Chapter 5: Alternative finger mark development techniques

5.1 Alternative blood enhancement techniques

1. History

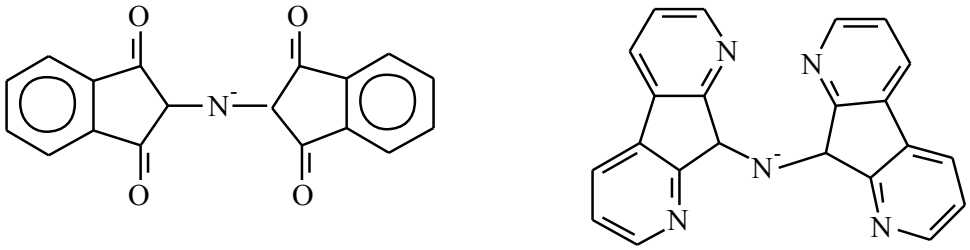
1.1 The history of the development of blood dyes has been outlined in Chapter 3.1, Acid dyes, of this *Fingerprint Source Book*.

2. Theory

2.1 General theory

2.1.1 The theory associated with the action of protein stains (in particular the acid dyes), in enhancing traces of blood is described in Chapter 3.1, Acid dyes (acid black 1, acid violet 17, acid yellow 7).

2.1.2 There are other reagents that react with the amines present in blood to give coloured or fluorescent products, the most well known of these being ninhydrin and 1,8-diazafluoren-9-one (DFO). They both react similarly with amino acids to form products that contain two deoxygenated molecules of the starting product bridged by a nitrogen atom, which is donated from the amine [1,2].



The reaction products with ninhydrin (left) and 1,8-diazafluoren-9-one (right) and amines.

2.1.3 While the reaction mechanisms and products have similarities, the method of their visualisation is entirely different. Ninhydrin, under the right conditions, produces an intensely coloured product called 'Ruhemann's purple' after the discoverer and DFO a pale pink, extremely fluorescent product. Ruhemann's purple can be made to fluoresce by complexing it with metal salts but this additional process is still not as sensitive as DFO [3]. DFO requires heat for the reaction to proceed [4] while ninhydrin will react at room temperature provided moisture is available, although the process proceeds much faster at elevated temperatures and humidities. These techniques are not specific to blood and will detect other amine-containing substances, including latent fingerprint deposits.

2.1.4 There are several ways of positively identifying blood using spectroscopic methods [5,6] but they are all carried out ex situ, so are of no use in the enhancement of blood-contaminated fingerprints.

2.1.5 Haemoglobin strongly absorbs light throughout the ultraviolet, visible and near infra-red parts of spectrum and this property can be utilised to detect and enhance blood, although once again this cannot be regarded as a way of confirming that it is blood that is present. Where deposits of blood are heavy or are present on light coloured surfaces a good white light may suffice to enable enough detail to be observed. However for pale or insubstantial deposits it may be necessary to use high-intensity light sources to enhance the contrast between the blood and the surface.

2.1.6 The use of fluorescence to enhance fingerprints in blood can be extremely effective in these circumstances. There are two ways this may be achieved:

- by exciting fluorescence of the background surface on which the blood is deposited;
- by treatment with a process that either breaks the haem group or turns the blood into a fluorescent species, or does both of these.

2.1.7 Many materials fluoresce when excited by high-intensity light in the ultraviolet and violet regions of the spectrum. This is coincidently where the haem group is most absorbent, with a peak around 421nm (known as the Soret Band) [5,7,8] and why blood-contaminated fingerprints will appear dark against a light background. Fluorescence examination may be used before any other fingerprint enhancement techniques as it is non-destructive and if long-wave ultraviolet or violet/blue light (350–450nm) [9] is used then DNA typing is also unaffected [10]. The use of ninhydrin, acid black 1 or acid violet 17 can further intensify the contrast between the fingerprint and the background by increasing the light absorption properties of the blood.

2.1.8 The use of a strong organic acid in conjunction with hydrogen peroxide [11,12] breaks up the haem group so that it is no longer effective at absorbing light. After such treatment, blood will fluoresce orange when excited by green light (500–550nm). This effect has also been noted as blood ages.

2.1.9 DFO produces fluorescent species with blood, which can be excited by green (510–570nm) light. This can be less effective on heavy deposits of blood as the haem group retains its ability to absorb both the excitation light and that emitted as fluorescence.

2.1.10 There are three kinds of tests for blood detection that use the haem group in haemoglobin: crystal tests; catalytic tests; and antibody tests. The sensitivity of these techniques is limited by their effectiveness to lyse blood cells, so releasing the haem-containing proteins that are only present within the red blood cells.

2.1.11 The Teichmann test [13] results in the formation of brown rhombohedral crystals of haematin and the Takayama test [14] in red-pink crystals of pyridine haemochromogen. Both of these tests have to

be carried out ex situ so are of no use for fingerprint enhancement as the ridge detail is inevitably destroyed as the blood is removed, unless an area containing no ridge detail, such as a smear, alongside the fingermark is used.

2.1.12 There are a number of advantages to the Takayama test, as compared with the Teichmann test. Heating is not required to obtain results within a reasonable amount of time in the Takayama test; and even if heat is applied, the test is not subject to being ruined by over-heating. The test also yields positive results under some of the circumstances where the Teichmann test fails.

2.1.13 The catalytic tests are only presumptive or infer the presence of haem, as they only use the haem to facilitate another reaction and are subject to both false positive and false negative reactions caused by a variety of non-blood substances. Consequently individual results require careful interpretation by experts.

2.1.14 These tests all rely on the 'peroxidase activity' of the haem group. Enzymes that catalyse the peroxide-mediated oxidation of organic compounds in vivo are called peroxidases; haemoglobin and the other compounds that show this catalytic property are thus said to have 'peroxidase activity'. This peroxidase activity may be utilised to cause the oxidation of colourless reduced dyes, such as phenolphthalein, leucocrystal violet, tetramethyl-benzidine and fluorescein, which when oxidised form their coloured, or in the case of the latter, fluorescent, counterparts.

$$H_2O_2 + \text{colourless reduced dye} \rightarrow H_2O + \text{coloured oxidised dye} \quad [15]$$

2.1.15 The luminol test also relies on the peroxidase activity of the haem group, but can be used with either hydrogen peroxide [16] or sodium perborate [17]. Then in the presence of blood a product which chemiluminesces is produced. The bluish-white chemiluminescence is faint and must be viewed in the dark by an operator who is fully dark-adapted to gain the best evidence from this test. However, even with careful application of luminol it is extremely easy to damage the fine detail of the blood-contaminated fingerprint ridges on both porous and non-porous surfaces. Therefore this technique should only be used when fine detail is not required and when other techniques might be compromised by surface type or impracticality, such as dark or patterned carpets [11].

2.1.16 The major concern with the catalytic tests for blood is that they can produce false-positive results in the presence of chemical oxidants and catalysts, salts of heavy metals such as copper, nickel and iron, and plant peroxidases such as those found in horseradish, citrus fruits, and numerous root vegetables [18]. A two-stage test can help to stop false positives from true peroxidases. The reduced colourless dye is applied initially and if no colour change is observed then the hydrogen peroxide

added. A colour change at this point is more likely to indicate the presence of blood rather than a peroxidase, although contamination by metal salts is not distinguished.

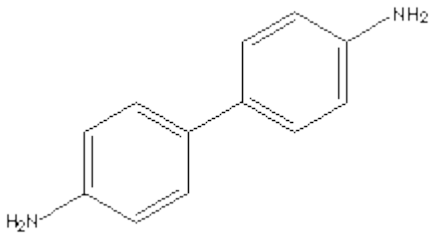
2.1.17 It is generally accepted that a negative result with a catalytic test proves the absence of blood, however strong reducing agents such as ascorbic acid [19] and active oxygen cleaning products [20] may inhibit such tests.

2.1.18 The antibody tests [21, 22] like the crystal tests are confirmatory for blood, but as they use anti-human Hb antibodies they are also specific for human blood. Currently (2011), they have to be used ex situ so are of no use for fingerprint enhancement, and it remains to be seen whether these tests can be used after the more effective enhancement techniques [22] to prove that what is being enhanced is human blood.

2.2 Specific reagents

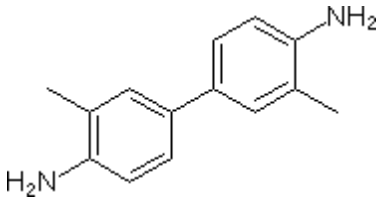
2.2.1 A review of blood enhancement agents has recently been conducted by Powell [23,24] and the relevant information below is extracted from these documents. Although the purpose of the review was for footwear enhancement, there is direct read-across to fingerprints because the contaminant being targeted is the same.

2.2.2 Benzidine: Benzidine was first used in 1904 and was the first reagent that utilised the peroxidase activity of haem. Benzidine is colourless in its reduced form and will turn dark blue when oxidised in the presence of haem or haem derivatives. It caused the entire surface being treated to be stained a light brown colour but was used on a variety of porous and non-porous surfaces. Due to its high sensitivity and dramatic colour change benzidine found widespread operational applications until health and safety concerns curtailed its use.



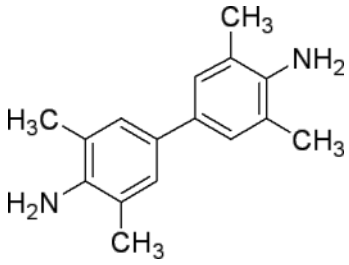
Structure of benzidine.

2.2.3 Ortho-tolidine: Ortho-tolidine is structurally related to benzidine, and is also colourless in its reduced form and dark blue when oxidised. It was first used in 1912 and again was widely employed due to its sensitivity and pronounced colour change. It was initially suggested as a possible alternative to benzidine. A sensitivity comparison of blood enhancement techniques rated ortho-tolidine second only to benzidine and suggested that it could be used providing that all health and safety precautions are taken.



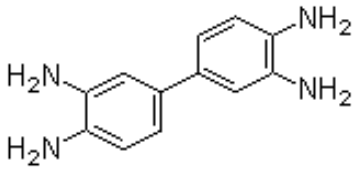
Structure of ortho-tolidine.

2.2.4 Tetramethyl-benzidine: As the commonly used reagents such as benzidine and ortho-tolidine were found to be carcinogenic thoughts were turned to find a new reagent of equal specificity but without the associated health and safety problems. There was some evidence that the issue was the participation of ortho-hydroxy derivatives of aromatic amines in the carcinogenic action, therefore the use of 3,5,3',5-tetramethylbenzidine (TMB) was suggested where ortho-hydroxylation is impossible. A print developed by TMB would turn green/blue.



Structure of 3,5,3',5- tetramethyl-benzidine).

2.2.5 Diaminobenzidine: Diaminobenzidine (DAB) undergoes a chemical polymerase reaction converting blood marks to an insoluble brown product. Its alternative name is tetraamino-biphenyl (TAB).



Structure of diaminobenzidine.

2.2.6 DAB is a derivative of benzidine and was thought to be a suitable substitute reagent for the enhancement of blood marks, as it is used as an aqueous solution and does not employ any organic solvents. The working solution is mixed just prior to use and involves the addition of a phosphate buffer solution to an aqueous solution of DAB. The reaction is initiated by hydrogen peroxide.

2.2.7 A widely used formulation is given below and involves the addition of a phosphate buffer working solution to the aqueous solution of DAB.

