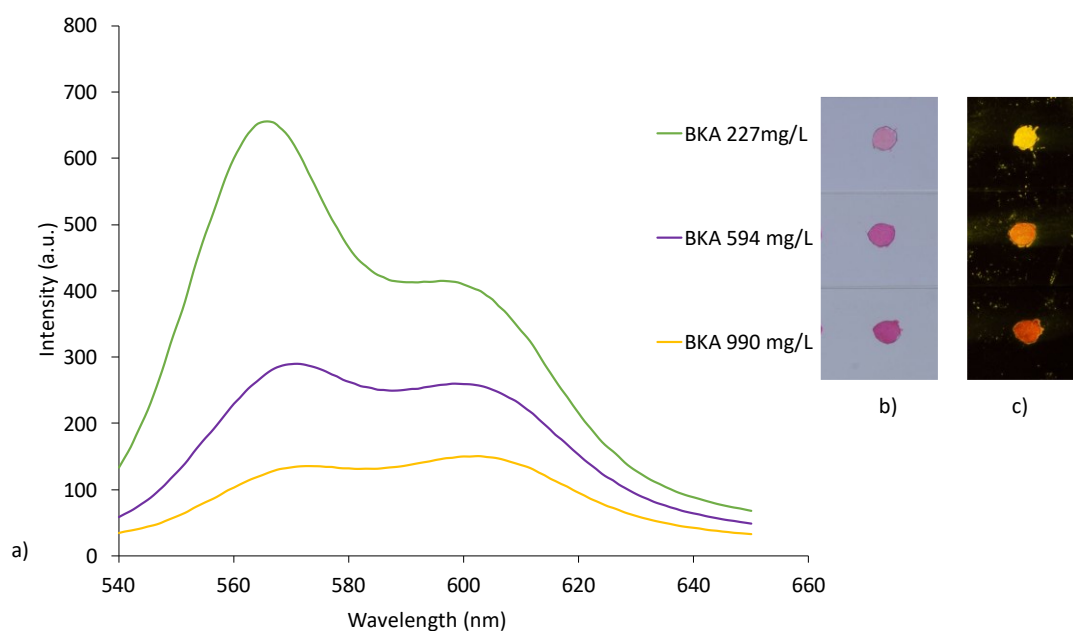
To verify this observation, spectroscopy studies were conducted. The results of the A4 the copy paper is depicted in Figure 3.12, with BKA 277 mg/L producing stronger fluorescence, which aligns with the previous observations. Examination of the amino

acid spots showed that BKA 990 mg/L (highest concentration) exhibited a deep purple-pink colour under visible light and a dark orange colour under Polilight at 505 nm. This changed to light purple-pink and bright yellow, respectively, with BKA 227mg/L (lowest concentration). These findings reinforce the suggestion that excess 1,2-indanedione forms non-fluorescent oligomeric side products that mask JP luminescence. It is worth noting that the dual visualisation of fingermarks in absorbance and luminescence modes, provides a valuable advantage to practitioners to quickly identify where the fingermark is located on an item, leading to better preservation and quicker processing time.

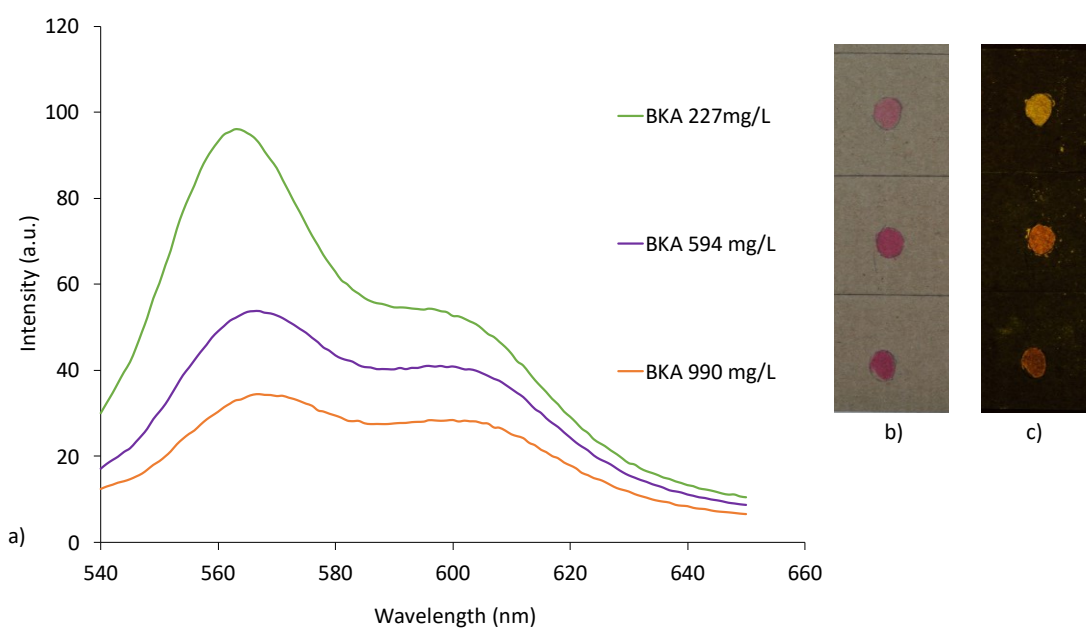
The results for cardboard are depicted in Figure 3.13, indicating a similar outcome, although less pronounced as seen in earlier observation. The experiment was also repeated, reducing the amount of amino acid impregnated per spot from 10 µg to 5 µg, 1.0 µg and 0.5 µg to mimic a depletion series, and similar results were observed.

#### **Frugal forensics analysis of 1,2-indanedione concentration in the BKA formulation**

As discussed above, the BKA formulation has four times more 1,2-indanedione than the CAST formulation, which adds a considerable cost to the reagent. Additionally, the dual visualisation of fingermarks using visible absorbance and luminescence examination is advantageous in casework. Therefore, it was decided to reduce the 1,2-indanedione concentration in the BKA formulation to 594 mg/L - similar to the NCFS formulation. This lowers the cost of 1,2-indanedione in the formulation from \$71.49 to \$43.07 USD per litre of working solution (40 % decrease) and strikes a balance between cost-effectiveness and dual visualisation.



**Figure 3.12:** Amino acid spots on copy paper treated with BKA IND/Zn formulations containing different 1,2-indanedione concentrations, (a) emission spectrum excited at 505 nm illustrating the difference in luminescence intensity (b) difference in colour under visible light, (c) difference in colour under Polilight at 505 nm.



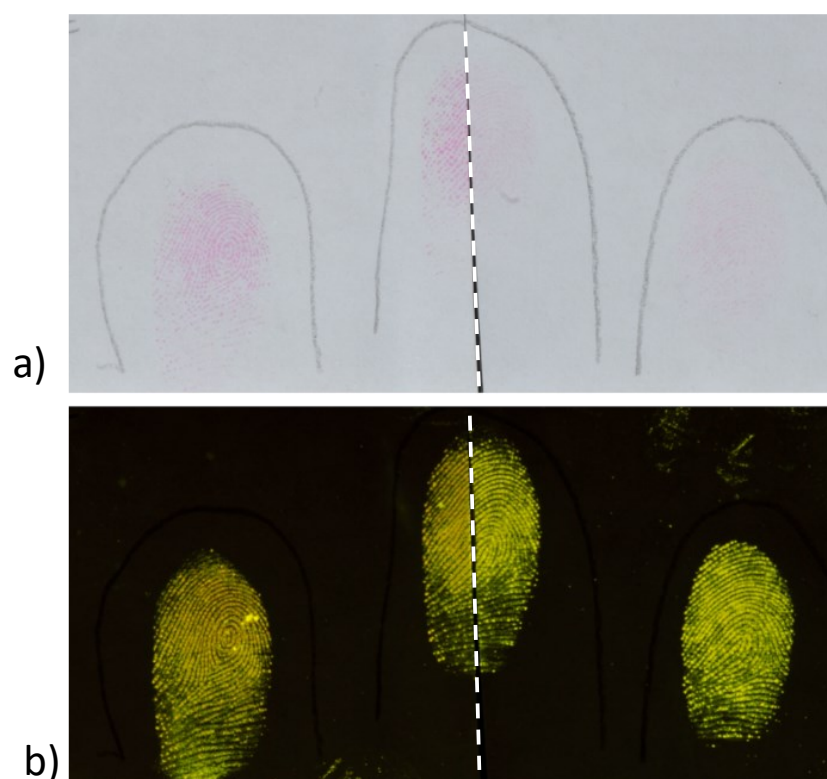
**Figure 3.13:** Amino acid spots on cardboard treated with BKA IND/Zn formulations containing different 1,2-indanedione concentrations, (a) emission spectrum excited at 505 nm illustrating the difference in luminescence intensity (b) difference in colour under visible light, (c) difference in colour under Polilight at 505 nm.

### 3.3.3 Carrier solvent - Solstice® PF as an alternative

#### 3.3.3.1 Overall performance of Solstice® PF

##### General observation

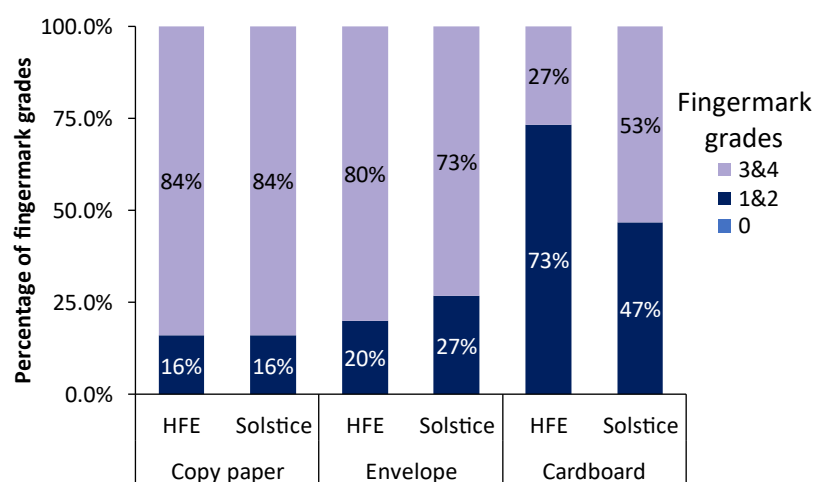
In this part of the study, the inverse correlation between visible absorbance and luminescence of developed fingermarks was also observed, depicted in Figure 3.14. HFE-IND/Zn produced a deep purple-pink colour under visible light, while Sol-IND/Zn exhibited a light purple-pink colour and stronger luminescence. This suggests that the use of Solstice® PF as a carrier solvent can have an impact on the chemical interaction between 1,2-indanedione and the target amino acids, resulting in subtle differences in product colour and luminescence. While similar observations have been made regarding a variety of carrier solvents,<sup>116, 122, 227</sup> the relevant mechanisms have yet to be identified.



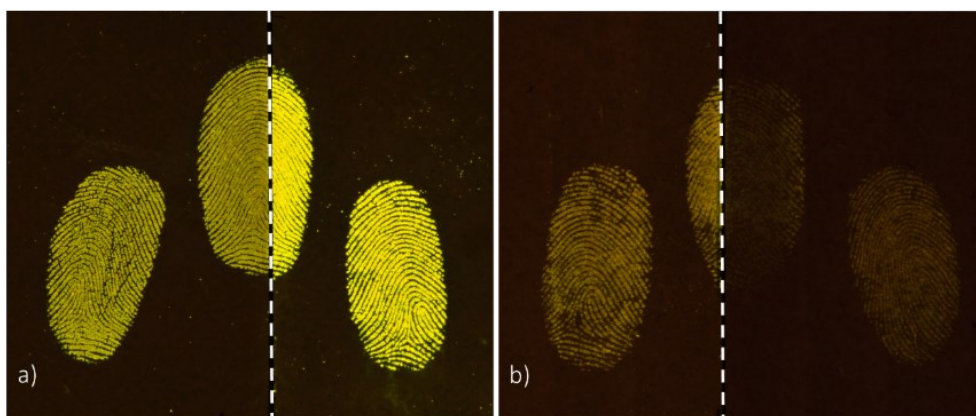
**Figure 3.14:** Latent fingermarks on copy paper treated with HFE-IND/Zn (left) and Sol-IND/Zn (right), (a) viewed under white light and (b) in luminescence mode.

## Overall performance and sensitivity

Overall, the two formulations showed similar levels of effectiveness and sensitivity, except for slight differences on cardboard surfaces (Figure 3.15). As discussed earlier, IND/Zn performance is negatively impacted by less porous substrates that do not effectively absorb fingermark residue, as well as substrates with lower cellulose content.<sup>254, 255</sup> Sol-IND/Zn was more effective on cardboard, developing more useful fingermarks than HFE-IND/Zn (Figure 3.16). This is consistent with results reported by Zhao *et al.*<sup>256</sup> on brown paper and newspaper with CAST IND/Zn.<sup>257</sup> The stronger luminescence exhibited by Sol-IND/Zn treated fingermarks may provide better contrast on cardboard surfaces.



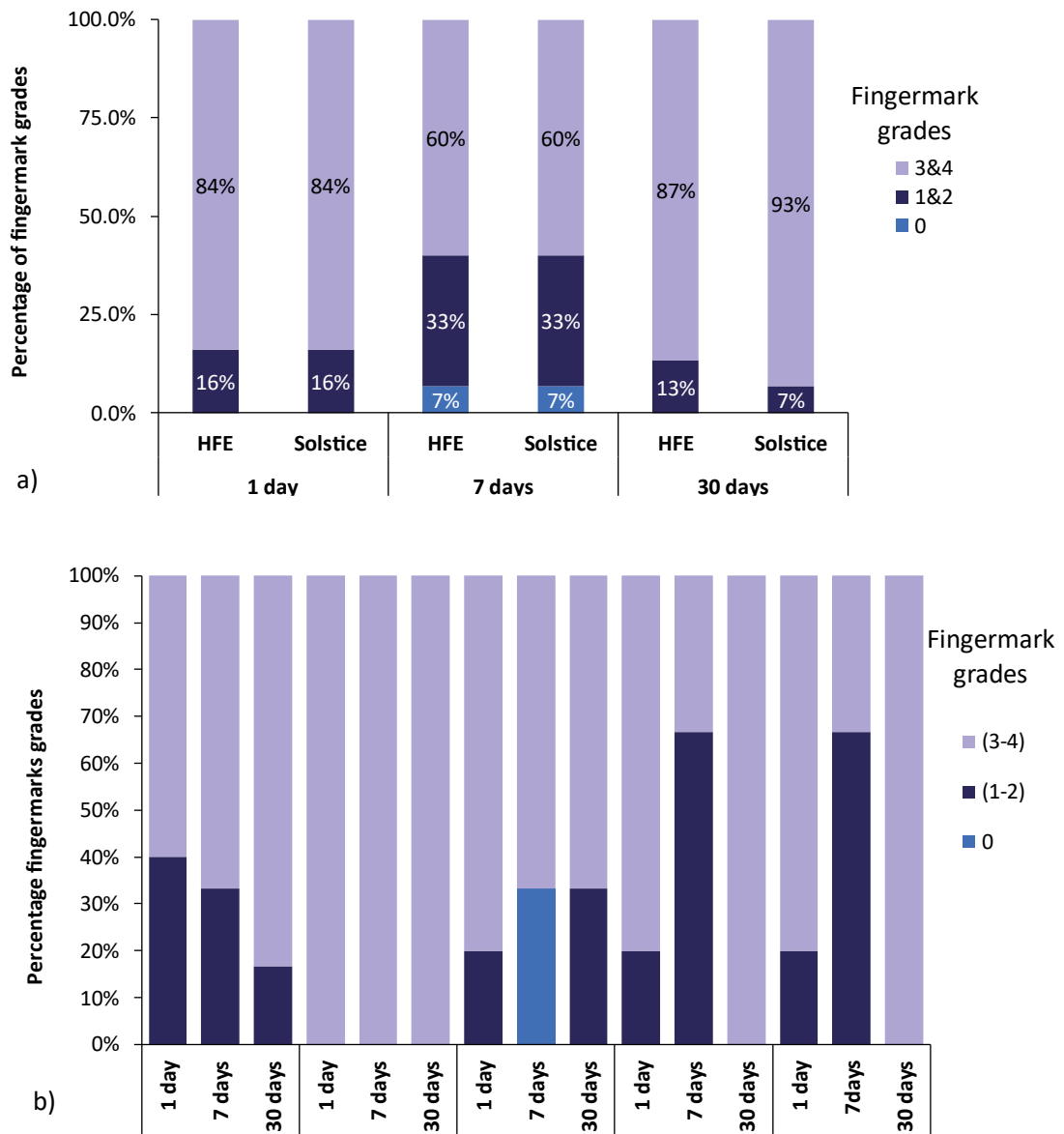
**Figure 3.15:** Comparison of percentage fingermark grades assigned to fingermarks treated with HFE-IND/Zn and Sol-IND/Zn on white copy paper, white envelope and cardboard.



**Figure 3.16:** Fingerprint halves developed on (a) white envelope treated with HFE-IND/Zn (left) and Sol-IND/Zn (right) and (b) cardboard treated with Sol-IND/Zn (left) and HFE-IND/Zn (right).

### Ageing samples

Sol-IND/Zn and HFE-IND/Zn were found to perform comparably on samples up to 30 days old developed on white copy paper (Figure 3.17). Overall, the quality of IND/Zn treated fingermarks for both reagents declined initially from 1 day to 7 days and then improved with increasing age over 30 days. While this could be a result of intra-donor variation in sample composition, a similar trend was noted by Zhao *et al.*<sup>256</sup> when comparing CAST HFE-IND/Zn and Sol-IND/Zn on copy paper, suggesting ageing mechanisms may also play a part.<sup>257</sup> Other reports indicate that IND/Zn efficiency declines with fingermark age over 3-4 weeks following deposition.<sup>258, 259</sup> Further analysis of individual donors shows that the difference could be attributed to donors 4 and 5 as indicated by Figure 3.17b.

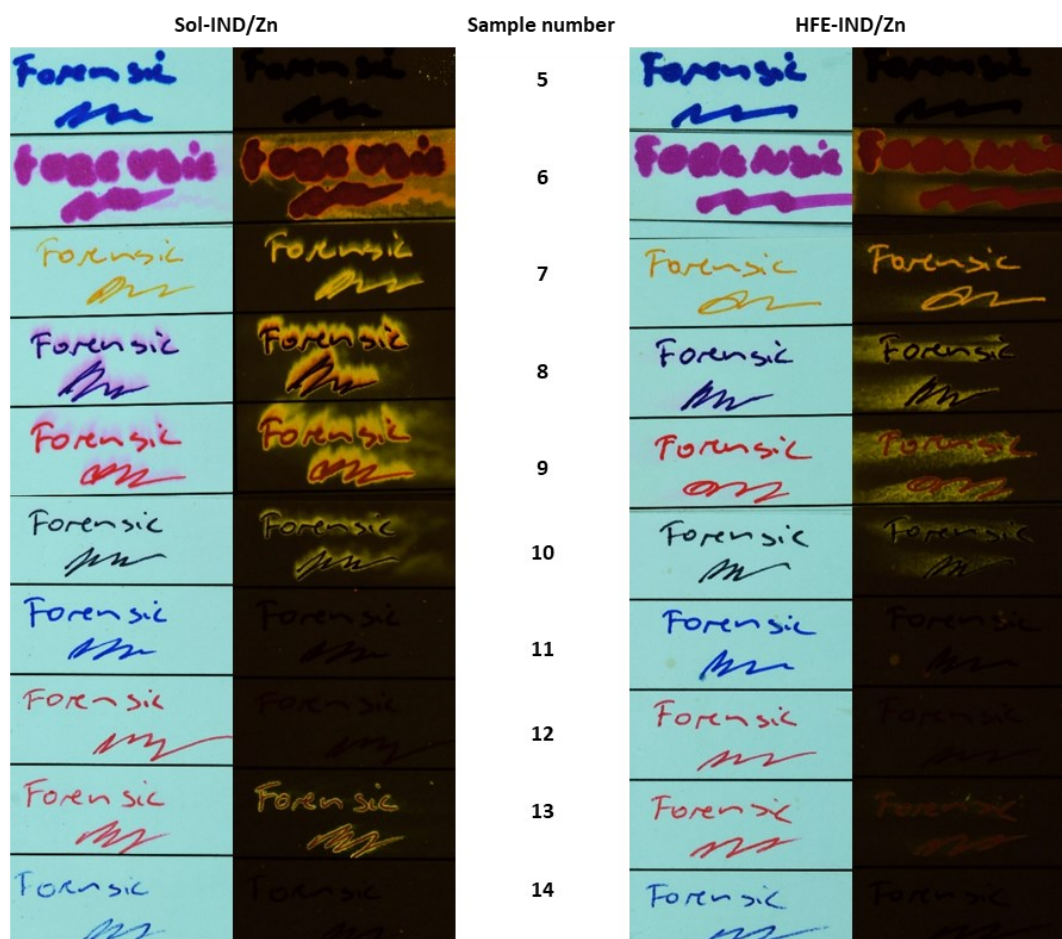


**Figure 3.17:** Comparison of percentage fingermark grades assigned to a) fingermarks aged 1, 7 and 30 days b) fingermarks aged for each donor developed on white copy paper and treated with HFE-IND/Zn and Sol-IND/Zn.

### 3.3.3.2 Compatibility with inks

Following immersion in the IND/Zn working solutions, visible and/or luminescent diffusion was exhibited by a number of inks, which can be attributed to differences in their composition (Figure 3.18).<sup>260, 261</sup> Most of the pens contained inks that were alcohol- or water-based (according to manufacturer information), with the exception of the ballpoint pens, which were based on glycol and benzyl alcohol, or oil. Red ballpoint pens, red and purple gel pens and pink permanent markers appeared to be

particularly affected, which may be due to the solubility of red colourants such as the dye eosin.<sup>261</sup> Significantly greater diffusion was exhibited by ink samples immersed in the Sol-IND/Zn reagent, which was unexpected given the results reported by Olszowska *et al.*<sup>233</sup> and the less polar nature of the NCFS formulation compared to that used by CAST.



**Figure 3.18:** Comparison of the effect of Sol-IND/Zn and HFE-IND/Zn reagents on inks, photographed under ambient light and luminescent mode.

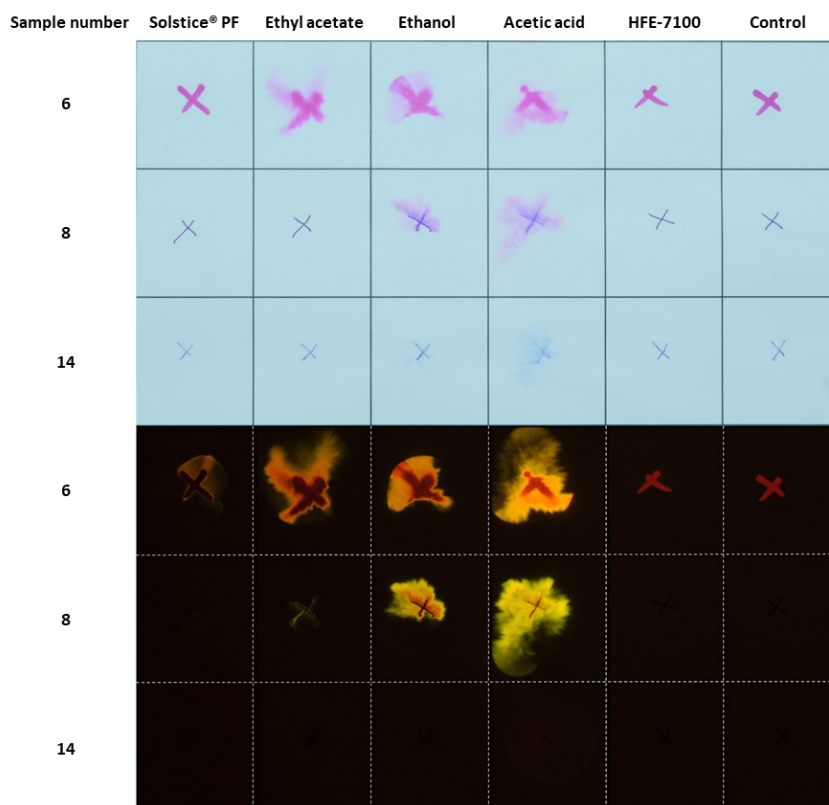
Olszowska *et al.*<sup>233</sup> conducted a similar investigation, primarily using blue and black ballpoint pens as examples of commonly used pens.<sup>262</sup> Their results showed that similar, minor diffusion effects were caused by CAST IND/Zn, regardless of carrier solvent type. By total volume, the NCFS IND/Zn reagent contains a significantly lower volume of highly polar co-solvents: about 20 % of that of the CAST formulation. Additionally, the CAST reagent contains methanol, a solvent not used in the NCFS formulation. The greater proportion of polar co-solvents in the CAST formulation,



particularly the inclusion of methanol, is likely to have a greater effect than any differences in polarity between Solstice® PF and HFE-7100. Methanol is used in the CAST formulation to increase luminescence intensity; however, increased methanol concentration is known to cause diffuse ridge detail as well as unwanted ink bleeding.<sup>116, 263</sup>

### **3.3.3.3 Effect of co-solvents on ink diffusion**

Following these results, the cause of the excess diffusion during Sol-IND/Zn treatment was investigated by spotting neat solvent and a series of 'knockout' solvent mixes (replacing one or two of the co-solvents with an equivalent volume of Solstice® PF) onto a subset of inks. It was found that Solstice® PF and all three co-solvents are sufficiently polar to dissolve at least one type of ink, while HFE-7100 alone had no visible effect (Figure 3.19). Similar results were observed when the experiment was repeated on 5 day old ink samples, indicating minimal effect of ink drying time on the results. However, it should be noted that longer term trials on a larger number of inks are required.



**Figure 3.19:** Inks spotted with neat solvent and photographed under ambient light and luminescence mode.

Further investigations utilising solvent mixes (equivalent to concentration in the IND/Zn reagent) were inconclusive regarding the exact cause(s) of diffusion for some ink types. Though greater ink diffusion was seen with Sol-IND/Zn compared to HFE-IND/Zn, the differences between reagent formulations cannot be solely attributed to the carrier solvent. As an example, ink 8 (a gel ink) did not appear to be affected by Solstice® PF alone but exhibited diffusion when treated with solvent mixes containing acetic acid. Gel inks are typically thickened with agents such as xanthan gum,<sup>264</sup> which is soluble in dilute acids.<sup>265</sup> It is unclear at this stage whether the differences in the degree of ink diffusion caused by the HFE-7100 and Solstice® PF formulations are due to a simple additive effect of Solstice® PF and the co-solvents, or whether there is additional interaction between the carrier solvent and the more polar co-solvents. The proportion of highly polar co-solvents in the NCFS formulation may simply be low enough that a change in carrier solvent polarity has a more noticeable effect. Noting from the HSP values in Table 3.2 the polarity and hydrogen bonding are stronger for Solstice® PF than HFE-7100.

### 3.3.3.4 Reagent stability

After 1.5 months in storage (refrigerator), the Sol-IND/Zn reagent was visually unchanged compared to its appearance immediately after preparation. However, after 2.5 months, the reagent appeared cloudy (Figure 3.20). A white precipitate (presumed to be 1,2-indanedione) was suspended in the solution, which could not be redissolved by allowing the reagent to come back to room temperature or by manual agitation. The precipitate continued to form during storage, such that the solution was almost completely opaque 4 months after preparation. In comparison, a batch of HFE-IND/Zn prepared and stored contemporaneously remained visually unchanged during this time. Further batches of Sol-IND/Zn were prepared to confirm these results, and in these cases, precipitation was first observed at 2-3 weeks. Regardless, the aged Sol-IND/Zn reagent still performed equally to fresh solution at all investigated time periods.



**Figure 3.20:** Sol-IND/Zn after 2.5 months in refrigerated storage, showing white precipitate.

These observations contrast significantly with those of Olszowska *et al.*<sup>233</sup> regarding CAST Sol-IND/Zn, which has been reported to remain stable for at least 6 months.<sup>233</sup> However, the standard HFE-7100 based NCFS and CAST IND/Zn formulations have

very different shelf lives. The CAST formulation has a reported shelf life of up to one year,<sup>127</sup> whereas the NCFS working solution has a shelf life of 3 months.<sup>17</sup> This is potentially due to the greater polarity of the UK formulation, which would enable 1,2-indanedione and zinc chloride to remain more stable in solution.

It remains unclear at this stage why Sol-IND/Zn appears to deteriorate faster than HFE-IND/Zn. The low storage temperature (4 °C) may be a contributing factor as room temperature storage is recommended for the NCFS formulation,<sup>17</sup> though both reagents were stored under the same conditions. Notably, the NCFS Workshop Manual recommends preparing separate stock solutions of 1,2-indanedione and zinc chloride, each with a shelf life of one year. This approach may present a solution for longer-term storage of Sol-IND/Zn, with the working solution prepared as needed.

#### **3.3.3.5 Frugal forensics analysis of Solstice® PF as a carrier solvent for fingerprint development**

These results indicate that Solstice® PF is a satisfactory alternative carrier solvent to HFE-7100 particularly on cardboard surfaces. However, from a frugal forensics perspective it presents several challenges in terms of transportation, storage and cost-effectiveness coupled with similar drawbacks as HFE-700.

Solstice® PF has a low boiling point of 19 °C, however it has a high heat of vaporisation of 194 kJ/kg, enabling it to maintain a liquid state in a climate-controlled laboratory environment.<sup>239</sup> However, because of this property, it must be stored in a pressurised cylinder. This can be problematic for small jurisdictions like Seychelles due to additional freight charges and use of more storage space for pressurised cylinders. Additionally, Solstice® PF is also a single source chemical from Honeywell,<sup>239</sup> consequently suffering from similar supply chain risks as HFE-7100. Therefore, it was determined that Solstice® PF was not the best alternative carrier solvent. Instead, the BKA formulation containing petroleum ether 40-60 °C and a modified 1,2-indanedione concentration of 594 mg/L was brought forward as a more sustainable option for laboratory trials.

### 3.4 Conclusions

This chapter investigated existing 1,2-indanedione zinc formulations to develop a sustainable version suitable for use in Seychelles and Global South jurisdictions. The study began with a head-to-head comparative assessment of three operational formulations used in Australia (NCFS), Germany (BKA) and the UK (CAST) on representative substrates commonly encountered in Seychelles. Despite the composition variations primarily associated with the difference in the carrier solvents, all three formulations were highly sensitive and gave similar performance. The BKA formulation was identified as the most cost-effective and supply-chain-friendly option, making it an ideal choice for resource-limited micro-jurisdictions that require minimal adaptation. The IND/Zn formulations were then modified using a frugal forensics approach, specifically focusing on modifying the co-solvents, active chemicals and carrier solvents.

Investigation into the role of polar solvents revealed that the more polar-modified IND/Zn formulations produce marginally stronger luminescence of the developed fingerprints, particularly with the use of methanol. The result supports the suggestion that boosting the co-solvents polarity can improve the luminescence of fingerprints, likely due to the increased solubility of 1,2-indanedione. However, an increase in polar solvent was also found to cause more background development and ridge diffusion, hence, it is crucial to have a balance between sensitivity and quality of fingerprints when altering polar solvents in the IND/Zn formulation. Consequently, the amount of co-solvents in the BKA formulation was maintained as it was seen to strike the right balance, and the use of ethanol was chosen over methanol for safety considerations, in line with the frugal forensic principles.

The concentration of 1,2-indanedione in the BKA formulation was found to be a significant cost factor, and that reducing it improves the effectiveness of the reagent. The result was attributed to excess 1,2-indanedione forming non-fluorescent oligomeric side products masking the luminescence of JP. Therefore, the formulation was modified to contain 594 mg/L of 1,2-indanedione, reducing the cost due to 1,2-indanedione by 40 %.

Replacing the expensive and supply chain-disruptive solvent HFE-7100 in IND-Zn formulation is the main challenge in developing a sustainable formulation. To address this challenge, Solstice® PF was investigated as a potential replacement and showed comparable effectiveness to HFE-7100 in the NCFS formulation and producing stronger luminescence, making it better for visualising fingerprints on surfaces such as cardboard. However, it can cause issues with the physical integrity of paper evidence due to relative higher diffusion of inks observed. Increased ink diffusion suggests chemical interactions with other reagent components that may affect stability. Solstice® PF-based reagent was found to form a precipitate within a month of storage, but it did not affect performance over 4 months. Moreover, Solstice® PF presented similar drawbacks as HFE-7100 in terms of supply chain risks.

Based on these results and considering the HSP values and comparable performance of the BKA-petroleum ether 40-60°C based formulation in the head-to-head comparative assessment, no further modification was made with respect to the carrier solvent. Although highly flammable, petroleum ether 40-60°C is an inexpensive and readily available solvent manufactured by multiple sources, providing a better alternative in managing the cost and exposure to supply chain risks associated with HFE-7100. For restricted-budget laboratories where cost and supply chain are key factors in deciding the formulation composition, petroleum ether 40-60°C presents a more sustainable carrier solvent.

As a result of the series of modifications using the frugal forensics framework, a sustainable method for further trials on its potential use in Seychelles was established and referred to as the Seychelles Police Force 1,2-indanedione zinc (SPF IND/Zn). The SPF IND/Zn formulation is as follows -

0.60 g of 1,2-indanedione

45 mL of ethyl acetate

10 mL of glacial acetic acid

900 mL of petroleum ether 40-60 °C

10 mL of zinc chloride stock solution

Zinc stock solution is as follows -

200 mg of zinc chloride

30 mL of ethanol

Processing conditions are as follows –

Heat press for 10 seconds at 160 °C

The SPF IND/Zn formulation was then subjected to laboratory trials to assess its suitability for operational implementation in Seychelles, as detailed in the next chapter. Furthermore, this investigation established a foundation for the experimental methodology employed in the laboratory trials.

## Chapter 4. SPF 1,2-indanedione zinc comparative assessment

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Portions of this chapter have been published or to be submitted for publication in the following articles:

Jemmy T. Bouzin, Jason Merendino, Stephen M. Bleay, Georgina Sauzier, and Simon W. Lewis. New light on old fingermarks: The detection of historic latent fingermarks on old paper documents using 1,2-indanedione/zinc. *Forensic Science International: Reports* **2020** 2 100145.

DOI: <https://doi.org/10.1016/j.fsir.2020.100145>

Jemmy T. Bouzin, Georgina Sauzier, and Simon W. Lewis. A sustainable 1,2-indanedione/zinc formulation for Seychelles. To be submitted.



## 4.1 Introduction

As discussed in Chapter 2, the IFRG guidelines<sup>24</sup> divide the research process into four phases, from the initial proof-of-concept (phase 1) to operational evaluation (phase 4). Each research phase follows specific guidelines regarding the type and number of substrates, donors, and fingermarks. As a method progresses to the latter phases, using more realistic fingermark samples to assess operational suitability becomes important. This is because, in real-life scenarios, fingermarks are not intentionally deposited. Therefore, the use of incidental fingermarks provides more representative samples that are likely to be encountered. In Chapter 3, a modified version of 1,2-indanedione/zinc chloride (IND/Zn) formulated for sustainable use by the Seychelles Police Force (SPF) was developed using deliberately deposited fingermarks in line with a phase 1/2 study. In this chapter, the adapted formulation (referred to as SPF IND/Zn) is evaluated through laboratory trials, including the use of incidental fingermarks in line with a phase 2/3 study.

Another IFRG recommendation is to test reagents under local conditions which accounts for available resources in terms of, personnel, equipment, substrates and environmental conditions. The use of local resources is particularly important in a frugal forensics approach as it acknowledges and adapts to local challenges, which is the cornerstone of the concept. This was demonstrated in Chapter 3 using a more economical carrier solvent and processing conditions for IND/Zn that are more robust to cost or supply chain limitations. The consideration of substrates and environmental conditions is more related to its impact on the latent fingermark detection effectiveness. In Chapter 3, the issue of substrate was addressed by using representative samples commonly encountered in Seychelles.

More importantly, the environmental conditions between Seychelles and the United Kingdom (UK), where most techniques adopted by SPF are developed, differ in terms of humidity and temperature as discussed in Chapter 1.<sup>191, 266</sup> For IND/Zn, the susceptibility to the environmental conditions seems reduced with the addition of zinc chloride to the formulation.<sup>104</sup> Additionally, on porous substrates, the eccrine residue absorbed into the substrate matrix provides a level of protection against

environmental factors. However, long-term exposure to humidity and temperature may affect the survivability of the latent fingermarks through an increased rate of degradation or diffusion processes. Studies have shown that humidity significantly affects the eccrine constituents; for example, chlorides have been determined to diffuse faster under high relative humidity.<sup>35, 70, 267</sup> In another study investigating the potential for degradation of selected amino acids present in eccrine residue, thermal degradation was observed after 3 minutes at 100 °C,<sup>268</sup> although this was not performed on an actual fingermark residue. The combined cyclic exposure to high humidity and temperature may influence the degradation and diffusion of eccrine components of fingermarks over time. This can potentially have consequences on the detection of old fingermarks between jurisdictions with significantly different climatic conditions.

Amino acid sensitive reagents such as IND/Zn, 1,8-diazafluoren-9-one (DFO), and ninhydrin have long been proposed for developing older fingermarks on paper<sup>36, 269</sup>. This has been practically demonstrated by Bleay *et al.*<sup>82</sup> where IND/Zn, DFO, and ninhydrin used in sequence were found effective in developing fingermarks on 32 year old cheques kept under controlled environments, while considerably less effective on older documents with an unknown history of environmental exposure. In contrast, Boudreault *et al.*<sup>69</sup> reported a significant decrease in the quality of developed fingermarks over time on white copy paper treated with IND/Zn, although this was not observed for fingermarks treated in sequence with ninhydrin. Together, these studies suggest that while the binding of amino acids to cellulose is likely to provide some stability, amino acids are not completely stable.

At the initial stage of this thesis, the trends in the effectiveness of the three active IND/Zn formulations (BKA, NCFs and CAST) for visualising latent fingermarks on paper substrates of various ages were explored under two different climatic conditions; those of Western Australia (WA) and the UK. This study was performed on incidentally handled documents representing three ageing periods (< 2, 35-40, 75-90 years) stored under UK and Western Australian conditions. The results indicated that the IND/Zn formulation is highly effective, developing fingermarks on 80 year old documents, and significantly increasing the established timescale for fingermark

detection with amino acid sensitive reagents. Comparison across the three ageing periods showed a common trend of a decrease in useful fingermarks due to progressive diffusion of the target amino acids occurring over time. Interestingly, the difference in climatic conditions appears to be more influential on fingermark development success rate than the formulation used. Exposure to environmental factors is likely to accelerate progressive diffusion of amino acids, though the natural moisture and aging of paper may also be influential. Therefore, the exposure and storage conditions and type of paper are important parameters to consider when assessing the success rate in developing old fingermarks on paper.