Solution A – fixing solution: Dissolve 20g 5-sulphosalicylic acid in 1 litre of distilled water.

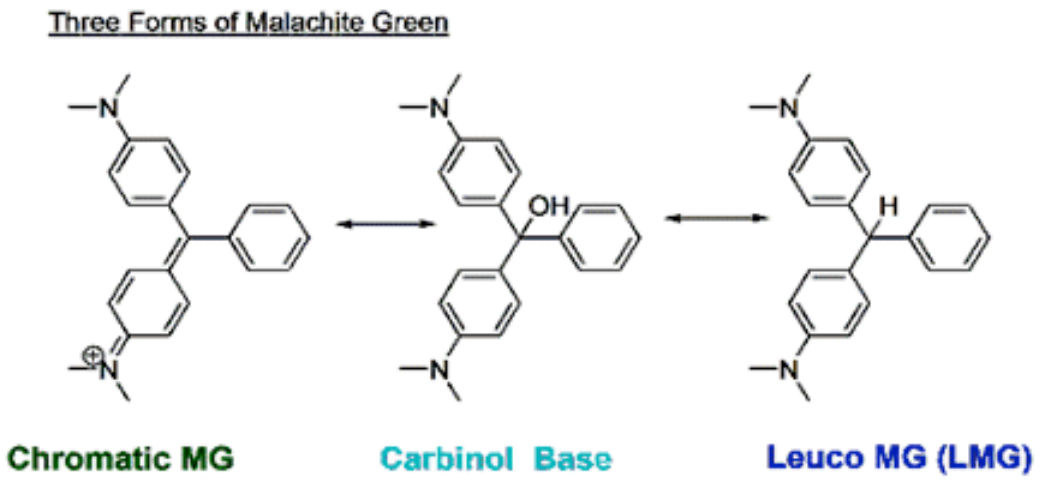
Solution B – buffer solution: Mix 100mL of 1M phosphate buffer (pH 7.4) with 100mLof distilled water.

Solution C – DAB: Dissolve 1g of 3,3'-diaminobenzidine tetrahydrochloride in 100mL of distilled water.

Working solution: Mix 180mL of solution B with 20mL of solution C and add 1mLof 30% hydrogen peroxide. The fixing solution is applied prior to the working solution.

2.2.8 Leuco-dyes: These are catalytic tests for blood and will bind with the proteins found in blood limiting the leaching and running of the developed impression. The hydrogen peroxide solutions will catalyse oxidation of the haemoglobin and its derivatives, producing a blue/green colour for leucomalachite green (LMG) and violet for leucocrystal violet (LCV).

2.2.9 Leucomalachite green: LMG is oxidised to form its coloured product when in contact with the haem group in blood.

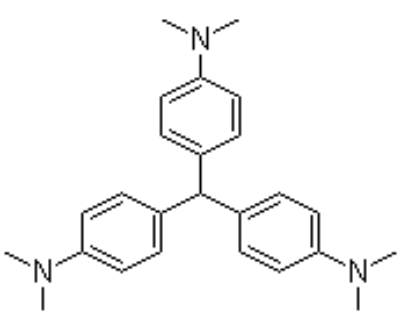


Changes occurring between the leuco- and coloured forms of malachite green

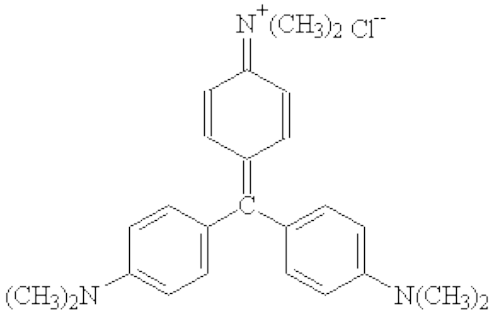
2.2.10 There are several formulations of LMG in the literature; they all contain LMG, diethyl ether, glacial acetic acid and hydrogen peroxide, the only difference being the quantity of each reagent. For optimum results the reagent must be prepared immediately prior to use. A green colour indicates that blood is present. The formulation given below is one used by the Royal Canadian Mounted Police.

Place 0.2g of LMG in a clean glass beaker, and add 67mL of methanol. Once the LMG is dissolved add 33mL of glacial acetic and 0.67g of sodium perborate and stir well until dissolved. Pour into a 1-litre beaker and add 300 ml of 1-methoxynonafluorobutane (HFE 7100). Store in a dark glass bottle until required. The prints can be fixed by submersion in ethanol.

2.2.11 Leucocrystal violet: LCV is the completely reduced form of crystal violet and is colourless. The reaction is initiated by hydrogen peroxide and when LCV comes into contact with the haem in blood the reaction is catalysed and the clear solution is converted to a purple/violet colour.



Crystal violet

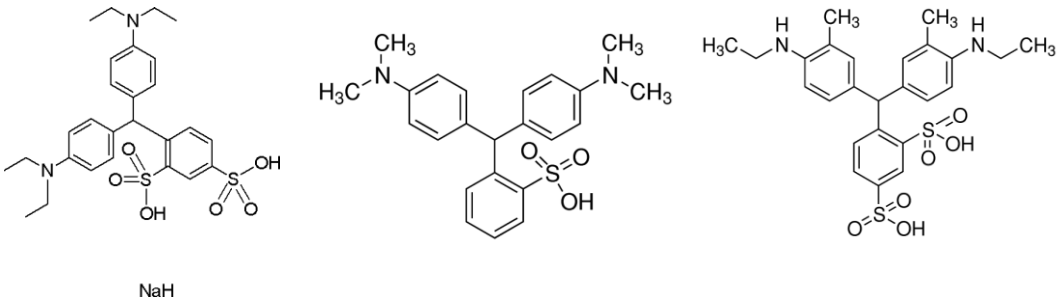


Leucocrystal violet

2.2.12 LCV is applied to the enhancement area via a spray method. The most common formulation is given below.

Dissolve 10g of 5-sulphosalicylic acid in 500mL of 3% hydrogen peroxide. Add and dissolve 3.7g sodium acetate. Add and dissolve 1g of leucocrystal violet with a magnetic stirrer. Store in dark-coloured glassware and refrigerate.

2.2.13 Alternative leuco dyes: Powell [24] studied a range of alternative leuco dyes to investigate whether issues with sensitivity and carcinogenicity of the existing leuco dyes could be overcome. The first alternative dye investigated was leuco patent blue (LPB). LPB is an acidic peroxidase dye compared with LCV, which is basic. As the fixing agent precipitates basic proteins, the acidic peroxidase reagent would then dye the basic proteins in a manner analogous to the protein stains. Two other similar systems, leuco berbelin blue (LBB) and leuco xylene cyanole (LXC) were also evaluated.



Structures of leuco patent blue, leuco berbelin blue , and leuco xylene cyanole.

2.2.14 Formulations for these reagents are given below.

0.1042g of leuco patent blue is dissolved in 10mL of water; 4mL of acetic acid and 1mL of 3% hydrogen peroxide are then added.

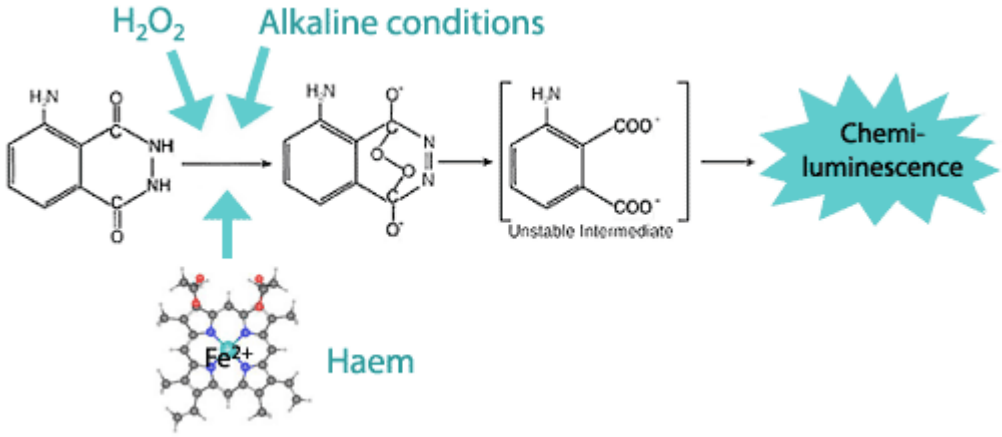
0.072g of leuco berbelin blue is dissolved in 10mL of water; 4mL of acetic acid and 1mL of 3% hydrogen peroxide are then added.

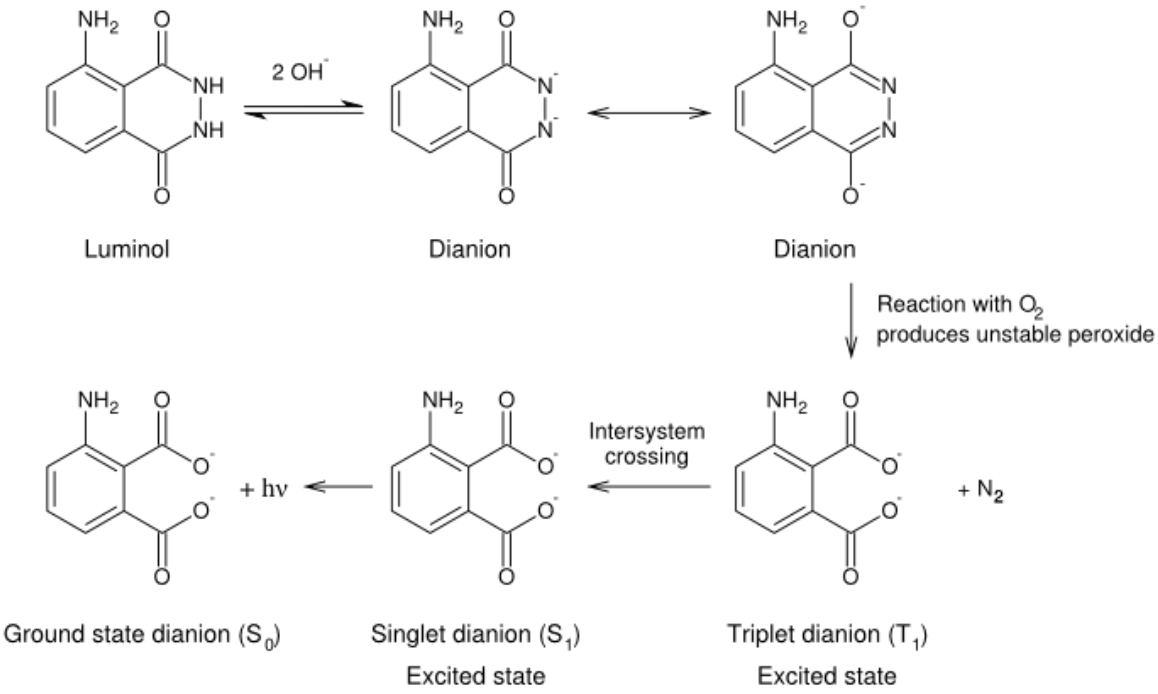
0.091g of leuco xylene cyanole is dissolved in 10mL of water; 10mL of acetic acid with 2mL of hydrogen peroxide are then being added.

2.2.15 Luminol: The active chemicals in this generic class of blood detection reagents are luminol ($C_8H_7O_3N_3$) and hydrogen peroxide (H_2O_2). The hydrogen peroxide and the luminol react in alkaline conditions to produce chemiluminescence (in this case a blue/white glow), with the reaction being catalysed by the iron present in haemoglobin.

2.2.16 In the resultant oxidation reaction, the luminol molecule loses nitrogen and hydrogen atoms and gains oxygen atoms, resulting in a compound called 3-aminophthalate. The reaction leaves the 3-aminophthalate in an excited state with the electrons in the oxygen atoms being promoted to

higher energy levels. The electrons quickly fall back to a lower energy level, emitting the extra energy as a light photon.





Schematic diagrams showing the mechanisms associated with the chemiluminescent reaction between luminol and blood.

2.2.17 This then produces a product that luminesces in the presence of blood. The bluish-white chemiluminescence is faint and must be viewed in the dark by an operator who is fully dark-adapted to gain the best evidence from this test. Even with careful application of luminol it is all too easy to damage the fine detail of blood-contaminated fingerprints. This technique should only be used when fine detail is not required and when other techniques might be compromised by surface type or impracticality, such as dark or patterned carpets. Two published formulations for luminol are

given below, and proprietary pre-prepared products (e.g. Bluestar) are also available.

Grodsky:

3.5g sodium perborate is dissolved in 500mL distilled water, 0.5g luminol and 25g sodium carbonate are added and dissolved. Solution is left to stand for five minutes before being used immediately.

Weber:

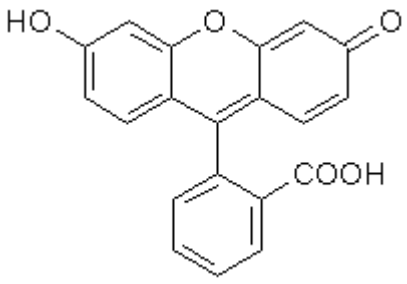
Stock solution A: 8g sodium hydroxide dissolved in 500mL distilled water.

Stock solution B: 10mL 30% hydrogen peroxide in 490mL distilled water.

Stock solution C: 0.354g luminol dissolved in 62.5mL of solution A and made up to final volume of 500mL with water.

Working solution: 10mL solution A + 10mL of solution B + 10mL of solution C + 70mL distilled water.

2.2.18 Fluorescein: Fluorescein is a presumptive test for blood that utilises the peroxidase activity of the haem group. The reduced form of the chemical, fluorescin, is colourless and when sprayed onto the target area it is oxidised to fluroscein, a coloured/fluorescent product, by the presence of blood associated proteins and iron ions found in the haemoglobin molecule. Even minute traces will fluoresce when excited with a light source between 425–485nm and viewed through a yellow to orange barrier filter.



Structure of fluorescein.

2.2.19 Fluorescein is usually applied in a two-step process – the application of fluorescein alone will develop the yellow coloration, however an overspray of hydrogen peroxide is also used to reduce background fluorescence and false-positive reactions.

2.2.20 The preparation of fluorescein is quite a lengthy process and the reduced fluorescin has a very short shelf life – the recommended usage is within 24 hours. The original formulation is as follows.

A 10% sodium hydroxide (NaOH) stock solution is prepared by dissolving 10g NaOH in 100mL deionised water.

1.0g fluorescein is dissolved in 100mLof the 10% NaOH stock solution and placed on a hot plate and heated gently.

10.0g zinc powder is then added and heated to a gentle boil.

The solution is allowed to cool and the un-dissolved zinc to settle.

The cooled solution is then decanted to remove any un-dissolved zinc.

The fluorescein reagent solution is then made by mixing 50mL of the decanted solution with 950mL of deionised water. This reagent must then be kept in dark glassware.

The hydrogen peroxide overspray is made by mixing 100mL of 30% hydrogen peroxide with 200mL deionised water.

3. Reasons technique is not recommended by CAST

3.1 The Home Office Centre for Applied Science and Technology (CAST) does not recommend the use of haem-specific, reactive blood dyes for general use because they are not as sensitive as the protein stains recommended in the *Manual of Fingerprint Development Techniques* [10]. This is intuitive – there is far more proteinaceous material present to interact with the dye than there is haem and therefore the protein stains will remain effective on far smaller quantities of blood residue than reactive dyes. This is supported by the sensitivity testing conducted by Sears *et al.* [11] when developing the formulations for acid black 1, acid yellow 7 and acid violet 17.

3.2 It is recognised that there will be circumstances where the use of haem-specific dyes will be preferable, e.g. where there is other proteinaceous contamination present and a more specific dye will more clearly identify the blood. Reactive dyes are also more suited to speculative searching of scenes, and can be more easily spray applied. However, this approach is more suited to footwear development than to fingerprints. A range of the alternative blood enhancement agents (protein stains and reactive dyes) is outlined below, with some comments on those most commonly proposed for operational use.

3.3 Benzidine: Benzidine was found to be a highly effective blood-enhancing reagent but was later recognised as a known carcinogen and there are reports in the literature stating forensic analysts developed bladder

cancer due to the use of this reagent. It is now known to be extremely hazardous and breathing its vapours or touching the chemical or its salts could cause cancer to develop. It is not recommended for use by CAST and is included in this review for historical purposes only

3.4 Ortho-tolidine: Although ortho-tolidine was originally proposed as a safer alternative to benzidine, there are several reports in literature stating that workers suffered from prolonged headaches and skin burns after using ortho-tolidine when safety precautions were not taken. Ortho-tolidine is now also a known carcinogen and its use is therefore not recommended by CAST.

3.5 3,5,3',5 Tetramethylbenzidine (TMB): Sensitivity studies carried out in comparison with acid black 1 show TMB to be significantly less sensitive. There are also concerns about TMB being a possible carcinogen and mutagen and its use is therefore not recommended by CAST.

3.6 3,3' Diaminobenzidine tetra hydrochloride dehydrate (DAB): Sensitivity studies carried out in comparison with acid black 1 show DAB to be significantly less sensitive. The colour formed during the reaction is light brown, which is similar to dried blood and not ideal for enhancement of bloody fingerprints, whereas a protein dye such as acid black 1 will stain the mark a dark colour, which will aid with contrast against the background. There are also reports on the suspected carcinogenic activity of DAB, and therefore it is not recommended for use by CAST.

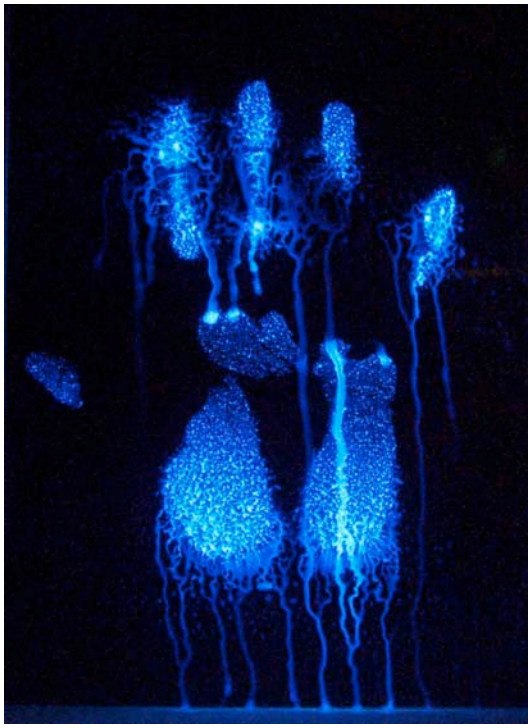
3.7 Leucomalachite green: LMG has been found to be less sensitive than acid black 1 and does not produce as vivid a colour change as some other reagents studied. It was also found to be less consistent in performance than LCV.

3.8 Leucocrystal violet: LCV has been shown to be an effective treatment for marks in blood, albeit less sensitive than protein stains. If a haem-specific reagent were to be recommended by CAST, LCV would be the preferred option but only under controlled conditions in a laboratory. The purple coloured form crystal violet is now classified a known carcinogen which makes large scale spraying at scenes undesirable.

3.9 Alternative leuco dyes: Of the alternative leuco dyes evaluated, LBB gave high background staining and although LPB and LXC were effective in preliminary studies, the cost of the dyes is prohibitive for operational use.

3.10 Luminol: Luminol and related compounds are not recommended for fingerprint detection because they are spray applied and could cause diffusion of marks. Because luminol relies on a chemiluminescent reaction to produce blue fluorescence that fades with time, multiple applications may be required to first locate and then photograph any fingerprints. However, it has been demonstrated that repeat applications

will ultimately cause diffusion of ridge detail and therefore the use of a reagent giving a coloured or conventionally fluorescent mark is preferred.



Palm print in blood on glass, with ridge detail diffused by excessive spraying.

3.11 Fluorescein: Fluorescein has been found to be lower in sensitivity to most of the other dyes outlined here and the acid dyes recommended in the *Manual of Fingerprint Development Techniques* [10].

3.12 Alternative protein stains: In addition to reactive dyes, CAST has considered a wide range of alternative protein stains that were evaluated in comparative studies with acid black 1, acid yellow 7 and acid violet 17 [25,26]. These dyes were rejected on the basis of lack of sensitivity, lack of availability or poor visibility of the developed mark. A summary of those systems evaluated is given in the table below.

Colour Index name	**Colour Index number**	**Comments**
Acid blue 92	13390	Plasma stain
Acid red 88	15620	Plasma stain
Acid red 29	16570	
Acid red 1	18050	Plasma stain
Acid yellow 23	19140	Collagen stain (protein)
Direct yellow 12	24895	Plasma stain (in pathology)
Acid red 71	27165	Cytoplasmic stain
Acid red 112	27195	Stain basic tissue elements

Acid blue 1/acid blue 3	42045/42051	
Basic violet 4	42600	
Acid blue 90	42655	Protein stain
Acid blue 83	42660	Protein stain
Acid violet 19	42685	Plasma stain
Acid dye	43535	
Basic blue 11	44040	
Acid red 87	45380	Plasma stain
Basic dye	51140, 51145	
Direct red 148	52005	
Acid blue 74	73015	
Quinacrine	-	
Lucifer Yellow (CH & VS)	-	
Rivanol	-	

Alternative protein stains evaluated by CAST but not recommended for operational use.

4. References

1. **McCaldin, D. J.** (1960) 'The Chemistry of Ninhydrin', *Chem. Rev.*, vol. 60, p 39.

2. **Wilkinson, D.** (2000) 'Study of the Reaction Mechanism of 1,8-Diazafluoren-9-one with the Amino-Acid L-Alanine', *Forens. Sci. Int.*, vol. 109, p 87.

3. **Stoilovic, M.** (1993) 'Improved Method for DFO Development of Latent Fingerprints', *Forens. Sci. Int.*, vol. 60, p 141.

4. **Hardwick, S., Kent, T., Sears, V. and Winfield, P.** (1993) 'Improvements to the formulation of DFO and the Effects of Heat on the Reaction with Latent Fingerprints', *Fingerprint Whorld*, vol. 19 (73), p 65.

5. **Soret, J. L.** (1883) 'Anallyse Spectrale: Sur le Spectre d'Absorption du Sang Dans la Partie violette et Ultra-Violette', *Comptes Rendus de l'Académie des Sciences*, vol. 97, p 1269.

6. **Sorby, H.C.** (1865) 'On the Application of Spectrum Analysis to Microscopical Investigations, and Especially to the Detection of Bloodstains', *Q. J. Sci.*, 2, p 198.