This chapter aims to evaluate the SPF IND/Zn formulation, taking into consideration the IFRG guidelines and the environmental considerations. It begins with a preliminary investigation of SPF IND/Zn followed by laboratory trials, including the use of incidental fingermarks.

## **4.2 Experimental**

### **4.2.1 Experimental consideration**

In line with the IFRG guidelines,<sup>24</sup> the laboratory trials were intended to be conducted in Seychelles; however, due to the COVID-19 pandemic all investigations were carried out in WA (discussed in section 2.2.3). Considerations were made to differences in local conditions between WA and Seychelles which may affect IND/Zn efficacy; namely climate (particularly humidity), as well as the composition of paper and other commonly available porous substrates.<sup>24, 115, 223, 227, 255, 270</sup> All substrates were sourced as representative of ones encountered in Seychelles, and additional experiments under humidity and elevated temperature to mimic the tropical conditions of Seychelles were conducted.

Given that the experiment was conducted in Australia, the National Centre of Forensic Science (NCFS) formulation was chosen for the comparative assessment with SPF IND/Zn.

## 4.2.2 Experimental methods

### 4.2.2.1 Substrate

Six substrates were used in this study, summarised in Table 4.1. All substrates were sourced from Officeworks Australia as representative of substrates encountered in Seychelles.

**Table 4.1:** Details of substrates used in the SPF IND/Zn formulation laboratory trials.

Substrate	Description	Colour	Weight (gsm)/Thickness
1	COS Premium Copy paper A4	White	80
2	OfficeMax Copy paper A4, 50% Recycled	White	80
3	Plain white Envelope D5	White	
4	JBurrows Notepad A4, 8mm ruled	Blue	70
5	PPS Cardboard	Brown	3.5 mm
6	Newsprint (The West Australian)	Grey	

### 4.2.2.2 Fingermark collection

Due to COVID-19 restricting the number of donors, natural fingermarks were collected from ten donors (six males and four females) between 22 and 60 years old. The comparative assessment was repeated using incidental fingermarks to increase the number of samples and obtain a reliable result. The fingermark collection procedure is outlined under each section.

### 4.2.2.3 Fingermark development

The IND-Zn working solution and sample development for NCFS was prepared as outlined by Stoilovic *et al.*<sup>221</sup> and described in section 2.5.1 together with that of SPF formulation.

### 4.2.2.4 Photography and grading of developed fingermarks

The photography conditions and grading are outlined in sections 2.6.1 and 2.7, respectively.

### **4.2.3 Preliminary investigations**

A series of preliminary investigations of the SPF formulation were conducted before the laboratory trials.

#### **4.2.3.1 Ink compatibility**

The compatibility of petroleum ether with inks was also investigated following the same procedure as the experiment in section 3.2.6.2, except that the ink templates were allowed to dry for 5 days before each half was treated with SPF or NCFS IND/Zn.

#### **4.2.3.2 Processing conditions**

Processing samples at ambient temperature by leaving samples overnight can be beneficial when a large number of samples need to be processed at any one time. The effectiveness of the process condition was investigated on white copy paper (COS premium) and cardboard using three donors. Each donor deposited two sets of five depletion series fingermarks on each substrate. Samples were aged for 24 hours prior to treatment with SPF IND/Zn. After treatment, one set was processed with a heat press at 160 °C for 10 seconds and the other with ambient heat (20-25 °C) in a closed box under office conditions. Both samples were photographed following the procedure outlined in section 2.6.1 on day 0, 1, 2, 3 and 4 after treatment.

#### **4.2.3.3 Simple comparative assessment**

Prior to laboratory trials, a preliminary comparative assessment was conducted using natural quartered fingermarks to compare SPF to the three active IND/Zn formulations using a similar collection procedure as section 2.3.2. Five donors deposited fingermarks on two substrates: white copy paper (COS Premium) and blue notepad (JBurrows), using the increased surface area of the right and left thumb. Each quarter was treated with a different IND/Zn formulation.

## **4.2.4 Laboratory trials**

### **4.2.4.1 Comparative assessment**

The developed SPF IND/Zn was compared to the NCFS formulation through a laboratory trial. Following a similar collection procedure as section 2.3.2, a total of ten donors deposited fingerprints on five substrates: white copy paper (COS Premium), white copy paper (OfficeMax 50% recycled), blue notepad (JBurrows), cardboard (PPS) and newspaper. Each donor deposited two sets of six depletion series fingerprints on each substrate. The samples were stored for 1 week following the procedure outlined in section 2.3.3. The collection was repeated, except that the samples were aged for 4 weeks, covering a timeframe in which casework samples are more likely to be processed.

### **4.2.4.2 Incidental fingerprints**

Incidental latent fingerprints were collected through an exercise where donors verified the content of an envelope, simulating activity in theft cases. This follows a previously developed method of using incidental fingerprints<sup>271</sup> for more realistic samples to enhance the accuracy of the result. Ten donors were instructed to verify the contents of a plain white envelope containing a 10 x 7 cm strip each of four substrates: white copy paper (COS Premium), white copy paper (OfficeMax 50% recycled), blue notepad (JBurrows) and cardboard (PPS). After verifying each item and placing it back into the envelope, all five paper substrates (including the envelope) were collected. Donors were asked to repeat the task after at least 30 minutes to allow skin secretions to replenish on the fingertips, resulting in two sets of samples and a total of ten items to be processed per donor.

All samples were stored between 24 to 30 hours in an office cupboard prior to development. One complete set of substrates from each donor was treated with SPF and the other with NCFS IND/Zn formulation.

#### **4.2.4.3 Environmental considerations**

To take into consideration the differences in local conditions between WA and Seychelles that may impact the efficacy of SPF IND/Zn, an experiment was conducted to compare fingerprint samples stored in an office setting to those stored in a simulated Seychelles dwelling. This would provide insight into the effectiveness of using SPF in Seychelles. To simulate conditions in Seychelles, the attic (level 5) of the Curtin University Chemistry Building 500 was used. This non-air-conditioned space contains louvre blades that facilitate ventilation, which are typical of the dwellings in Seychelles. Ten donors donated a single set of fingerprints on two substrates: copy paper (OfficeMax 50% recycled) and cardboard. Following procedure 2.3.2, a total of four sets of fingerprints were collected on each substrate, one for each aging period of 1 day, 1 week, 1 month, and 3 months. Each half of the split fingerprints was placed in a carton box and stored/aged either in office conditions or in the level 5 space. Data loggers were placed at each site to monitor the temperature and humidity of the storage locations. The experiment was conducted in the late summer to take advantage of the high level of humidity during this season in WA.<sup>272</sup>

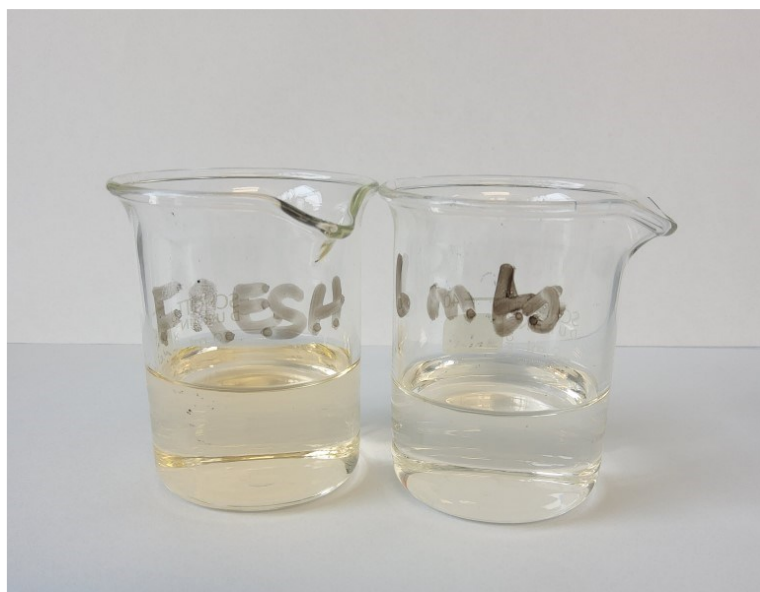
#### **4.2.4.4 Reagent stability**

To assess the reagent stability, SPF IND/Zn solution was used to treat latent fingerprints on copy paper (COS Premium) at 1, 2, 3 and 6 months after preparation. Five split depletion series fingerprints were collected from five donors at each time interval of testing. All samples were stored for at least 24 hours in an office cupboard prior to development. Sample halves treated with the aged reagent were compared to those treated with 'fresh' (less than 7 days old) SPF IND/Zn solution. Visual inspection of the solution was also conducted at these times.

#### **4.2.4.5 UV-visible spectroscopy**

Further investigation was performed using visible spectroscopy to assess changes in 1,2-indanedione peak absorbance due to observed differences in the SPF reagent's colour over time (Figure 4.1). SPF IND/Zn working solutions of more than 6, 12 and 18 months since preparation, together with a fresh solution (less than 1 day old) and

a reference blank were analysed using an Agilent Cary 60 UV-Vis Spectrophotometer as outlined in section 2.6.3.



**Figure 4.1:** SPF 1,2-indanedione working solution less than 1 day old (left) and more than 6 months old (right) showing difference in colour.

#### 4.2.5 Statistical analysis

Statistical analysis software R version 4.3.0 was utilised to assess whether there was significant difference in the fingerprint grades between the IND/Zn treatments. The non-parametric Mann-Whitney U test was used for laboratory trials comparing SPF and NCFS ( $n = 1200$ ). The laboratory trials and incidental fingerprints data ( $n=100$ ) were subjected to a chi-squared test to assess whether the treatments have different distributions in fingerprint grade frequencies. Additionally, a difference in proportion test was also performed on the laboratory trial data, as it is one of the acceptable parametric tests<sup>226</sup> that can be applied to fingerprint grading comparison. All statistical tests were carried out at 95% confidence level ( $p < 0.05$ ).

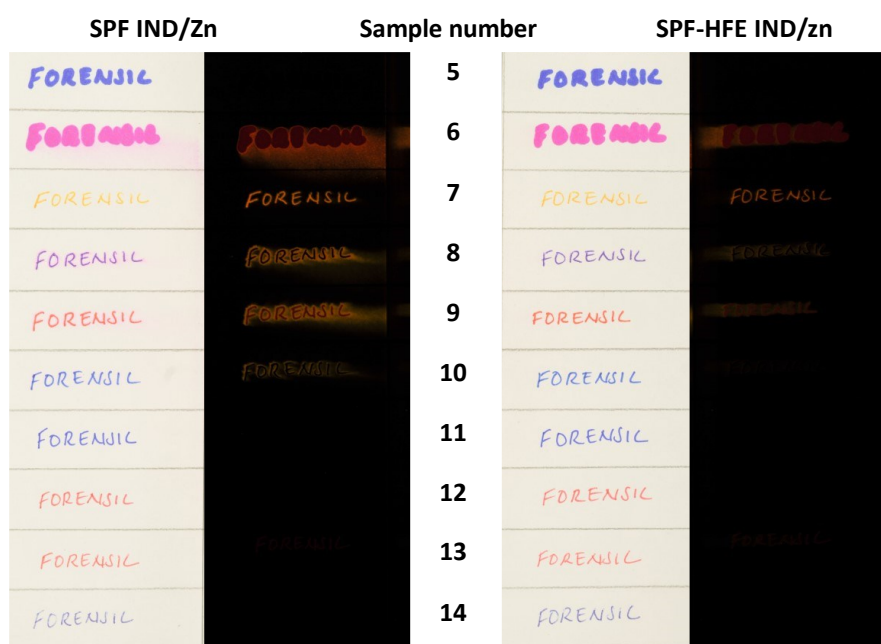


## 4.3 Results and discussion

### 4.3.1 Preliminary investigations

#### 4.3.1.1 Inks compatibility

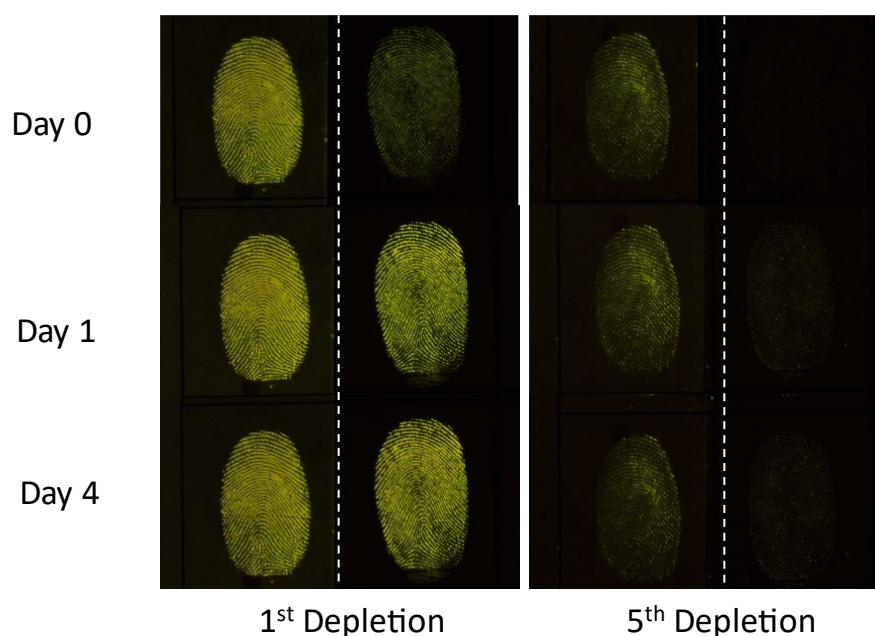
The inks compatibility experiment revealed that the petroleum ether carrier solvent in SPF IND/Zn and HFE-7100 used in the NCFS formulation caused diffusion on the same type of inks. The effect was slightly more pronounced with petroleum ether, as shown in Figure 4.2, but less compared to Solstice® PF evaluated in Chapter 3. Diffusion was more evident with the pink permanent marker (sample 6), which typically contains non-polar colourants and resins in a volatile solvent. Non-polar colourants are used to prevent dissolving of the inks when in contact with water and the resin secures the ink colourant to the paper once the solvent evaporates.<sup>273</sup> The high alkane content in petroleum ether 40-60 °C is likely to increase diffusion of the non-polar constituents in permanent marker. Heptane, the major constituent of petroleum ether, is highly non-polar with a high dispersion force, as indicated by the HSP values (Table 3.2, Chapter 3). In general, most pens including ballpoints, did not show diffusion, indicating petroleum ether's effectiveness comparable to HFE-7100.



**Figure 4.2:** Comparison of the effect of SPF IND/Zn and NCFS IND/Zn reagents on inks, photographed under ambient light and luminescent mode.

#### 4.3.1.2 Ambient temperature processing conditions

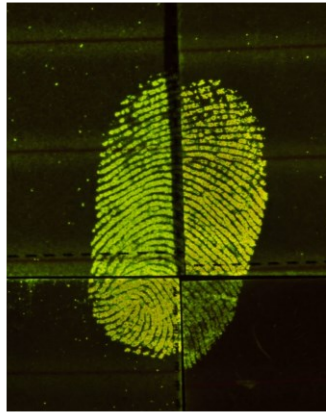
Figure 4.3 shows that fingermarks are fully developed after 24 hours following ambient temperature processing conditions. However, weaker fingermark deposits (5<sup>th</sup> depletions) showed differences in luminescence even after an extended time period of 4 days. This is likely due to the rate of reaction to form fluorophoric Joullie's pink being slower compared to using a heat press. Therefore, it is important to exercise caution when using ambient temperature, as weak fingermarks may be undetected due to reduced sensitivity. It is possible that using elevated outside ambient temperature (> 25 °C) in tropical countries like Seychelles could provide better sensitivity, but further investigation is necessary.



**Figure 4.3:** Comparison of fingermarks treated with SPF IND/Zn and processed with heat press at 160 °C (left) and ambient temperature of 25 °C (right) on copy paper.

#### 4.3.1.3 Simple comparative assessment

A comparison of quartered fingermarks indicated that the SPF formulation was effective when compared to the three active IND/Zn formulations on both substrates. No background staining or ridge diffusion was observed, as shown in Figure 4.4. Based on the preliminary investigation results and findings from Chapter 3, the SPF IND/Zn formulation was brought forward for laboratory trials.

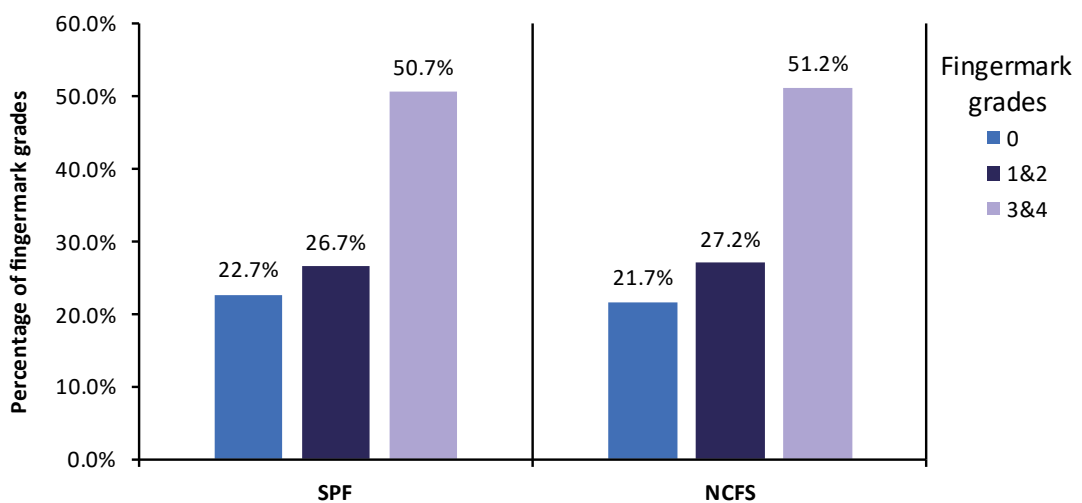


**Figure 4.4:** Comparison of quartered fingerprints on blue notepad paper, developed with SPF IND/Zn (top left), NCFS IND/Zn (top right), and BKA IND/ZN (bottom left) and CAST IND/Zn (bottom right).

### 4.3.2 Laboratory trials

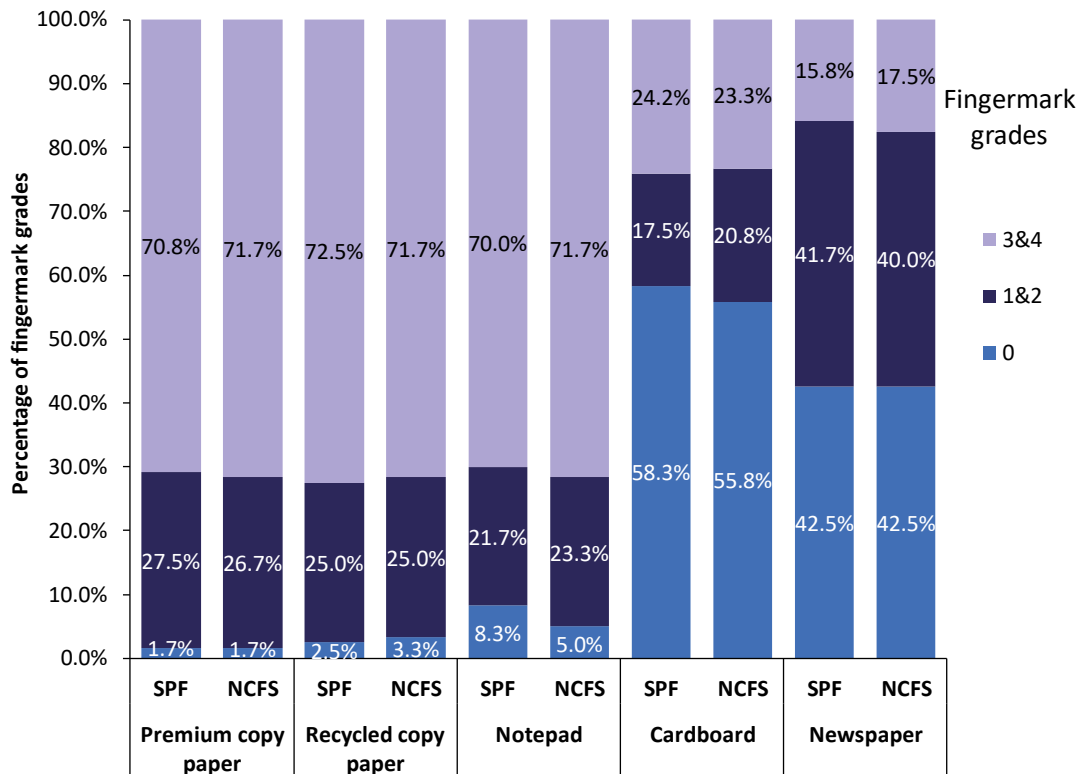
#### 4.3.2.1 Comparative assessment

The comparative assessment of SPF IND/Zn to NCFS generated a total of 1200 fingerprint halves, providing significant data points for analysis. Figure 4.5 shows the overall fingerprint grades and clearly indicates that the SPF formulation is equally sensitive and effective as NCFS IND/Zn, developing 50.7 % and 51.2 % useful fingerprints, respectively. The detection rate for both formulations was lower when compared to the head-to-head comparison of IND/Zn formulations in Chapter 3. This was attributed to the addition of newspaper as a difficult substrate.



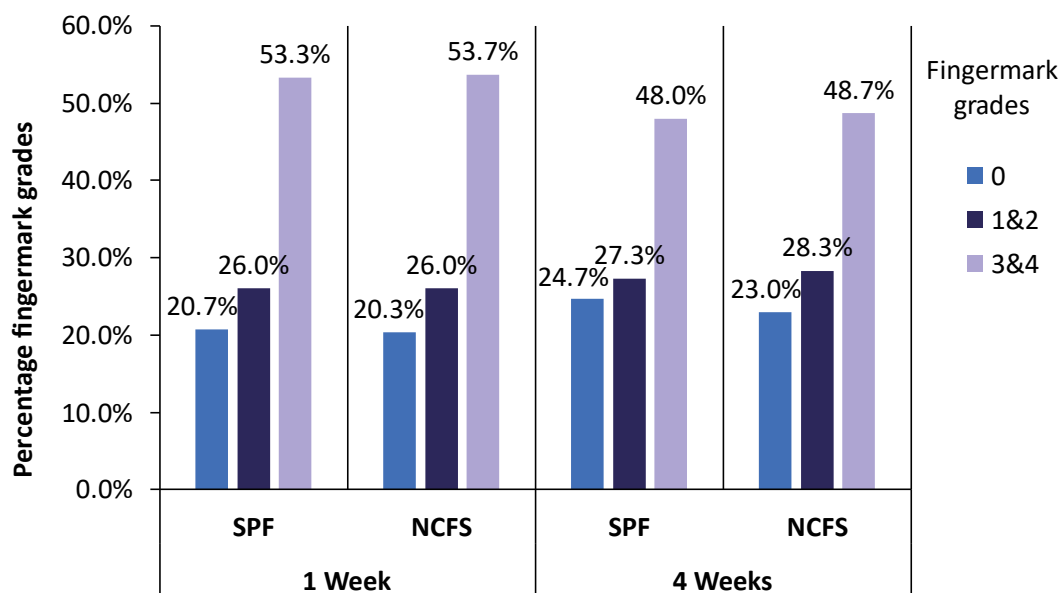
**Figure 4.5:** Percentage of fingerprints graded (3&4), (1&2), and (0) out of 1200 fingerprint halves developed with SPF and NCFS IND/Zn formulations on five different paper substrates.

Two main trends were observed in the grades of fingermarks developed between the different substrates used (Figure 4.6). High recovery trends were observed on white copy paper (Premium and Recycled) and notepad paper, while cardboard and newspaper gave low recovery rates. Both formulations were least effective on newspapers, due to the low-quality paper and fluorescent background. Newspaper is produced using 100% recycled fibres through a mechanical pulping method, which differs from the chemical process employed in paper production.<sup>249, 250, 274</sup> The mechanical treatment of the pulp results in shorter fibre lengths, decreasing the paper's smoothness. Additionally, printed papers like newspapers can exhibit high fluorescence, making any fingermarks on these pages hard to discern from the background due to the poor contrast between the fingermark and substrate, as depicted in Figure 4.8e. Consistent results were observed on cardboard for both treatments. Nevertheless, the performance was inferior when compared to the head-to-head comparison in Chapter 3, particularly for grades 0 and 1&2. This may be due to the deposition pressure, which is evident from the consistent results between SPF and NCFS, given splint fingermarks were used. Overall, SPF had similar trends to NCFS IND/ZN, indicating that SPF performs comparably to NCFS IND/Zn on each type of substrate.



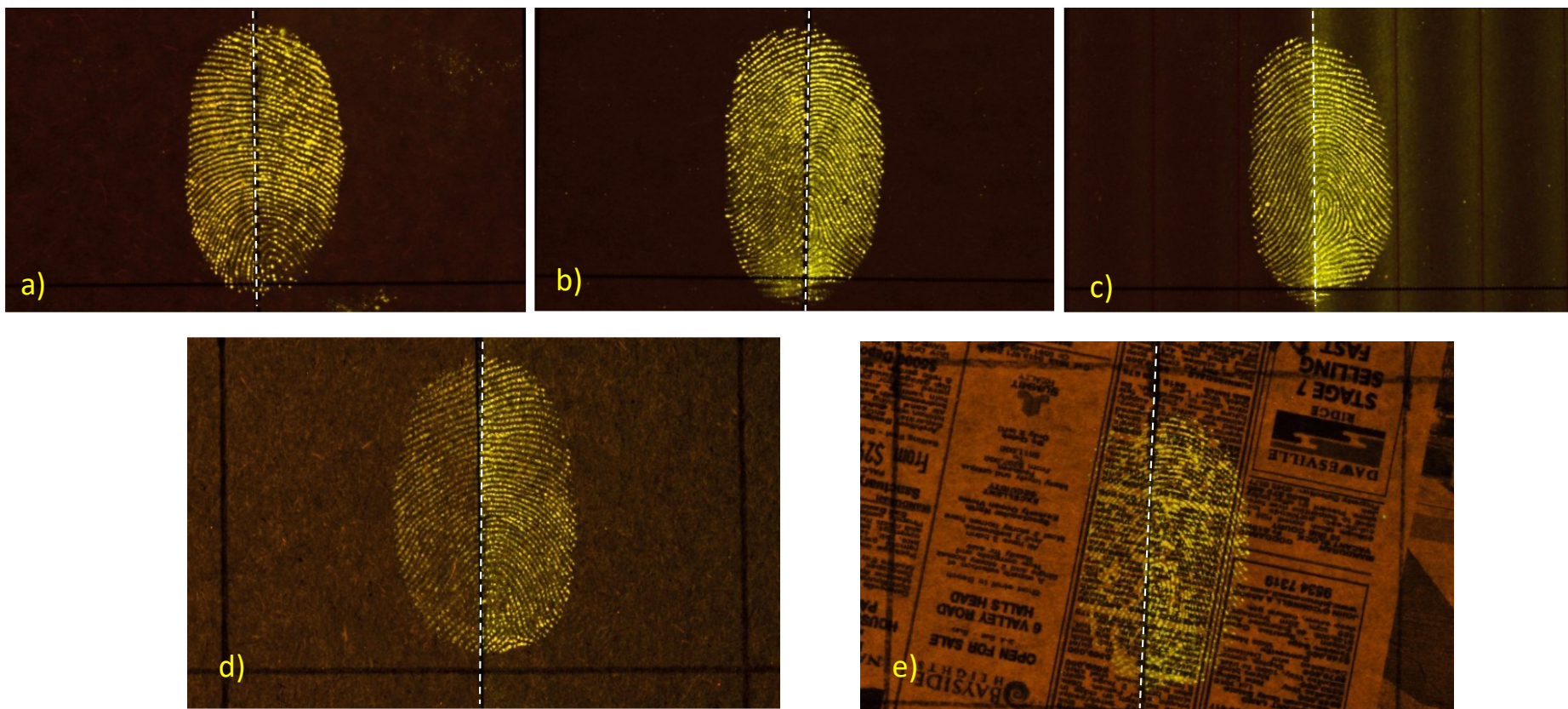
**Figure 4.6:** Comparison of percentage fingermarks grades across five paper substrates, developed with SPF and NCFS IND/Zn formulation.

The two formulations also showed similar effectiveness on samples that were 1 week and 4 weeks old. Figure 4.7 shows a comparable distribution in fingermark grades with a slight reduction in the percentage of useful fingermarks developed for the 4-week samples. This difference is not considered significant as it was expected that the triangle of interaction between fingermark deposit, substrate, and environment over this period could cause slight changes in the fingermark composition. More importantly, the results indicate that the SPF formulation is effective on both 1 week and 4 weeks samples, covering a timeframe during which most casework samples are likely to be processed in Seychelles.



**Figure 4.7:** Percentage of fingermarks graded (3&4), (1&2), and (0) developed for 1 week and 4 weeks samples, treated with SPF and NCFS IND/Zn formulation.

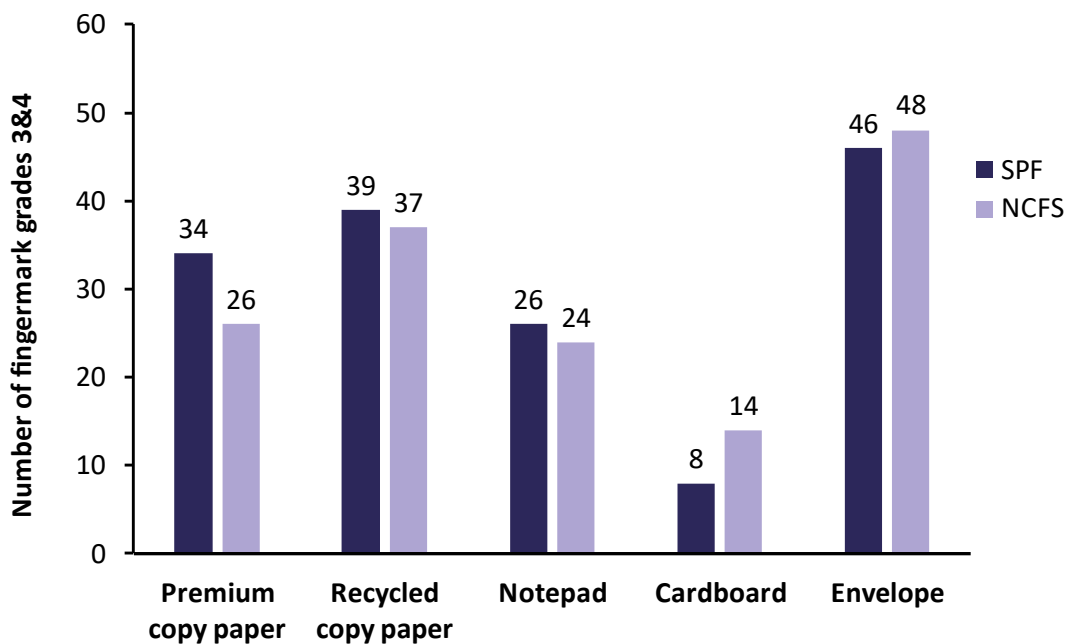
Examination of ridge details and quality of fingermarks developed revealed that those developed using the SPF formulation showed slightly stronger luminescence compared to the NCFS IND/Zn (Figure 4.8). It is worth noting that both formulations have similar concentrations of 1,2-indanedione. Hence, the difference in luminescence is likely due to the SPF formulation having a more polar co-solvent, as discussed in section 3.3.2. The observed difference was more noticeable on cardboard and newspaper compared to white copy paper. On notepad paper, a slightly brighter background was also observed with the SPF formulation. However, these variations did not affect the quality of the developed fingermarks.



**Figure 4.8:** Examples of fingermark developed with NCFS IND/Zn (left half) and SPF IND/Zn (right half) on (a) COS Premium white copy paper, (b) OfficeMax recycled white copy paper, (c) JBurrows blue notepad, (d) PPS cardboard, (e) The West Australian newspaper.

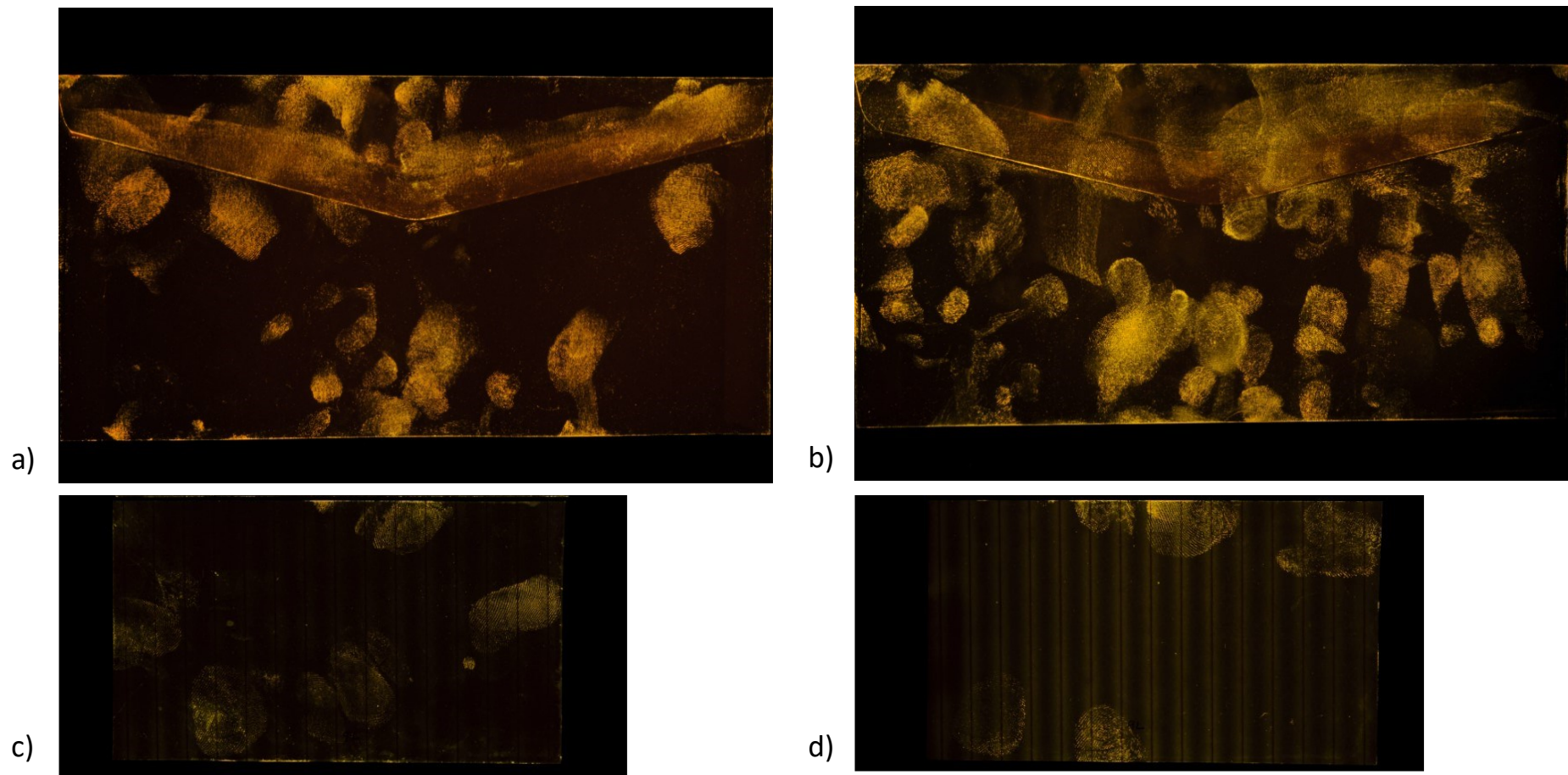
#### 4.3.2.2 Incidental fingermarks

A trial involving incidental fingermarks enabled the performances of the SPF and NCFS IND/Zn formulations to be compared across a range of substrates handled 'realistically' through a simulated exercise. Figure 4.9 shows the number of useful fingermarks developed across the various substrates and reveals that both formulations are highly effective at developing large numbers of incidental fingermarks. Some well-developed fingermarks were not graded as useful due to overlap of ridge impressions as shown in Figure 4.10. As expected, the highest number of fingermarks were developed on the envelope due to being the substrate that was the most handled during the exercise. Most fingermarks were concentrated at the edges of the substrates, which is associated with the activity,<sup>271, 275</sup> as most donors handled the edge of the substrate while inspecting it. A similar quality of fingermarks was developed by both formulations and no ink diffusion was observed on the blue ruled notepad paper. The results based on developing incidental fingermarks on 100 items examined on both sides provide valuable insight into its operational suitability and reinforce the comparable performance between the two IND/Zn formulations observed during the laboratory trials.



**Figure 4.9:** Comparison of total number of useful incidental fingermarks detected with SPF and NCFS IND/Zn treatment on five different substrates.





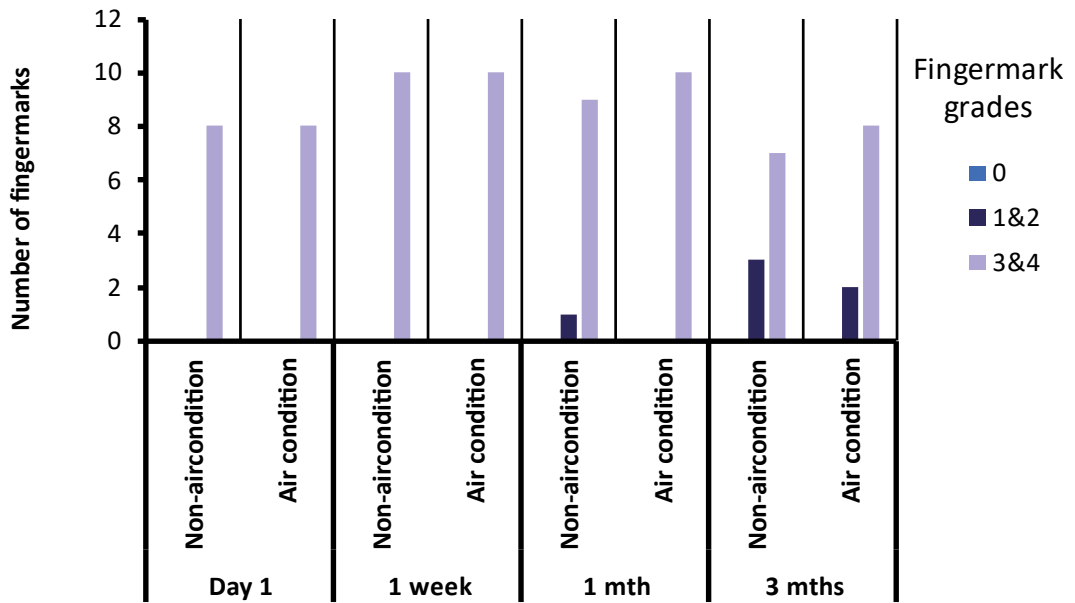
**Figure 4.10:** Examples of incidental fingerprints developed with NCFS IND/Zn (left) and SPF IND/Zn (right half) on (a & b) white plain envelope, (c & d) JBurrows blue notepad. Fingerprints were deposited by the same donor on each type of substrate.