7. **Kotowski, T. M. and Grieve, M. C.** (1986) 'The Use of Microspectrophotometry to Characterize Microscopic Amounts of Blood', *J. Forens. Sci.*, vol. 31 (3), p 1079.

8. **Stoilovic, M.** (1991) 'Detection of Semen and Blood Stains Using Polilight as a Light Source', *Forens. Sci. Int.*, vol. 51, p 289.

9. **Hardwick, S. A., Kent, T. and Sears, V.G.** (1990) *Fingerprint Detection by Fluorescence Examination; A Guide to Operational Implementation*, ISBN 0 86252 554 3. London: Home Office.

10. **Bowman, V. (Ed)** (1998, revised 2002, 2004, 2009) *Manual of Fingerprint Development Techniques*, 2nd edition, ISBN 1 85893 972 0. London: Home Office.

11. **Sears, V., Butcher, C. and Fitzgerald, L. (**2005) 'Enhancement of Fingerprints in Blood – Part 3 Reactive Techniques, Acid Yellow 7 and Process Sequences', *J. Forens. Ident*., vol. 55 (6), p 741.

12. **Fischer, J. F. and Miller, W. G. (**1984) 'The Enhancement of Blood Prints by Chemical Methods and Laser Induced Fluorescence', *Ident. News*, vol. 34 (7), p 2.

13. **Teichmann, L.** (1853) 'Ueber die Krystallisation des Orpnischen Bestandtheile des Blutes', *Z. Ration. Med*., vol. 3, p 375.

14. **Takayama, M.** (1912) 'A Method for Identifying Blood by Hemochromogen Crystallization', *Kokka Igakkai Zasshi*, vol. 306, p 463.

15. **Seigel, J. (ed-in-chief)** (2000) *Encyclopedia of Forensic Sciences*, ISBN 0-12-227215-3, p 1333. London: Academic Press.

16. **Weber, K.** (1966) 'Die Anwendung der Chemiluminescenz des Luminols in der Gerichtlichen Medizin und Toxikologie', *Deutsche Zeitschrift für Gerichtliche Medizin*, vol. 57, p 10.

17. **Grodsky, M., Wright, K. and Kirk, P.** (1951) 'Simplified Preliminary Blood Testing', *J. Crim. L. Criminol. & Police Sci*., vol. 42, p 95.

18. **Seigel, J. (ed-in-chief)** (2000) *Encyclopedia of Forensic Sciences*, ISBN 0-12-227215-3, p 1334. London: Academic Press.

19. **Eckert, W. G. and James, S. H.** (1989) 'Interpretation of Bloodstain Evidence at Crime Scenes', p 121. New York: Elsevier Science Publishing.

20. **Castelló, A., Francés, F., Corella, D. and Verdú, F.** (2009) 'Active oxygen doctors the evidence', *Die Naturwissenschaften*, vol. 96 (2), p 303.

21. **Hochmeister, M. N., Budowle, B., Sparkes, R., Rudin, O., Gehrig, C., Thali, M., Schmidt, L., Cordier, A. and Dirnhofer, R.** (1999) 'Validation Studies of an Immunochromatographic 1-Step Test for the Forensic Identification of Human Blood', *J. Forens. Sci.,* vol. 44 (3), p 597.

22. **Johnston, E., Ames, C. E., Dagnell, K. E., Foster, J. and Daniel, B. E.** (2008) 'Comparison of Presumptive Blood Test Kits including Hexagon OBTI', *J. Forens. Sci.,* vol. 53 (3), p 687.

23. **Powell, L.** (2008) 'Enhancement of footwear marks made in blood', literature review submitted in part fulfilment of MSci degree, June. University of Strathclyde.

24. **Powell, L.** (2008) *Enhancing footwear marks in blood*, HOSDB Student Placement Report, July.

25. **Prizeman, T.** (1998) *Enhancement of Fingerprints in Blood – Optimization of Amido Black Formulations*, PSDB Student Placement Report.

26. **Mutch, M.** (1996) *Evaluation of Reagents for Enhancing Fingerprints in Blood on Porous Surfaces,* PSDB Student Placement Report,

5.2 4-Dimethylaminocinnamaldehyde (DMAC)

1. History

1.1 4-Dimethylaminocinnamaldehyde (DMAC) was first proposed as a fingerprint development reagent in the UK by Morris *et al.* in 1973 [1] and was believed to react with the urea present in eccrine fingerprint secretions. In the initial work conducted at AWRE, DMAC appeared to be more sensitive than the ninhydrin formulations and processing conditions then in use, and it was decided to proceed to operational trials in 1973. For operational use DMAC was dissolved in a mixed ethanol/ chlorofluorocarbon (CFC) solvent and the articles to be treated immersed in the solution until visible marks developed. When DMAC reacts with urea under acidic conditions it gives a magenta coloured product within two minutes, the developed mark providing good contrast with the background.

Palm print developed using 4-dimethylaminocinnamaldehyde solution.

1.2 The operational trials in the UK were conducted in a limited number of police forces and abandoned after only a few months as the performance of DMAC was found to be poor in terms of finger mark yield compared to ninhydrin. Many of the marks that were developed were also diffuse and lacking in ridge detail. As a consequence the use of DMAC as a solution dipping process was discontinued in the UK by the mid 1970s.

1.3 Van Enckevort [2] found DMAC, when sprayed or dipped, to be useful on a wide range of substrates in laboratory trials, particularly those that showed a high background development with ninhydrin. However, he too

found the reagent to be less successful in operational trials with the prints visualised showing blurred ridge detail, which was attributed to the diffusion of urea. He observed that useful prints were only obtained up to three to ten days after deposition and consequently found little use for the reagent.

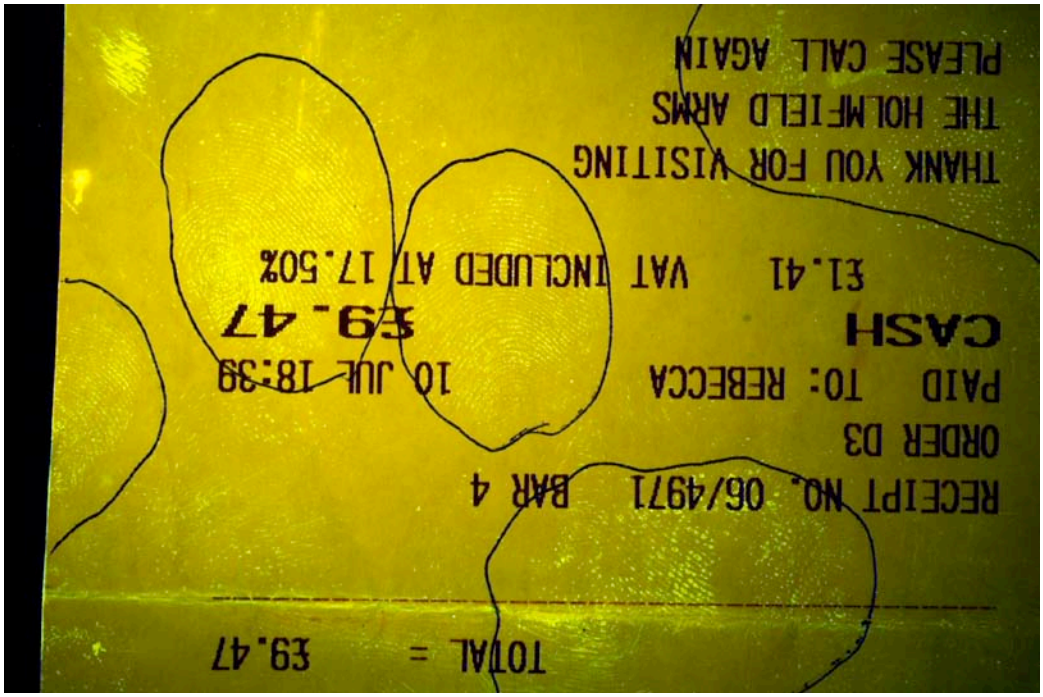
1.4 DMAC was later investigated as a fuming agent and was found by Brennan *et al.* [3] to give good ridge detail visualisation on a wide selection of substrates, with potential to be included in routine sequential examination procedures. Katzung [4] reported that prints developed using DMAC fuming showed yellow fluorescence under excitation using 360nm light sources and that he had managed to detect four-week-old prints using this method.

1.5 Although vapour phase fuming can offer an answer to problems associated with solvent based fingerprint techniques, some researchers have described the limitations and scope of the reagent's ability to produce visible prints. Sasson and Almog [5] concluded that although ninhydrin was a more general and versatile reagent, DMAC was preferable to ninhydrin on fresh prints (up to 72 hours old) in situations where the application of heat is not possible. Brennan [6] reported that for cases involving porous items other than thermal papers, all the prints developed by DMAC were subsequently developed by 1,8-diazafluoren-9-one (DFO), ninhydrin or physical developer and concluded that DMAC fuming was less effective than existing processes on such articles. On thermal papers, however, prints were developed on the thermal surface that would otherwise have been lost using other methods. This study was further reported by the Metropolitan Police Serious Crimes Unit [7] which emphasised the potential of vapour phase fuming with DMAC and subsequent visualisation of the fluorescence using a laser as a powerful non-destructive technique that does not interfere with following sequential treatments. It was regarded as having particular potential for detecting marks on thermal papers.

1.6 In the mid 1990s, the use of DMAC as a 'contact transfer' development process was proposed by Ramotowski [8] for development of fingerprints on paper. This approach involves pressing an exhibit between two sheets of paper that have been soaked with DMAC solution and subsequently dried, resulting in a pale yellow colouration to the paper and barely visible prints that give yellow fluorescence when illuminated with green light.

1.7 Experiments have also been carried out to investigate the use of the contact transfer process on the polymer banknotes used in Australia , looking at different temperatures and exposure times. Results indicated that contact transfer at room temperature was not particularly successful, with results demonstrating poor contrast between the notes and prints treated up to four hours. They also found that heat contact transfer at various temperatures using an ironing press for 20 seconds developed high background luminescence and the contrast between the developed

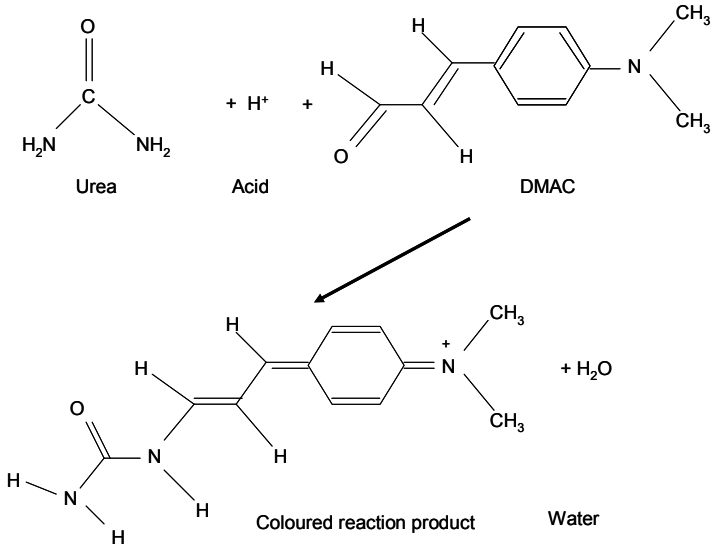
fingerprint and background was very low. The contact transfer technique has since been proposed for development of fingerprints on thermal papers with the stated advantages that it leaves the printed text intact and does not cause the thermal receipt to blacken during processing.



Fingerprints developed using contact transfer 4-dimethylaminocinnamaldehyde process.

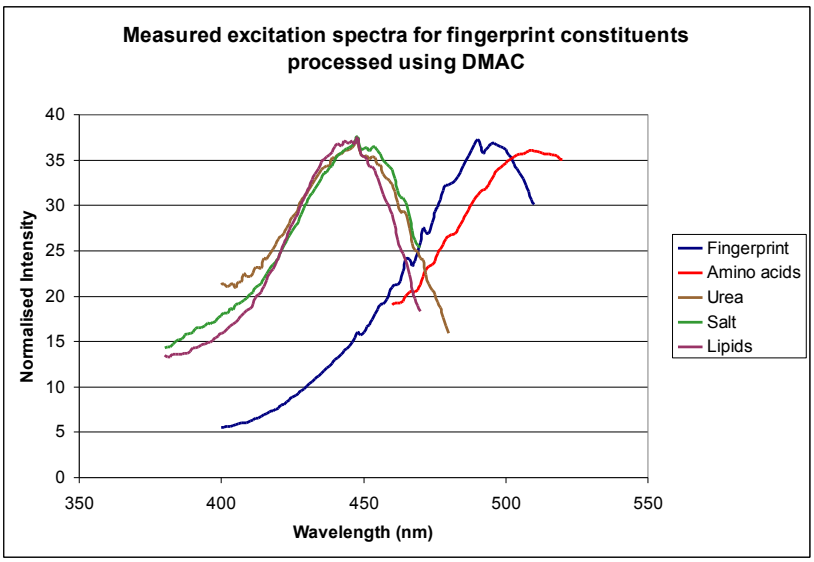
2. Theory

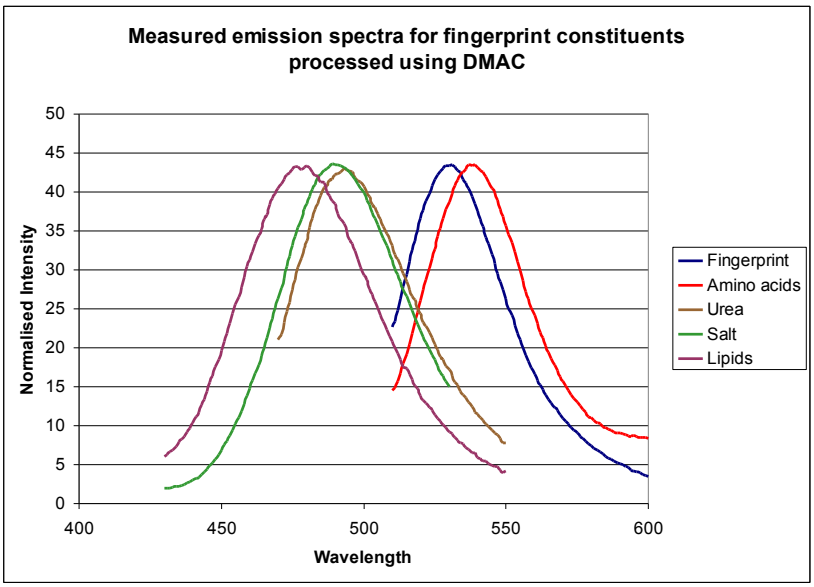
2.1 The reaction mechanism for the original solution treatment form of DMAC was the formation of a coloured Schiff base by the reaction between DMAC and urea under acidic conditions.



Proposed mechanism for formation of coloured product from reaction between 4-dimethylaminocinnamaldehyde and urea under acid conditions.

2.2 The precise mechanism by which fluorescence occurs in the contact transfer process is not known, but spectroscopy has been carried out by the Home Office Centre for Applied Science and Technology (CAST), which indicates that when used as a contact transfer process DMAC interacts with amino acid constituents in the fingerprint rather than urea. The nature of the fluorescent reaction products has not been determined.





Excitation and emission spectra obtained for filter paper pad impregnated with fingerprint deposits and model fingerprint constituents, then treated with the 4-dimethylaminocinnamaldehyde contact transfer process.

a) b)

Reaction products formed between 4- dimethylaminocinnamaldehyde and 0.1M solutions of amino acids and other fingerprint constituents a) visible and b) fluorescence.

2.3 The formulation originally used for solution dipping was a two-part system made up as follows.

Solution A: mix 650mL of 1,1,2-trifluorotrichloroethane (CFC113) with 350mL of absolute ethanol. Take 750mL of the mixed solvent, add 5g of DMAC and stir until dissolved, then make up to 1 litre with remainder of solvent, filter and store in a brown bottle.

Solution B: mix 650mL of CFC113 with 350mL of absolute ethanol. Add 20g of 5-sulphosalicylic acid and stir until dissolved.

2.4 A working solution is made by mixing together equal proportions of solutions A and B, and articles are then dipped. Spray application is possible, but in this case the surface to be treated is first sprayed with solution A, followed by a second spray of solution B.

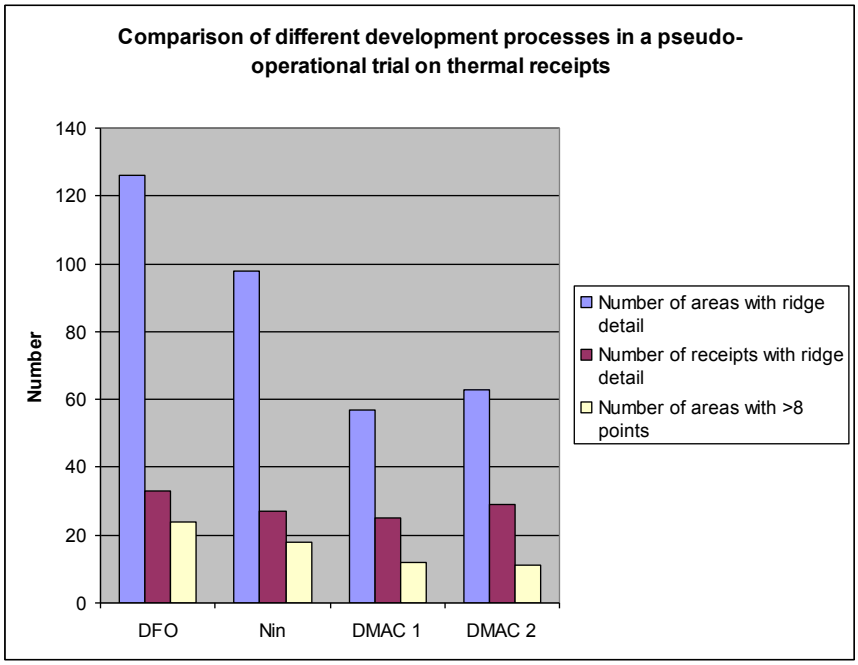
2.5 The contact transfer process utilises sheets of paper immersed in a solution of 0.25g of DMAC dissolved in 100mL of ethanol. The sheets are then allowed to dry. The article to be treated is sandwiched between two sheets of impregnated paper, placed in a press and left overnight.

3. Reasons technique is not recommended by CAST

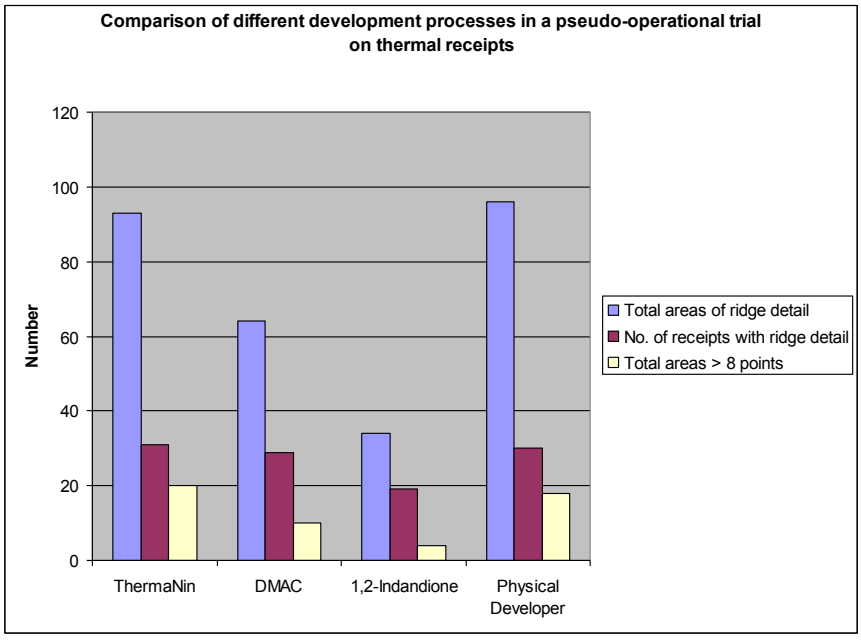
3.1 DMAC is not recommended by CAST in either solution dipping or contact transfer form. Operational experience in the 1970s demonstrated that the solution dipping process was not suitable for marks more than a few days old because of the rapid diffusion of the urea constituent. The solution dipping formulation is based on CFCs and would not be acceptable for use without reformulation to a less ozone-depleting solvent.

3.2 More recently, CAST conducted experiments to compare the effectiveness of DMAC against DFO and ninhydrin for cases where it is not necessary to retain printed text on thermal receipts. A further

comparison was conducted with ThermaNin, 1,2 indandione and physical developer for cases where it is necessary to retain printed text on thermal receipts. In both these cases, pseudo-operational trials confirmed laboratory experiments, and in neither case was DMAC found to be as effective as processes currently recommended by CAST [10,11,12].



a)



b)

Results of pseudo-operational trials conducted on batches of thermal receipts comparing the effectiveness of the contact transfer 4-dimethylaminocinnamaldehyde process with a) techniques removing printed text and b) techniques leaving printed text visible.

4. References

1. **Morris, J. R., Goode, G. C. and Godsell, J. W.** (1973) 'Some new developments in the chemical development of latent fingerprints', *Pol. Resid. Bull.*, AWRE and PSDB, 21 p 31. London: Home Office.

2. **Van Enckevort, H. J.** (1987) 'The detection and visualisation of latent fingerprints: a review', *Report No. CD 2380, Chemistry Division*. New Zealand: Department of Scientific and Industrial Research.

3. **Brennan, J., Bramble, S., Crabtree, S. and Wright, G.** (1995) 'Fuming of latent fingerprints using dimethylaminocinnamaldehyde', *J. Forens. Ident*., 45, pp 373–380.

4. **Katzung, W**. (1985) 'New reagents for the chemical development of latent fingerprints on paper and their possible applications', *Krim. und Forens. Wissenschaften*, No. 57, 58, pp 82–89.

5. **Sasson, Y. and Almog, J.** (1978) 'Chemical reagents for the development of latent fingerprints 1: Scope and limitations of the reagent 4-dimethylaminocinnamaldehyde', *J. Forens. Sci.* vol. 23, p 852.

6. **Brennan, J. S.** (1995) 'The development of fingerprints by fuming with dimethylaminocinnamaldehyde (DMAC)', *Proceedings of the International Symposium on Fingerprint Detection and Identification*, , June 1995, pp 85–90. Israel: Ne'urim.

7. **Metropolitan Police Serious Crimes Unit** (1995) 'Turning a concept into a successful aid to major crime scene management', *11th International ICPO–Interpol Forensic Science Symposium, Talking Paper*, 1995. Metropolitan Police Forensic Science Laboratory.