#### 4.3.2.3 Environmental considerations

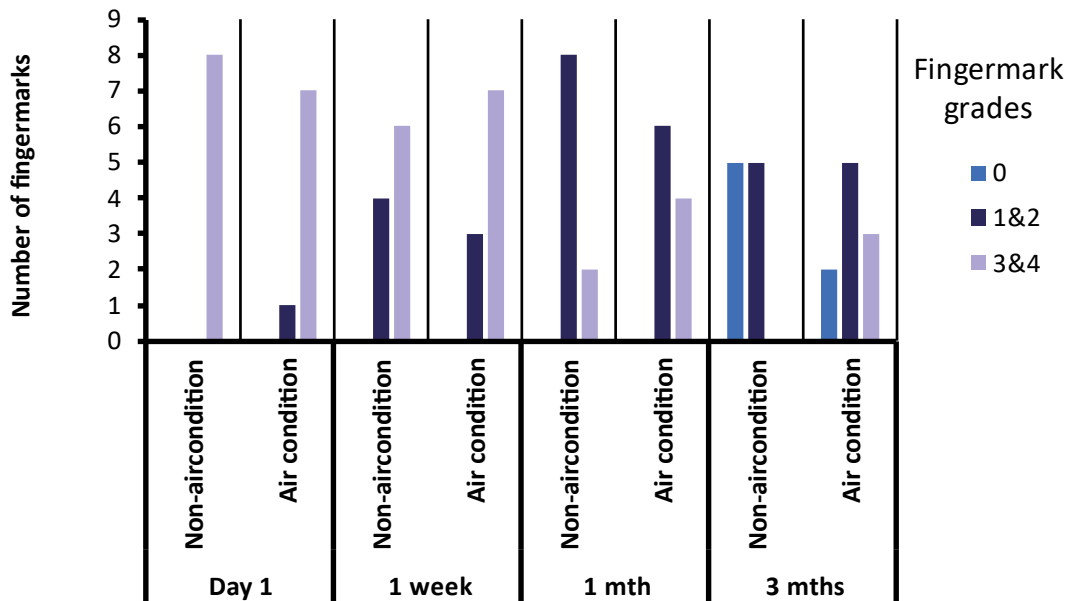
The conditions of the two storage locations registered on the data logger are provided in Appendix B. The air-conditioned location registered a relatively constant temperature of 21 °C and a relative humidity (RH) between 33 % to 67 %, while the non-air-conditioned storage registered a range of temperatures from 14-32 °C and 30-81 % RH. SPF IND/Zn treatment was effective on both samples despite the differences in temperature and RH between the two locations (Figure 4.11 and Figure 4.12). However, slight differences were noted in the quality of fingermarks and background development for samples that were 3 months old, as depicted in Figure 4.13. Fingermarks stored under a non-air-conditioned environment appear fainter with a darker background.

These differences were attributed to the more extreme conditions of the non-air-conditioned environment impacting the fingermark deposit and substrates physicochemical properties. Although amino acids are well preserved in the paper matrix,<sup>82</sup> exposure to cyclic humidity and temperature can cause gradual diffusion of amino acids over time. It has been suggested that water-soluble components such as urea and chlorides can migrate faster under high relative humidity,<sup>14</sup> although this process would be expected to occur at a slower rate for amino acids. Additionally, changes in the natural moisture content of the paper may have a combined effect on the diffusion mechanism and the composition of the paper. It has been demonstrated that various cycle conditions of humidity and temperature change the hydrogen bonding network in paper and its capacity for moisture absorption and desorption.<sup>276</sup> These physicochemical changes over time are likely to impact the diffusion of water-soluble components of latent fingermark residue, including amino acids.

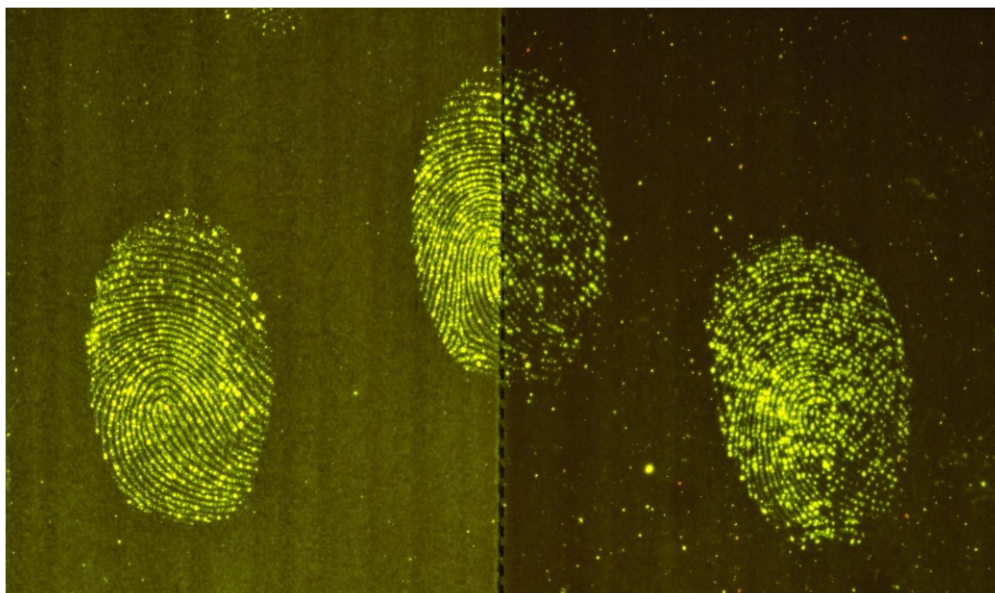
The findings indicate that SPF IND/Zn is effective in high temperature and humidity conditions similar to those in Seychelles for samples less than 3 months old. The consistent performance of SPF IND/Zn on samples stored under two different environmental conditions suggests that the variation in samples over 3 months old is more likely due to the environment rather than the IND/Zn reagent.



**Figure 4.11:** Comparison of fingermarks on white copy paper stored under non-air-conditioned and air-conditioned environments and treated with SPF IND/Zn.



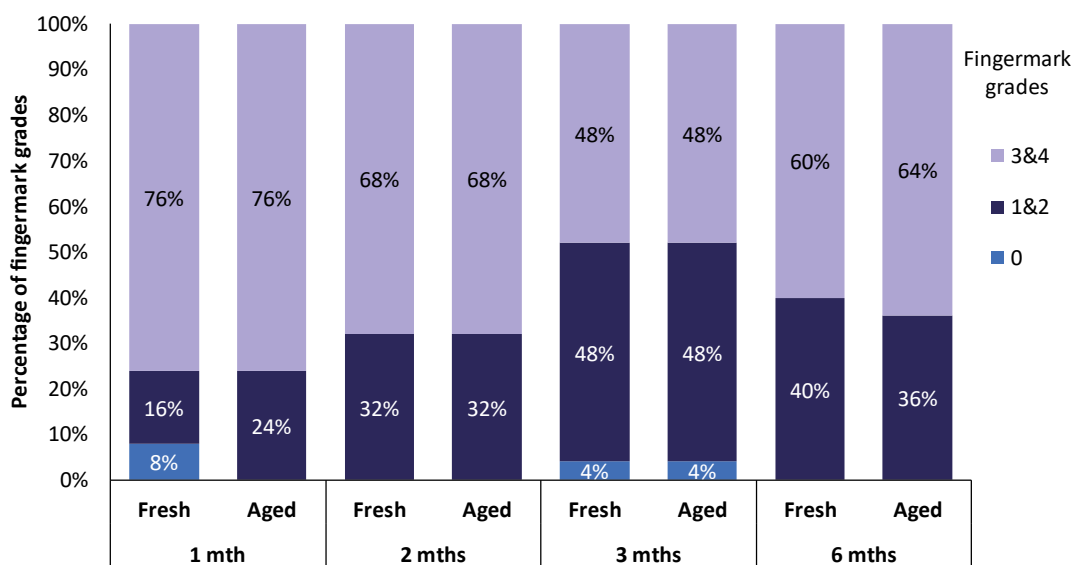
**Figure 4.12:** Comparison of fingermarks on cardboard stored under non-air-conditioned and air-conditioned environments and treated with SPF IND/Zn.



**Figure 4.13:** Comparison of fingerprint ridge details on recycled copy paper stored under an air-conditioned environment (left), non-air-conditioned environment (right) and treated with SPF IND/Zn.

#### **4.3.2.4 Reagent stability**

Investigation into the SPF working solution over 6 months suggested it to be stable, developing a comparable percentage of useful fingerprints as fresh SPF solutions (Figure 4.14). However, the effectiveness decreased for samples developed with 3 months old reagent. This is possibly due to fingerprint variability, since split fingerprints were used, and both fresh and aged SPF formulations showed a decrease in effectiveness. Additionally, the SPF working solution gradually became colourless, as seen in Figure 4.1, indicating possible chemical interactions. Further investigation was performed using visible spectroscopy to assess changes in the 1,2-indanedione peak absorbance.

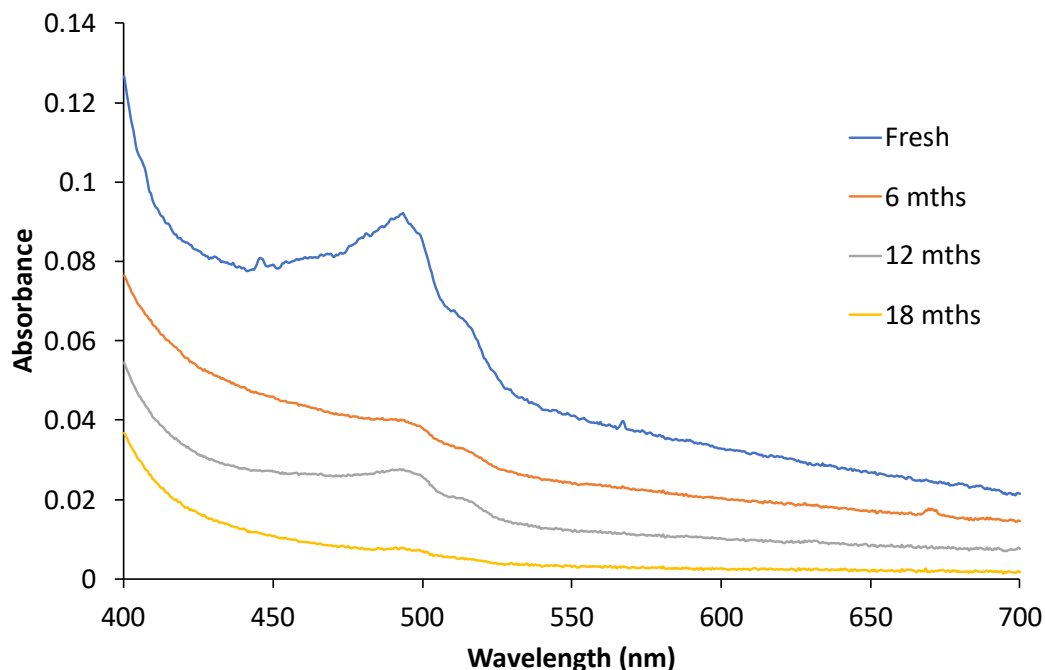


**Figure 4.14:** Percentage of useful fingermarks detected on white copy paper and treated with SPF IND/Zn with increased reagent age.

#### 4.3.2.5 UV-vis spectroscopy

Examination of the spectra of each working solution shows a small peak at 490 nm for fresh working solutions, which correlates with 1,2-indanedione absorption.<sup>17</sup> However, this peak gradually decreased in size as the solution aged (Figure 4.15). 1,2-indanedione is likely responsible for the faint-yellow colour, and the hypochromic effect has been suggested to be due to the formation of a hemiketal structure in the presence of ethanol, which makes the structure less conjugated.<sup>127, 228</sup>

In section 3.3.3, it was observed that a decrease in 1,2-indanedione improves luminescence. However, this effect is only expected to reach a threshold point. Any further decrease in 1,2-indanedione would result in a decrease in luminescence. The SPF formulation containing 594 mg/L of 1,2-indanedione appears to remain above this threshold for working solutions aged more than 6 months. This is based on the comparative performance with fresh working solutions. Therefore, a shelf-life of 6 months is suggested for the SPF formulation. However, it must be noted that working solutions should be made when required and not stored for long durations since it is known that 1,2-indanedione is not stable in non-polar solvents and can interact with other solvents to form compounds that do not visualise latent marks.



**Figure 4.15:** Absorption spectra for SPF IND/Zn working solutions of less than 7 days old, 6, 12 and 18 months since preparation, showing the decrease in 1,2-indanedione peak absorbance.

### 4.3.3 Statistical Analysis

A Mann-Whitney U test performed on the comparative assessment data showed that there is no significant difference in the fingerprint grades between SPF and NCFS IND/Zn treatments ( $p = 0.56$ ). Similarly, a chi-squared test showed that there is no significant difference in the distributions of fingerprint grade frequencies between the two treatments when performed on comparative assessment data ( $p = 0.87$ ) and also when assessing the distributions of useful fingerprint frequencies for the incidental fingerprint data ( $p = 0.58$ ). Additionally, the difference in proportion test showed no significant difference in the proportion of useful fingerprints (grade 3&4) developed by SPF and NCFS IND/Zn treatments in the laboratory trial data ( $p = 0.86$ ).

The statistical findings reinforce the above results, demonstrating that the developed SPF IND/Zn is as effective as active formulations used by different police agencies around the world. The R code and results of statistical analysis are provided in Appendix A-3.

## 4.4 Conclusions

In this chapter, the developed SPF formulation was assessed by comparing it to the NCFS formulation. The assessment was carried out through laboratory and incidental fingerprints trials across five substrates, two ageing periods of 1 and 4 weeks, and a range of ten donors in line with the IFRG guidelines.<sup>24</sup> The findings demonstrated the comparable performance between SPF and NCFS formulations, developing 50.7 % and 51.2 % of suitable fingerprints, respectively. Both formulations exhibited high sensitivity and effectively developed fingerprints that were 1 week and 4 weeks old. Comparison of incidental fingerprints on 100 items gave similar results and statistical analysis reinforced comparable performance. The study concluded that the SPF formulation is stable for at least 6 months, despite spectroscopy studies showing a gradual decrease in 1,2-indanedione peak absorbance over an 18 month period.

This study demonstrated the successful approach of modifying existing techniques within the frugal forensics framework. As a result, this can increase accessibility and facilitate the use of latent fingerprint methods in operational laboratories to improve the recoverability of latent fingerprints. The conclusions above must be qualified with the recognition that the studies were conducted on limited number of fingerprint samples and substrates.

## Chapter 5. Modification of WET UCIO powder suspension for fingerprint detection on pressure sensitive adhesive tape

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Portions of this chapter to be submitted for publication in the following article:

Jemmy T. Bouzin, Georgina Sauzier, and Simon W. Lewis. Modification of Wet UCIO powder suspension for fingerprint development on adhesive tapes for use in Seychelles. *Forensic Chemistry* **2023**. To be submitted.

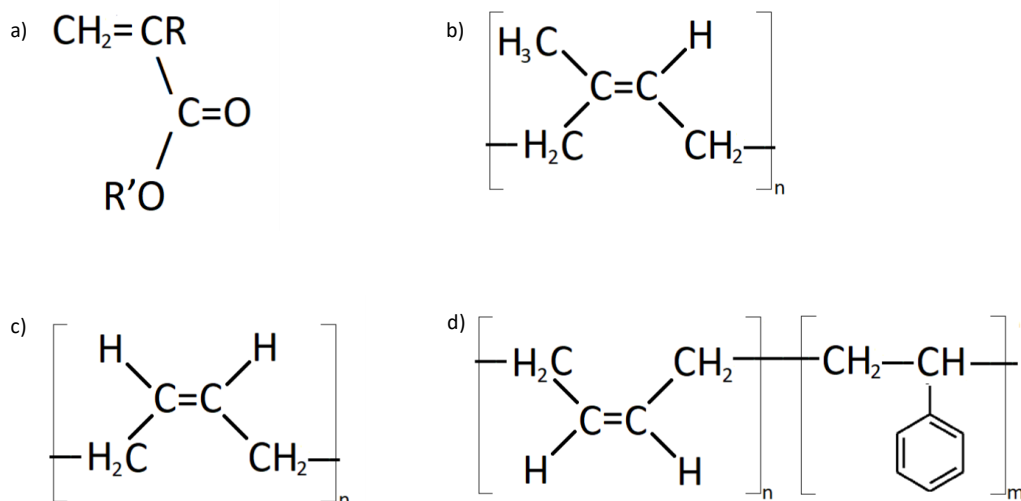


## 5.1 Introduction

This chapter explores the adaptation of a carbon-based powder suspension for latent fingerprint detection on non-porous pressure-sensitive adhesive (PSA) tape. The study demonstrates how the frugal forensics concept can be applied to a recently published method.

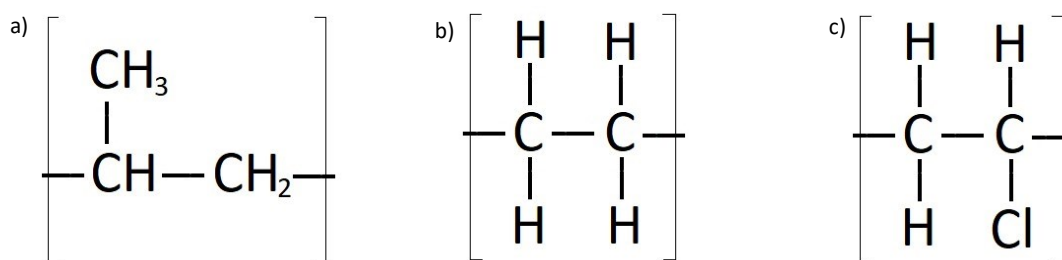
PSA tape is a commonly accessible everyday material that is often the subject of forensic analysis. PSA tapes are used in crimes to immobilise or gag victims in kidnapping and homicide cases, seal drug packaging, and construct improvised explosive devices.<sup>160, 277-279</sup> When handled for any purpose, the sticky-side surface of the adhesive tape serves as a substrate for trace evidence such as fingerprints<sup>279-281</sup>. However, the detection of these fingerprints can be challenging due to the intrinsic adhesive nature of PSA tapes. Moreover, the cost of conventionally recommended fingerprint detection techniques may limit their accessibility in some jurisdictions.

PSA tapes generally have two primary components: a pressure-sensitive adhesive coated on a backing material. Acrylic and rubber-based (natural or synthetic) forms are the most common adhesives.<sup>282 283</sup> Acrylic PSAs are generally manufactured through polymerisation of acrylate monomer with the general structure shown in Figure 5.1a. Various monomers are used, such as methacrylate ( $R' = \text{methyl group}$ ), 2-ethylhexyl acrylate ( $R' = 2\text{-ethyl hexyl}$ ), and copolymer blends.<sup>282</sup> Natural rubber PSA can be based on natural rubber with composition largely corresponding to cis-1,4-polyisoprene; however, due to the high production cost, most are made from synthetic rubber such as poly-cis-1,4-butadiene and styrene-butadiene copolymers.<sup>283</sup> The chemical structures of the polymers are shown in Figure 5.1b-d.

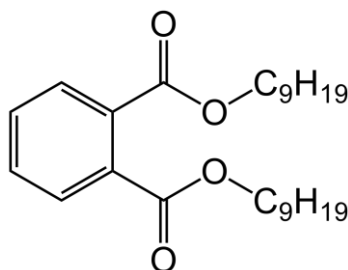


**Figure 5.1:** The molecular structure of materials used in the adhesive of PSA tapes, a) acrylate monomer, b) natural rubber polyisoprene, c) synthetic rubber poly-cis-1,4-butadiene, d) synthetic rubber polystyrene-butadiene.

Depending on the tape application, various backing materials are used. These backings include paper for easy-to-tear tape, polyester for high-strength tapes, and other plastic polymers (Figure 5.2) such as polypropylene, polyethylene, and polyvinyl chloride (PVC).<sup>282</sup> Modifying additives, such as tackifier resins, plasticisers and fillers, may also be incorporated.<sup>283, 284</sup> For example, PVC insulating tape typically contains phthalate ester such as diisononyl phthalate (Figure 5.3) as a plasticiser to impart flexibility along with inorganic materials such as titanium dioxide, calcium carbonate, barium sulfate, talc, kaolin, and antimony oxide; used as fillers, flame retardants, or stabilisers.<sup>284</sup> The variation in chemical composition and the intrinsic adhesive nature, which is key to pressure-sensitive tape functions, makes fingerprint development without background surface staining challenging.



**Figure 5.2:** The molecular structure of materials used in the backing of PSA tapes, a) polypropylene, b) polyethylene, c) polyvinyl chloride.

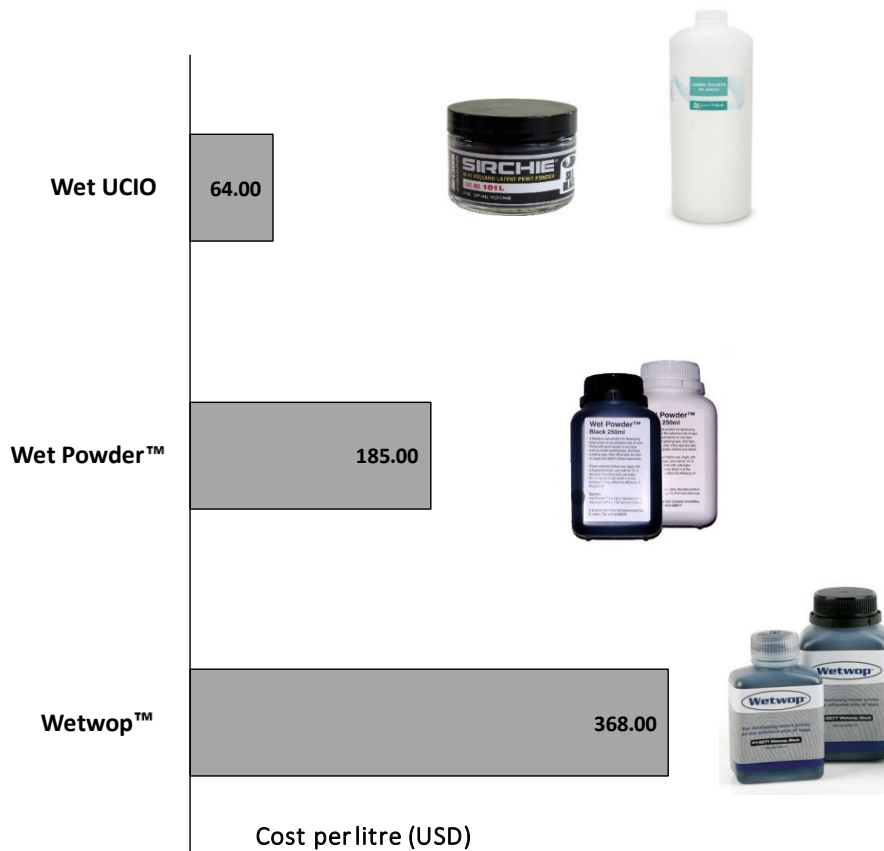


**Figure 5.3:** The molecular structure of diisononyl phthalate, a common plasticiser used in adhesive tape to impart flexibility.<sup>285</sup>

Powder suspension is an effective and recommended method for developing latent fingerprints on the adhesive side of non-porous tapes.<sup>16</sup> It consists of a powder suspended by a surfactant in water to form a paste that is then lightly painted over the adhesive tape using a brush, followed by washing with tap water. The effectiveness of powder suspensions has been reported to depend on several factors. The surfactant critical micelle concentration (c.m.c), a concentration above which surfactant micelle structures are spontaneously formed,<sup>286</sup> has been shown to influence powder suspension effectiveness. In a study by Downham *et al.*<sup>166</sup>, a formulation containing 1 x c.m.c of Triton X-100 surfactant was found to cause background staining, whilst formulations with 2 x c.m.c exhibited discriminate powder deposition on fingerprint, suggesting the possible role of surfactant micelles in the powder deposition mechanism.

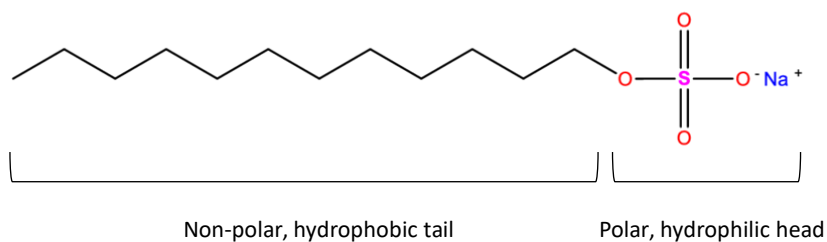
Other factors include the physio-chemical properties of the PSA tape surface. The variation in chemical composition can have a direct impact on the effectiveness of powder suspension. Studies by Bacon *et al.*<sup>287</sup> using carbon-based suspension powder on various polymer-based substrates, found that titanium dioxide pigments in polymer substrates cause surface-wide background staining. This was not observed for similar polymer substrates without titanium dioxide pigments or where the pigments were 30 nm below the surface. Additionally, the type of powder has also been suggested to play a critical role. In a study on different samples of iron oxide (II/III) powders with varying distributions of size, Downham *et al.*<sup>164</sup> reported better performance with powder samples exhibiting higher sub-micrometre particle populations. This suggests that the particle size and shape likely influence powder suspension performance. As a result of the variation, various types of black and white powders have been trialled, leading to several pre-mixed commercial proprietary powder suspension formulations. Black powder suspension formulation is either carbon or iron oxide-based. Commercial carbon-based formulations include Wetwop™ Black from Lightning Powder Company<sup>171</sup> and Wet Powder Black from Kjell Carlsson,<sup>172</sup> whereas Adhesive Side Powder Dark from Sirchie<sup>173</sup> is iron-oxide-based. White titanium dioxide-based powder suspension is also available for treating dark-coloured non-porous adhesive tape.<sup>174</sup>

Carbon-based powder suspension is currently the recommended method for treating light-coloured adhesive side of non-porous tape.<sup>16</sup> However, its operational use may be limited in small laboratories with restricted budgets due to its high cost (Wetwop™ costs \$340 USD per litre). Another consideration when using a pre-mixed product is the general lack of control over formulation quality and shelf-life.<sup>168</sup> This has led to interest in developing cheaper in-house alternatives. A recent example is the development of Wet UCIO powder suspension,<sup>175</sup> consisting of carbon black Sirchie powder mixed with a commercial Gran Velada surfactant solution. This formulation has been reported to be more effective than commercial Wetwop™ on eight types of adhesive tape.<sup>175</sup> while only costing c.a \$64 USD per litre (at least five times cheaper than Wetwop™) (Figure 5.4).



**Figure 5.4:** The products and cost of Wet UCIO and two commercial powder suspension formulations Wet Powder black Wetwop™ black.

However, the Gran Velada surfactant solution is not readily accessible outside Europe,<sup>288</sup> limiting its use in Seychelles or other non-European jurisdictions. Nonetheless, the commercial Gran Velada surfactant solution contains 27 % w/v of sodium dodecyl sulfate (SDS),<sup>175</sup> an anionic surfactant consisting of a hydrophobic alkyl tail of 12 carbon atoms attached to a hydrophilic sulfate group (Figure 5.5).



**Figure 5.5:** The molecular structure of sodium dodecyl sulfate (SDS).

SDS is a low-cost and common surfactant used in various detergent, cosmetic, pharmaceutical and food products.<sup>289-292</sup> Given the accessibility, cost-effectiveness and low toxicity,<sup>293</sup> SDS presents a sustainable alternative to the Gran Velada surfactant solution. Lab-grade SDS is available in different forms, including aqueous solutions of concentration 10 % or 20 % w/v. Another option is lab-grade SDS powder to produce in-house surfactant solution. The latter offers better transportation, storage, and quality control. Additionally, preparing fresh or small-volume stock solutions, as and when required, provides better shelf-life and reduces wastage, which aligns with a frugal forensics approach.

SDS is relatively hydrophilic with a water solubility for commercial salts ranging between 130 to 150 g/L at 20 °C,<sup>293 294</sup> and a generally accepted c.m.c of 8.1 mM at 25 °C.<sup>295-298</sup> To obtain a high SDS surfactant concentration similar to that of Gran Velada solution (27 % w/v), organic solvents, (e.g. ethanol, glycols or glycol ethers) are typically used as additives in the composition.<sup>299</sup> The use of organic solvent has been shown to influence the micellisation process and wettability properties of the surfactant,<sup>298-301</sup> which may impact the powder suspension effectiveness.

This study aims to enhance the accessibility of the WET UCIO powder suspension by exploring readily available SDS salts to produce an in-house surfactant solution as a substitute for the Gran Velada solution. The first part of the study involves characterisation of pressure-sensitive tapes using ATR-FTIR analysis to assess and consider the surface's chemical composition and possible impact. The second consists of modifying the WET UCIO formulation and comparative assessment, which can be categorised into three stages.

- Stage A: Formulation modification and simple comparison to WET UCIO powder suspension.
- Stage B: Supplementary studies investigating the role of additives and background staining on insulating tapes.
- Stage C: Comparative assessment of modified formulation with commercial Wetwop™ black powder suspension.

## 5.2 Experimental

### 5.2.1 Experimental methods

#### 5.2.1.1 Powder suspension preparation

The preparation of the various powder suspension formulations used in this study (Wet UCIO, Wetwop™ black, Wet Powder black, and modified Wet UCIO formulations) are described in section 2.5.2. The composition and approximate c.m.c of the modified formulations are specified in Table 5.1. In this study, Wetwop™ black and Wet Powder black is referred to as Wetwop™ and Wet Powder unless otherwise stated.

**Table 5.1:** *Composition and approximate c.m.c of in-house surfactant solution used in this study. The c.m.c of SDS was taken to be 8.2 mM.<sup>297</sup>*

Designation	Surfactant solution composition	SDS c.m.c (approx.)
1 % SDS	1 g SDS in 100 ml deionised water	4 x c.m.c
5 % SDS	5 g SDS in 100 ml deionised water	20 x c.m.c
10 % SDS	10 g SDS in 100 ml deionised water	40 x c.m.c
27 % SDS	27 g SDS in 5 ml ethanol and 95 ml deionised water	100 x c.m.c
15 % SDS (Wet SPF)	15 g SDS in 5 mL ethanol and 95 mL deionised water	60 x c.m.c
10 % Ethanol	15 g SDS in 10 mL ethanol and 90 mL deionised water	
5 % PEG-400	15 g SDS in 5 mL PEG-400 and 95 mL deionised water	
10 % PEG-400	15 g SDS in 10 mL PEG-400 and 90 mL deionised water	

#### 5.2.1.2 Substrates

Twelve PSA tapes of various brands and types were purchased in new condition for this study (Table 5.2). Investigation primarily made use of six tapes (#1 to #6), with additional insulating tapes (#7 to #12) subsequently purchased for supplementary studies, investigating background staining observed on this type of tape. Before sample preparation, the length circumference of each tape was discarded to ensure a clean surface for IR (infrared) analysis and fingerprint deposition.

**Table 5.2:** *Details of tapes used in this study.*

Sample Code	Tape Type	Brand	Colour	Source
Tape #1	Packaging	Scotch 3M	Transparent	Officeworks
Tape #2	Packaging	Scotch 3M	Transparent	Officeworks
Tape #3	Packaging	Bear	Brown	Bunnings
Tape #4	Duct Tape	Paint Partner	Silver	Bunnings
Tape #5	Masking	PPS	White	Officeworks
Tape #6	Insulating	Nitto	Yellow	Bunnings
Tape #7	Insulating	Click	Red	Bunnings
Tape #8	Insulating	Click	Blue	Bunnings
Tape #9	Insulating	Click	Black	Bunnings
Tape #10	Insulating	Click	White	Bunnings
Tape #11	Insulating	Deta	Yellow	Bunnings
Tape #12	Insulating	Nitto	White	Bunnings

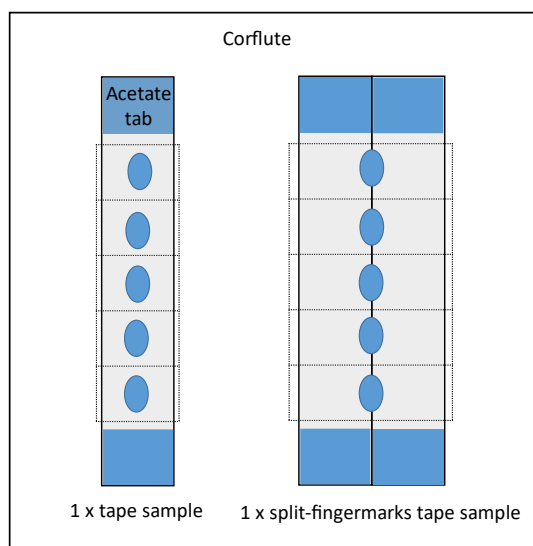
#### 5.2.1.3 ATR-FTIR Tape Analysis

The backing and adhesive of each PSA tape was analysed using Thermo Scientific Nicolet iS50 Fourier Transform Infrared (FTIR) spectrophotometer with a single-bounce attenuated total reflectance (ATR). The methodology is described in section 2.6.4.

#### 5.2.1.4 Fingerprint collection, photography, and grading

Tape samples of approximately 20 cm strips were cut, and acetate backing tabs were placed at each end on the adhesive side for labelling and handling purposes. The tape strips were secured with the adhesive side facing up on a corflute sheet with a background grid to indicate position to deposit fingerprints. Each tape sample consisted of a set of five fingerprint depositions. Two samples of the same tape were secured side by side for split fingerprint deposition as illustrated in Figure 5.6.





**Figure 5.6:** Schematic of latent fingerprint deposition on tape samples.

Fingermarks were collected from a range of ten donors (four females and six males aged between, 22-60) following procedure outlined in section 2.3.2. The number of donors and tapes used for each experiment are specified in Table 5.3. Samples were developed following the procedure outlined in section 2.5.2. Visual examination and photography are outlined in section 2.6.1 and grading of fingerprints is described in section 2.7.

**Table 5.3:** Details of the number of donors, tapes and powder suspension formulations used in each experiment.

Experiment stage	Experiment type	Number of Donors	Tapes used	Powder suspension used
A	SDS solubility	5	#1, #2	1% SDS, 5% SDS, 10% SDS, 27% SDS
	Wet SPF formulation	3	#1, #2	Wet SPF, 27% SDS
B	Comparison Wet SPF to Wet UCIO	5	#2, #4, #6	Wet SPF, Wet UCIO
C	Role of additives	5	#3, #5, #6, #7	Wet SPF, Wet SPF - 10 % Ethanol, Wet SPF - 5 % PEG-400, Wet SPF - 10 % PEG-400
	Insulating tape background staining	5	#6, #7, #8, #9, #10	Wet SPF, Wet UCIO, Wet Powder
D	Comparative assessment Wet SPF to Wetwop	10	#2, #3, #4, #5, #6, #10, #11, #12	Wet SPF, Wetwop
	Sensitivity test	5	#1, #4, #7	Wet SPF, Wetwop

### **5.2.2 Stage A – Formulation modification**

Experiments were conducted to explore the effectiveness of modified WET UCIO powder suspension prepared with SDS concentration below the water solubility value, specifically 1 %, 5 %, and 10 % SDS surfactant solutions. Due to limited Gran Velada SDS surfactant solution available for this study, an in-house modified Wet UCIO powder suspension produced from 27 % SDS was used for the preliminary comparison. To achieve 27 % w/v SDS concentration, the surfactant was dissolved in 5 % v/v ethanol/deionised water mixture. Each donor deposited four sets (one for each formulation) of single fingermarks on each tape (acrylic and rubber-based adhesive tapes).

Additionally, a modified Wet UCIO formulation produced from 15 % w/v SDS formulation, designated “Wet SPF” was compared with the 27 % w/v SDS formulation, using the same tapes as above. Each donor deposited two sets of split five-depletion series fingermarks on each tape to provide an initial insight into the sensitivity of this formulation.

The Wet SPF formulation was subsequently compared to WET UCIO in line with phase 1 of the IFRG guidelines. Each donor deposited two sets (one for each formulation) of five-depletion series fingermark on each tape (packaging-, duct- and insulating tapes).

### **5.2.3 Stage B – Supplementary studies**

To investigate the effect of additives on powder deposition, the Wet SPF formulation was compared to three modified formulations with varying concentrations and types of additives; namely 10 % ethanol, 5 % PEG-400 and 10 % PEG-400 (Table 5.3). Each donor deposited four sets (one for each formulation) of single fingermarks on each tape (brown packaging, Nitto insulating, Click insulating, and masking tape).

Background staining was investigated using three different powder suspensions (Wet SPF, WET UCIO and commercial Wet Powder) on five insulating tapes. The same

fingermark procedure as above was followed, except that three sets of fingermarks were collected, one for each formulation.

#### **5.2.4 Stage C - Comparative assessment to Wetwop™**

An assessment was made of the relative performance of Wet SPF powder suspension with the commercial Wetwop™, which is the current operational technique in Seychelles. The assessment was carried out using ten donors on eight adhesive tapes and two ageing periods (1 week and 4 weeks) in line with phase 2/3 of the IFRG guidelines.<sup>24</sup> Each donor deposited two sets (one for each ageing period) of single split fingermarks on each tape. Following fingermark collection, each strip was stuck to the backing of another strip of the same tape to simulate the condition of tape packages operationally encountered. The extra strip was removed before fingermark development.

To assess sensitivity, ten depletion series of natural split fingermarks were collected from each donor on three tapes. The samples were aged for 1 week following a similar procedure as above.

#### **5.2.5 Statistical analysis**

Statistical analysis software R version 4.3.0 was utilised to assess whether there was significant difference in the fingermark grades developed by different powder suspension formulation treatments. The non-parametric, Mann-Whitney U and chi-squared tests were used to assess the overall performance ( $n= 320$ ) and sensitivity ( $n= 300$ ) between Wet SPF and Wetwop™. Additionally, a difference in proportion test was also performed on both data sets to assess whether the proportion of grade 3s and 4s (useful fingermarks) of one treatment differs from the other. All statistical tests were carried out at 95 % confidence level ( $p < 0.05$ ).

## 5.3 Results and discussion

### 5.3.1 Tape Characterisation

As discussed in section 5.1, the chemical composition of PSA tapes may affect powder suspension performance, hence characterisation of the tapes was carried out. The IR spectra obtained allowed the backing and adhesive of the 12 tapes used in this study to be grouped based on the main polymer present. The backing was divided into four groups: polypropylene, polyethylene, cellulose acetate, and PVC whereas the adhesive was differentiated into two groups, namely acrylic- and synthetic rubber-based polymer (Table 5.4). The characteristic peak assignment for the different polymers is provided in Table 5.5.

**Table 5.4** Classification of adhesive and backing composition based on IR analysis of the main polymer type.

Sample Code	Tape Type	Brand	Colour	Adhesive Composition	Backing Composition	Source
Tape #1	Packaging	Scotch 3M	Transparent	Acrylic	Polypropylene	Officeworks
Tape #2	Packaging	Scotch 3M	Transparent	Rubber	Polypropylene	Officeworks
Tape #3	Packaging	Bear	Brown	Acrylic	Polypropylene	Bunnings
Tape #4	Duct Tape	Paint Partner	Silver	Rubber	Polyethylene	Bunnings
Tape #5	Masking	PPS	White	Rubber	Cellulose	Officeworks
Tape #6	Insulating	Nitto	Yellow	Rubber	PVC	Bunnings
Tape #7	Insulating	Click	Red	Rubber	PVC	Bunnings
Tape #8	Insulating	Click	Blue	Rubber	PVC	Bunnings
Tape #9	Insulating	Click	Black	Rubber	PVC	Bunnings
Tape #10	Insulating	Click	White	Rubber	PVC	Bunnings
Tape #11	Insulating	Deta	Yellow	Rubber	PVC	Bunnings
Tape #12	Insulating	Nitto	White	Rubber	PVC	Bunnings

#### 5.3.1.1 PSA tape backing

The spectra of the backing of packaging tape #1, #2 and #3 all show the characteristic polypropylene profile (Figure 5.7) with quadruplet absorption bands at 2950, 2868  $\text{cm}^{-1}$  and 2916, 2840  $\text{cm}^{-1}$  attributed to C-H stretching of  $\text{CH}_3$  and  $\text{CH}_2$  groups, peaks at 1452, 1377  $\text{cm}^{-1}$  attributed to C-H bending of  $\text{CH}_2$  and  $\text{CH}_3$  groups, and minor peaks at 1168, 974  $\text{cm}^{-1}$  assigned to C-C stretching and C-H rocking of  $\text{CH}_3$  and at 840  $\text{cm}^{-1}$  assigned to C-H rocking of  $\text{CH}_3$  groups.<sup>302, 303</sup>

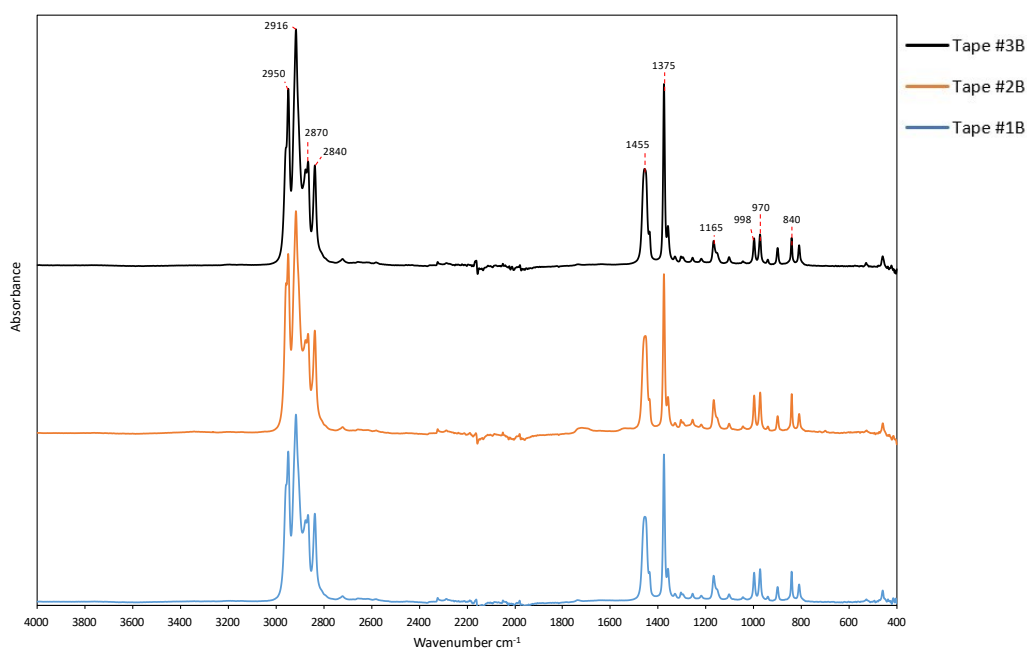
The spectrum of the backing of duct tape #4 shows the typical pattern of linear low-density polyethylene (LLDPE) (Figure 5.8a) with peaks at 2914 and 2846  $\text{cm}^{-1}$  attributed to C-H stretching and characteristic peaks of both high-density polyethylene (HDPE) pair of splitting peaks at 1468,1462  $\text{cm}^{-1}$  and 726,716  $\text{cm}^{-1}$  attributed to  $\text{CH}_2$  in-phase and out-of-phase rocking, and low-density polyethylene (LDPE) single peak at 1379  $\text{cm}^{-1}$  (attributed to methyl umbrella mode from the side chain).<sup>304, 305</sup>

**Table 5.5:** PSA tapes characteristic IR peak assignment for PSA tapes backing and adhesive.<sup>302-311</sup>

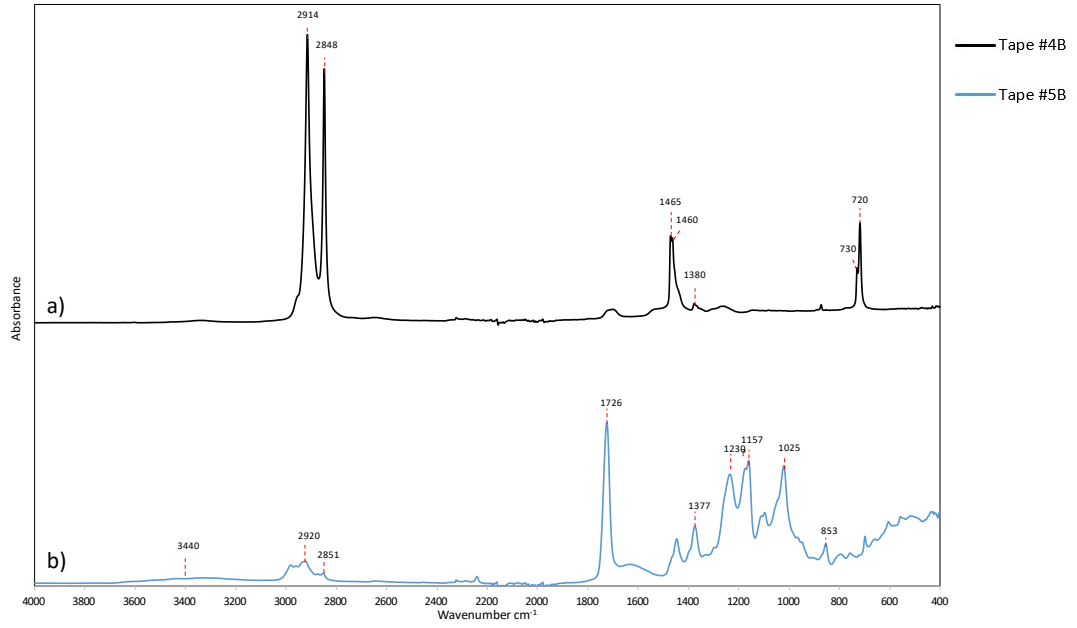
Tape side	Polymer type	Absorption band ( $\text{cm}^{-1}$ )	Assignment
Backing	Polypropylene	840	CH rocking
		974	CH <sub>3</sub> rocking
		1168	C-C stretching
		1377	CH bending symmetrical
		1452	CH bending symmetrical
		2950, 2868 2916, 2840	C-H stretching of CH <sub>2</sub> and CH <sub>3</sub>
	Polyethylene	726	CH <sub>2</sub> rocking
		1468	CH <sub>2</sub> scissoring
		2914	C-H stretching Symmetrical
		2846	C-H stretching Symmetrical
	Cellulose acetate	1026	C-O stretching
		1230	C-O-C stretching
		1377	CH <sub>2</sub> deformation
		1726	C=O stretching
		3400 (weak)	O-H stretching
	Polyvinylchloride	615, 698	Stretching
		955	CH <sub>2</sub> rocking
		1316, 1252	C-H bending of CHCl
		1423	CH <sub>2</sub> bending
		2915, 2850	CH <sub>2</sub> stretching
Adhesive	Acrylate	1160	-C-O-C stretching asymmetrical
		1240	C-O stretching
		1730	- C=O stretching
		2958, 2930, 2872	C-H stretching
	Styrene-isoprene copolymer	750, 695	C H deformation in ring (polystyrene)
1451, 1376		C-H deformation	
1600, 1490 (weak)		Ring vibrations	
2956, 2930, 2854		C-H stretching	

The backing of masking tape #5 conforms to a cellulose acetate polymer (Figure 5.8b) with a characteristic weak broad peak at 3400 attributed to O-H stretching, peaks between 2920 and 2851  $\text{cm}^{-1}$  assigned to CH stretching, typical strong peak at 1726  $\text{cm}^{-1}$  assigned to C=O stretch of the ester group, 1377  $\text{cm}^{-1}$  assigned to  $\text{CH}_2$  deformation vibration, typical peaks 1230, 1026  $\text{cm}^{-1}$  attributed to C-O-C stretching of the acetyl group and C-O stretching of pyranose ring respectively.<sup>303, 306, 307</sup>

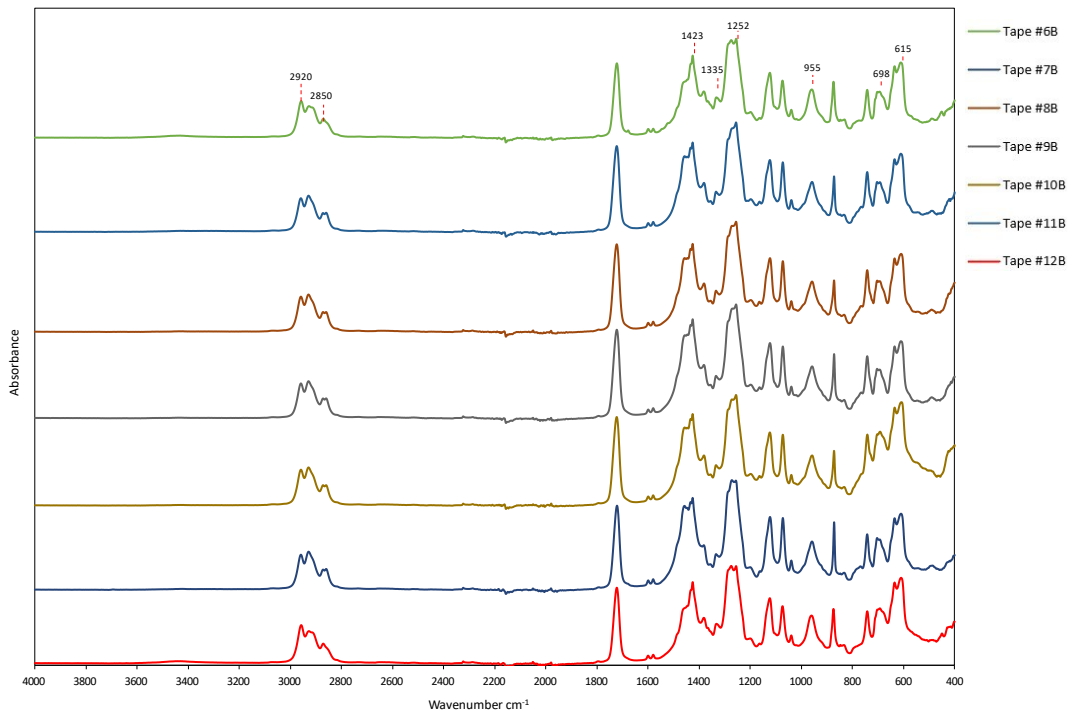
The insulating tapes #6-12 were identified as PVC backing (Figure 5.9) with characteristic peaks at 2915 and 2850  $\text{cm}^{-1}$  assigned to  $\text{CH}_2$  stretching, 1423  $\text{cm}^{-1}$  attributed to  $\text{CH}_2$  bending, 1316  $\text{cm}^{-1}$  and 1252  $\text{cm}^{-1}$  to C-H bending of CH-Cl, 955  $\text{cm}^{-1}$  assigned to  $\text{CH}_2$  rocking and peaks at 698  $\text{cm}^{-1}$  and 615  $\text{cm}^{-1}$  to C-Cl stretching.<sup>306, 307</sup> Additional absorption bands were attributed to phthalates plasticiser, particularly weak doublet at 1591 and 1582  $\text{cm}^{-1}$  and stronger peak at 744  $\text{cm}^{-1}$  assigned to phthalate aromatic ring vibration. Other peaks include 1726  $\text{cm}^{-1}$  and 1120  $\text{cm}^{-1}$ , 1068  $\text{cm}^{-1}$  attributed to the ester group.<sup>304, 308, 309</sup>



**Figure 5.7:** ATR-FTIR spectra of polypropylene backing of packaging tape #1-3. Spectra have been offset for clarity.



**Figure 5.8:** ATR-FTIR spectra of (a) polyethylene backing of duct tape #4 and (b) cellulose acetate backing of masking tape #5. Spectra have been offset for clarity.

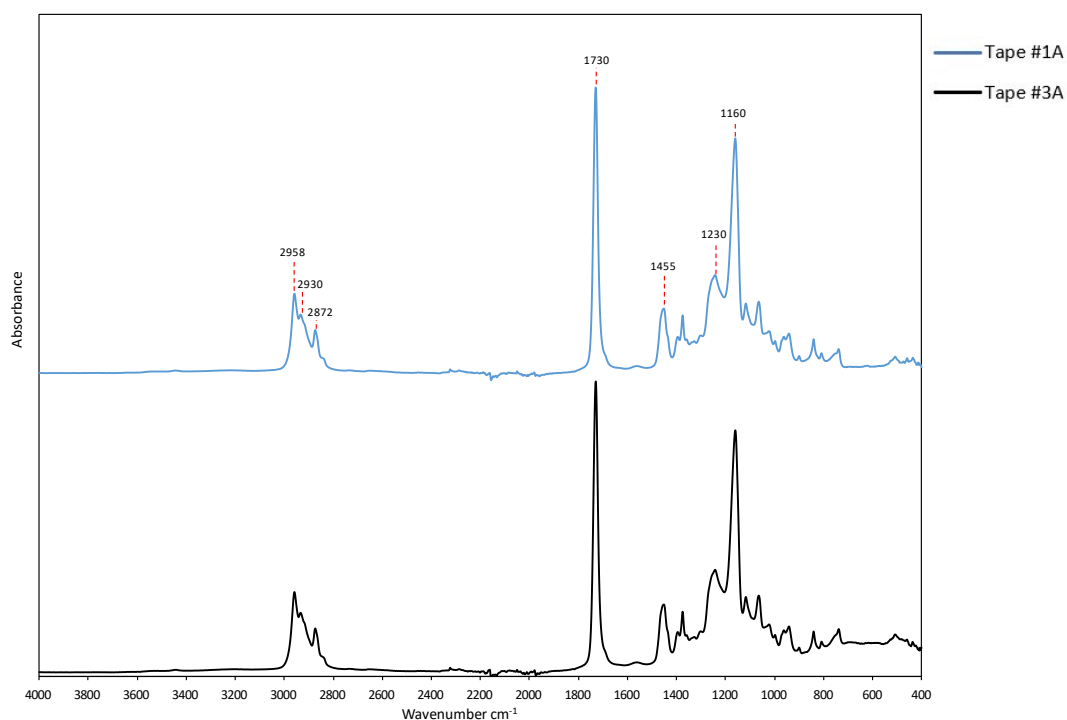


**Figure 5.9:** ATR-FTIR spectra of polyvinylchloride backing of insulating tapes #6-#12. Spectra have been offset for clarity.

### 5.3.1.2 PSA tape adhesive

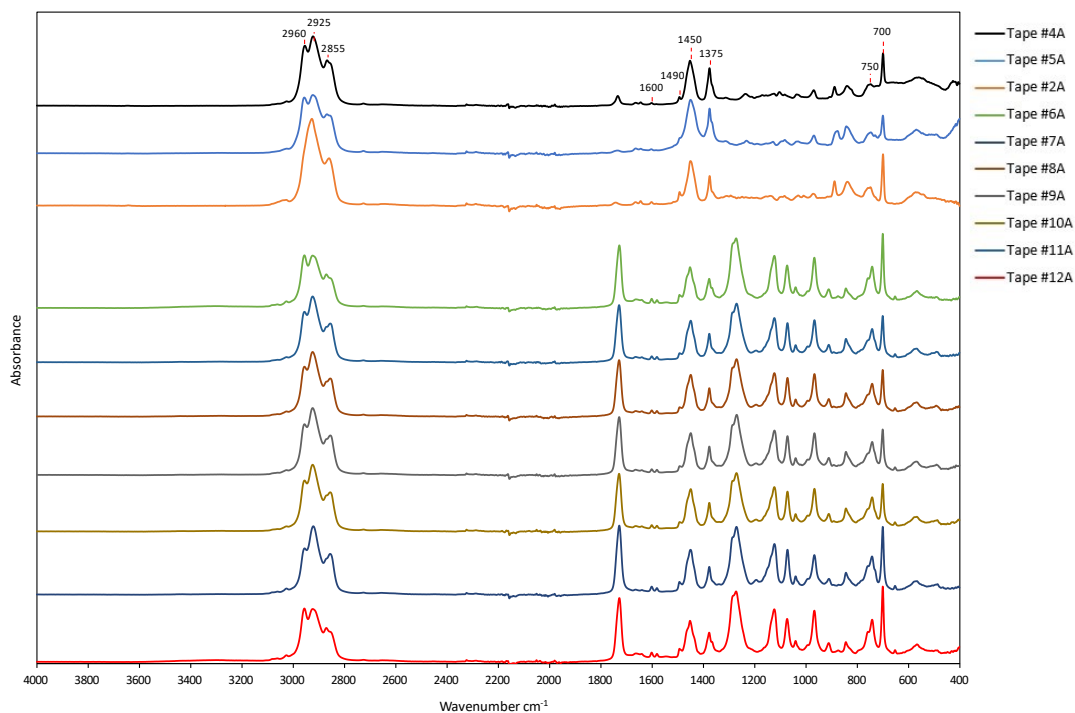
The spectra of adhesive for packaging tape #1 and #3 show typical pattern of acrylic-based polymer (Figure 5.10), with peaks at 2958, 2930  $\text{cm}^{-1}$  and 2872  $\text{cm}^{-1}$  assigned to C-H stretching of  $\text{CH}_2$  and  $\text{CH}_3$  groups, a characteristic strong absorption band at 1730  $\text{cm}^{-1}$  attributed to C=O stretching of ester group, a broad peak at 1240  $\text{cm}^{-1}$  and the strong absorption at 1160  $\text{cm}^{-1}$  assigned to stretching vibration of the ester group.<sup>303, 306</sup>

The adhesive of packaging tape #2, duct tape #4, masking tape #5 and insulating tapes #6-12 were all identified as synthetic rubber (styrene-isoprene copolymer) (Figure 5.11), showing aliphatic absorption bands at 2956, 2930, 2854  $\text{cm}^{-1}$  attributed to C-H stretching, at 1451, 1376  $\text{cm}^{-1}$  assigned to C-H deformation, and aromatic diagnostic bands at 1600, 1490  $\text{cm}^{-1}$  assigned to ring vibration, and peaks at 750, 695  $\text{cm}^{-1}$  assigned to bending of aromatic =C-H and C=C groups of polystyrene.<sup>306, 310</sup>



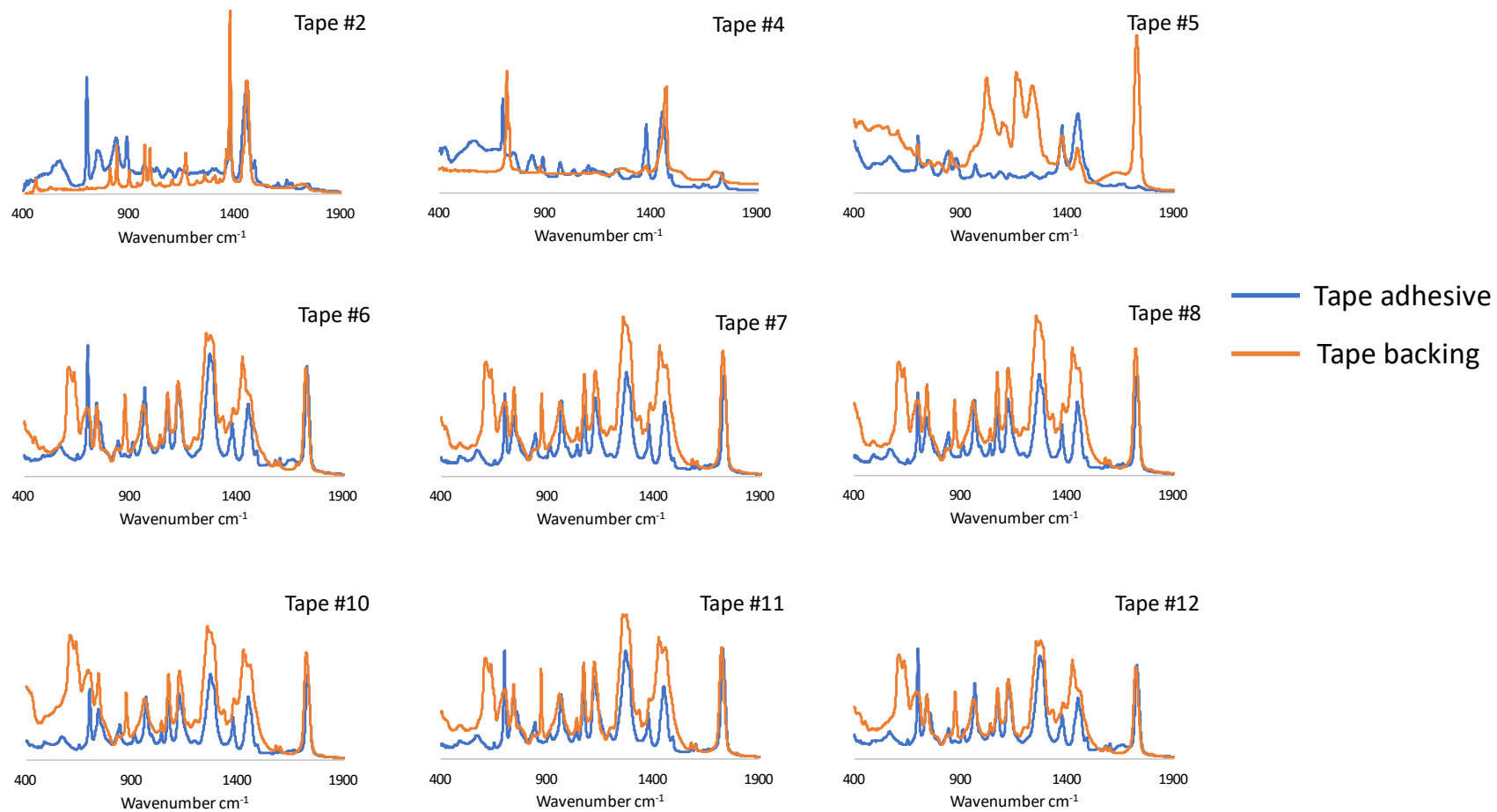
**Figure 5.10:** ATR-FTIR spectra of acrylic adhesive packaging tape #1 and #2.



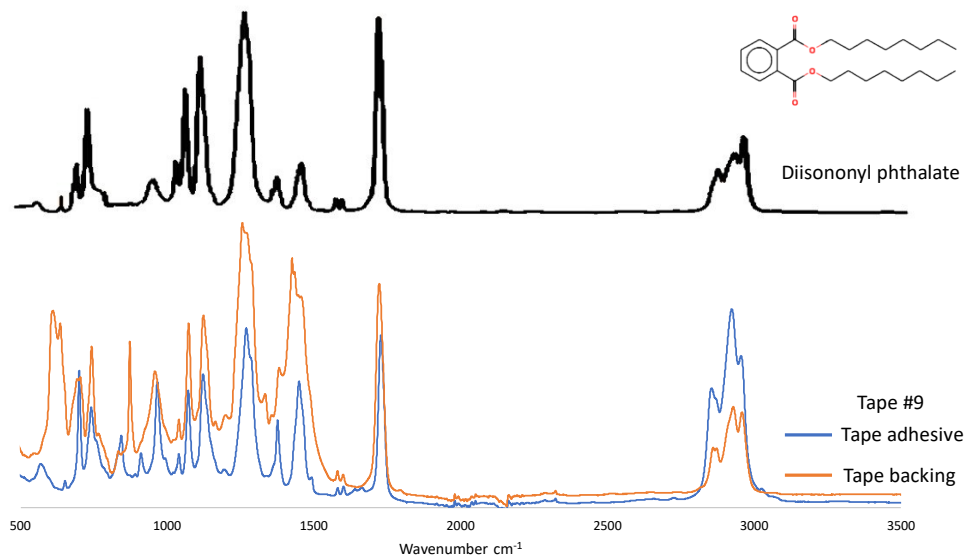


**Figure 5.11:** ATR-FTIR spectra of synthetic rubber adhesive for packaging tapes #2, duct tape #4, masking tape #5 and insulating tape #6-#12. Spectra have been offset for clarity.

Compared to other synthetic rubber adhesive tapes, insulating tapes #6-12 displayed additional peaks in their spectra, suggesting possible contributions from the tape backing in the adhesive spectra. Figure 5.12 shows the overlay of the fingerprint region of both backing and adhesive spectra of synthetic rubber adhesive tapes. Examination of the spectra of packaging tapes #2, duct tape #4, and masking tape #5 showed no significant contributions from backing in the adhesive spectra. However, with all insulating tapes, striking similarity was observed between the tape and adhesive spectra, especially the carboxylate bands. This was attributed to the leaching of the plasticizer diisononyl phthalate into the adhesive glue, illustrated in Figure 5.13. Diisononyl phthalate possesses an ortho-substituted aromatic ring, which results in a strong absorbance at  $745\text{ cm}^{-1}$  associated with the ortho aromatic group. Additionally, the characteristic weak doublet peaks at  $1600\text{ cm}^{-1}$  and  $1585\text{ cm}^{-1}$  are present due to the aromatic ring quadrant stretching vibration.<sup>311</sup>

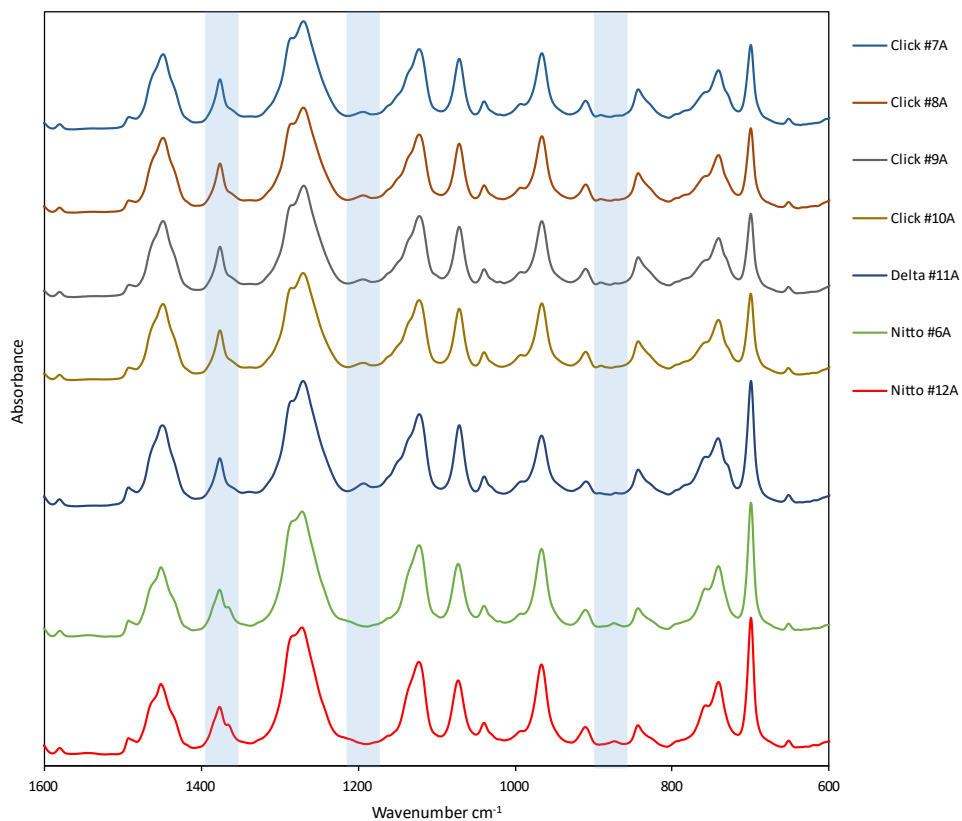


**Figure 5.12:** Overlay of the fingerprint region of both backing and adhesive IR spectrum of rubber adhesive tapes showing significant contribution of the backing in the adhesive spectrum of tape#6 to #12.



**Figure 5.13:** *Diisononyl phthalate IR spectrum<sup>312</sup> offset over rubber adhesive tape's backing and adhesive spectrum of tape #9. Showing the contribution of diisononyl phthalate in the backing and adhesive tape spectra.*

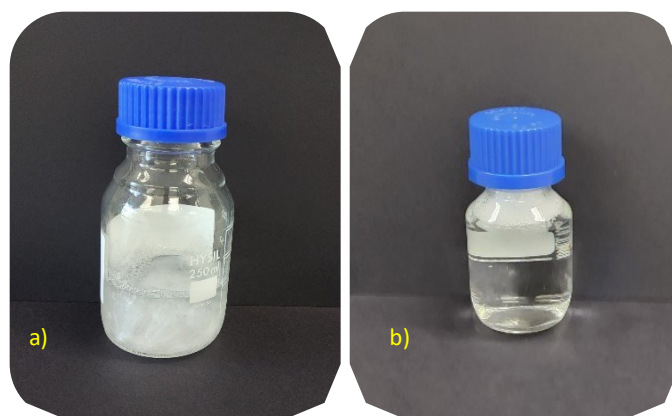
A visual examination of the spectra showed that despite originating from three distinct manufacturers, the insulating tapes displayed high similarities with only slight variations in the fingerprint region as shown in Figure 5.14. Notably, the Nitto tapes #6 and #12 were distinguishable from the other brands based on these differences in chemical composition, which are attributed to titanium dioxide and carbonates pigments.<sup>304, 309, 313</sup> As discussed earlier these differences may cause variation in the effectiveness of powder suspension between the different tapes.



**Figure 5.14:** Comparison of infrared spectra of different insulating tape synthetic rubber adhesive. The main differences are highlighted (blue shaded regions).

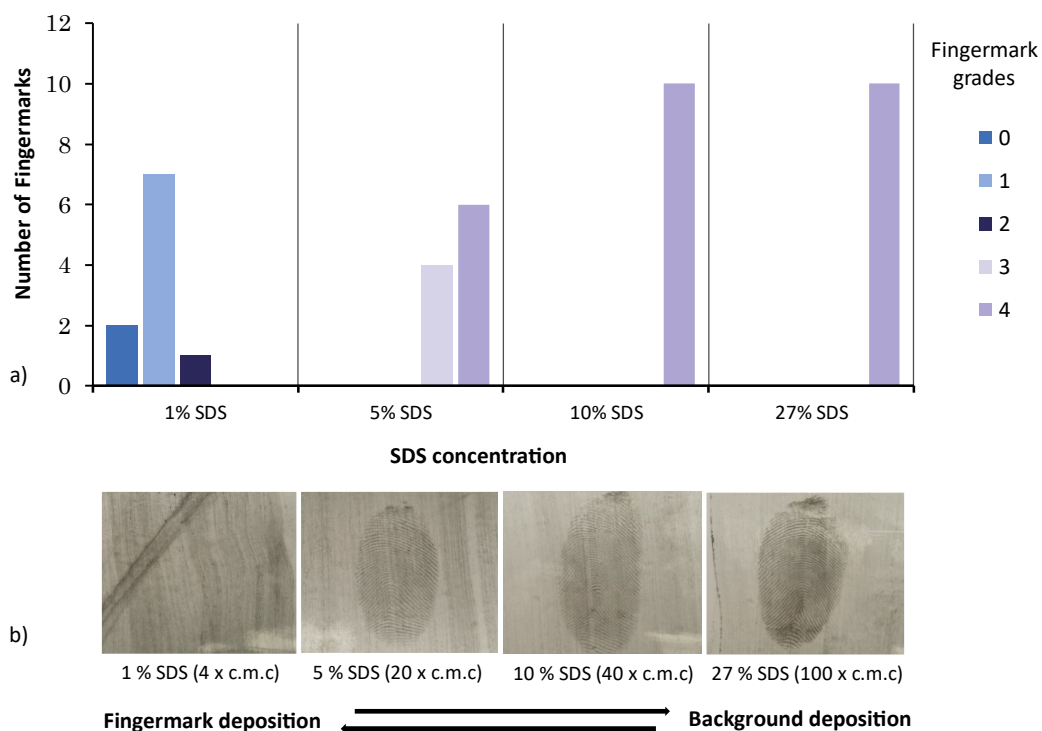
### 5.3.2 Formulation modification

The water solubility of SDS is generally reported in the range of 130 to 150 g/L (13 % to 15 % SDS w/v) at 20 °C.<sup>293 294</sup> Preparation of in-house 27 % w/v SDS surfactant water mixture (equivalent concentration to the Gran Velada solution) formed crystalline structures a day after preparation as shown in Figure 5.15. This indicates that the Gran Velada surfactant solution likely contains an additive to bring the high concentration of SDS into solution. However, this was not stated on material and safety data sheet.<sup>314</sup>



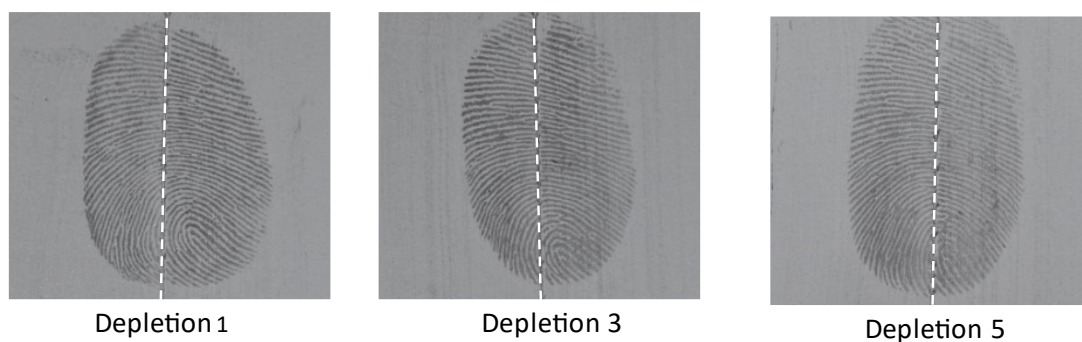
**Figure 5.15:** Sodium dodecyl sulfate solutions (a) insoluble solution of 27 % w/v in deionised, (b) clear solution of 15 % w/v in 5 % ethanol deionised water mixture.

Powder suspensions prepared with differing concentrations of SDS were thus investigated, with the results presented in Figure 5.16. Formulations containing 5 % (~20 x c.m.c) SDS surfactant concentration and above were effective in developing fingermarks with grades 3&4 (useful fingermarks). In contrast, no useful fingermarks were developed with powder suspension prepared from 1 % SDS (~ 4 x c.m.c). The variation in fingermark quality with SDS concentration was observable when examining the degree of powder deposition, as shown in Figure 5.16b. The powder suspension with 1 % SDS exhibited low contrast due to high background staining and minimal powder deposition on the fingermarks. Increasing the SDS concentration reversed the trend, with the 27 % SDS powder suspension resulting in high deposition on the fingermarks and a relatively clean background. This pattern was observed on both acrylic and rubber-based tapes but was more noticeable on the former. The result underlines the critical role that surfactant concentration plays in powder suspension effectiveness and is consistent with a previous study by Downham *et al.*,<sup>165</sup> which showed that using Triton X-100 surfactant in an iron oxide powder suspension requires a surfactant concentration above the critical micelle concentration (2 x c.m.c) to prevent indiscriminate staining. This study indicates that a surfactant concentration above 4 x c.m.c of SDS is necessary for carbon-based powder suspension. Compared to the non-ionic Triton X-100, the ionic nature of SDS surfactant likely increases its interaction with the background surface, resulting in a greater disruption of the surfactant micelles, hence necessitating a higher concentration.



**Figure 5.16:** Comparison of (a) the number and grades of fingermarks developed (b) the degree of powder deposition on fingermarks and background after treatment with modified WET UCIO powder suspensions prepared with 1 %, 5 %, 10 % and 27 % w/v SDS surfactant solution.

Whilst 27 % SDS formulation developed highest quality fingermarks, the difference with the 10 % SDS formulation was minor. Moreover, 27 g of SDS to produce 100 mL of surfactant solution was a sizeable amount given the low density of SDS ( $1.01 \text{ g/cm}^3$ ).<sup>315</sup> Additionally, reducing the surfactant concentration would be more cost-effective and relatively less likely to precipitate over time. Therefore, a 15 % w/v surfactant solution containing 5 % v/v ethanol/deionised water mixture (Wet SPF) was investigated. Ethanol, a readily available laboratory reagent, was used as an additive to avoid precipitation of SDS. Wet SPF was found to be equally effective and sensitive in developing fingermarks as the formulation containing 27 % SDS, on both acrylic- and rubber-based PSA tapes as shown in Figure 5.17.