8. **Ramotowski, R.** (1995) 'Fluorescence Visualisation of Latent Fingerprints on Paper Using p-Dimethylaminocinnamaldehyde (PDMAC)', *Proceedings Of the International Symposium on Fingerprint Detection and Identification*, June 1995, pp 91–94. Israel: Ne'urim.

9. **Flynn, J., Stoilovic, M. and Lennard, C.** (1999) 'Detection and enhancement of latent fingerprints on polymer banknotes: a preliminary study', *J. Forens. Ident*., vol. 49, pp 594–613.

10. **HOSDB** (2006) *Fingerprint and Footwear Forensics Newsletter*, HOSDB Publication No. 58/06, October. London: Home Office.

11. **Lee, J.** (2007) *Evaluation of ThermaNin*, BSc Final Year Project Report. University of Lincoln.

12. **Lee, J. L., Bleay, S. M., Sears, V. G., Mehmet, S. and Croxton, R.** (2009) 'Evaluation of the Dimethylcinnamaldehyde contact transfer process and its application to fingerprint development on thermal papers', *J. Forens. Ident*., vol. 59 (5), pp 544–569.

5.3 Electrochemical techniques

5.3.1 Etching and electrodeposition

1. History

1.1 Untreated metal surfaces present an unusual problem for fingerprint development. While the majority of non-porous surfaces received in laboratories are effectively inert, in the case of metals there is the potential for chemical reactions to occur between constituents of the fingerprint (e.g. salts) and the metal surface. In extreme circumstances this can result in a permanent record of the fingerprint being etched into the metal surface. However, the interactions that occur are very dependent on the metals present and the particular constituents in the fingerprint, and reactions will only occur if conditions are favourable. In many cases the metal will be alloyed with other elements to inhibit such corrosion reactions occurring, e.g. 'stainless steel'.

1.2 It is possible to utilise the chemical reactions that can occur between a metal, the fingerprint constituents and a chemical solution to visualise fingerprints on this type of surface. Essentially, there are two generic types of technique that can be applied, etching and electrodeposition. In etching techniques, material is selectively dissolved from the surface and into solution. If the fingerprint constituents either enhance or inhibit the rate of etching at the fingerprint ridge relative to that of the background, there may be sufficient contrast produced to enable the fingerprint to be visualised. In electrodeposition the reverse is true. Metal is deposited from solution onto the surface and if the presence of the fingerprint constituents inhibits or accelerates growth of the deposit on the ridges relative to the rate of growth on the background, contrast will again be produced.

1.3 The primary sources of untreated metal surfaces are cartridge cases, which have always presented a problem for fingerprint development because of the conditions they are exposed to. High temperatures, abrasion and deposition of propellant residue all reduce the chances of recovering fingerprints and a variety of techniques have been considered. Given [1] investigated powdering techniques on brass and nickel-plated cartridges, but also included nitric acid fuming as a technique for selectively etching the metal. It was considered that sebaceous prints would protect the metal surface from corrosion, thus producing contrast.

1.4 Around the same time, Belcher was experimenting with techniques for developing fingerprints of different metals after heating [2,3]. He proposed dipping copper into solutions of brown photographic toner, and steel samples into liquid gun-blueing solution [2], later recommending potassium permanganate solution for cartridge casings with thin copper coatings [3]. In 1977 Belcher wrote to New Scotland Yard to propose the operational use of these techniques on articles recovered from terrorist

incidents and this prompted an investigation by the Police Scientific Development Branch (PSDB) into related techniques [4]. Among the chemicals investigated were: nitric acid, which showed some preferential etching of nickel-based cases; 5% selenic acid, which gave the 'gun blueing' effect on brass with some results on steel and nickel; copper sulphate, which etched nickel; sodium sulphide, which gave reasonable results on brass; and a solution of antimony in hydrochloric acid, which plated antimony onto the metal surfaces. Hydrochloric, sulphuric and hydroiodic acids gave no useful results. Vacuum metal deposition was noted to give reasonable results on most metal surfaces.

1.5 Interest in techniques for development of fingerprints on cartridges revived in the mid-1990s, with several papers on the subject being presented at the International Symposium on Fingerprint Detection and Identification in Israel in 1995. Saunders and Cantu [5] investigated the use of a modified physical developer, acidified silver nitrate and gun blueing for unfired cartridge casings and also compared superglue and gun blueing on a range of fired cases. It was found that the most effective combination was superglue, followed by gun blueing, although success rates on operational work were not as good as those observed experimentally.

1.6 Wiesner *et al.* [6] considered the effects of firing conditions on fingerprint development and compared gun-blueing, silver nitrate and superglue. The effects of gunpowder residue, friction and heating to high temperatures were studied. Of the techniques investigated gun blueing again exhibited most promise.

1.7 Migron *et al*. [7,8] considered the electrodeposition of palladium for the development of latent fingerprints and assessed a range of palladium compounds for this purpose. Good results were obtained for fingerprints on unfired cartridges and in some cases a preliminary etch of the surface using iodine also produced good images of the fingerprint. However, it proved difficult to develop marks on fired cartridges using this technique.

1.8 Bentsen *et al.* [9] tested a variety of electrodeposition techniques on fired cartridge cases using solutions of copper, nickel, chromium and tin sulphate at different concentrations and compared the results with those obtained by other techniques, including 4% selenious acid (the principal constituent of gun blueing solutions). Selenious acid had a higher sensitivity than the other electrodeposition techniques and therefore these were not studied further.

1.9 One issue sometimes experienced with the use of gun blue solutions was the overdevelopment of the blue surface coating formed. Cantu *et al.* [10] demonstrated that acidified hydrogen peroxide could be used to prevent overdevelopment and that the same solution could also be used to visualise sebaceous prints on metal surfaces by selectively etching the background.

1.10 There was a general consensus among researchers that gun blueing, either used singly or in combination with other processes such as superglue, was one of the most effective processes in revealing marks on brass surfaces. For other types of metal surface, such as aluminium, alternative formulations such as aluminium black were investigated [11,12]. These still contain selenious acid as the principal active constituent, but with a range of other chemicals making them more suited for use on aluminium.

1.11 More recent studies involving electrochemical techniques include an extensive comparative investigation conducted by the Bundeskriminalamt (BKA) [13] and an investigation conducted in the laboratories of Strathclyde Police [14]. The conclusions from both these studies indicate that optimum treatments may vary from metal to metal and that there may be some merit in combining techniques such as superglue and palladium deposition.

2. Theory

2.1 The chemical reactions associated with the principal electrochemical techniques are outlined below.

2.2 Silver nitrate: For silver nitrate on brass or copper surfaces, a reaction occurs between the silver in solution and the copper in exposed regions of the surface. This results in deposition of silver (as a grey deposit) on the surface.

$$2Ag^{+} + Cu \leftrightarrow 2Ag + Cu^{2+}$$

2.3 Gun blueing: The principal reaction occurring with gun blueing is associated with the reaction of selenious acid with metals, shown below.

$$H_2SeO_3 + 4H^{+} + 4e^{-} \leftrightarrow Se + 3H_2O \text{ (general reaction)}$$

2.4 Although selenious acid will work on a range of different metals, it is most suited to brass where parallel reactions occur between copper and zinc, and zinc and selenious acid, resulting in the formation of the black CuSe product on the surface.

$$Cu^{2+} + Zn \rightarrow Cu + Zn^{2+}$$
$$H_2SeO_3 + 4H^{+} + 2Zn \rightarrow Se + 3H_2O + 2Zn^{2+}$$
$$Se + Cu \rightarrow CuSe_{(black)}$$

2.5 Palladium deposition: Several different palladium compounds were investigated for palladium deposition and the reactions of those found most suited for this purpose with brass are shown below. In both cases a coating of grey palladium metal is formed on the surface.

$Zn/Cu + 2Na_2PdCl_4 \rightarrow Pd + ZnCl_2 + CuCl_2 + 4NaCl$
$2Zn/Cu + 2K_2PdCl_6 \rightarrow Pd + 2ZnCl_2 + 2CuCl_2 + 4KCl$

3. Reasons technique is not recommended by CAST

3.1 CAST does not currently (2011) recommend any electrochemical processes for fingerprint detection because their relative effectiveness has not been established. In addition, some of the chemicals used in the processes are highly corrosive and there are health and safety issues associated with their use. However, such processes may prove to be more effective than the techniques currently recommended and it is hoped that a planned comparative study between electrochemical techniques, scanning Kelvin probe and current techniques, such as superglue and vacuum metal deposition, will enable more detailed advice to be given.

4.References

1. **Given, B. W.** (1976) 'Latent Fingerprints on Cartridges and Expended Cartridge Casings', *J. Forens. Sci*, vol. 21 (3), pp 587–594.

2. **Belcher, G. L.** (1977) 'Methods of Casting and Latent Print Recovery', *Fingerprint Ident. Mag.*, vol. 59 (1), pp 14–15.

3. **Belcher, G. L.** (1980) 'Developing Latents on Copper-coated Casings', *Fingerprint Whorld*, October, p 39.

4. **Jones, R.** (1978) *Fingerprints on Heated Metals – Composite Report to 2 August 1978*, unpublished internal PSDB note. London: Home Office.

5. **Saunders, G. C. and Cantu, A. A.** (1995) 'Evaluation of Several Techniques for Developing Latent Fingerprints on Unfired and Fired Cartridge Cases', *Proceedings of the International Symposium on Fingerprint Detection and Identification*, June 26–30, 1995, pp 155–160. Israel: Ne'urim.

6. **Wiesner, S., Springer, E. and Argaman, U.** (1995) 'A Closer Look at the Effects of the Shooting Process on Fingerprint Development on Fired Cartridge Cases', *Proceedings of the International Symposium on Fingerprint Detection and Identification*, June 26–30, 1995, pp 161–178. Israel: Ne'urim.

7. **Migron, Y., Mandler, D., Frank, A., Springer, E. and Almog, J.** (1995) 'Is a Fingerprint Left on a Fired Cartridge? The Development of Latent Fingerprints on Metallic Surfaces by Palladium Deposition', *Proceedings of the International Symposium on Fingerprint Detection and Identification*, June 26–30, 1995, pp 217–226. Israel: Ne'urim.

8. **Migron, Y. and Mandler, D.** (1997) 'Development of Latent Fingerprints on Unfired Cartridges by Palladium Deposition: A Surface Study', *J. Forens. Sci*, vol. 42 (6), pp 986–992.

9. **Bentsen, R. K., Brown, J. K., Dinsmore, A., Harvey, K. K. and Kee, T. G.** (1996) 'Post Firing Visualisation of Fingerprints on Spent Cartridge Cases', *Sci. and Jus.*, vol. 36, pp 3–8.

10. **Cantu, A. A., Leben, D. A., Ramotowski, R., Kopera, J. Simms, J. R.** (1998) 'Use of Acidified Hydrogen Peroxide to Remove Excess Gun Blue from Gun Blue-Treated Cartridge Cases and to Develop Latent Prints on Untreated Cartridge Cases', *J. Forens. Sci*, vol. 43 (2), pp 294–298.

11. **Smith, K. and Kauffman, C.** (2001) 'Enhancement of Latent Prints on Metal Surfaces'*, J. Forens. Ident*., vol. 51 (1), pp 9–15.

12. **Perdue, J.** (undated) *The Use of Aluminium Black for the Development of Latent Prints*, USA: Mississipi Crime Laboratory Report.