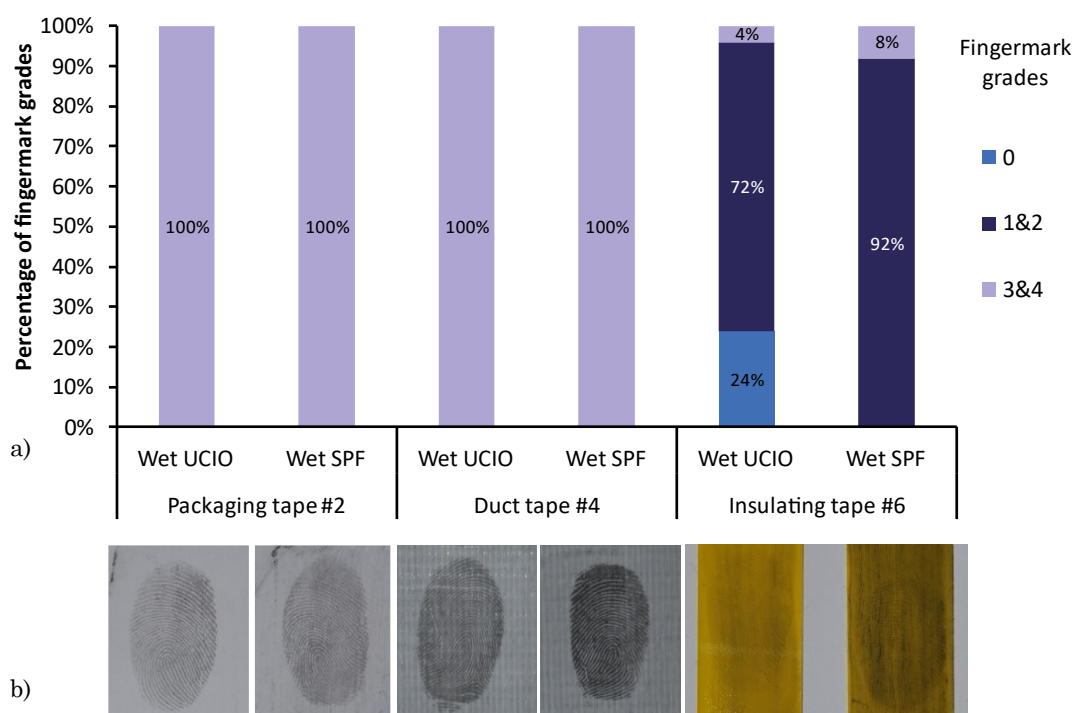


**Figure 5.17:** Comparable performance and sensitivity of Wet SPF formulation (left) and 27 % SDS-modified WET UCIO formulation (right) in developing fingermarks on rubber-based adhesive tape.

The Wet SPF was then compared to Wet UCIO and both formulations were found to be highly effective on packaging tape #2 and duct tape #4, with all fingermarks graded as useful (Figure 5.18a). Overall, the percentage of useful fingermarks developed was 68 % for Wet SPF and 60 % for WET UCIO, slightly lower than the 86.5 % effectiveness reported by Claveria *et al.*<sup>175</sup> using WET UCIO on five rubber-based adhesives. The reduced effectiveness was attributed to the indiscriminate background staining observed on Nitto insulating tape #6 (Figure 5.18b). This is likely due to the variations in substrate chemical composition, as a result of titanium dioxide in the adhesive tape, as indicated by the visual examination of the IR spectra. Pigments such as titanium dioxide have been reported to cause widespread background staining with carbon-based powder suspension.<sup>287</sup>

Another observation was that the powder deposition was more intense with Wet SPF than with WET UCIO formulation (Figure 5.18b). This may explain why 24 % of the fingermarks developed with WET UCIO on insulating tape #6 were undetected (grade 0). In contrast, all fingermarks were detected with Wet SPF formulation, but a high percentage were graded as not useful, due to low contrast caused by background staining. Although subtle, the differential powder deposition on fingermarks was also observed on packaging tape #2 and duct tape #4 with Wet SPF formulation exhibiting darker fingermarks compared to WET UCIO. One possible cause is the difference in SDS concentration. Wet SPF has a concentration of 15 % w/v while WET UCIO has 27 % w/v. During these experiments, no significant differences were observed between

the two SDS concentrations, indicating that other factors, such as the presence of additives, may be influencing the results.

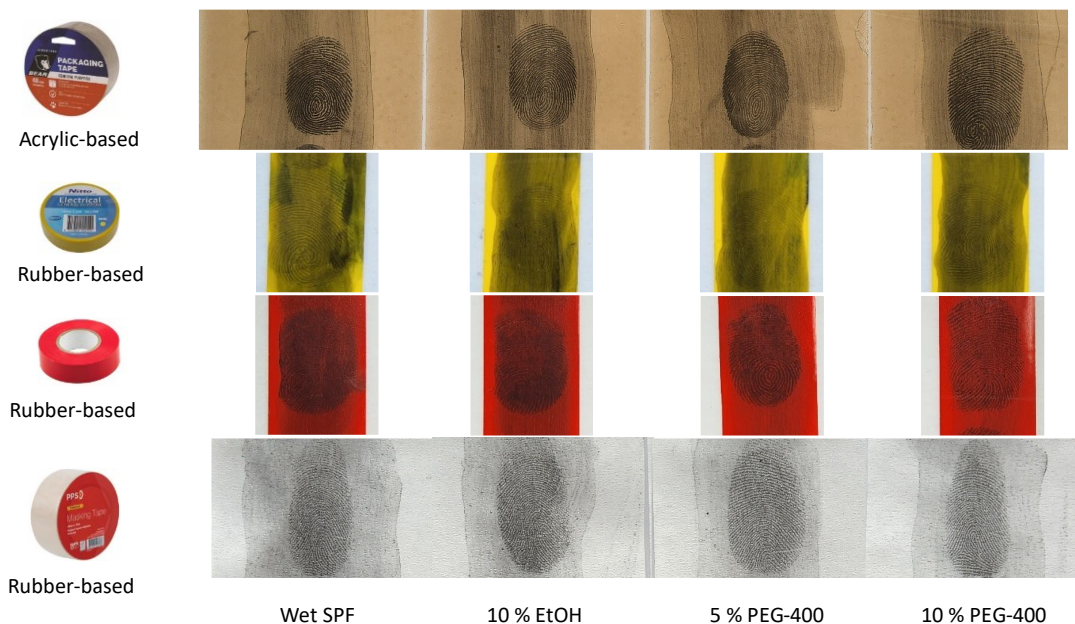


**Figure 5.18:** Comparison of (a) percentage of fingerprints graded (3&4), (1&2), and (0) out of 150 fingerprints (b) the degree of powder deposition on fingerprints and background after treatment with WET UCIO and Wet SPF powder suspensions on packaging tape, duct tape and insulating tape.

### 5.3.3 Supplementary studies

To investigate the role of additives, a set of modified Wet SPF formulations was evaluated using ethanol and PEG-400 as additives. Both additives gave similar results in terms of the number and quality of developed fingerprints across four tapes (Figure 5.19). This indicates that the two additives appeared to have minimal impact on the effectiveness of Wet SPF formulation, although this does not refute the possibility that the additive in Wet UCIO may cause a minor reduction in powder deposition. In line with frugal forensics, ethanol is a readily available and economical laboratory reagent, almost half the price of PEG-400. Nonetheless, the results suggest that PEG-400 is equally effective as an alternative additive.



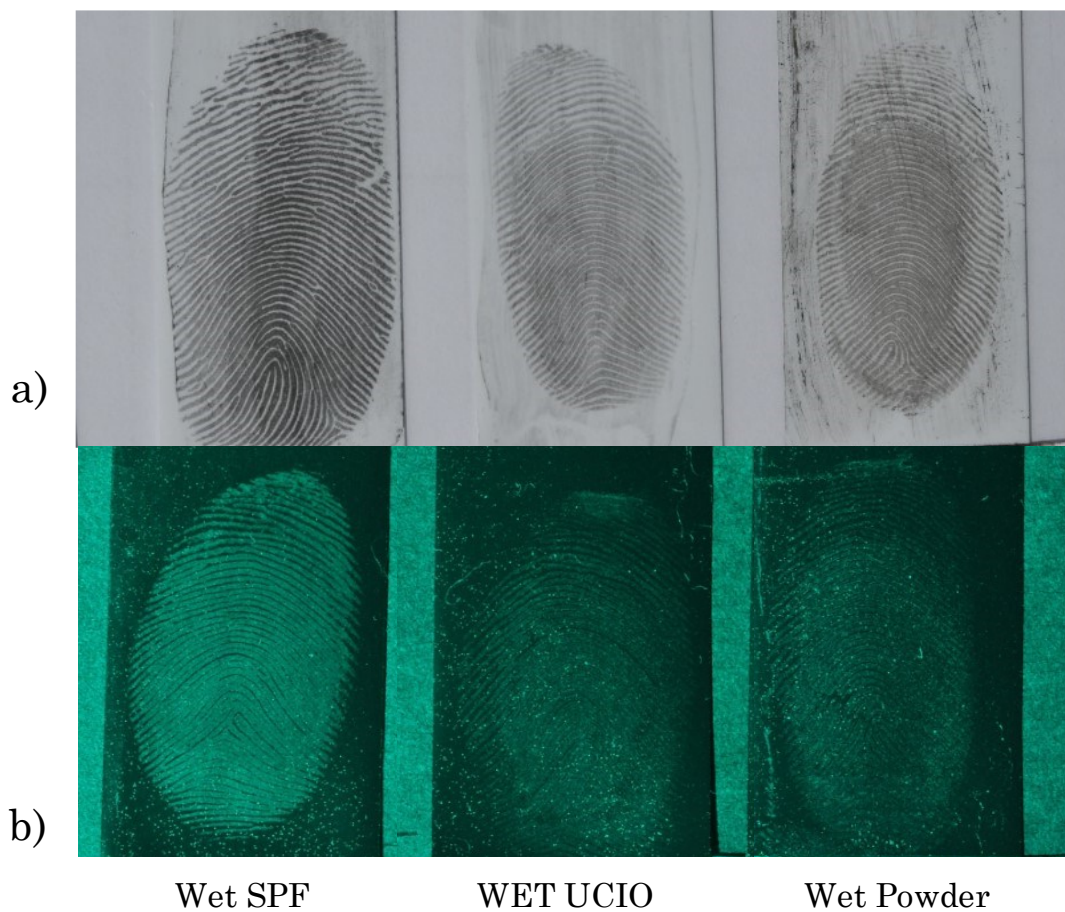


**Figure 5.19:** Comparison of fingerprint ridge details and contrast developed on four different tapes after treatment with Wet SPF powder suspensions prepared with different additives; 5 % ethanol, 5 % PEG-400, and 10 % PEG-400 v/v.

Investigation into the five insulating tapes revealed that the background staining was specific to Nitto tape #6, with all formulations exhibiting similar results. The IR analysis showed that all insulating tapes were of PVC backing and rubber-based adhesive, but Nitto tapes showed slight difference in chemical composition compared to other brands. This strongly suggests that background staining is substrate related and more likely due to additives in the adhesive as discussed above. Additionally, the earlier observation that powder deposition on fingerprints was higher with Wet SPF formulation, was also seen when compared to the commercial Wet Powder formulation. The findings suggest that Wet SPF may be more effective than Wet Powder in developing weaker fingerprints on rubber-based insulating tapes.

Interestingly, the insulating tape #9 used in this study was black-coloured, which is generally processed with white powder suspension for better contrast. However, in this study, the carbon-black-based powder was used, and visualisation was achieved by exploiting the difference in absorbance between the treated fingerprint and tape surfaces.<sup>17</sup> The treated black-coloured tape was examined using a Polilight under

different excitation wavelengths and filters to obtain the best viewing conditions. The combination of 505 nm excitation and 550 nm filter was found to be optimum, as the visualised marks closely resemble black and white fingermarks, which are generally preferred by practitioners<sup>17</sup> (Figure 5.20). The result indicates that carbon-based powder suspension may be used for light and dark tapes, offering a cheaper alternative to commercial white powder suspensions. However, further investigation is required on a larger subset of samples of various types of dark-coloured tapes.

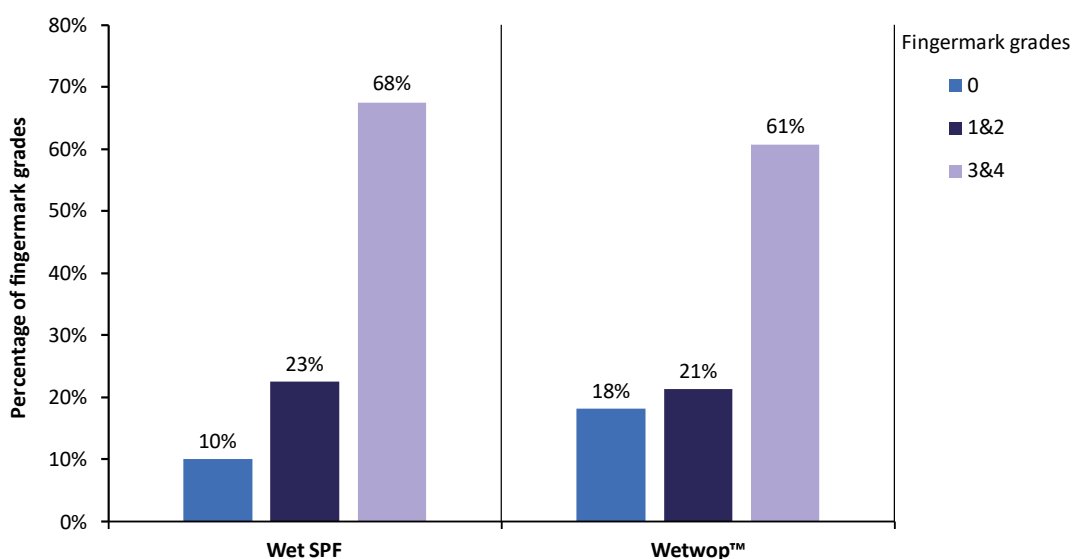


**Figure 5.20:** Fingermark ridge details and contrast after treatment with Wet SPF, WET UCIO and Wet Powder on (a) Click white insulating tape #10 under white light, (b) Click black insulating tape #9 under 505 nm excitation and 550 nm filter.

### 5.3.4 Comparative assessment to Wetwop™

#### 5.3.4.1 Overall performance

The comparative assessment of Wet SPF and Wetwop™ powder suspension in developing 1 week and 4 weeks old fingermarks deposited on eight adhesive tapes by a range of 10 donors are summarised in Figure 5.21. In total, 320 split fingermarks were graded to provide insight into overall performance with Wet SPF formulation developing 68 % of useful fingermarks and Wetwop™ 61 %. The result clearly shows the comparative effectiveness of Wet SPF to the commercial Wetwop™ powder suspension on both acrylic and rubber-based PSA tapes.



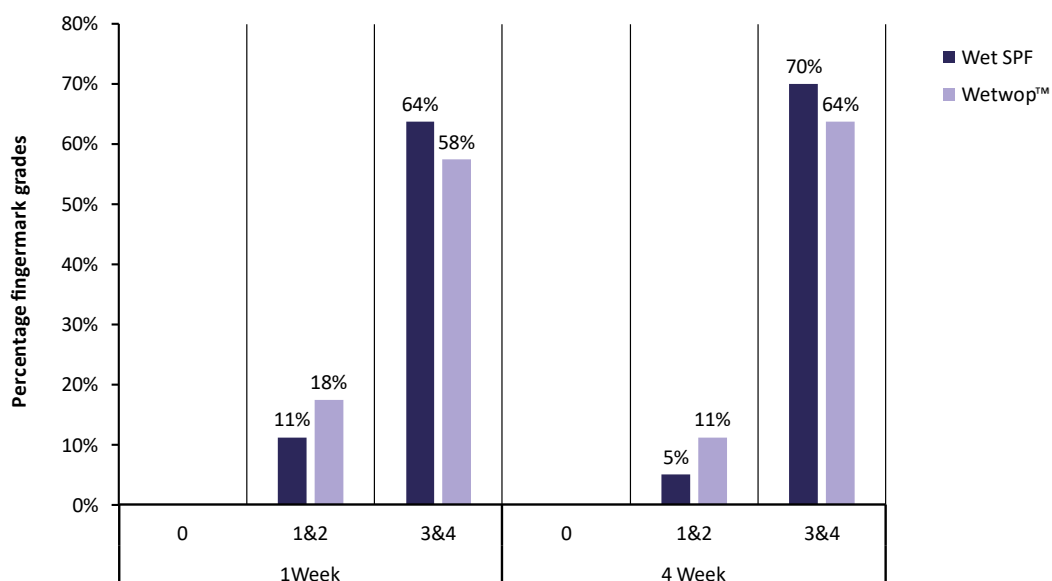
**Figure 5.21:** Percentage of fingermarks graded (3&4), (1&2), and (0) out of 320 fingermarks developed with Wet UCIO and Wet SPF on eight different types of tape.

The results for the two aged samples of 1 week and 4 weeks are presented in Figure 5.22. Both formulations appeared to be more effective on older fingermarks, with Wet SPF increasing from 65 % to 70 % and Wetwop™ from 58 % to 64 %. The effectiveness of powder suspension on older fingermarks has been suggested to be due to the loss of the water-soluble eccrine component over time, exposing and reducing the interaction distance between the eccrine components encapsulated within the non-water soluble constituents and particles of powder suspension.<sup>16</sup> The

result demonstrates the applicability of Wet SPF on aged fingermarks that are likely to be encountered in casework.

In terms of substrates, both formulations exhibited high effectiveness in developing fingermarks on acrylic and rubber-based PSA tapes (Table 5.6), with exception of Nitto rubber-based insulating tapes #6 and #12, and rubber-based masking tape #5. As expected, due to differences in chemical composition, Nitto insulating tapes #6 and #12 exhibited background staining, resulting in poor effectiveness. In contrast, both formulations were highly effective in developing useful fingermarks on similarly coloured insulating tapes #10 and #11 but from a different manufacturer. The background staining on Nitto insulating tapes has been discussed above and these results reinforced the suggestion that this is due to the chemical composition of the Nitto adhesive PSA tapes. Excluding the Nitto insulating tapes, Wet SPF and Wetwop™ percentage effectiveness increases to 89 % and 81 % respectively, close to the reported 92 % effectiveness of Wet UCIO method.<sup>175</sup>

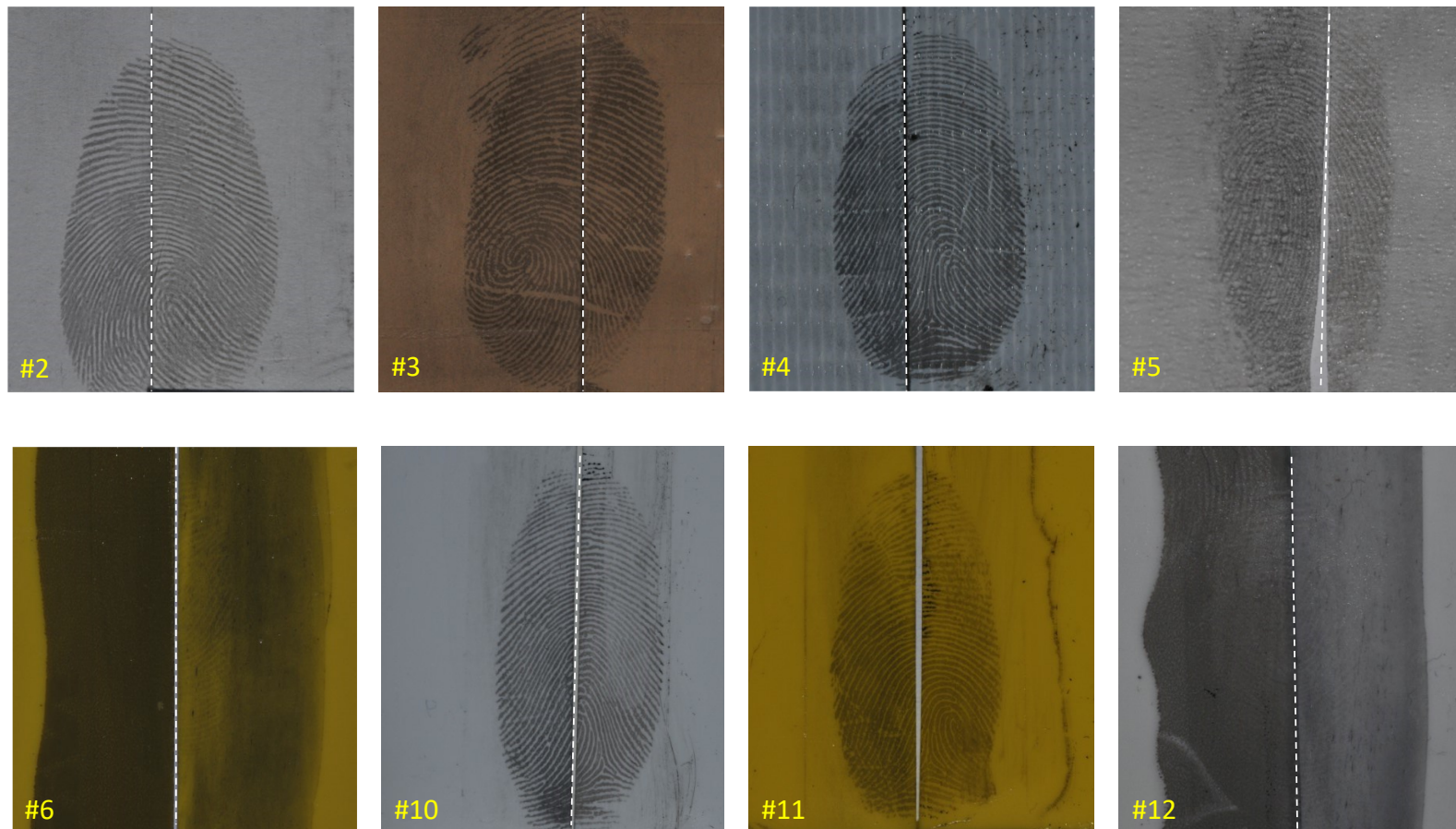
On the rubber-based masking tape #5, Wet SPF and Wetwop™ were found to be 40 % and 25 % effective, respectively. A similar effectiveness of 37.5 % was reported for rubber-based masking tape with WET UCIO powder suspension.<sup>175</sup> Examination of the developed fingermarks showed that despite a relatively clean background, poor contrast was observed since the space between ridges was partly stained (Figure 5.23). This is likely because during the drying process, the solution partitioned into the adhesive layer and diffuses in the cellulose matrix to stain the backing, reducing the overall quality of the developed marks.



**Figure 5.22:** Percentage of fingermarks graded (3 & 4), (1 & 2), and (0) out of 320 fingermarks developed with Wet UCIO and Wet SPF on eight different types of tape aged for 1 week and 4 weeks.

**Table 5.6:** Percentage of fingermarks graded 3&4 on each of the eight types of adhesive tape.

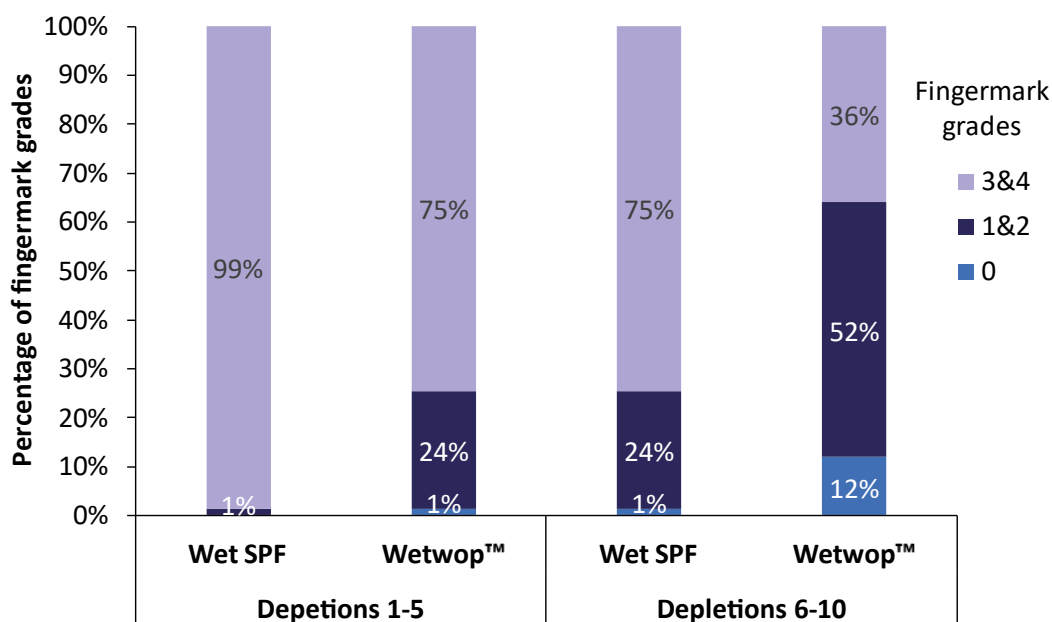
Sample Code	Tape Type and colour	Brand	Adhesive Composition	Wet SPF	Wetwop
Tape #2	Packaging (Transparent)	Scotch 3M	Rubber	100%	85%
Tape #3	Packaging (Transparent)	Bear	Acrylic	100%	100%
Tape #4	Duct Tape (Silver)	Paint Partner	Rubber	100%	90%
Tape #5	Masking (White)	PPS	Rubber	40%	25%
Tape #6	Insulating (Yellow)	Nitto	Rubber	5%	0%
Tape #10	Insulating (White)	Click	Rubber	100%	95%
Tape #11	Insulating (Yellow)	Deta	Rubber	95%	90%
Tape #12	Insulating (White)	Nitto	Rubber	0%	0%



**Figure 5.23:** Examples of fingerprint developed with Wet SPF (left) and Wetwop™ (right) on eight pressure sensitive tapes #2 Scotch 3M clear packaging tape, #3 Scotch 3M brown packaging tape, #4 Paint Partner silver duct tape, #5 PPS white masking tape, #6 Nitto yellow insulating tape, #10 Click white insulating tape, #11 Delta yellow insulating tape, #12 Nitto white insulating tape.

### 5.3.4.2 Sensitivity

A total of 300 split fingermarks comprising up to 10<sup>th</sup> depletion were processed to assess the sensitivity of Wet SPF to Wetwop™. Figure 5.24 shows the sensitivity results with grouping of 1st to 5th depletions and 6th to 10th depletions. The result indicates Wet SPF to show superior sensitivity particularly for the 6<sup>th</sup> to 10<sup>th</sup> depletion series. As discussed earlier, this is due to higher powder deposition on fingermarks exhibited by Wet SPF resulting in better effectiveness in developing weaker fingermarks.



**Figure 5.24:** Percentage of fingermarks grades out of 300 fingermark halves developed with Wet UCIO and Wet SPF on three different types of tape grouped into depletion series 1-5 and 6-10.

Regarding reagent stability, Wet SPF that has been prepared with SDS solution over 6 months old was found to be as effective as suspension powder prepared with fresh SDS solution. It has also been reported that aqueous SDS solution can remain stable for up to three years.<sup>316</sup> Although further studies are required to test the shelf life of the Wet SPF suspension powder mixtures, preparing the formulation fresh and when needed is recommended to avoid wastage, given its ease of preparation.

### 5.3.5 Statistical analysis

A Mann-Whitney U, chi-squared and difference in proportion tests performed on the overall performance data, showed no significant difference in the fingermark grades ( $p = 0.051$ ), its distribution of frequencies ( $p = 0.057$ ), or proportion of grades 3s and 4s ( $p = 0.2$ ) between Wet SPF and Wetwop™. For the sensitivity data, all three tests revealed significant differences ( $p = 0.2 \times 10^9$ ,  $0.7 \times 10^8$ ,  $0.2 \times 10^8$  respectively). The statistical results reinforce the observation that Wet SPF's performance is comparable and shows better sensitivity to commercial Wetwop™ black powder suspension. The R code and results of all statistical analysis are provided in Appendix A-4.

## 5.4 Conclusion

This chapter demonstrates how the frugal forensics concept can be applied to a recently published method through the adaptation of a carbon-based powder suspension for latent fingermark detection on non-porous PSA tapes. Successful modification of the recently published WET UCIO powder formulation by replacing the inaccessible Gran Velada surfactant solution with an in-house sodium dodecyl sulfate surfactant solution has been shown to be effective in the recovery of latent fingermarks on PSA adhesive tapes.

The Wet SPF powder suspension containing 15 % SDS in a 5 % ethanol/water mixture has been shown to be as effective as the commercial Wetwop™ powder suspension on eight different tapes using ten donors. Other benefits include an inexpensive formulation and readily available surfactant, accessible from multiple suppliers. In respect of remote-location jurisdictions, the use of salts compared to commercial solution surfactants allows for easy transportation, storage and better control of quality and shelf-life.

The use of Wet SPF combined with forensic light sources appeared effective in visualising fingermarks on challenging surfaces of dark-coloured adhesive tapes. Further studies on the comparative assessment with commercial white powder



suspension are suggested, as this may allow the use of a single formulation suited to both light- and dark-coloured substrates.

Based on this successful demonstration in line with IFRG phase 2/3 studies, the Wet SPF method is recommended for validation test as an operational technique that can be used in Seychelles for developing latent fingerprints on adhesive tapes. The formulation is as follows:

1.5 grams Sirchie silk black fingerprint powder

10 mL of 15 % sodium dodecyl sulfate solution

15 % sodium dodecyl sulfate solution is as follows –

15 grams of sodium dodecyl sulfate

100 mL of 5 % ethanol/deionised water mixture.

## **Chapter 6. Investigation into acid Nile blue as an emerging lipid-sensitive and post-cyanoacrylate detection method**

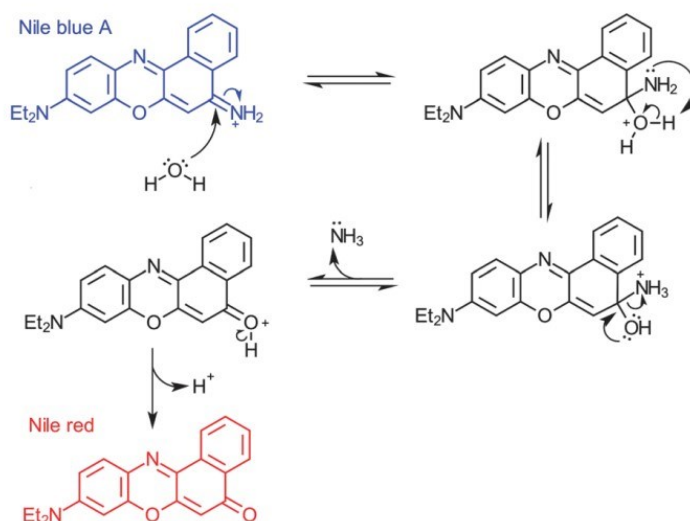
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## 6.1 Introduction

Chapters 3 and 4 investigated a well-established amino acid sensitive reagent for porous surfaces, and Chapter 5 a recently published method on non-porous surfaces. This chapter explores the benzophenoxazine dye Nile blue, as an emerging lipid-sensitive latent fingerprint detection technique within the construct of a frugal forensics approach.

Nile blue, as a lipid-sensitive reagent has several benefits in a frugal forensics context. It is a non-toxic water-based solution that is simple to use, low-cost (ca. \$3.20 USD per gram)<sup>153</sup> and an easily accessible forensic method. This makes it a promising option for jurisdictions with limited resources.

As discussed in Chapter 1, Nile blue is a cationic dye used for staining neutral fats, phospholipids, and fatty acids in histology.<sup>149</sup> Its behaviour in water is influenced by its acid form, which can form hydrogen aggregates that do not fluoresce.<sup>142, 317</sup> However, a small amount of Nile blue undergoes spontaneous hydrolysis to Nile red<sup>142, 143</sup> as shown in Figure 6.1. Nile red is also used in histology for staining triacylglycerols, phospholipids, esteryl esters, and lipoproteins. It is a neutral molecule that is highly fluorescent, particularly in non-polar solvents.<sup>142</sup>



**Figure 6.1:** Reaction pathway proposed for the spontaneous hydrolysis of Nile Blue to Nile red in aqueous solution.<sup>146</sup>

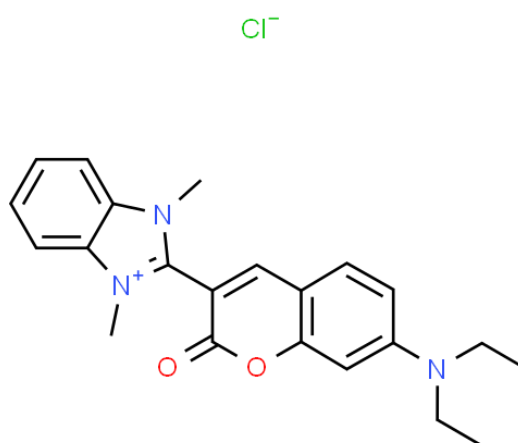
The presence of the two stains creates a dual-purpose fingerprint detection reagent. Nile red dissolves preferentially in neutral lipids,<sup>148</sup> producing photoluminescence when excited at 505 nm and viewed through a 550 nm long pass barrier filter.<sup>146, 318</sup> Nile blue interacts with the acidic components, resulting in visible dark blue development and suppression of background fluorescence.<sup>146</sup> The properties of Nile red and Nile blue depend on the solvent polarity and dielectric constant. In non-polar media, they fluoresce brightly in the visible to near-infrared range, whilst in polar solvents, their fluorescence redshifts with lower intensity due to hydrogen bonding.<sup>142, 319</sup>

Recent studies by Frick *et al.*<sup>146</sup> demonstrated aqueous Nile blue's effectiveness in developing lipid-rich fingerprints on a range of wet porous and non-porous surfaces. Further studies by Crocker *et al.*<sup>154</sup> have shown that modifying the aqueous Nile blue with sulfuric acid increases fingerprint fluorescence intensity due to increased hydrolysis of Nile blue into Nile red. However, comparative assessment of acid Nile with an operational lipid-sensitive technique, such as Oil red O (ORO), is yet to be carried out. This can establish the feasibility of using Nile blue as an operational lipid-sensitive technique for porous surfaces.

Another potential application of acid Nile blue is as a post-cyanoacrylate stain. In an initial investigation conducted by Chester *et al.*,<sup>151</sup> the use of Nile blue as a stain after cyanoacrylate treatment was found to have limited effectiveness. However, this was likely due to the short processing time of only 20 seconds. The underlying mechanism of post-treatment stain was not discussed, nor does it commonly appear in the literature. Other preliminary studies<sup>187, 188</sup> compared Nile blue with other luminescent dyes including rhodamine 6G (R6G). However, this did not include Basic Yellow 40 (BY40), which is the current operational method used by the Seychelles Police for enhancement of cyanoacrylate fumed fingerprints.

Basic yellow 40 (7-(diethylamino)-3-(1,3-dimethylbenzimidazol-3-ium-2-yl)chromen-2-one;chloride) is a common basic fluorescent dye introduced as an alternative to R6G due to suspected carcinogenic risk of R6G.<sup>68</sup> BY40 generally refers to several dyes, of which the recommended dye for fingerprint development is the chloride

base (Figure 6.2). Recently there has been concern over the industry's lack of clarity around individual powders' specifications, due to reports of ethanolic-BY40 solution becoming cloudy.<sup>168</sup> These were based on a sulfate salt of BY40 dyes (CAS No: 35869-60-4) that does not dissolve completely. Moreover, the mechanism of post-treatment stains is poorly understood as it is unclear whether the stain partitions into the fingerprint residue or only stains the polycyanoacrylate structure. One explanation is that the fibrous network of polycyanoacrylate works like a sieve, trapping the dye molecules. Another possibility is that the cyanide (CN<sup>-</sup>) anions of polycyanoacrylate may weakly interact with the cations of basic dyes through van der Waals forces.<sup>68</sup> A fundamental understanding of the staining process could direct the use of new dyes and the treatment sequencing process.



**Figure 6.2:** *The molecular structure of Basic Yellow 40 (CAS Number 29556-33-0) recommended dye for fingerprint enhancement.*

This study has two main objectives, firstly to conduct a comparative assessment of acid Nile blue to ORO as a lipid-sensitive reagent on wet porous substrates. Secondly, to assess the potential of acid Nile blue as a post-cyanoacrylate fingerprint stain and explore the underlying staining mechanism using confocal Raman microscopy. This will provide information on the frugal forensics use of Nile blue dye as a fingerprint enhancement technique.

## 6.2 Experimental

### 6.2.1 Experimental methods

#### 6.2.1.1 Reagent preparation and sample development

Acid Nile blue was prepared, and samples were developed as per Crocker *et al.*'s<sup>154</sup> method outlined in section 2.5.5. ORO was prepared and samples were developed as per the modified formulations in Frick *et al.*,<sup>137</sup> outlined in section 2.5.3.

Cyanoacrylate fumed prints were prepared as outlined in section 2.5.4. The samples were allowed to cure for 24 hours before post-cyanoacrylate staining with acid Nile blue or BY40 following the procedure in sections 2.5.5 and 2.5.6 respectively.

#### 6.2.1.2 Substrates

For the comparative assessment to ORO study, four types of substrates commonly encountered by the Seychelles Police were used and are summarised in Table 6.1.

For the post-cyanoacrylate stain study, three types of substrates (glass, metal, and plastic) that are generally processed with cyanoacrylate were used and summarised in Table 6.2.

**Table 6.1:** Detail of substrates used in the comparative assessment to ORO.

Substrate	Description	Colour	Weight (gsm)/Thickness
1	Winc copy paper A4	White	80
2	OfficeMax copy paper A4, 50% recycled	White	80
3	PPS Brown kraft paper	Brown	50
4	Newsprint (The West Australian)	Grey	

**Table 6.2:** Details of substrates used in the post-cyanoacrylate study.

Substrate	Type	Description	Source
1	Glass	Borosilicate microscope slides 1 mm	VWR International, Leuven
2	Metal	Aluminium sheet 0.5 mm	Bunnings
3	Plastic	Polypropylene plastic lid	Red Dot

### **6.2.1.3 Fingerprint collection**

Single split fingerprints were collected from a range of six donors (three females and three males, aged 22-60) following procedure outlined in section 2.3.2. Samples were stored in cupboard in office space under ambient average temperature of 21 °C and RH of 56 %.

### **6.2.1.4 Sample visualisation and evaluation**

Visual examination and photography are outlined in section 2.6.1 and grading of fingerprints is described in section 2.7.

## **6.2.2 As a lipid-sensitive reagent on porous substrates**

### **6.2.2.1 Preliminary Study**

Crocker *et al.*<sup>154</sup> reported issues with Nile blue not dissolving completely, during the development of acid Nile blue. The excess Nile blue reacted with sulfuric acid, leading to yellow staining that could impact fluorescence intensity. However, this issue appears less problematic with 0.3 M sulfuric acid. To investigate the issue of yellow staining, 50 mL of acid Nile blue solution was filtered through filter paper to remove any sediments and then compared with unfiltered acid Nile blue solution.

Additionally, the quartered fingerprint methodology was used to determine the optimal processing time for acid Nile blue treatment. Sample treatment was performed as described by Crocker *et al.*<sup>154</sup> with each quarter of a fingerprint sample immersed for different time intervals of 5 mins, 10 mins, 20 mins or 30 mins.

All preliminary studies were conducted using Winc copy paper. While natural fingerprints were initially used, the experiments were repeated with charged fingerprints due to poor results. Consequently, charged fingerprints were also used for comparative assessments with ORO.

### **6.2.2.2 Comparison to Oil red O**

Acid Nile blue was compared to ORO under dry and wet conditions as well as three aging periods. Two sets of split fingermarks from each donor on recycled copy paper were collected, with one set submerged in tap water and the other stored under dry conditions in an enclosed box under office environment. Both dry and wet samples were aged for 1 day before each half of the split fingermarks was processed with either acid Nile blue or ORO. To minimise damage while handling paper substrates under wet conditions, all split fingermarks were cut into halves before storage and wet samples were allowed to dry before treatment. The experiment was repeated for aging periods of 10 days and 30 days.

For substrate investigation, donors deposited split fingermarks on four paper types listed in Table 6.1. The fingerprint samples were stored for 7 days under wet conditions following a similar procedure to the wet samples as above.

### **6.2.3 As a post-cyanoacrylate stain**

#### **6.2.3.1 Comparison to Basic Yellow 40**

To compare acid Nile blue and BY40, natural fingermarks were collected from five donors on three substrates listed in Table 6.2. To investigate donor effects, charged fingermarks were collected under the same conditions.

#### **6.2.3.2 Confocal Raman microscopy investigation into post-cyanoacrylate staining mechanism.**

As an initial study using confocal Raman microscopy, three sets of charged single split fingermarks were collected from a good donor on microscope slides. To ensure the integrity of the fingerprint ridges during development, the side-by-side microscope slides for split fingerprint collection were secured with cellulose tape at each end. After cyanoacrylate fuming and curing time, one set was stained with acid Nile blue, one with BY40, and one left unstained. The microscope slides were carefully separated allowing lateral and transverse examination of the stained and unstained



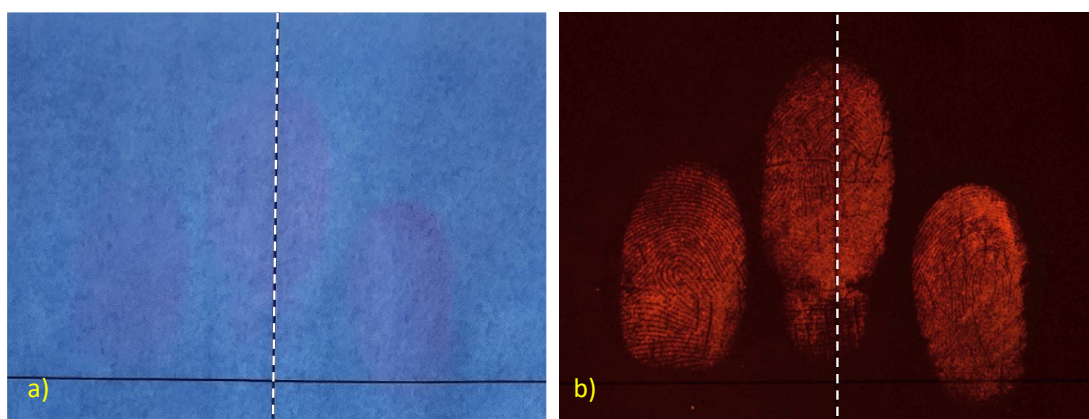
fingermarks. The samples were examined with confocal Raman microscopy as outlined in section 2.6.5.

## 6.3 Results and discussion

### 6.3.1 As a lipid-sensitive reagent on wet porous substrates

#### 6.3.1.1 Preliminary investigations

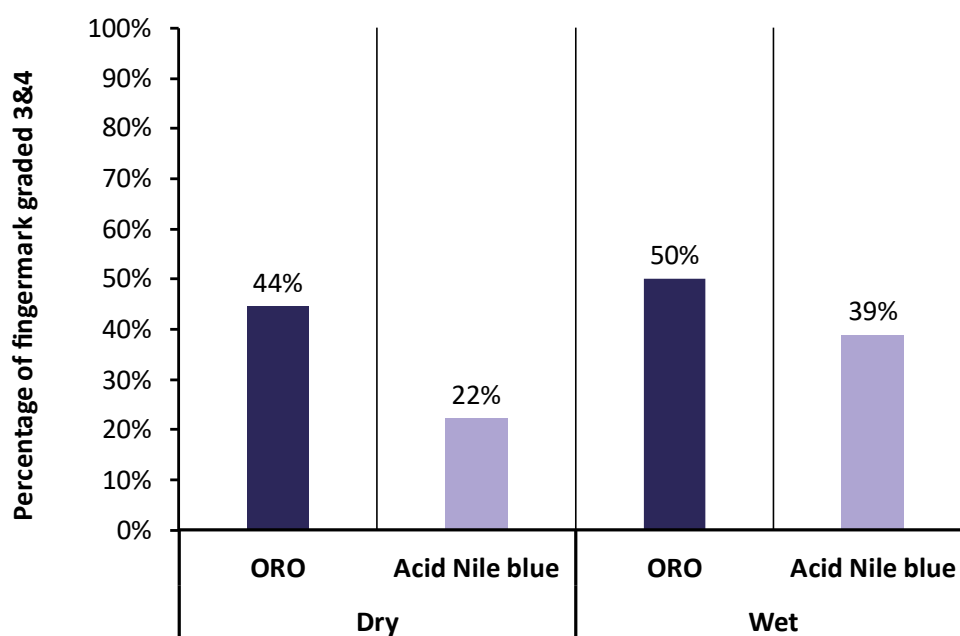
The preliminary investigation revealed that whilst Nile blue residue was observed on the filter paper there was no major difference between filtered and unfiltered solution. More importantly, no yellow staining was observed (Figure 6.3). This indicates that using 0.3 M sulphuric acid solution is sufficient to dissolve Nile blue and not cause yellow staining. For all donors, the 20 minutes immersion time developed the best quality fingermarks. For the processing time, the 5 minutes immersion developed the least number of fingermarks whilst the 20 minutes immersion time consistently developed the best quality fingermarks. This suggested that the optimal processing time for the Nile blue to partition into the fingerprint residue is 20 minutes. Therefore, the processing time was kept at 20 minutes for all other experiments.



**Figure 6.3:** Comparison of fingermarks developed by filtered acid Nile blue solution (left half) and unfiltered acid Nile blue solution (right half): (a) under white light with no observable yellow stains and (b) at 450 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.

### 6.3.1.2 Comparison to Oil red O

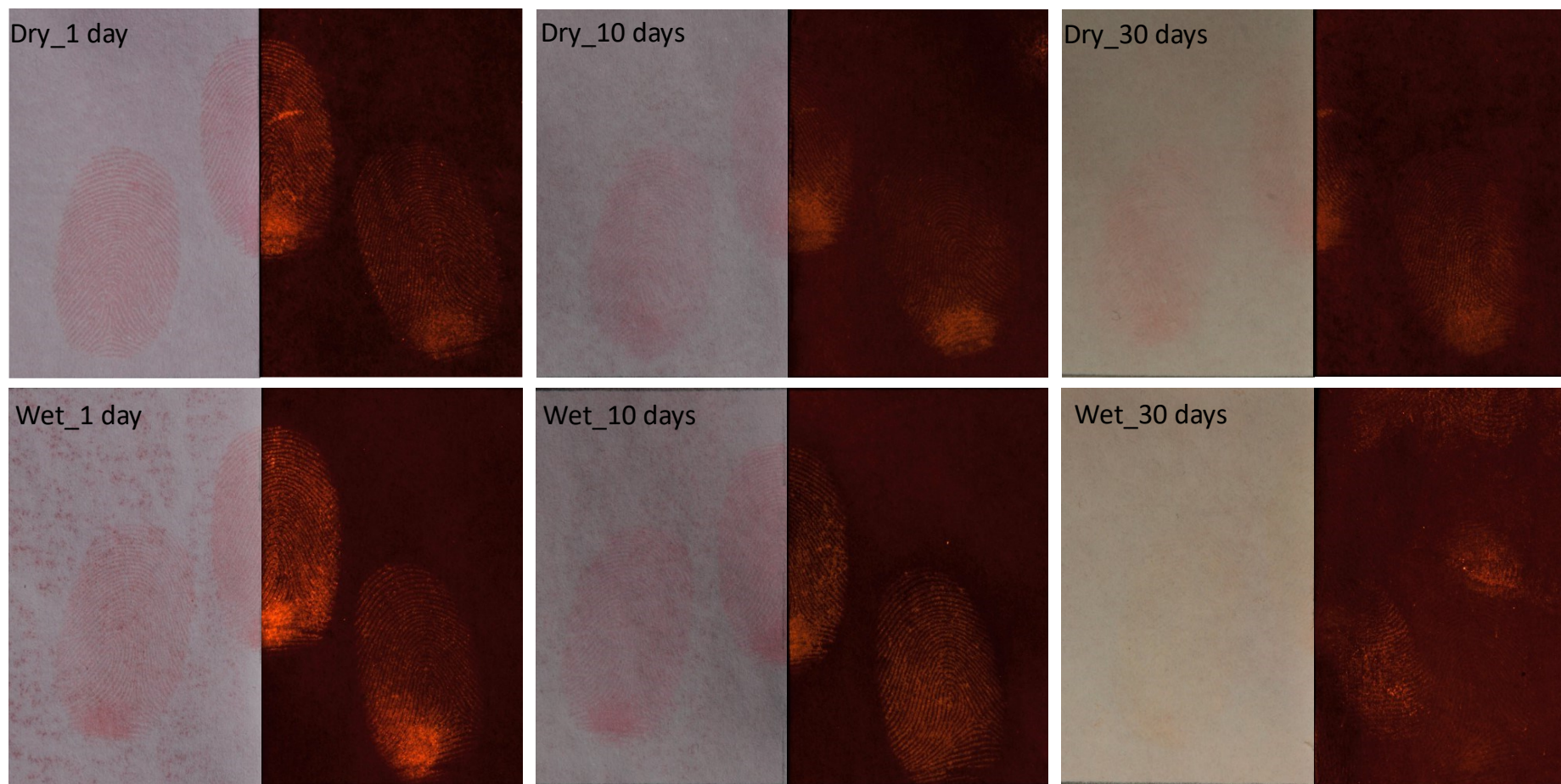
Frick *et al.*<sup>137</sup> modified Beaudoin's ORO formulation, resulting in a simple, non-toxic, low-cost lipid-sensitive fingerprint detection method that aligns with the frugal forensics concept similar to acid Nile blue. The comparative assessment of the two methods was investigated using split fingerprints and three aging periods (1, 10 and 30 days) under dry and wet conditions. The results shown in Figure 6.4 and Figure 6.5, indicate that ORO outperforms acid Nile blue in developing fingerprints subjected to wet or dry conditions.



**Figure 6.4:** Percentage of fingerprints graded 3&4 out of 72 fingerprint halves developed with ORO or acid Nile blue under wet and dry conditions.

It was observed that with increasing aging period both reagents were less effective, with no fingerprints detected after 30 days. The result is consistent with other studies that have shown ORO and Nile blue target the 'fragile fraction' of the lipid component and hence are only effective on fingerprints that are less than a few weeks old.<sup>107, 139, 154, 320</sup> As expected, the two reagents appeared to be more effective on wet fingerprints, likely due to reduced exposure to air for the lipid component

when submerged, slowing degradation of the fragile fraction.<sup>15, 35</sup> This result suggests that like ORO, acid Nile blue should be applied to fingerprints less than 4 weeks old.

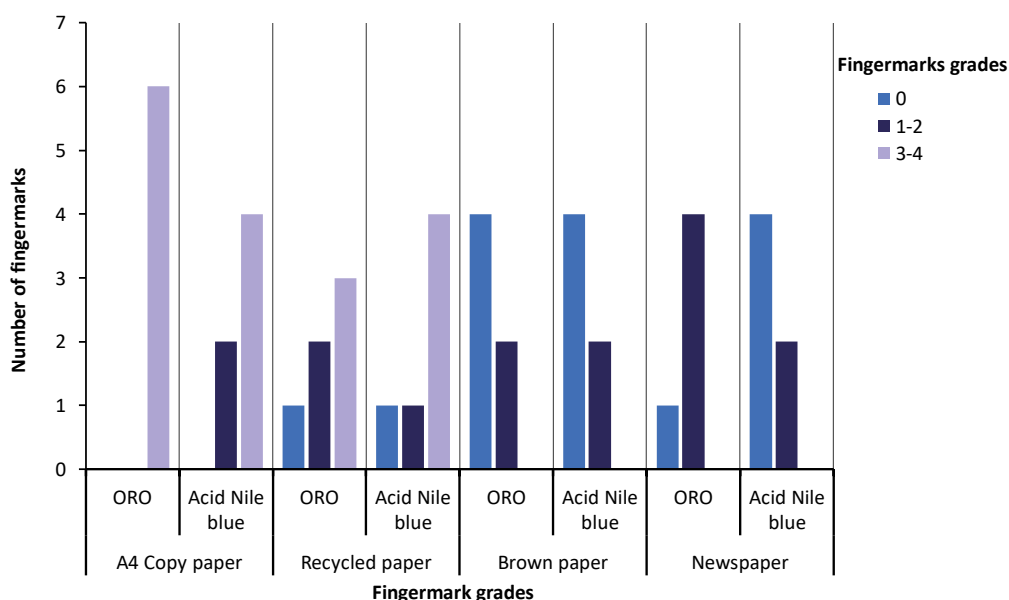


**Figure 6.5:** Comparison of fingerprints aged 1 day, 10 days and 30 days developed by ORO (left half) under white light and acid Nile blue (right half) at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.

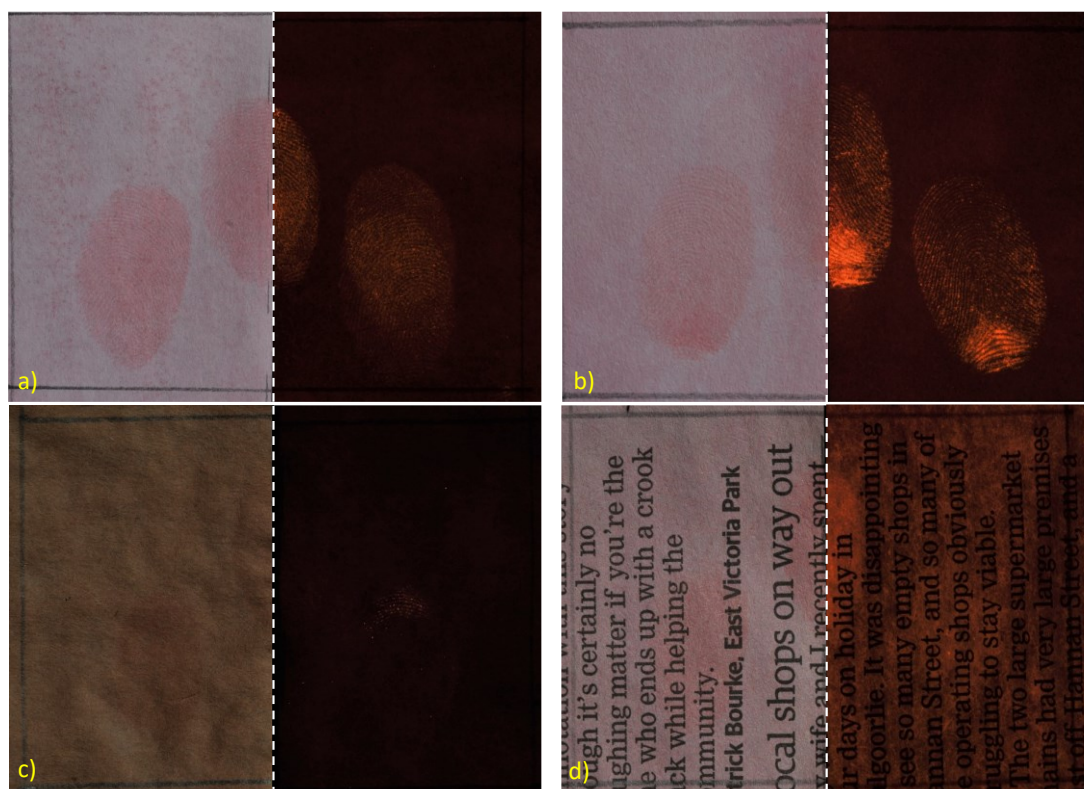
An investigation was performed on four substrates using split fingermarks submerged in tap water for 1 week to simulate casework conditions where lipid-sensitive reagents are likely to be used. The results are shown in Figure 6.6, and reinforce the slight superiority of ORO except on recycled paper, which showed a reversed trend compared to the previous result. This could originate from intra-donor variation in sample composition, potentially compounded by the limited number of donors.

Both reagents were most effective in developing fingermarks on A4 premium copy paper and recycled paper with the least effective development seen on brown paper and newspaper (Figure 6.6 and Figure 6.7). The low porosity of the brown paper and newspaper is less likely to absorb and retain the lipid fragile fraction, leaving it more susceptible to being washed off over 1 week. Previous work by Fritz *et al.*<sup>321</sup> reported a decrease in fingermark quality with increasing water submersion periods.

Although ORO seems more sensitive as a lipid reagent, the added value of the photoluminescent properties of acid Nile blue prompted further investigation as a post-cyanoacrylate stain on non-porous substrates.



**Figure 6.6:** Number and grades of fingermarks developed by ORO and acid Nile blue on A4 copy paper, recycled A4 copy paper, kraft brown paper and newspaper after 1 week of submersion in tap water.



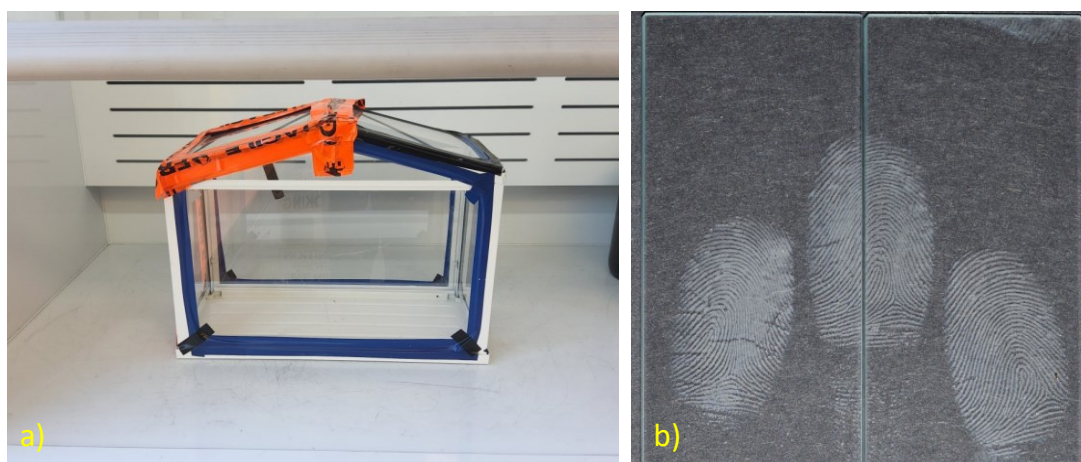
**Figure 6.7:** Comparison of fingerprints on different substrates submerged in tap water for 1 week and developed by Oil red O (left half) under white light and acid Nile blue (right half) at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter: (a) white copy paper (b) 50 % recycled copy paper (c) brown kraft paper and (d) newspaper.

### 6.3.2 Suitability as a post-cyanoacrylate stain and exploring the staining mechanism

#### 6.3.2.1 Preliminary investigation

Due to the COVID-19 pandemic, an improvised fuming chamber was used instead of the intended commercial fuming chamber at the Western Australia Police Force facility, which was inaccessible due to access restrictions. Previous work by McGann *et al.*<sup>187</sup> and Boseley *et al.*<sup>188</sup> at Curtin University, used an Ikea Sokker Greenhouse (35cm x 45 cm x 22 cm) as a sealed terrarium container (Figure 6.8a) with a cup-shaped aluminium foil to hold the superglue (~1.5 g) in the centre as a fuming chamber. The quality of cyanoacrylate marks developed after 2 hours of fuming was on par with fingerprints developed at the Western Australia Police Force facility.<sup>187</sup>

Preliminary investigation using this method was conducted and reproducible results were obtained (**Figure 6.8b**)



**Figure 6.8:** (a) *Improvised cyanoacrylate fuming chamber* (b) *quality of cyanoacrylate fingermarks developed after 2 hours of fuming on borosilicate microscope slides.*

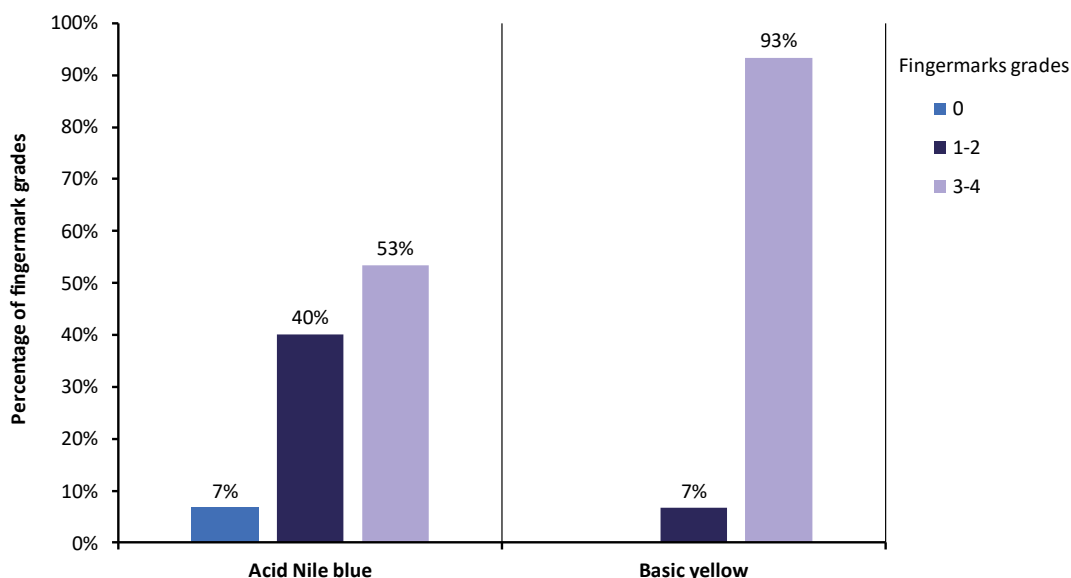
The processing condition for staining with acid Nile blue follows the procedure of simple immersion in solution as when used as a lipid-sensitive reagent on porous substrate. As part of the method development, the staining processing time was reduced to ten minutes since it gave comparable results to the suggested twenty minutes processing time in Frick *et al.*<sup>146</sup> (Figure 6.9). The reduced processing time is likely due to fingerprint deposits on non-porous surfaces being more accessible for Nile blue to partition into, compared to fingerprint residues in a cellulose matrix for porous surfaces.



**Figure 6.9:** *Comparison of cyanoacrylate fumed fingerprints stained by acid Nile blue for ten minutes (left half) and twenty minutes (right half), at 505 nm excitation wavelength of Polilight and viewed with 550 nm long-pass barrier filter.*

### 6.3.2.2 Comparison to Basic Yellow 40

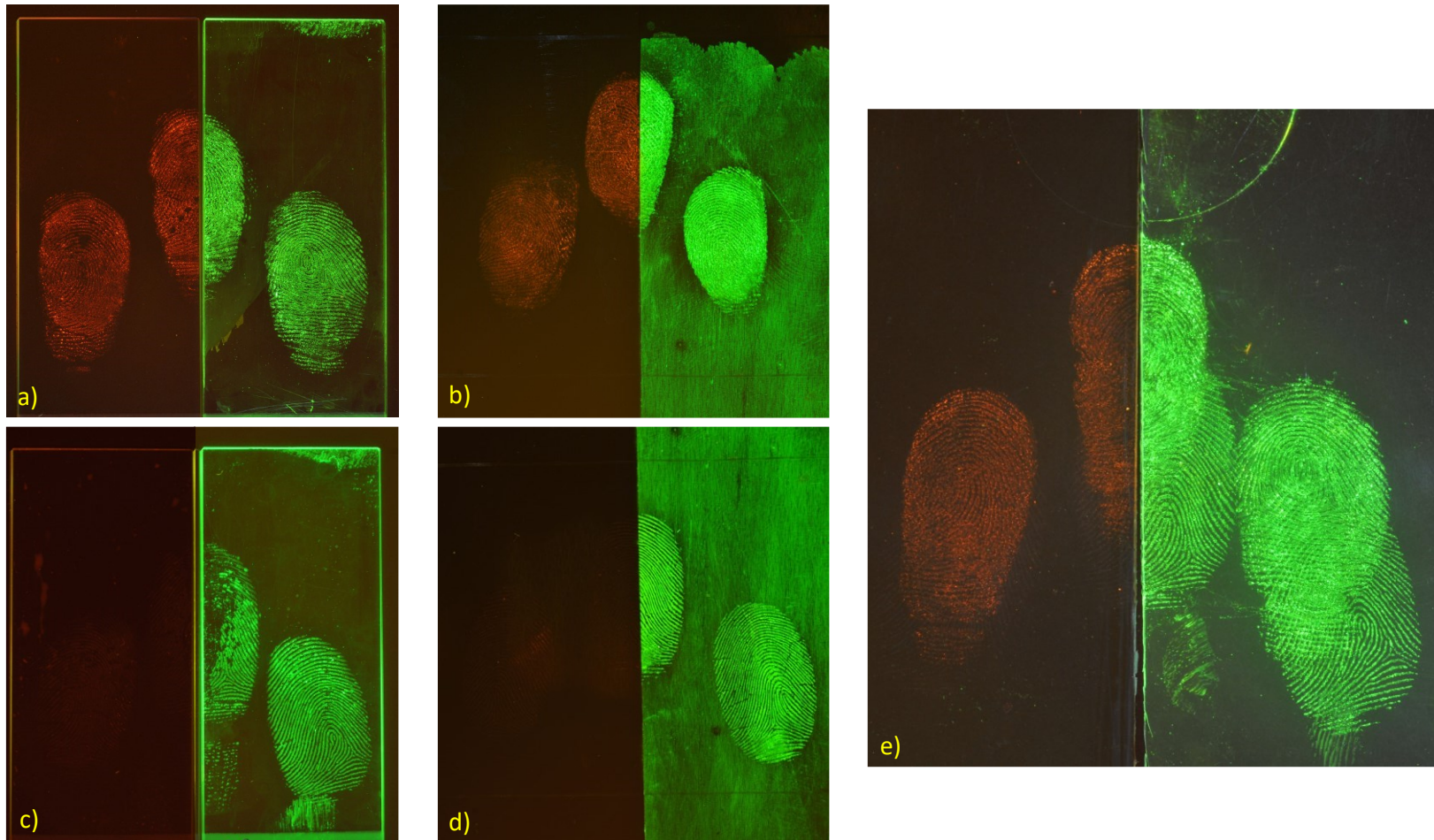
To directly compare acid Nile blue and BY40, natural split fingermarks from five donors on three substrates were treated with both methods. The results for all substrates and donors are summarised in Figure 6.10. These results show that acid Nile blue has potential as post-cyanoacrylate stains with over 50 % of fingermarks graded as identifiable, although BY40 is a more effective option.



**Figure 6.10:** Percentage of fingermarks grades developed out of 30 fingermark halves with acid Nile blue and Basic Yellow 40 on borosilicate glass, aluminium sheet, and polypropylene plastic lid.

It was observed that BY40 consistently performed well across all donors and substrates, while acid Nile blue varied across donors, exhibiting weak development with some donors. This is illustrated in Figure 6.11 which shows a comparison of donor 1 and donor 3 across all three substrates. BY40 developed all fingermarks, however with acid Nile blue, no useful fingermarks were developed for donor 3. Interestingly on plastic, donor 1 inadvertently overlaid fingermarks on a sample deposited by donor 3. The development of this sample is shown in Figure 6.11e, demonstrating the donor effect, as only donor 1's fingermarks were developed using acid Nile blue. Further investigation into natural and charged fingermarks for all donors showed that fingermarks treated with acid Nile blue indicated preferential staining of lipid-rich fingermarks, whereas no donor effect was observed with BY40.





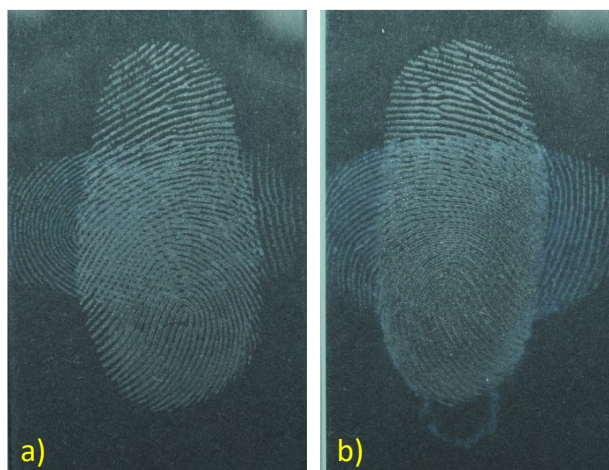
**Figure 6.11:** Comparison of fingermarks developed with acid Nile blue (left half) and Basic Yellow 40 (right half) illustrating donor effect: (a) on borosilicate glass for donor 1 (b) on aluminium sheet for donor 1 (c) borosilicate glass for donor 3 (d) aluminium sheet for donor 3 (e) on polypropylene superimposed of donor 1 (top) and donor 3 (bottom).

These results suggest that staining with acid Nile blue involves the stain partitioning into the sebaceous component of the fingerprint deposit. A possible avenue to increase the technique's sensitivity is to explore the more hydrophobic derivatives of Nile red and Nile blue, such as dialkylated derivatives. These have been demonstrated through initial studies<sup>322</sup> to produce more luminescent and visible fingerprints. It must be noted that specific photographic conditions were set for the experiment and that the weak development with acid Nile blue can be further enhanced by increasing exposure time or using enhancement software.

Another inference is that two different stain mechanisms are at play. In contrast to the general idea that the dyes stain the polymer structure, the results seem to suggest that in the case of acid Nile blue, the dye is likely slip through the polymer structure to interact with the fingerprint deposit. This prompted further investigation into the staining mechanism and the benefit of sequencing acid Nile blue with BY40 to improve detection of overlapping fingerprints, in addition to the initial results (Figure 6.11e).

### **6.3.2.3 Sequencing to enhance detection of overlapping fingerprints**

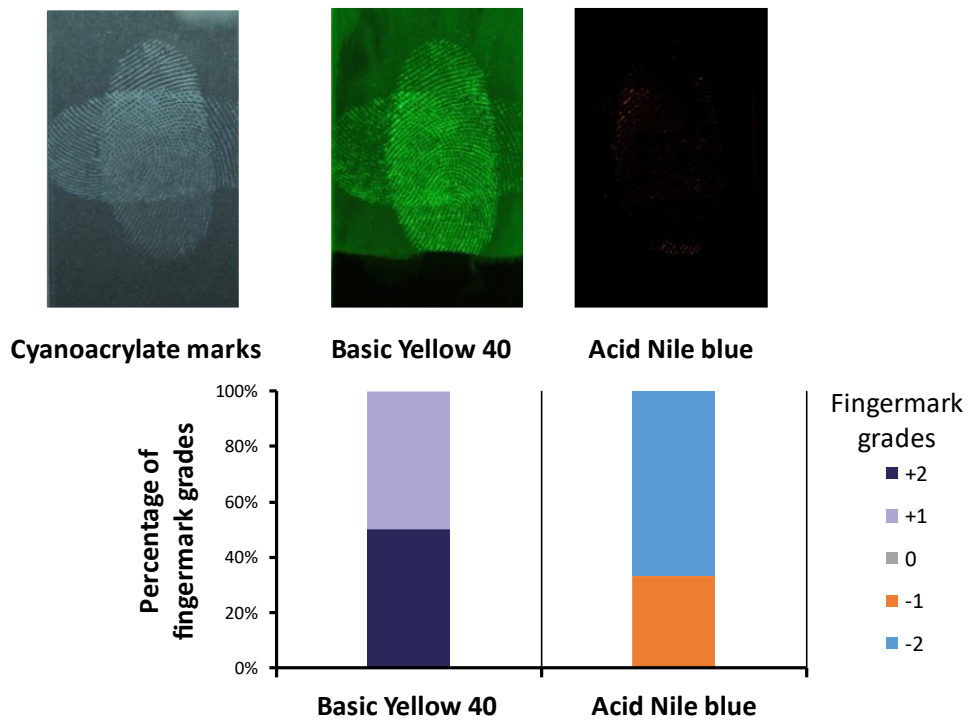
Overlapping fingerprints is known to reduce their recoverability. This portion of the study explored whether using two different staining mechanisms could enhance detection through sequencing. Two types of overlapping fingerprints were collected on microscope slides (Figure 6.12); one set having natural fingerprints (right thumb) overlapping a charged one (left thumb), and the other in the reverse position, with each set collected twice. Fingerprints were collected from three donors and developed using the methodology outlined in section 6.2.3. Each type of overlapping cyanoacrylate fumed fingerprints was stained using the sequence acid Nile blue → BY40 and the reverse sequence.



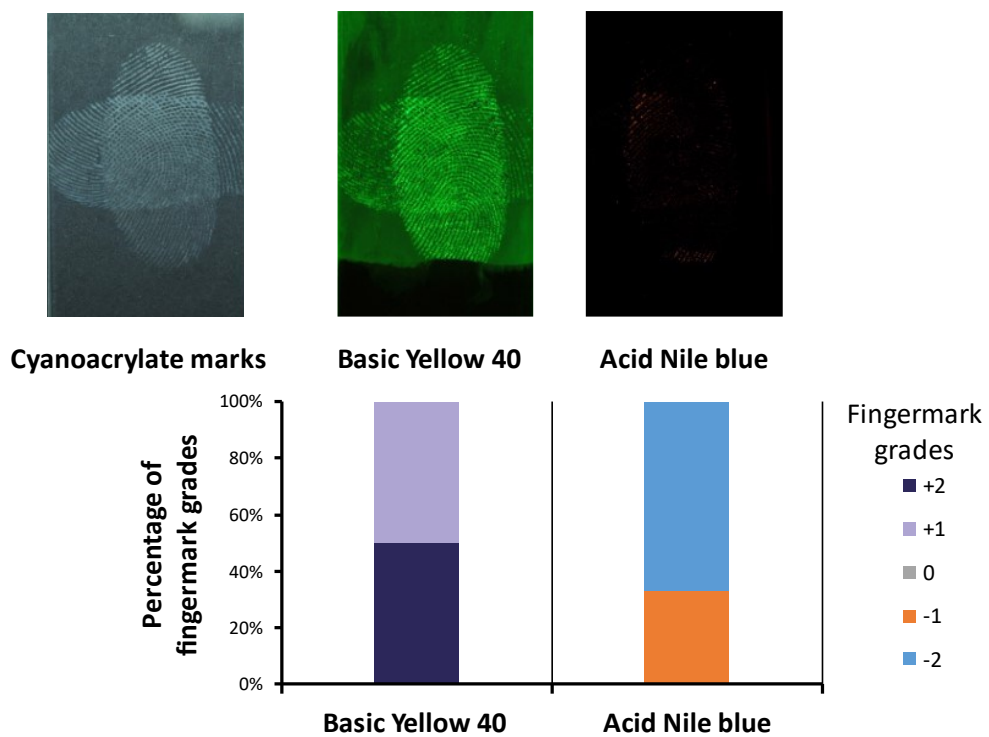
**Figure 6.12:** *Overlapping fingerprint collection (a) natural over charged fingerprints (b) charged over natural fingerprints.*

Given that fingerprint quality would depend on the sequence of the two techniques, the University of Canberra (UC) scale was adapted to provide a more relevant evaluation. Under treatment 1, UC scores of (+2) were given if both charged and natural fingerprints were developed or (+1) if only one fingerprint was developed, which would be classified as a useful fingerprint under the CAST scale. If both fingerprints are only partially developed, a score of (0) is given, while a score of (-1) is assigned if only one fingerprint is partially developed and (-2) if no fingerprints are developed. In treatment 2, scores are adjusted based on whether the quality of fingerprints improved or deteriorated. The results of this study are summarised in Figure 6.13 and Figure 6.14.

As can be seen for both types of overlapping fingerprints, using the sequence BY40 → acid Nile blue resulted in the first treatment (BY40) being effective in staining natural and charged fingerprints, but the second (acid Nile blue) not being as successful. This may be due to BY40 staining the polycyanoacrylate polymer structure, as it has been suggested that the cyanide ( $\text{CN}^-$ ) anions of polycyanoacrylate form weak van der Waals bonds with basic dye cations.<sup>68</sup> Consequently, this could prevent the acid Nile blue, as the second treatment from partitioning into the underlying fingerprint deposit.



**Figure 6.13:** Overlapped fingerprints developed with cyanoacrylate and stained using the sequence BY40 → acid Nile blue, (top) image of results at different development stages and (bottom) UC scale percentage scores assessment result.



**Figure 6.14:** Overlapped fingerprints developed with cyanoacrylate and stained using the sequence acid Nile blue → BY40, (top) image of results at different development stages and (bottom) UC Scale percentage scores assessment result.