13. **Hilgert, M.** (2005) 'Latent Fingerprints on Cartridge Cases', *Presented at International Fingerprint Research Group Meeting*, 11–15 April 2005. The Hague, Netherlands: Netherlands Forensic Institute.

14. **Dominick, A.** 2006 *Recovery of Latent Fingerprints and DNA from Unfired and Fired Cartridge Cases*, MSc Thesis. University of Strathclyde.

5.3.2 Heating and electrostatic powdering

1. History

1.1 A recent addition to the range of techniques that can be utilised for the visualisation of fingerprints on cartridge casing (and other metals) is the method developed by Dr John Bond at Northamptonshire Police [1-4]. This visualisation technique utilises the fact that salts and other components of fingerprint residue are capable of causing metals and their alloys to corrode. In the technique the metal surface is heated to promote further corrosion and oxidation of the surface, the combination of which may produce sufficient distinction in colour between the fingerprint ridges and the uncorroded metal for the mark to be seen without any further treatment. Further enhancement of the mark can be obtained by applying an electrostatic charge of 2.5kV to the surface, then applying carbon-coated spherical beads, as used in the electrostatic detection apparatus (ESDA) process (see Chapter 5.4), to the surface.

1.2 The technique was shown to work for a range of different metals and alloys [1,2] and to continue to develop marks after surfaces had been cleaned with water and acetone. The technique has attracted much

interest worldwide and has been used on operational casework dating back several years [3]. Research is ongoing to determine the corrosion mechanisms that operate in producing the fingerprint images [2,4], to look at the physical and chemical changes occurring at the surface, and also to measure anion and cation concentrations in eccrine sweat.

2. Theory

2.1 The theory associated with the process is that corrosion is locally initiated on the metal surface by the action of chloride ions in the fingerprint residues. In general, the process operating is:

$$M^{Z+} + ZCl^- + ZH_2O \rightarrow M(OH)_Z + Z(H^+ + Cl^-)$$

2.2 This process results in pitting corrosion penetrating into the metal surface. This localised pitting corrosion is then enhanced by the subsequent exposure to heat, where the colour change of the metal surface caused by oxidation may also aid visualisation of the fingerprint.

2.3 The corroded areas of the metal surface also have a surface potential to the uncorroded metal, and it is these differences that are exploited by electrostatic charging and subsequent powdering.

3. Reasons technique is not recommended by CAST

3.1 The Home Office Centre for Applied Science and Technology (CAST) does not currently (2011) recommend the process because its relative effectiveness has not been compared with both currently recommended processes and processes that are still under development, including the scanning Kelvin probe. Comparative studies are planned in the near future and more informed advice will then be given

3.2 When the process was used on operational casework in the UK, it was observed to cause detrimental effects to the striations in the surface that are used for ballistic analysis. This resulted in the use of the technique being suspended. These detrimental effects were attributed to the high temperature used for 'developing' the corrosion in the surface, and this temperature has subsequently been reduced to overcome this issue.

4. References

1. **Bond, J. W.** (2008) 'Visualisation of latent fingerprint corrosion of metallic surfaces', *J. Forens. Sci*, vol. 53 (4), pp 812–822.

2. **Bond, J. W.** (2008) 'The Thermodynamics of Latent Fingerprint Corrosion of Metal Elements and Alloys', *J. Forens. Sci*, vol. 53 (6), pp 1344–1352.

3. **Bond, J. W. and Heidel, C.** (2009) 'Visualisation of latent fingerprint corrosion on a discharged brass shell casing', J*. Forens. Sci*, vol. 54 (4), pp 892–894.

4. **Bond, J. W.** (2009) 'Visualisation of latent fingerprint corrosion of brass', *J. Forens. Sci*, vol. 54 (5), pp 1034–1041.

5.4 Electrostatic detection apparatus (ESDA)

1. History

1.1 The Police Scientific Development Branch (PSDB) set up a general investigative contract with the London College of Printing in the early 1970s, with the purpose of exploring novel fingerprint detection methods and also methods for taking the fingerprints of prisoners. During this contract the electrostatic detection apparatus (ESDA) was proposed, originally for the detection of finger marks on fabrics [1-3]. In-house work at PSDB had indicated that the decay time for charged fingerprints on most materials was very short and that this precluded the use of direct charging and toning as an effective detection technique. The researchers at the London College of Printing overcame this by covering the surface being examined with a thin layer of Mylar (a polyester) and producing the charge image on this thin polymer film. The thin film was exposed to a corona charging device and then treated with an electrostatic image developer, in this case carrier beads mixed with a cascade toner.

1.2 At around the same time, Japanese researchers also demonstrated that electrostatic images of fingerprints could be transferred to thin polymer films from paper exhibits by sandwiching the paper between the polymer films and holding them in a steel press [4]. Upon separation, the electrostatic image on the polymer sheet was developed by scattering dielectric powders of sulphur, lead oxide and toner over the surface. However, this approach does not appear to have been progressed further and no practical apparatus appeared from this research.

1.3 Further PSDB-sponsored research demonstrated that the process was capable of developing fingerprints on surfaces, including papers and fabrics, but this was confined to fresh marks and those over 24 hours old did not generally produce acceptable images. Attempts to improve sensitivity were unsuccessful and therefore the work on fingerprints was terminated. However, during the course of these studies it had been observed that the technique was capable of revealing indented writing on paper and could give results superior to techniques then available, such as oblique lighting [5,6]. A further contract was placed by PSDB to develop apparatus specifically for enhancement of indenting writing and this was subsequently developed and manufactured as a commercial system by Foster and Freeman in the UK, with other manufacturers taking up the concept worldwide.

1.4 HO SRDB did revisit the electrostatic detection apparatus (ESDA) in the early 1980s to establish whether it was possible to explain some of the phenomena associated with the process and also to see if any advances in technology could be used to improve the speed or sensitivity of the process [7]. An experimental system utilising a scanning probe was developed during the course of these studies but was not progressed further. A large format ESDA system was also built with the intention of investigating the technique to screen large areas of fabric for contact

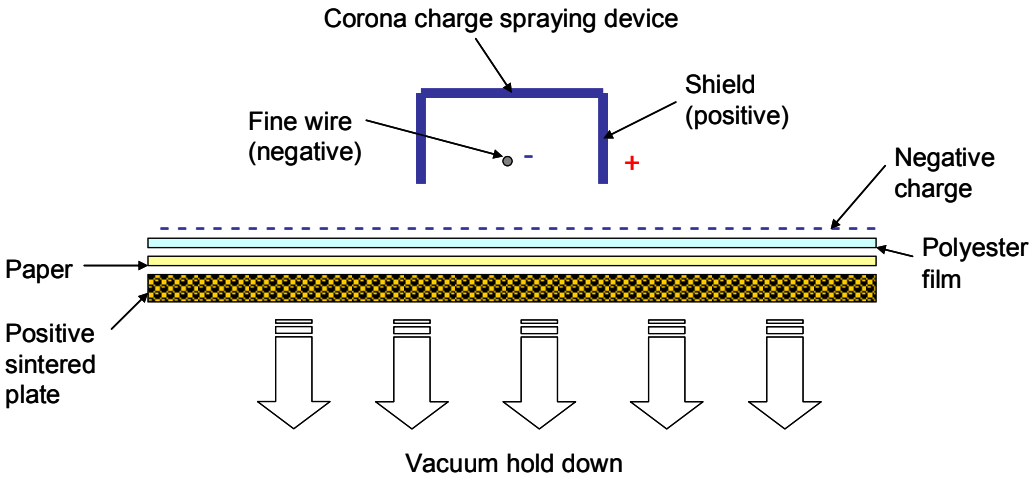
areas that could then be targeted using other, more sensitive techniques such as radioactive sulphur dioxide. This had limited success and was not taken forwards to production.

1.5 Although the ESDA system was primarily adopted for document analysis, research was carried out to establish an integrated forensic approach for document examination by examining whether treatment with ESDA could be detrimental to subsequent development of fingerprints. Initial results by Heath in 1983 [8] appeared to indicate that ESDA in general was detrimental to subsequent treatment with ninhydrin and that pre-humidification for five minutes prior to ESDA and ninhydrin treatment actually improved the quality of the fingerprints. This was contradicted in later studies by Moore [9] who found that pre-humidification of documents was detrimental to the development of fingerprints with ninhydrin, and that exposures for longer than 5–15 minutes were to be avoided. The pre-humidification effect was thought to be cumulative and repeat exposures of documents to pre-humidification and ESDA were to be avoided where possible. When it became known that pre-humidification enhanced the performance of ESDA for indented writing, HO SRDB almost immediately issued warnings that this could be detrimental to the detection of amino acids in fingerprints, particularly on some types of paper. A later study by Azoury *et al.* [10] looked at the effects of pre-humidification on fingerprint development by other amino acid reagents, including 1,8-diazafluoren-9-one (DFO) and 1,2 indandione. The results of Moore were confirmed and it was also shown that pre-humidification was detrimental to subsequent treatment with 1,2 indandione and less so to DFO, although exposures of over 60 minutes also began to degrade DFO development.

1.6 Although ESDA is found today in most UK police fingerprint laboratories, it is primarily used as a document analysis technique and if fingerprints are detected by the technique during document processing this is regarded as a bonus rather than an expected outcome.

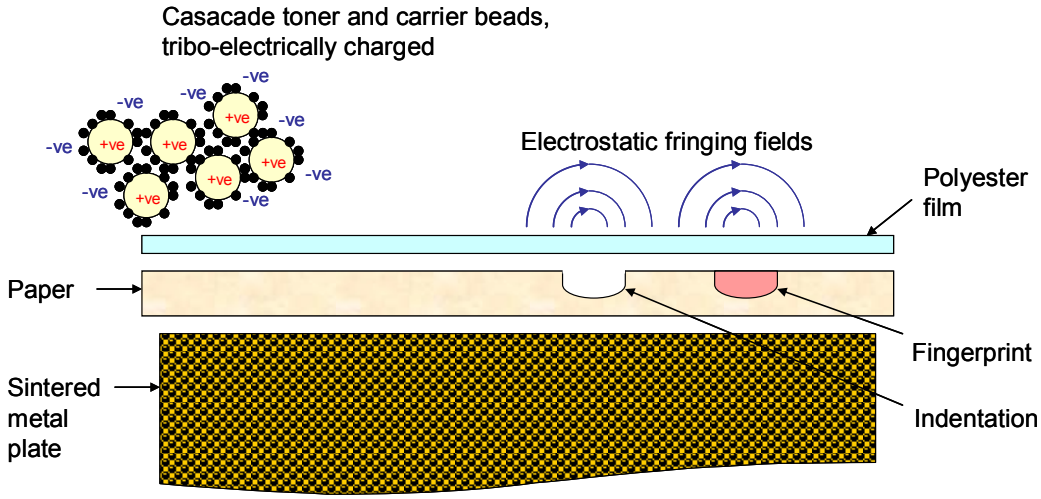
2. Theory

2.1 The mechanism of ESDA has not been conclusively established, but it is possible to describe the stages in the process. The porous exhibit to be treated is first held down on a sintered plate using a vacuum, and a thin (~3.5μm) film of Mylar laid over the top of it. This film is then negatively charged by passing a charge spraying device known as a corotron above the surface.