When using sequence acid Nile blue → BY40, as expected acid Nile blue exhibited preferential staining of the charged fingermarks, providing better resolution with the overlapped natural fingermarks (Figure 6.14). The use of BY40 as a second treatment, in most instances, led to the development of both natural and charged fingermarks. This reinforces the suggestion that two different mechanisms are at play, with acid Nile blue staining the underlying fingermark deposit and not preventing the staining of the polymer structure by BY40. Based on this result, acid Nile blue would be recommended in the sequence acid Nile blue → BY40.

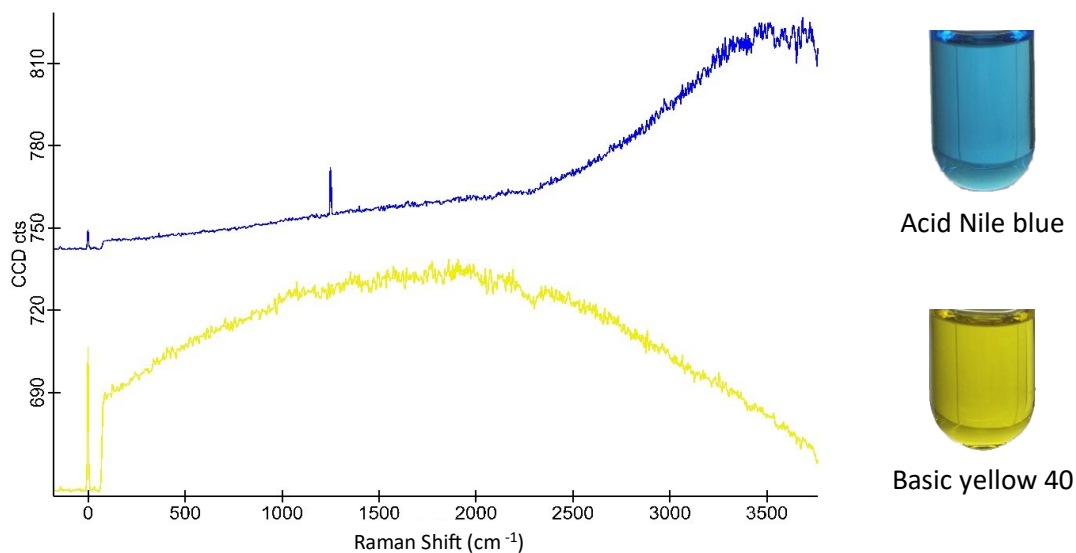
#### **6.3.2.4 Confocal Raman microscopy investigation into post-cyanoacrylate staining mechanism.**

The staining mechanism was investigated using confocal Raman microscopy to explore where the different staining dye is sitting in post-cyanoacrylate stained fingermark. The fundamental understanding has the benefit of directing the development of post-cyanoacrylate staining and its position in the sequencing process.

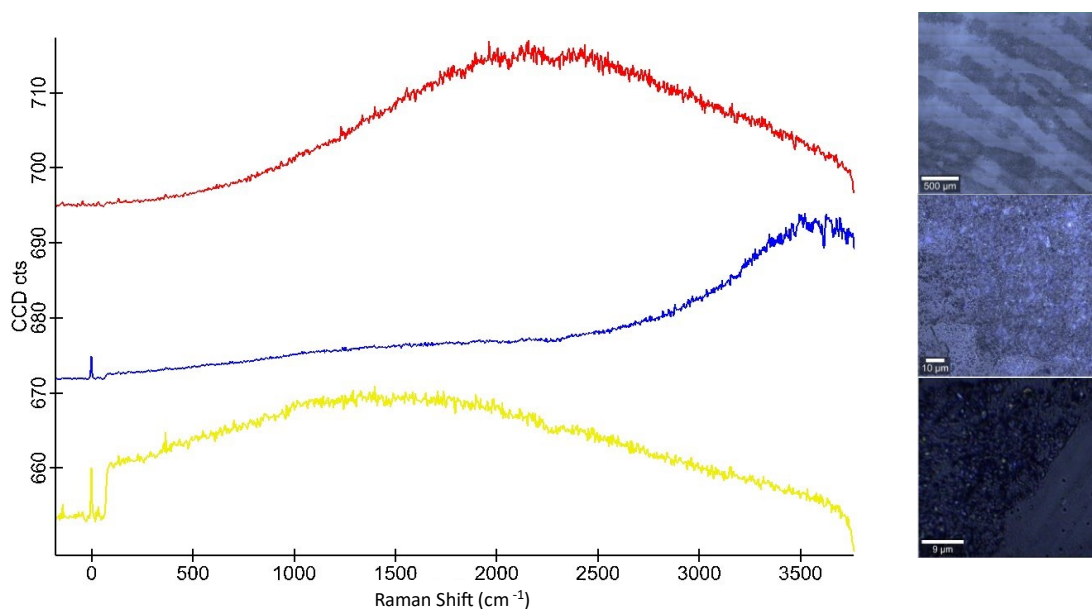
To begin with, an assessment was conducted to ensure that both dyes gave a response with the 532 nm laser of the confocal Raman microscope. This was accomplished by scanning a drop of each dye on a microscope slide. The results depicted in Figure 6.15 showed that both dyes produced a fluorescence response, which was characterised by a broad peak compared to sharper peaks observed during Raman scattering.<sup>323</sup> More importantly, the maximum intensity of each peak occurred at a distinct wavenumber, making it possible to differentiate between the two dyes.

Subsequently, a fumed cyanoacrylate fingermark was analysed to investigate possible interference of the cyanoacrylate polymer by comparing unstained cyanoacrylate fingermarks with stained ones. All three types of polycyanoacrylate marks were distinguishable as shown in Figure 6.16. The unstained fingermark shows a maxima at  $2300\text{ cm}^{-1}$ , while the BY40 stain mark has a maxima at  $1400\text{ cm}^{-1}$ . On the other hand, acid Nile blue has a maxima at  $3500\text{ cm}^{-1}$ , which corresponds to a Raman

shift wavelength of 650 nm, the expected maxima for Nile red. This outcome was to be expected since Nile red is responsible for the photoluminescence properties exhibited by Nile blue, as discussed previously.



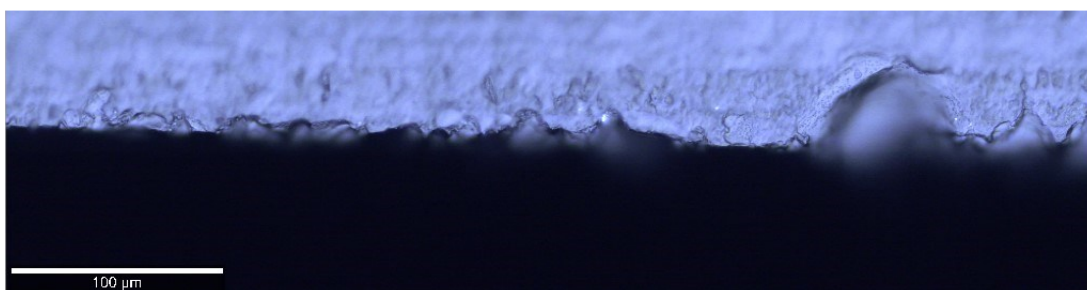
**Figure 6.15:** Raman spectra of acid Nile blue and Basic Yellow 40 liquid smear on glass slide, acquired using WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser.



**Figure 6.16:** Raman spectra of unstained cyanoacrylate fingermark (red), cyanoacrylate fingermark stained with acid Nile blue (blue) and cyanoacrylate fingermark stained with Basic Yellow 40 (yellow)

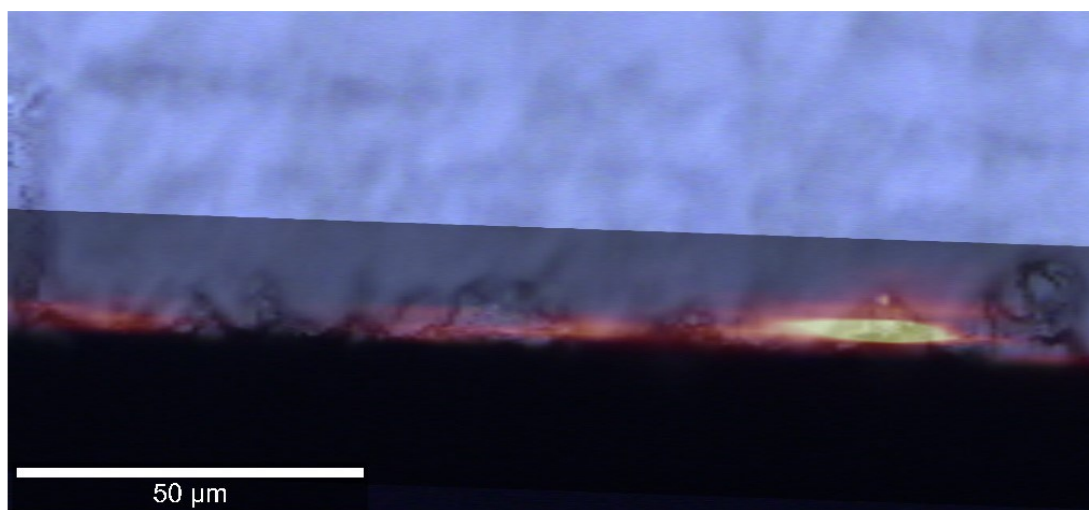
on a glass slide, acquired using WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser.

However, examination of post-cyanoacrylate-stained fingermarks did not provide conclusive information about the staining mechanism. The cross-section of the fingermarks did not reveal the structure of the polymer (Figure 6.17), possibly due to reasons such as the fingermark not being in focus, the instrument's spatial resolution, or the polymer's transparent nature. However, overlay Raman images show fluorescent and non-fluorescent regions, indicating the fingermark ridges and furrows (Figure 6.18).

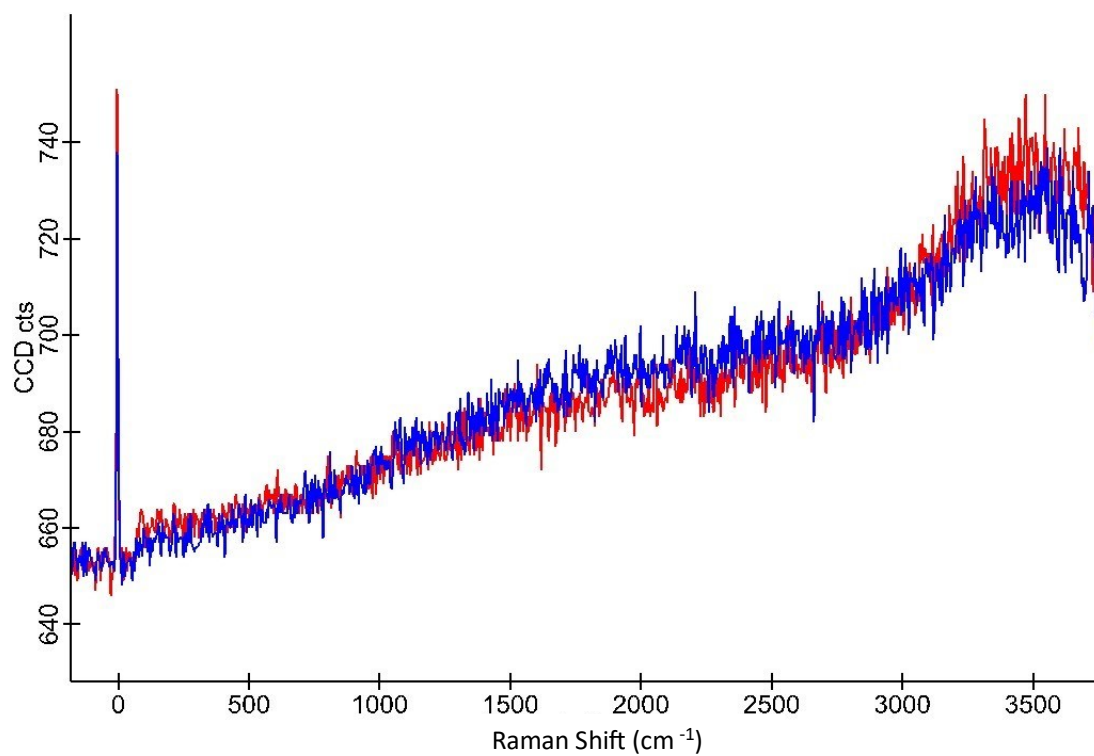


**Figure 6.17:** Visual image of the cross-section of post-cyanoacrylate stain fingermark on glass slide. The fingermark and polymer structure not apparent. The glass side was mounted edge-up.

Spectra taken across different positions of the fingermarks' cross-sections gave similar profiles, and the cyanoacrylate polymer structure could not be distinguished from the fluorescent stain of acid Nile blue (Figure 6.19). The fluorescence distribution of the fumed fingermark suggested a flat morphology, likely due to the non-humidity-controlled chamber used for the cyanoacrylate process in this study. Previous studies have shown that the microstructures formed during the development of the marks depend on the relative humidity, with 70-90 % producing noodle-like structures while 60 % produced flat, film-like structures. The film-like structure is expected to be shorter in height than the noodle-like structure, possibly too small and beyond the instrument's spatial resolution. Nonetheless, the results in section 6.3.2.2 suggest that acid Nile blue and BY40 stained cyanoacrylate fingermarks via two different mechanisms.



**Figure 6.18:** Visual and Raman image overlay of the cross-section of post-cyanoacrylate stain fingerprint on glass slide showing fluorescent and non-fluorescent regions.



**Figure 6.19:** Overlay of Raman spectra at two opposite ends of a post-cyanoacrylate fingerprint stained with acid Nile blue on a glass slide, acquired using a WITec alpha 300SAR confocal Raman microscope equipped with a 532 nm NdYAG laser.



## 6.4 Conclusion

This chapter explored acid Nile blue as an emerging fingerprint detection technique that is low-cost, non-toxic, easily accessible, and easy to use, making it ideal as a frugal forensics method. It was compared to ORO as a lipid-sensitive reagent and investigated as a potential post-cyanoacrylate fingerprint stain.

Like ORO, acid Nile blue was found to successfully develop fingerprints on wet porous substrates, with its effectiveness decreasing as the fingerprint ages, resulting in no fingerprints detected after 30 days. The result reinforces the suggestion that Nile blue targets the 'fragile fraction' of the sebaceous component and should be applied to fingerprints less than 4 weeks old.

As a post-cyanoacrylate stain, acid Nile blue was effective in developing lipid-rich fingerprints whereas no preferential development was observed with BY40. These findings indicate that acid Nile blue interacts with the sebaceous component of the fingerprint deposit. It appears that acid Nile blue penetrates through the polymer structure to interact with the fingerprint residue, contrary to the belief that dyes stain the polymer structure.

Due to the limited sensitivity of acid Nile blue, it is unlikely to replace the Seychelles Police's current operational techniques. However, its frugal forensics attributes can be applied as a training tool and in outreach programs given that it is low-cost, water-based, and low toxicity, particularly when using the non-acidified version. Moreover, the insight gained from understanding the post-cyanoacrylate staining mechanism can aid in developing improved post-cyanoacrylate stains. It is important to note that the aforementioned conclusions must be qualified with the recognition that the studies were conducted on a limited number of fingerprint samples and substrates. Therefore, further analysis is required to establish generalisation.

## Chapter 7. Frugal forensics sustainability assessment tool

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Portions of this chapter have been published in the following articles:

Jemmy T. Bouzin, Thais López, Anna L. Heavey, Jessie. Parrish, Georgina Sauzier, and Simon W. Lewis. Mind the gap: The challenges of sustainable forensic science service provision. *Forensic Science International Synergy* **2023** 6 100318.

DOI: <https://doi.org/10.1016/j.fsisyn.2023.100318>

## 7.1 Introduction

In Chapters 3, 4, 5 and 6, various methods were identified and modified to achieve sustainability in line with the frugal forensic approach. Sustainability was evaluated based on a particular attribute of frugal forensics. For example, the SPF 1,2-indanedione zinc (IND/Zn) was found to be more sustainable than the UK CAST formulation due to the cost of the working solution and the difference in availability and accessibility of the carrier solvents.<sup>233, 324</sup> However, the counterargument is that in terms of safety, the CAST formulation is more sustainable as it uses a non-flammable solvent HFE-7100, compared to SPF IND/Zn which uses the flammable solvent petroleum ether 40-60 °C.<sup>201, 241</sup> This raises the question of how to assess the sustainability of a method comprehensively and objectively within the frugal forensics framework.

As discussed in Chapter 1, frugal forensics focuses on three core principles (Resilient, Economical and Quality) and six attributes (Performance, Accessibility, Availability, Cost, Simplicity and Safety) which take into consideration jurisdictional local challenges to develop sustainable forensic science provisions. Regardless of the local challenges, the development and assessment of forensic science methods must be carried out in a manner reflecting international best practices and minimum requirements.<sup>2, 325</sup> For latent fingerprint detection, the IFRG guidelines<sup>24</sup> provide such an evaluation and reporting framework for the performance assessment of new or modified methods. The guidelines specifically recommend testing methods in their local operational environment. The frugal forensics approach can be viewed as an extension of this recommendation by assessing performance and sustainability in a local context.

To address the aforementioned question, an assessment tool was developed that draws upon the IFRG fingerprint performance framework through the use of a similar scoring system as the fingerprint grading scale.<sup>19, 224</sup> This tool consists of two components (depicted in Table 7.1 and Table 7.2) to assess the sustainability between two methods. Table 7.1 provides a comparative scoring system to assess the sustainability of a new or modified method against an existing method referred

to as “alternative” and “control” methods, respectively. Table 7.2 provides a guide on how likely the alternative method would be recommended for implementation.

**Table 7.1:** *Frugal forensics sustainability comparative score assessment tool*

PAACSS comparative score	
Score	Description
2	Major benefit of alternative method
1	Significant benefit of alternative method
0.5	Minor benefit of alternative method
0	No difference between methods
-0.5	Minor benefit of control method
-1	Significant benefit of control method
-2	Major benefit of control method

**Table 7.2:** *Frugal forensics method recommendation score assessment tool*

Recommendation score	
Total Score	Description
$\geq 3$	Strongly recommended
2	Recommended
0.5	Maybe recommended
$\leq 0$	Not Recommended

The assessment works by evaluating each of the six frugal forensics attributes between the alternative and control method using Table 7.1. The sum of scores (total score) are calculated and based on this score, Table 7.2 provides a guide of how likely the alternative method would be recommended for implementation with respect to the control method. The attributes are classified into minor, significant, and major benefits, with their relevance being evaluated according to the local circumstances of each jurisdiction. For instance, a jurisdiction with a reliable supply chain may assign less weight to the availability factor. In other words, the weights are not uniform but rather contingent upon a jurisdiction's unique challenges. This approach enables a

comprehensive assessment of the six attributes of frugal forensics and, consequently, the sustainability of the method for a specific jurisdiction.

This chapter evaluates the sustainability of the methods developed in previous chapters using the developed assessment tool. The application of the tool, results, and its benefits are discussed.

## **7.2 Experimental**

The assessment using the evaluative tool was conducted as follows:

The SPF IND/Zn formulation, investigated in Chapters 3 and 4, was evaluated using the CAST formulation as a control method. CAST IND/Zn was chosen as the control since it is the method expected to be used by default in Seychelles.

The Wet SPF method, developed in Chapter 5, was evaluated using Wetwop™ as a control method. Acid Nile blue as a post-cyanoacrylate stain, investigated in Chapter 6, was evaluated using Basic Yellow 40 (BY40) as a control method. These two control methods were chosen because they are the current operational techniques used in Seychelles.

## **7.3 Results and Discussion**

Table 7.3 provides a summary of the assessment tool results for the sustainability evaluation of the various methods explored in this study under Seychelles conditions and limitations. Based on the results, SPF IND/Zn and Wet SPF powder suspension are recommended as replacements for CAST IND/Zn and Wetwop™, respectively. However, acid Nile blue is not recommended as a substitute for BY40. The reasoning for each detection technique's score is explained below.

In Chapters 3 and 4, it was shown that the SPF IND/ZN and CAST formulations were equally effective compared to the Australian NCFS formulation. Therefore, a score of 0 was attributed to the performance of SPF against the CAST formulation. However, the main difference between these two formulations is the carrier solvent. CAST formulation uses HFE-7100, which poses a significant supply chain risk, while the SPF

formulation uses petroleum ether 40-60 °C, which is more easily accessible, thus reducing this risk. The latter solvent hence provides a major advantage in managing the supply chain risk, resulting in a score of +2 attributed to both Availability and Accessibility. The difference in carrier solvents also results in a variation in cost, with one litre of working solution costing \$267 USD for CAST and \$122 USD for the SPF formulation. Inexpensive formulation is a major economic advantage for laboratories with limited budgets and gives the SPF formulation a score of +2 for cost. Petroleum ether is a highly flammable solvent and is a significant safety risk. However, the risk is reduced when used in a properly fitted laboratory with safety protocols in place, hence a score of -1 was attributed to safety. Both methods are quite similar in terms of equipment and processing, except that the CAST formulation requires oven processing while the SPF formulation uses a heat press. The latter is a less expensive and simpler process, so it was given a simplicity score of +0.5. Based on the total score of +5.5, SPF IND/Zn is the strongly recommended option over the CAST formulation under Seychelles conditions and limitations.

**Table 7.3:** Comparative score and recommendation results for SPF IND/Zn with CAST formulation, Wet SPF powder suspension with Wetwop™ formulation and post-cyanoacrylate acid Nile blue method with BY40.

Fingermark detection method			Frugal forensics attributes assessed using Table 6.1						Recommendation assessed using Table 6.2	
Type	Alternative method	Control method	Performance	Availability	Accessibility	Cost	Safety	Simplicity	Total score	Recommendation
1,2-indanedione zinc	SPF	CAST	0	+2	+2	+2	-1	+0.5	+5.5	Strongly recommended
Powder suspension	Wet SPF	Wetwop™	0	0	+1	+2	0	-1	+2	Recommended
Post-cyanoacrylate staining	Acid Nile blue	BY40	-2	0	0	+1	0	0	-1	Not recommended

In Chapter 5, the comparative effectiveness of Wet SPF to the commercial Wetwop™ powder suspension on both acrylic and rubber-based pressure-sensitive adhesive tapes was clearly shown. Hence, a score of 0 was attributed to the performance of Wet SPF against Wetwop™ formulation. The Wet SPF formulation was a modification of the WET UCIO powder formulation<sup>175</sup> with the inaccessible Gran Velada surfactant being replaced by an in-house surfactant solution prepared from a readily available

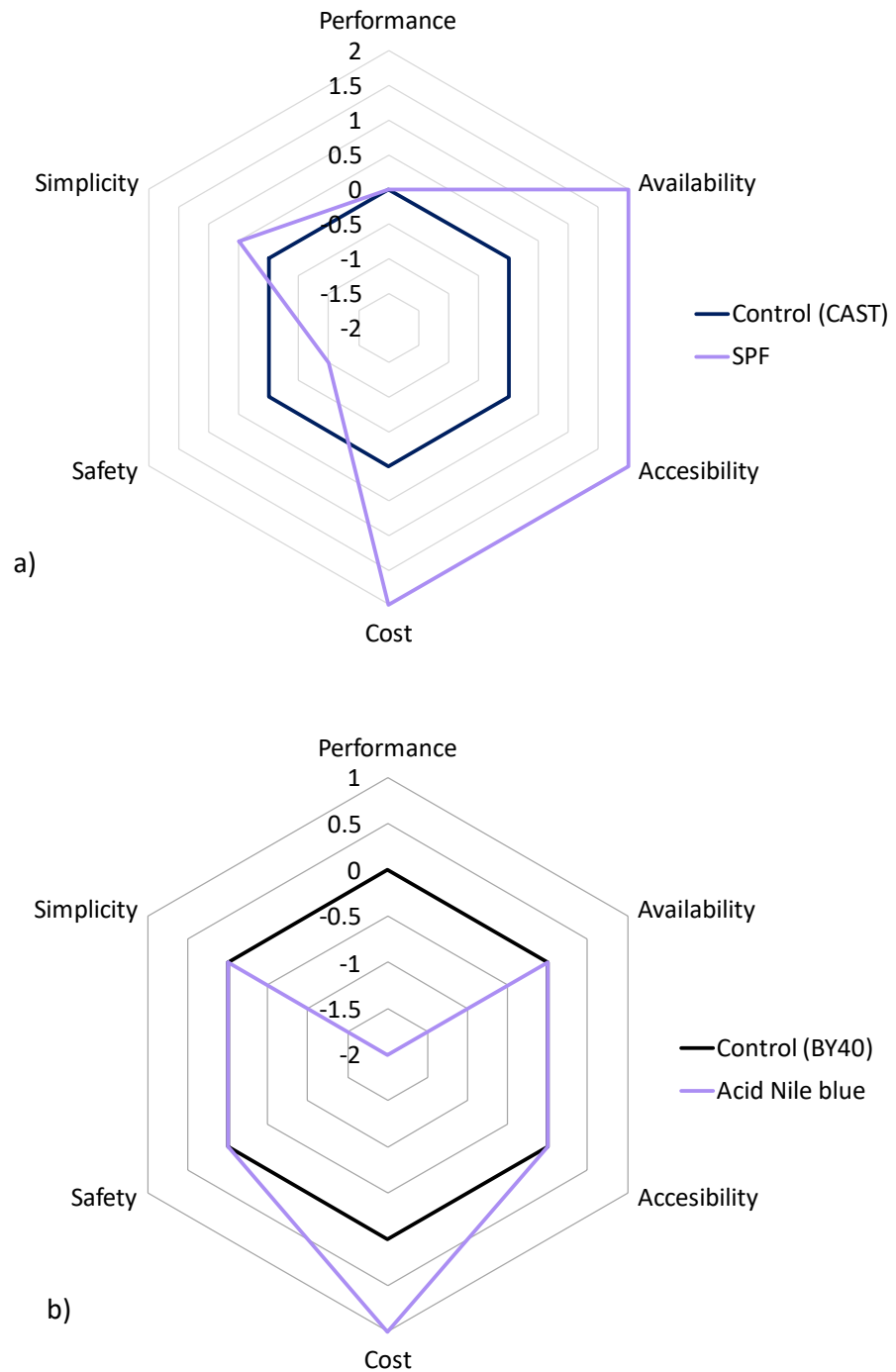
SDS salt, accessible from multiple suppliers. In remote locations such as Seychelles, using in-house powder suspension (Wet SPF) compared to commercial formulation (Wetwop™) allows for easier transportation, storage, and better control of quality and shelf-life. This means that although both formulations are readily available, Wet SPF is more accessible. Therefore, a score of 0 and +1 was attributed to Availability and Accessibility, respectively. In terms of cost, the Wet SPF formulation presents a major advantage, being four times cheaper than Wetwop™, hence a score of +2 was attributed to Cost. Both formulations are low-risk procedures but Wetwop™ is a simpler method as it does not involve preparation. Therefore, Safety and Simplicity were rated 0 and -1, respectively. Based on the total score of +2, the Wet SPF powder suspension formulation is the recommended option over the commercial Wetwop™ formulation for operational use in Seychelles.

In Chapter 6, although acid Nile blue was shown to be effective as a post-cyanoacrylate stain – particularly for lipid-rich fingerprint deposits - BY40 was found to be superior in developing all types of fingerprints. Therefore, a score of -2 was attributed to performance. The approximate cost of BY40 dye in one litre of working solution is \$6.32 USD, while Nile blue costs only \$0.21 USD. This represents a significant economic saving, hence a score of +1 was given for Cost. Both dyes are readily available and accessible, safe and simple, resulting in a score of 0 for all of these attributes. Based on the total score of -1, acid Nile blue is not recommended as a replacement for BY40 under Seychelles conditions.

In essence, the assessment tool provides a more objective and holistic evaluation for the sustainability of latent fingerprint methods in consideration of the jurisdiction-specific challenges. It provides a transparent process to evaluate and select methods for implementation. This is crucial to demonstrate the validity and weighting of results provided to end-users of forensic information and forms a critical component of quality assurance in forensic science service provision.

Furthermore, the assessment tool has benefits beyond decision-making, as it can also identify areas for sustainability improvement. For example, a graphical representation of the scores for the IND/Zn and acid Nile blue methods, with the

control method given a base score of zero (Figure 7.1), reveals safety as an area for improvement for SPF IND/Zn and performance for acid Nile blue.



**Figure 7.1:** Comparison of frugal forensics attributes scores for (a) the SPF IND/Zn formulation with CAST as control method and (b) acid Nile blue as a post-cyanoacrylate stain with BY40 as control. All control methods were given a base score of zero.



Similarly, to the hierarchy of controls in managing health and safety risks,<sup>326, 327</sup> a framework could be put in place to address the factors that contribute to the identified areas or negative scores. Subsequently, the modified method could be reevaluated using the sustainability tool, providing a methodical process for developing sustainable methods. Hence the assessment tool can be used both as decision-making and a planning tool. It is important to acknowledge that while the current tool is tailored towards latent fingerprint methods in micro and remote jurisdictions, it can also be adjusted to suit other jurisdictions and other forensic science service provisions.

## **7.4 Conclusion**

This chapter addresses the question of how to comprehensively and objectively evaluate the sustainability of a method using the frugal forensics approach. Frugal forensics focuses on three principles and six attributes to develop sustainable forensic science provisions. International best practices and minimum requirements must be followed for developing and accessing forensic science methods. For latent fingerprint detection, the IFRG guidelines provide an evaluation and reporting structure. In consideration of these frameworks, a grading tool was developed to assess the sustainability of new or modified forensic science methods.

This approach uses a scoring system to evaluate the sustainability of a new or modified method based on the six attributes of frugal forensics and provides a recommendation for implementation. Based on the evaluative tool results in the context of Seychelles, the SPF IND/Zn was strongly recommended over the CAST formulation. Similarly, the Wet SPF powder suspension was recommended over the commercial Wetwop™ formulation, while acid Nile blue was not recommended as a replacement for BY40.

Overall, the tool offers a more objective and comprehensive assessment that considers the unique challenges of each jurisdiction. This approach is both rigorous and thorough, ensuring a holistic evaluation of the frugal forensics sustainability

attributes. Moreover, the objective approach promotes transparency in method selection and adheres to the quality assurance requirements.

## Chapter 8. Conclusion and future work

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Portions of this chapter have been published in the following articles:

Jemmy T. Bouzin, Aaron J. Horrocks, Georgina Sauzier, Stephen M. Bleay, and Simon W. Lewis. Comparison of three active 1,2-indanedione-zinc formulations for fingerprint detection in the context of limited resources and supply chain risks in Seychelles. *Forensic Chemistry* **2022** 30 100439.

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Jemmy T. Bouzin, Thais Lópes, Anna L. Heavey, Jessie. Parrish, Georgina Sauzier, and Simon W. Lewis. Mind the gap: The challenges of sustainable forensic science service provision. *Forensic Science International Synergy* **2023** 6 100318.

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## 8.1 Conclusion

This thesis aimed to improve latent fingerprint detection protocols in remote and resource-limited jurisdictions, focusing on Seychelles as a small island state. In this project, the frugal forensics framework has been proposed as a means of enhancing the accessibility of latent fingerprint detection techniques and enabling their use in the Global South. This approach emphasises the development of sustainable methods that are adapted to local challenges for a resilient and cost-effective forensic science service. Through the successful application of this concept to three different latent fingerprint methods, two sustainable Seychelles Police Force (SPF) formulations (SPF 1,2-indanedione/zinc and Wet SPF powder suspension) that are well-suited for use in Seychelles were developed. Additionally, an evaluation tool was developed to facilitate a holistic assessment of the sustainability of methods to be implemented while satisfying the quality assurance requirements. Its utility as a strategic planning instrument for the systematic enhancement of the sustainability of a particular methodology is an added advantage.

In essence, this thesis presents the first systematic framework for sustainable forensic science service provision. In the context of latent fingerprint provision, this project will improve the detection capabilities of latent fingerprints and facilitate their use in operational laboratories in the Global South, particularly jurisdictions with limited resources such as the Seychelles. The development of sustainable methods ensures service continuity where otherwise it would have been limited or absent. Implementation of effective latent fingerprint detection techniques has the potential to increase the recovery of fingerprint evidence and improve their effectiveness in criminal investigations. Altogether, this research will have a direct impact on the forensic reform in Seychelles, improving the forensic capacity and fostering research and development. The outcomes may also be used as a benchmark by jurisdictions facing similar circumstances, such as Mauritius, Madagascar, Philippines, and Timor-Leste. Hence, creating more consistency in the quality of forensic service provision across different jurisdictions. The generated knowledge will likely have a global impact, particularly as other regions search for alternative

carrier solvents for amino acid sensitive reagents.<sup>324</sup> Future work recommendations are presented below, in addition to the chapter-specific conclusions.

## **8.2 Alternative carrier solvent for amino acid sensitive reagent**

In Chapter 3, the frugal forensics approach was used to improve an existing method for fingerprint enhancement on porous surfaces. This was achieved by modifying the co-solvents, active chemicals, and carrier solvents of three active 1,2-indanedione/zinc (IND/Zn) formulations to develop a sustainable formulation (SPF IND/Zn). The main challenge was to replace the expensive and supply chain-disruptive solvent, HFE-7100, in the IND/Zn formulation.<sup>233</sup> Through the frugal forensics approach, petroleum ether 40-60 °C in the BKA IND/Zn formulation was found to be a better alternative for managing costs and reducing supply chain risks. Solstice® PF was also considered as an alternative carrier solvent, but it presented similar supply chain challenges to HFE-7100 whilst causing higher ink diffusion and possible chemical interactions with other reagent components that could affect stability. The benefit of frugal forensics was further demonstrated by reducing excess 1,2-indanedione in the BKA formulation, improving reagent effectiveness and cost. The importance of achieving the right balance of the polar solvent to ensure optimal luminescence while minimising background development and ridge diffusion was highlighted.

Future work should explore other alternative carrier solvents, especially with the recent development that the production of the standard carrier solvent for amino acid sensitive reagents, HFE-7100, will be discontinued by 3M in 2025.<sup>324</sup> From a frugal forensics perspective, it would be beneficial to investigate other readily available and affordable solvents that have similar properties to a suitable carrier solvent. Additionally, it is important to gain a better understanding of how the carrier solvent Solstice® PF interacts with reagent components and how it affects reagent performance. This information will be valuable in the search for alternative carrier solvents.

### 8.3 Powder suspension and acid Nile blue improvements

The versatility of the frugal forensics approach was demonstrated on non-porous surfaces through its application to a recently published method. Chapter 5 focused on the development of an effective and accessible method for detecting latent fingerprints on pressure-sensitive adhesive (PSA) tape. The method involves modifying a recently published<sup>175</sup> powder suspension formulation by substituting the commercial surfactant solution with an in-house SDS solution, resulting in a more affordable and easily accessible reagent. The effectiveness of the modified formulation was shown to be comparable to that of the commercial Wetwop™ powder suspension.

Future work should focus on re-assessing the effective ratio of powder to the modified surfactant solution. The in-house SDS surfactant solution was produced using a 5 % ethanol/water mixture as an additive. Variation in the concentration and type of additives was found to play a minimal role in the powder suspension performance. However, it is important to investigate if this holds true for other additives. Moreover, the modified formulation demonstrated efficacy in developing latent fingerprints on both light and dark-coloured tapes when used with a forensic light source. This suggests the possibility of using a single formulation for different coloured tapes. As this study was confined to carbon-based powder, it is suggested that further research be conducted to compare results with white powder suspension. Currently, there is ongoing research at Curtin University on improvised powders under the banner of frugal forensics. Combining the Wet SPF surfactant with viable improvised powders presents another avenue for future research.

In Chapter 6, acid Nile blue as an emerging lipid-sensitive fingerprint detection technique with frugal forensics attributes was investigated. Through comparative analysis with Oil red O, the study revealed that acid Nile blue is most effective in detecting charged fingerprints that are less than 4 weeks old. Additionally, acid Nile blue is selective in its sensitivity towards sebaceous material when used as a post-cyanoacrylate stain, suggesting a different staining mechanism compared to Basic

Yellow 40. However, further research is needed to enhance acid Nile blue's sensitivity, which may involve exploring other benzophenoxazine dyes.

#### **8.4 IFRG guidelines requirements**

The IND/Zn formulation and powder suspension method developed in Chapters 3-5, were investigated in line with phases 2/3 of the IFRG guidelines.<sup>24</sup> Comparative assessment of each method against an operational technique was carried out through laboratory trials using split fingermarks on a range of ten donors and multiple substrates. The evaluation included incidental fingermarks and simulated casework substrates for meaningful results using more realistic samples. The sensitivity of the methods was tested using split depletion fingermarks.

Overall, the effectiveness of the methods was demonstrated using 1200 fingermark halves on five different porous substrates for SPF IND/Zn and 320 fingermarks on eight different PSA tapes for Wet SPF. Additionally, both formulations were also shown to be effective on 1 week and 4 weeks old samples, which is the general timeframe for processing casework samples in Seychelles. However, additional IFRG requirements must be satisfied before either method's application in casework. Further operational validation is needed on casework samples to test their operational suitability and this phase should be carried out at the operational laboratory.

#### **8.5 Sustainable forensic science service provision**

Chapter 7 addressed the question of how to systematically assess the sustainability of a method. An assessment tool was developed drawing on the same concept utilised in the fingermark grading scale. By objectively assessing all six attributes of frugal forensics, this tool promotes transparency in method selection and adheres to the quality assurance requirements. Not only does it aid in decision-making, but it also acts as a valuable planning tool by pinpointing areas in need of improvement. The application of the tool was demonstrated through the successful assessment of the three methods investigated in Chapters 3-6. Furthermore, the frugal forensics

concept provides an opportunity to revisit current methods from a distinct perspective.

In summary, this thesis has demonstrated how frugal forensics can be applied and evaluated to develop sustainable latent fingerprint detection techniques. While the focus is on fingerprint detection, the economic, resilient, and quality-driven principles of frugal forensics can be extended to the broader forensic science service in the Global South. Through awareness of frugal forensics, investigation into and application of attainable, fit-for-purpose methodologies may bridge the gap in service provision in the Global South, promoting justice and aiding the public good, for present and future generations. The role of forensic science in achieving the United Nations Sustainable Development Goals is significant and calls for future research to promote an inclusive and outward-looking approach to forensic science, especially as the forensic community<sup>328</sup> re-evaluates the current state and definition of this field.



## Chapter 9. References

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## Appendices

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## **Appendix A-1: Statistical analysis test assumptions.**

### **Mann-Whitney U Test**

#### Assumptions

- # Data points should be independent
- # Only two categorical dependent variables (Treatment A and B)
- # Ordinal independent variable (CAST grades)

#### Hypothesis

- # Null hypothesis: The mean ranks of the groups are the same ( $H_0$ )
- # Alternate hypothesis: The mean ranks of the groups are not the same ( $H_A$ )

### **Kruskal-Wallis Test**

#### Assumptions

- # Data points should be independent
- # Two or more categorical dependent variables (Treatment A, B, C)
- # Ordinal independent variable (CAST grades)

#### Hypothesis

- # Null hypothesis: The mean ranks of the groups are the same ( $H_0$ )
- # Alternate hypothesis: The mean ranks of the groups are not the same ( $H_A$ )

### **Chi-squared Test**

#### Assumptions

# Data points should be independent

# At least ten values recorded in each cell

Hypothesis

# Null hypothesis: The mean ranks of the groups are the same ( $H_0$ )

# Alternate hypothesis: The mean ranks of the groups are not the same ( $H_A$ )

### **Difference of Proportions – Treatment A vs. Treatment B**

Assumptions

# Data points should be independent

# At least ten values recorded in each cell

Hypothesis

# Null hypothesis: The proportions are the same ( $H_0$ )

# Alternate hypothesis: The proportions are not the same ( $H_A$ )

## Appendix A-2: Statistical analysis - Chapter 3 R codes and results.

### HeadtoHead\_IND/Zn\_Statistical test

#### Grade frequency table

	NCFS (tA)	BKA (tB)	CAST (tC)
Grade 0	10	5	8
Grade 1	28	32	38
Grade 2	40	42	34
Grade 3	50	50	44
Grade 4	22	21	26

### HeadtoHead\_IND/Zn (Kruskal-Wallis Test)

#### R Code

```
tA = c(rep(0,10), rep(1,28), rep(2,40), rep(3,50), rep(4,22))
```

```
tB = c(rep(0,5), rep(1,32), rep(2,42), rep(3,50), rep(4,21))
```

```
tC = c(rep(0,8), rep(1,38), rep(2,34), rep(3,44), rep(4,26))
```

```
Nsamp = 450
```

```
data_matrix = as.data.frame(matrix(0, Nsamp, 2))
```

```
colnames(data_matrix) = c("Treatment", "Grade")
```

```
data_matrix[1:150,1] = "tA"
```

```
data_matrix[151:300,1] = "tB"
```

```
data_matrix[301:450,1] = "tC"

data_matrix[,1] = as.factor(data_matrix[,1])

data_matrix[1:150,2] = tA

data_matrix[151:300,2] = tB

data_matrix[301:450,2] = tC

kruskal.test(Grade~Treatment, data = data_matrix)
```

### **Result**

Kruskal-Wallis chi-squared = 0.12693, df = 2, p-value = 0.9385

### **Conclusion**

At a 95% level of confidence there not enough evidence to support that a randomly drawn value from one treatment is more likely to be larger or smaller on average than a randomly drawn value from a different treatment.

### **HeadtoHead\_IND/Zn (chi-square)**

R Code

```
tA = c(10,28,40,50,22)

tB = c(5,32,42,50,21)

tC = c(8,38,34,44,26)

Ntreatment = 3

data_matrix = matrix(0, length(tA), Ntreatment)

data_matrix[,1] = tA
```

```
data_matrix[,2] = tB
```

```
data_matrix[,3] = tC
```

```
chisq.test(data_matrix)
```

### **Result**

X-squared = 5.2084, df = 8, p-value = 0.7351

### **Conclusion**

At a 95% level of confidence there not enough evidence to support that there is some dependency between the grades and the treatments.

## Appendix A-3: Statistical analysis - Chapter 4 R codes and results.

### LabTrial\_IND/Zn\_Statistical test

#### Grade frequency table

	SPF (tA)	NCFS (tB)
Grade 0	136	130
Grade 1	67	69
Grade 2	93	94
Grade 3	202	192
Grade 4	102	115

### LabTrial\_IND/Zn (Difference in proportion)

#### R Code

uA = 304

uB = 307

nA = 600

nB = 600

```
prop.test(c(uA,uB), c(nA,nB), alternative="two.sided", conf.level = 0.95, correct = FALSE)
```

#### Result

X-squared = 0.03001, df = 1, p-value = 0.8625

95 percent confidence interval:

-0.06156907 0.05156907

sample estimates:

prop 1 prop 2

0.5066667 0.5116667

### **Conclusion**

At a 95% level of confidence there not enough evidence to support that the proportion of Grade 3&4 under Treatment A is not equal to the proportion of Grade 3&4 under Treatment B.

### **LabTrial\_IND/Zn (Mann-Whitney U)**

#### **R Code**

```
tA = c(rep(0,136), rep(1,67), rep(2,93), rep(3,202), rep(4,102))
```

```
tB = c(rep(0,130), rep(1,69), rep(2,94), rep(3,192), rep(4,115))
```

```
wilcox.test(tA, tB, paired = FALSE, alternative = "two.sided")
```

#### **Result**

W = 176653, p-value = 0.5657

### **Conclusion**

At a 95% level of confidence there not enough evidence to support that a randomly drawn value from one treatment is more likely to be larger or smaller on average than a randomly drawn value from a different treatment.

## LabTrial\_IND/Zn (Chi-squared Test)

### R Code

```
tA = c(136,67,93,202,102)
```

```
tB = c(130,69,94,192,115)
```

```
Ntreatment = 2
```

```
data_matrix = matrix(0, length(tA), Ntreatment)
```

```
data_matrix[,1] = tA
```

```
data_matrix[,2] = tB
```

```
chisq.test(data_matrix)
```

### Result

X-squared = 1.2027, df = 4, p-value = 0.8777

### Conclusion

At a 95% level of confidence there not enough evidence to support that there is some dependency between the grades and the treatments.

## IncidentalTrial\_IND/Zn\_Statistical test

### Number of (3&4) fingermarks frequency table

	SPF (tA)	NCFS (tB)
Substrate 1	34	26
Substrate 2	39	37



Substrate 3	26	24
Substrate 4	8	14
Substrate 5	46	48

---

### **IncidentalTrial\_IND/Zn (Chi-squared Test)**

#### **R Code**

```
tA = c(136,67,93,202,102)
```

```
tB = c(130,69,94,192,115)
```

```
Ntreatment = 2
```

```
data_matrix = matrix(0, length(tA), Ntreatment)
```

```
data_matrix[,1] = tA
```

```
data_matrix[,2] = tB
```

```
chisq.test(data_matrix)
```

#### **Result**

Kruskal-Wallis chi-squared = 0.12693, df = 2, p-value = 0.9385

#### **Conclusion**

At a 95% level of confidence there not enough evidence to support that a randomly drawn value from one treatment is more likely to be larger or smaller on average than a randomly drawn value from a different treatment.

## Appendix A-4: Statistical analysis - Chapter 5 R codes and results.

LabTrial\_CPS\_Statistical test

Grades frequency table\_LabTrial\_CPS

---

	Wet SPF	Wetwop
Grade 0	16	29
Grade 1	26	19
Grade 2	10	15
Grade 3	14	21
Grade 4	94	76

---

Grades frequency table\_Sensitivity\_CPS

---

	Wet SPF	Wetwop
Grade 0	1	10
Grade 1	7	21
Grade 2	12	36
Grade 3	32	35
Grade 4	98	48

---

LabTrial\_CPS (Mann-Whitney U Test)

R Code

```
tA = c(rep(0,16), rep(1,26), rep(2,10), rep(3,14), rep(4,94))
```

```
tB = c(rep(0,29), rep(1,19), rep(2,15), rep(3,21), rep(4,76))
```

```
wilcox.test(tA, tB, paired = FALSE, alternative = "two.sided")
```

### **Result**

W = 14285, p-value = 0.05069

### **Conclusion**

At a 95% level of confidence there not enough evidence to support that a randomly drawn value from one treatment is more likely to be larger or smaller on average than a randomly drawn value from a different treatment.

### **Sensitivity\_CPS (Mann-Whitney U Test)**

#### **R Code**

```
tA = c(rep(0,1), rep(1,7), rep(2,12), rep(3,32), rep(4,98))
```

```
tB = c(rep(0,10), rep(1,21), rep(2,26), rep(3,35), rep(4,48))
```

```
wilcox.test(tA, tB, paired = FALSE, alternative = "two.sided")
```

### **Result**

W = 14428, p-value = 2.742e-09

### **Conclusion**

At a 95% level of confidence there is enough evidence to support that a randomly drawn value from one treatment is more likely to be larger or smaller on average than a randomly drawn value from a different treatment.

### **LabTrial\_CPS (Chi-squared Test)**

```
tA = c(16,26,10,14,94)
```

```
tB = c(29,19,15,21,76)
```

```
Ntreatment = 2
```

```
data_matrix = matrix(0, length(tA), Ntreatment)
```

```
data_matrix[,1] = tA
```

```
data_matrix[,2] = tB
```

```
chisq.test(data_matrix)
```

### **Result**

X-squared = 9.1503, df = 4, p-value = 0.05745

### **Conclusion**

At a 95% level of confidence there is not enough evidence to support that there is some dependency between the grades and the treatments.

### **Sensitivity\_CPS (Chi-squared Test)**

```
tA = c(1,7,12,32,98)
```

```
tB = c(10,21,36,35,48)
```

```
Ntreatment = 2
```

```
data_matrix = matrix(0, length(tA), Ntreatment)
```

```
data_matrix[,1] = tA
```

```
data_matrix[,2] = tB
```

```
chisq.test(data_matrix)
```

## Result

X-squared = 43.621, df = 4, p-value = 7.69e-09

## Conclusion

At a 95% level of confidence there is enough evidence to support that there is some dependency between the grades and the treatments.

## LabTrial\_CPS (Difference in proportion)

### R Code

```
uA = 108
```

```
uB = 97
```

```
nA = 160
```

```
nB = 160
```

```
prop.test(c(uA,uB), c(nA,nB), alternative="two.sided", conf.level = 0.95, correct = FALSE)
```

## Result

X-squared = 1.6424, df = 1, p-value = 0.2

## Conclusion

At a 95% level of confidence there not enough evidence to support that the proportion of Grade 3&4 under Treatment A is not equal to the proportion of Grade 3&4 under Treatment B.

## Sensitivity\_CPS (Difference in proportion)

### R Code

$u_A = 130$

$u_B = 83$

$n_A = 150$

$n_B = 150$

`prop.test(c(uA,uB), c(nA,nB), alternative="two.sided", conf.level = 0.95, correct = FALSE)`

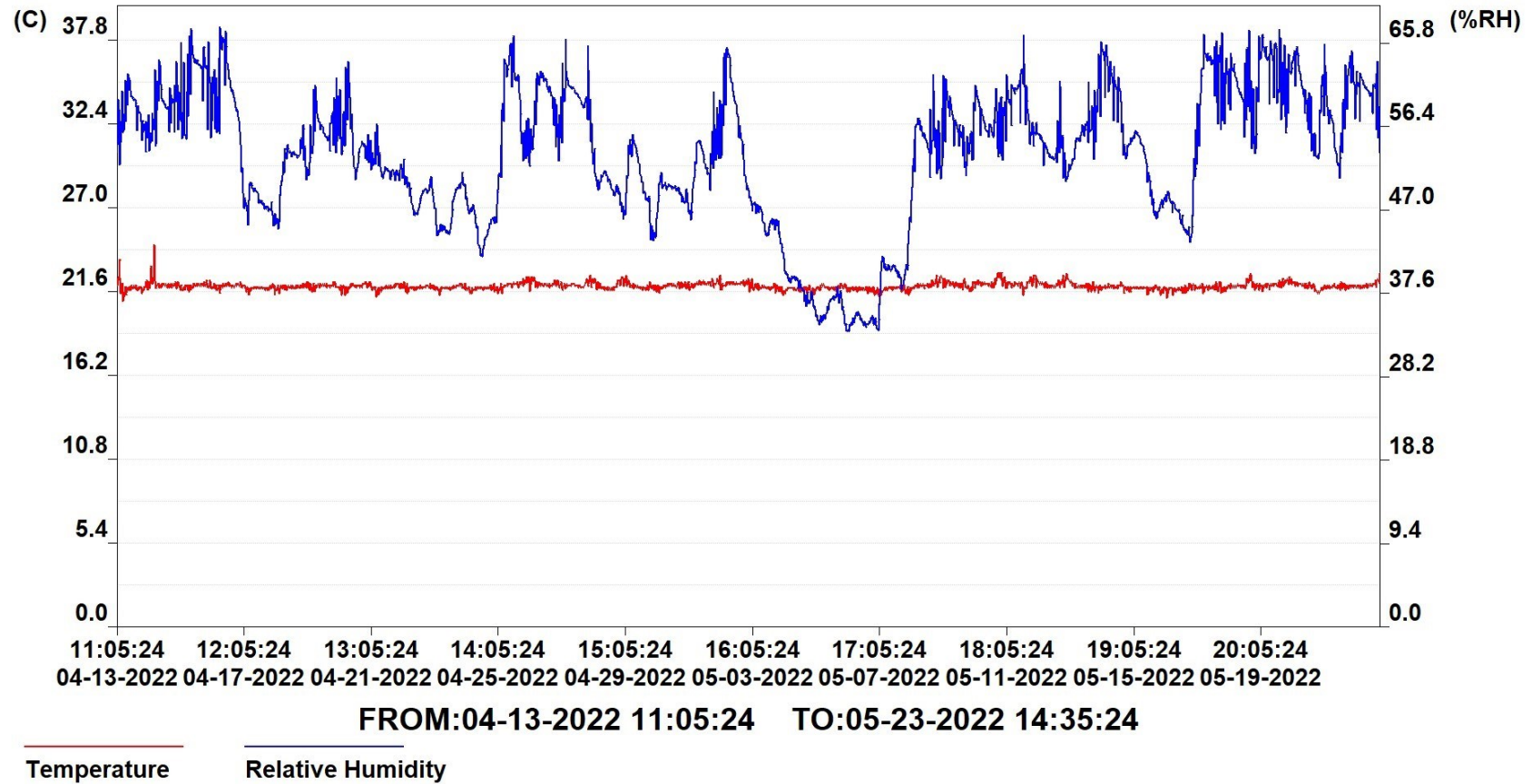
### **Result**

X-squared = 35.762, df = 1, p-value = 2.23e-09

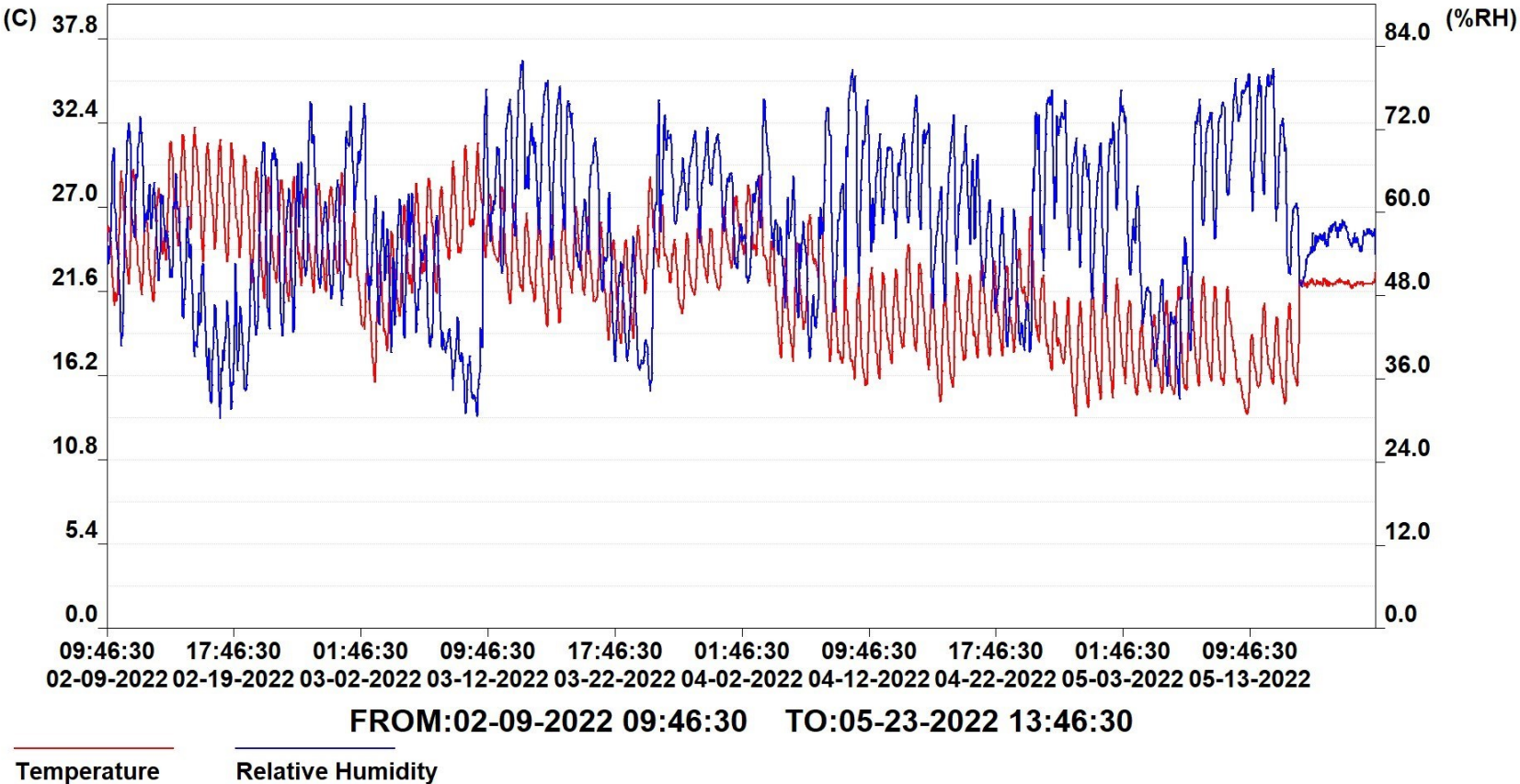
### **Conclusion**

At a 95% level of confidence, there is enough evidence to support that the proportion of Grade 3&4 under Treatment A is not equal to the proportion of Grade 3&4 under Treatment.

**Appendix B-1: Air-condition office datalogger temperature and relative humidity records.**



**Appendix B-2: Non-air-condition Level 5 space datalogger temperature and relative humidity records.**





## Appendix C-1: 3M Announcement letter (PFAS Discontinuation).

3M Electronics Materials Solutions Division

3M Center, Building 223-3S-32  
St. Paul, MN, USA 55144-1000



December 20, 2022

Dear Valued Customer,

3M today announced it plans to exit per- and polyfluoroalkyl substance (PFAS) manufacturing and work to discontinue use of PFAS in its product portfolio by the end of 2025. This portfolio decision is based on careful consideration and a thorough evaluation of the external landscape, including multiple factors such as accelerating regulatory trends focused on reducing or eliminating the presence of PFAS in the environment and changing stakeholder expectations.


This impacts products/product families you purchase from 3M listed in Appendix 1. Unfortunately, supply of most of these products continues to be impacted by global supply challenges and strong industry demand exceeding current capacity. Some of the products are also impacted by ongoing force majeure events. We will continue to take all reasonable steps to avoid supply disruptions, but we anticipate that our ability to supply will continue to be constrained.

3M will continue to fulfill contractual obligations during the transition period. Please note that regardless of any order acknowledgement, 3M may not be able to supply your orders due to continuing force majeure events and supply constraints, and we strongly urge customers to find alternative sources of supply.

3M will manage customer last time buy requests based on availability of supply, and we will work with customers to determine specific needs and put in place an action plan to help ensure an orderly transition. 3M does not currently anticipate stockpiling these products.

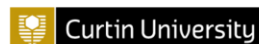
We understand you may have questions concerning this announcement. Please contact your 3M sales representative, who will work with you to the best of their ability to help support.

Sincerely,

  
David N. Schneider  
Vice President, Chemicals and Semiconductor  
3M Company, Electronics Materials Solutions Division

## Appendix D-1: Human ethics participant information form (English).

Investigation into improved protocols for latent fingerprint detection techniques for jurisdictions with limited resources and tropical climates



### PARTICIPANT INFORMATION STATEMENT

<b>HREC Project Number:</b>	<i>HRE2020-0162</i>
<b>Project Title:</b>	Investigation into improved protocols for latent fingerprint detection techniques for jurisdictions with limited resources and tropical climates
<b>Chief Investigator:</b>	Professor Simon Lewis
<b>Student researcher:</b>	Jemmy Bouzin
<b>Version Number:</b>	01
<b>Version Date:</b>	06/Apr/2020

#### **1. What is the Project About?**

Fingermarks are a valuable tool in criminal investigations. This research project will investigate improved protocols for fingerprint detection techniques and facilitate their use in operational laboratories, particularly in countries with limited resources and/or tropical climates. Increased scientific capabilities in fingerprint detection techniques have the potential to increase recovery rate of fingerprint evidence and improve their effectiveness in criminal investigations.

#### **2. Who is doing the Research?**

The project is being conducted by Jemmy Bouzin under the supervision of Professor Simon Lewis and is partially funded by the Government of Seychelles. The results of this research project will be used by Jemmy Bouzin to obtain a Doctor of Philosophy at Curtin University.

#### **3. Why am I being asked to take part and what will I have to do?**

This research depends upon collecting a number of sample latent fingerprints to analyse. We are looking for volunteers to allow the research team to collect a sample of your fingerprints for this research. For this research, we are asking you to manually handle and/or intentionally deposit latent fingerprints on a variety of objects. These latent fingerprints will be developed using chemical detection methods and digitally recorded using photography.

#### **4. Are there any benefits' to being in the research project?**

There is no direct benefit to you from participating in this research. However, by participating you will be assisting us to increase the recovery of fingerprint evidence and improve their effectiveness in criminal investigations to benefit the community in general.

#### **5. Are there any risks, side-effects, discomforts or inconveniences from participating?**

Apart from giving up a small portion of your time, we do not expect that there will be any risks or inconveniences associated with taking part in this study. You will handle different objects to deposit fingerprints, however there are no foreseeable physical risks.

**6. Who will have access to my information?**

The information collected in this research will be re-identifiable (coded). This means that we will collect data that can identify you, but will then remove identifying information on any data or sample and replace it with a code when we analyse the data. The data will not be used for automated verification or biometric identification. Only the research team have access to the code to match with your name, if it is necessary to do so. Any information we collect will be treated as confidential and used only in this project unless otherwise specified. The following people will have access to the information we collect in this research: the research team and, in the event of an audit or investigation, staff from the Curtin University Office of Research and Development. The information we collect in this study will be kept under secure conditions at Curtin University for 7 years after the research is published, before a decision is made as to whether it should be destroyed. The results of this research may be presented at conferences or published in professional journals. You will not be identified in any results that are published or presented. The research data may be used in future related projects.

**7. Will you tell me the results of the research?**

If you are interested in obtaining a summary of the results, please contact the researchers after February 2024. Results will not be individual but based on all the information we collect and review as part of the research.

**8. Do I have to take part in the research project?**

Taking part in this research project is entirely voluntary and you do not have to agree if you do not want to. You also have the right to withdraw from the project at any time without it affecting your relationship with the University, the funding body or any staff member. If you choose to withdraw from the project, any data collected from you will be destroyed. You will be informed in a timely manner if any information becomes available that may affect your consent to continue participation.

**9. What happens next and who can I contact about the research?**

If you decide to take part in this research, we will ask you to sign a consent form. By signing the consent form, you are telling us that you understand what you have read and what has been discussed. Signing the consent form indicates that you agree to participate in the research project. Please take your time and ask any questions you have before you decide what to do. You will be given a copy of this information and the consent form to keep.

**10. Further Information:**

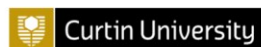
Curtin University Human Research Ethics Committee (HREC) has approved this study (HREC number 2020-0162). Should you wish to discuss the study with someone not directly involved; in particular, any matters concerning the conduct of the study or your rights as a participant, or you wish to make a confidential complaint, you may contact the Ethics Officer on (08) 9266 9223 or the Manager, Research Integrity on (08) 9266 7093 or email [hrec@curtin.edu.au](mailto:hrec@curtin.edu.au).

For further information about the study, please contact Professor Simon Lewis on (08) 92662484 or email [s.lewis@curtin.edu.au](mailto:s.lewis@curtin.edu.au) or Jemmy Bouzin at [j.bouzin@postgrad.curtin.edu.au](mailto:j.bouzin@postgrad.curtin.edu.au)

Thank you very much for your involvement in this research.

Your participation is greatly appreciated.

## Appendix D-2: Human ethics consent form (English).



Investigation into improved protocols for latent fingerprint detection techniques for jurisdictions with limited resources and tropical climates

### CONSENT FORM

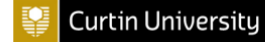
<b>HREC Project Number:</b>	<i>HRE2020-0162</i>
<b>Project Title:</b>	Investigation into improved protocols for latent fingerprint detection techniques for jurisdictions with limited resources and tropical climates
<b>Chief Investigator:</b>	Professor Simon Lewis
<b>Student researcher:</b>	Jemmy Bouzin
<b>Version Number:</b>	01
<b>Version Date:</b>	06/April/2020

1. I have read, the information statement version listed above and I understand its contents.
2. I believe I understand the purpose, extent and possible risks of my involvement in this project.
3. I voluntarily consent to take part in this research project.
4. I understand that I have the right to withdraw my participation in this research project at any time without it affecting my relationship with Curtin University or any staff member.
5. I understand that my data collected will not be used for automated verification or biometric identification.
6. I have had an opportunity to ask questions and I am satisfied with the answers I have received.
7. I give permission for my contribution to this research to be included in peer and non-peer reviewed journals and conferences, provided that I am not identified in any way.
8. I understand that my contribution to this research may be used in future related projects.
9. I understand that this project has been approved by Curtin University Human Research Ethics Committee and will be carried out in line with the National Statement on Ethical Conduct in Human Research (2007).
10. I understand I will receive a copy of this Information Statement and Consent Form.

Participant Name	
Participant Signature	
Date	

## Appendix D-3: Human ethics participant information form (Creole).

L'investigation dan amelyorasyon protokol teknik pou deteksyon lanprent-ledrwa pou bann zirediksyon avek resous limite e klima tropik



### DEKLARATION LENFORMASYON POU PARTISIPAN

<b>HREC Nimero Proze:</b>	HRE2020-0162
<b>Tit Proze:</b>	L'investigation dan amelyorasyon protokol teknik pou deteksyon lanprent-ledrwa pou ban zirediksyon avek resous limite e klima topik
<b>Sef Resers:</b>	Profeser Simon Lewis
<b>Etidyan ki per fer resers:</b>	Jemmy Bouzin
<b>Nimero Verzion</b>	01
<b>Dat Verzion</b>	06/Apr/2020

#### **1. Lo kwa sa resers i baze?**

Lanprent-ledrwa i en zouti enportan dan bann lenvestigation kriminel. Sa proze pou fer resers lo lenvestigation dan amelyorasyon bann protokol teknik pou deteksyon lanprent-ledrwa e fasilite zot l'itilizasyon dan laboratwar, sirtou pou bann pei avek resous limite ek klima tropik. Amelyorasyon dan teknik deteksyon lanprent-ledrwa i ogmant posibilite pou dekouver plis levidans lanprent-ledrwa e ogmant son lefikasite dan bann ka kriminel.

#### **2. Lekel ki pe fer sa Resers?**

Sa proze resers pe ganny fer par Jemmy Bouzin anba sipervizyon Professor Simon Lewis e ganny parselman finanse par Gouvernman Sesel. Rezilta sa proze resers pou ganny servi par Jemmy Bouzin pou obtenir en degree Doktora kot Liniversite Curtin.

#### **3. Akoz mon pe ganny demande pou pran par e ki mon pou bezwen fer?**

Sa resers i depann lo koleksyon en serten kantite lanprent-ledrwa pou analize. Nou pe rod volonter pou donn zot lanprent-ledrwa pou sa resers. Nou pou demann ou pou tous bann lobze pou depoz ou lanprent-ledrwa entonsyonelman ou non-entonsyonelman. Sa bann lanprent-ledrwa pou ganny devlope avek bann metod kemikal e apre ganny rikorde par fotografi.

#### **4. Eski i annan okenn benefis pou pran par dan sa resers?**

Napa okenn benefis direk dan pran par dan sa resers. Me si ou partisepe, ou pou ede amelyor teknik deteksyon e lefikasite levidans lanprent-ledrwa dan bann ka kriminel, e sa i anmenn gran benefis pou lasosyte aneneral.

#### **5. Eski l'annan okenn risk ou lenkonvenyans dan partisip dan sa resers?**

Otreman ki donn ou letan, nou pa vwar okenn risk ou lenkonvenyans dan pran par dan sa resers. Ou pou bezwen tous bann lobze pou depoz ou lanprent-ledrwa, me nou pa antisip okenn risk fizik.

#### **6. Lekel ki pou annan akse avek mon lenformasyon?**

Bann lenformasyon ki ganny kolekte dan sa resers pou ganny re-identifie. Sa l'vedir ki nou pou anmas lenformasyon ki kapab identifie ou, me nou pou ranplas sa lenformasyon avek en kod ler

nou pe analize. Ou lenformasyon pa pou ganny servi pou verifikasyon otomatik oubyen pou idantifikasyon. Zis bann endividi ki dan sa group resers ki pou konnen e annan akse avek ou lenformasyon ki korespond avek sa kod, e sa pou ganny akse zis dan ka kot i neserer. Tou lenformasyon ki nou kolekte pou konfidansyel e ganny servi zis dan sa proze, sof, si i ganny endike otreman. Sa bann dimoun swivan pou annan akse avek bann lenformasyon ki nou kolekte dan sa resers: tim resers e personel sorti kot lofis Resers e Delopman kot Liniversite Curtin dan ka en odit ou lenvestigasyon. Lenformasyon ki nou kolekte dan sa proze pou ganny garde an sekirite kot Liniversite Curtin pou 7 an apre ki sa resers i ganny piblibrye, e en desizyon pou ganny pran apre sa si sa bann lenformasyon pou ganny detri. Rezilta sa resers i kapab ganny prezante kot bann konferans ou piblibrye dan ban zournal profesyonel. Ou pa pou ganny idantifye dan okenn sa bann prezantasyon ou konferans. Sa bann lenformasyon resers i kapab ganny servi dan bann lezot proze relevan dan le fitir.

**7. Eski mon pou ganny enformen rezilta sa resers?**

Si ou entere pou konn rezilta sa resers, silvouple kontakte sa tim resers apre Fevriye 2024. Rezilta pa pou baze lo en endividi me lo tou lenformasyon kolektiv kin ganny anmase dan sa resers.

**8. Eski mon bezwen pran par dan sa resers?**

Pran par dan sa resers i volanter e ou pa bezwen agree pou partisipe si ou pa anvi. Ou annan drwa pou ou retir ou participation dan sa proze a okenn moman ki ou anvi san afekte ou relasyon avek Gouvernman Sesel, Liniversite Curtin ou okenn personnel sa bann lenstitisyon. Si ou retir ou partisipasyon, tou ou lenformasyon ki nou kolekte pou ganny detri. Ou pou ganny enformen o pli vit posib si okenn nouvo lenformasyon i afekte ou konsantman pou kontiyen partisip dan sa resers.

**9. Ki arrive apre e lekel ki mon kontakte pou lenformasyon lo sa resers?**

Si ou decide partisip dan sa resers nou pou demann ou pou sir en form konsantman. Par sir sa form konsantman i vedir ou konpran sa ki oun lir e as kin ganny diskite e ou endike ki ou agree pou partisip dan sa resers. Silvouple pran letan pou demann kekyon ki ou annan avan decide ki ou pou fer. Ou pou ganny donnen en kopi sa bann lenformasyon e form konsantman.

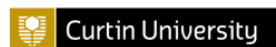
**10. Lezot Lenformasyon:**

Sa resers in ganny aprouve par Komite Etik pou Resers Imen kot Liniversite Curtin (HREC number 2020-0162). Si ou anvi diskite sa proze avek en endividi ki pa direktman enplike dan sa resers; partikilyerman okenn konsern ki annan pou fer avek la fason ki sa resers pe ganny fer , oubyen ou drwa koman en partisipan, oubyen si ou anvi fer en konplent konfidansyel, ou kapab kontakte en Zofisye Etik lo (+61 8 9266 9223) oubyen Manager, Entegrite Resers lo (+61 8 9266 7093) ou email [hrec@curtin.edu.au](mailto:hrec@curtin.edu.au). Pou plis lenformasyon lo sa resers, silvouple kontakte Professor Simon Lewis lo (+61 8 9266 2484) ou email [s.lewis@curtin.edu.au](mailto:s.lewis@curtin.edu.au) oubyen Jemmy Bouzin [j.bouzin@postgrad.curtin.edu.au](mailto:j.bouzin@postgrad.curtin.edu.au)

Mersi bokou pou ou lentere dan sa resers.

Ou partisipasyon i ganny vreman apresye.

## Appendix D-4: Human ethics consent form (Creole).



Lenvestigasyon dan amelyorasyon protokol teknik pou deteksyon lanprent-ledrwa pou bann zirediksyon avek resours limite e klima tropik

### FORM KONSANTMAN

<b>HREC Nimero Proze:</b>	<i>HRE2020-0162</i>
<b>Tit Proze:</b>	Lenvestigasyon dan amelyorasyon protokol teknik pou deteksyon lanprent-ledrwa pou bann zirediksyon avek resours limite e klima tropik
<b>Sef Resers:</b>	Profeser Simon Lewis
<b>Etidyan ki pe fer Resers:</b>	Jemmy Bouzin
<b>Nimero Verzion:</b>	01
<b>Dat Verzion:</b>	06/Apr/2020

1. Monn lir verzion deklarasyon lenformasyon parey i ganny liste anler e mon konpran son konteni.
2. Mon kwar ki mon konpran rezondet e posiblite bann risk dan pran par dan sa proze.
3. Mon volanterman donn mon konsantman pou pran par dan sa resers.
4. Mon konpran ki a okenn moman, mon annan drwa pou retir mon partisipasyon dan sa resers san afekte mon relationsyon avek Liniversite Curtin ou okenn manm.
5. Mon konpran ki mon lenformasyon ki ganny kolekte pa pou ganny servi pou fer verifikasyon otomatik ou identifikasyon.
6. Monn ganny loportinite pou demann kestyon, e mon satisfè avek larepons ki monn ganyen.
7. Mon donn permisyon pou mon kontribisyon dan sa resers ganny enkli dan bann publikasyon avek kondisyon ki mon pa ganny identifiye dan okenn fason.
8. Mon konpran ki mon kontribisyon dan sa resers i kapab ganny servi dan bann lezot proze relevan dan le fitir.
9. Mon konpran ki sa resers inn ganny aprouve par komite Resers lo Etik Imen kot Liniversite Curtin, e pou ganny fer an lir avek Declaration National lo "Conduite" Etik pou Resers Imen (2007).
10. Mon konpran ki mon pou resevwar en kopi Deklarasyon Lenformasyon e Form Konsantman.

Non Partisipan	
Sinyatir Partisipan	
Dat	

## **Appendix E-1: Contribution statement - New light on old fingerprints paper.**

To whom it may concern,

I, **Jemmy T. Bouzin**, contributed conceptualisation, investigation, methodology, visualization, writing-original draft, writing-review & editing to the following paper:

Jemmy T. Bouzin, Jason Merendino, Stephen M. Bleay, Georgina Sauzier, and Simon W. Lewis. New light on old fingerprints: The detection of historic latent fingerprints on old paper documents using 1,2-indanedione/zinc. *Forensic Science International: Reports* 2020, 2, 100145.

I as a Co-Author, endorse that this level of contribution by the candidate indicated above is appropriate.

**Jason Merendino**

**Stephen M. Bleay**

**Georgina Sauzier**

**Simon W. Lewis**



**Appendix E-2: Contribution statement – Forensic science in Seychelles paper.**

To whom it may concern,

I, **Jemmy T. Bouzin**, contributed conceptualisation, investigation, visualization, writing-original draft, writing-review & editing to the following paper:

Jemmy T. Bouzin, Georgina Sauzier, and Simon W. Lewis. Forensic science in Seychelles: An example of a micro-jurisdiction forensic delivery system. *Forensic Science International: Synergy* 2021, 3, 100139.

I as a Co-Author, endorse that this level of contribution by the candidate indicated above is appropriate.

**Georgina Sauzier**

**Simon W. Lewis**

**Appendix E-3: Contribution statement – IND/Zn Comparative assessment paper.**

To whom it may concern,

I, **Jemmy T. Bouzin**, contributed conceptualisation, investigation, methodology visualization, writing-original draft, writing-review & editing to the following paper:

Jemmy T. Bouzin, Aaron J. Horrocks, Georgina Sauzier, Stephen M. Bleay, and Simon W. Lewis. Comparison of three active 1,2-indanedione-zinc formulations for fingerprint detection in the context of limited resources and supply chain risks in Seychelles. *Forensic Chemistry* 2022, 30, 100439.

I as a Co-Author, endorse that this level of contribution by the candidate indicated above is appropriate.

**Aaron J. Horrocks**

**Georgina Sauzier**

**Stephen M. Bleay**

**Simon W. Lewis**

## **Appendix E-4: Contribution statement – Solstice® PF paper.**

To whom it may concern,

I, **Jemmy T. Bouzin**, contributed conceptualisation, investigation, methodology visualization, writing-original draft, writing-review & editing to the following paper:

Jemmy T. Bouzin, Amanda A. Frick, Georgina Sauzier, and Simon W. Lewis. Preliminary evaluation of Solstice® PF as a replacement carrier solvent for Australian fingerprint detection. *Forensic Science International* 2022, 340, 111465.

I as a Co-Author, endorse that this level of contribution by the candidate indicated above is appropriate.

**Amanda A. Frick**

**Georgina Sauzier**

**Simon W. Lewis**

## **Appendix E-5: Contribution statement – Mind the gap paper.**

To whom it may concern,

I, **Jemmy T. Bouzin**, contributed conceptualisation, investigation, visualization, writing-original draft, writing-review & editing to the following paper:

Jemmy T. Bouzin, Thais Lopes, Anna L. Heavey, Jessie. Parrish, Georgina Sauzier, and Simon W. Lewis. Mind the gap: The challenges of sustainable forensic science service provision. *Forensic Science International Synergy* 2023, 6, 100318.

I as a Co-Author, endorse that this level of contribution by the candidate indicated above is appropriate.

**Thais López**

**Anna Heavey**

**Jessie Parrish**

**Georgina Sauzier**

**Simon Lewis**