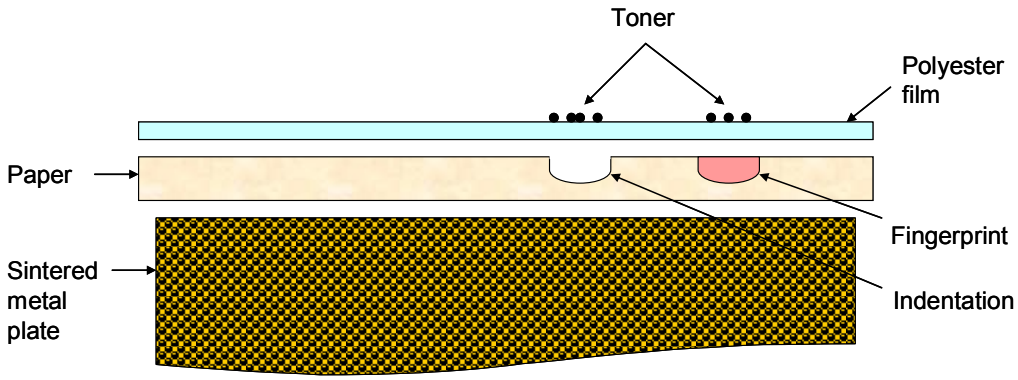


Schematic diagram of the general charging procedure for electrostatic detection apparatus.

2.2 The charging process sets up electrostatic fringing fields around features in the exhibit (the exact mechanisms of which are not precisely known). A mixture of carrier beads (fine glass spheres) and toner particles (carbon black) are cascaded across the surface, and the toner selectively adheres to areas where the fringing fields are present. This is illustrated schematically below.



a)



b)

Schematic diagrams showing toner development of electrostatic images a) development of electrostatic fringing fields on the polymer film and b) selective adherence of toner particles to regions where fields are present.

2.3 It was originally proposed that the fringing fields could be explained by a simple capacitance theory [6]. The indentations cause a local increase in capacitance due to a reduction in the distance between the charged surfaces and fingerprints, causing a local increase in capacitance because of the water in the fingerprint increasing the local dielectric constant. However, capacitance variations cannot be the only mechanism because it is noted that very deep impressions sometimes do not develop with ESDA.

2.4 It was later proposed that the indented writing effect could be explained by damage and abrasion of surface fibres caused by lateral movement between sheets of paper during the writing process [7]. The poor

performance often observed with glossy papers in the ESDA process may be explained by the fact that such papers are sized, calendared or highly loaded with inorganic filler.

2.5 Another theory proposed to explain the improved performance of ESDA often observed after pre-humidification of the article was termed 'surface variation theory' [11], which considered that after humidification the paper no longer behaved as a dielectric but as a conductor. In this theory the variation of electrostatic potential on the polymer film is a function of the degree of close contact between the polymer film and the paper, and also variation in surface features of the paper, such as glossiness and smoothness (which may also be modified by the presence of fingerprint residue). This could explain why deep indentations, where the film does not contact the paper, do not produce results using ESDA. As fingerprint residues are absorbed into the porous surface, their effect on the surface will reduce, which may explain the poor development observed on marks over 24 hours old. However, none of these mechanisms has been conclusively proven.

Fingerprints developed by electrostatic detection apparatus while processing a document.

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend the ESDA process as a primary fingerprint detection technique because it is less sensitive than other techniques for

developing fingerprints on porous surfaces, and is ineffective on marks more than 24 hours old. However, it may reveal fingerprints when used as part of an integrated strategy for retrieval of forensic evidence, ESDA being mostly non-destructive to fingerprint evidence unless pre-humidification is used. It is referred to as an additional development process in the 'Notes' section of the treatment chart for porous surfaces *Manual of Fingerprint Development Techniques* [12].

4. References

1. **Foster, D. J. and Morantz, D. J.** (1976) *Automatic Fingerprint Recording – A Report on the Machine Developed at The London College of Printing*, Report on PSDB contract London College of Printing. London: Home Office.

2. **Morantz, D. J., Freeman, R. M. and Foster, D. J.** (undated) *An Electrostatic Imaging Technique for the Detection of Fingerprints on Fabric*, Report on PSDB contract, London College of Printing. London: Home Office.

3. **Foster, D. J. and Morantz, D. J.** (1977) *The Detection of Fingerprints on Fabrics by the Development of Electrostatic Images*, Final Report on PSDB contract, London College of Printing. London: Home Office.

4. **Shirai, M., Hayashi, T. and Arai, S.** (1977) 'Transcription of Fingerprints on Papers by Electrostatic Dust Figure', *Reports of the Nat. Res. Inst. of Police Sci.*, vol. 30 (1), pp 45–50.

5. **Young, P. A.** (1979) *Equipment for the Detection of Indented Impressions*, HO PSDB Technical Memorandum No. 8/79. London: Home Office.

6. **Foster, D. J. and Morantz, D. J.** (1979) 'An Electrostatic Imaging Technique for the Detection of Indented Impressions in Documents', *Forens. Sci. Int*., vol. 13, pp 51—54.

7. **Kent, T.** (1986) *The Electrostatic Development of Fingerprints and Indented Writing – A Review*, unpublished HO SRDB paper. London: Home Office.

8. **Heath, J. S.** (1983) 'The Effects of ESDA Examination and Photocopying on the Recovery of latent Fingerprints on Documents', *presented to the 3rd Scientific Meeting of the Australian Society of Forensic Document Examiners*, 25/26 June, 1983. Australia: Melbourne, Victoria.

9. **Moore, D. S.** (1988) 'The Electrostatic Detection Apparatus (ESDA) and its Effects on Latent Prints on Paper', *J. Forens. Sci.*, vol. 33 (2), pp 357–377.

10. **Azoury, M., Gabbay, R., Cohen, D. and Almog, J.** (2003) 'ESDA Processing and Latent Fingerprint Development: The Humidity Effect', *J. Forens. Sci*, vol. 48 (3), pp 564–570.

11. **Wanxiang, L. and Xiaoling, C.** (1987) 'Electrostatic Imaging Technique: A Study of its Principle and the Effect of Experimental Condition on Imaging', *presented at IAFS Vancouver*, 1987. Canada: Vancouver.

12. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

5.5 Fuming techniques

1. History

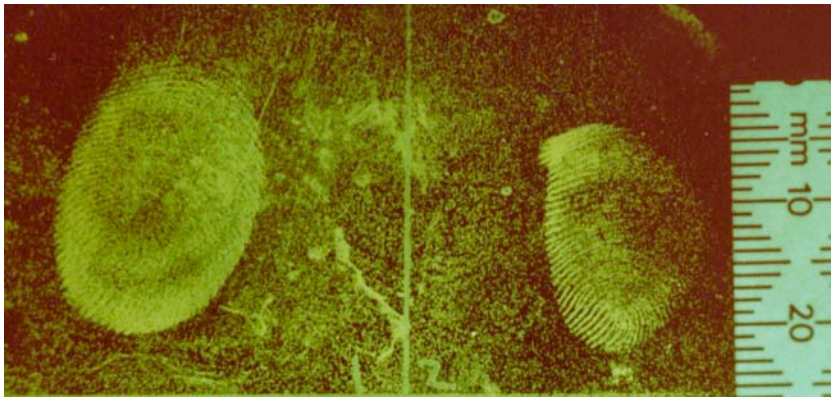
1.1 The development of fingerprints using fuming processes has been utilised since the early days of fingerprint identification. Iodine and osmium tetroxide were already known to develop fingerprints on porous surfaces by the 1920s and the fuming of a range of other substances has been investigated since then.

1.2 Several of the processes described in other chapters either involve fuming, e.g. superglue, or have been investigated as fuming techniques, e.g. 4-dimethylaminocinnamaldehyde (DMAC). Fuming has the potential advantages that it does not wet the article, which may be a benefit if subsequent document analysis is required, will permeate porous exhibits, and impinge upon all available surfaces for non-porous exhibits.

1.3 Fuming can be used to develop fingerprints in several ways, described in more detail in the section below. In a review of techniques for development of latent prints issued in 1974 [1], Micik lists three fuming techniques: iodine; hydrogen fluoride (for etching fingerprints on glass); and the burning of substances, including camphor and magnesium, to produce fumes that selectively deposited particles on fingerprint ridges.

1.4 Almog and Gabay [2] carried out an investigation into the development of fingerprints on paper by fuming several fluorescent chemicals. Good results were reported for anthranilic acid (for fresh marks), anthracene (for older marks) and antimony trichloride. In some cases the fluorescent chemical was selectively deposited on the ridges, in other cases deposition occurred on the background only.

1.5 The Home Office Scientific Research and Development Branch (HO SRDB) conducted a subsequent study into the anthracene fuming process [3], first investigating the optimisation of fuming conditions using fingerprints deposited on glass slides and then applying the optimised process to fingerprints on different types of plastic and metal surfaces. The potential benefits of vacuum deposition of anthracene were also explored. It was found that sublimation in air gave better results than vacuum deposition and although the process did develop fingerprints on plastics, it was not as effective as other processes already available. Results on metals were more promising and anthracene fuming was found to be more effective than iodine over a range of different metal surfaces.



Photograph of fingerprints on metal developed by anthracene fuming.

1.6 Haque [4] considered the fuming of naphthalene and camphor, followed by iodine fuming and dusting with magnetic powder. This multi-step process appeared to give excellent sensitivity on plastic bag substrates. The selective attack of polymer surfaces using the fumes of halogenated hydrocarbons such as dichloromethane and chloroform was also studied. The technique worked well on polystyrene, but was ineffective on vinyl and thermoset plastics, and did not work at all on polyethylene.

1.7 Fuming has also been reported in combination with other processes for the revelation of fingerprints. Meylan *et al.* described the fuming of ammonium hydrogen carbonate after exposure of a paper exhibit to a corona discharge [5]. This combined treatment produced fluorescent fingerprints that could be excited by ultraviolet light. This technique was further investigated by Davies *et al.* [6]; they carried out an analysis of the fluorescent products and suggested that lipid derivatives were responsible for the fluorescence observed.

1.8 In addition to the hydrofluoric acid fuming process mentioned by Micik for developing fingerprints on glass, other acid fuming techniques have been considered. Bentsen *et al*. [7] trialled nitric acid fuming for development of fingerprints on brass cartridge cases and Broniek and Knaap [8] proposed hydrochloric acid fuming as a technique for revealing fingerprints on thermal receipts. The highly corrosive nature of these substances meant that such techniques were not widely adopted for operational use because of the precautions required for their use.

1.9 A novel process that has been recently reported by Kelly *et. al*. is the use of disulphur dinitride, allowed to sublime under a static vacuum [9]. This has been shown to be capable of developing fingerprints on a wide range of surfaces, including paper, fabric, clingfilm and metals, possibly by formation of the blue-black sulphur-nitrogen backbone (SN_x) polymer.

2. Theory

2.1 Because many different types of substance have been used as fuming techniques for the development of fingerprints, there is no single mechanism that applies to all chemicals. A range of mechanisms may operate and some of these are outlined below.

2.2 Absorption of coloured vapours into fingerprint residues – this is the mechanism that occurs for iodine (and other halogens, such as bromine).

2.3 Chemical reaction between fumes and fingerprint residues to form a coloured or fluorescent reaction product, e.g. the black product formed by osmium tetroxide fumes.

2.4 Catalysis of a polymerisation reaction by fingerprint residues, promoting growth of a solid phase from gaseous fumes – this is the case for the superglue process and also possibly the recently reported disulphur dinitride process [9].

2.5 Selective deposition of particulates on fingerprint ridges (or background) – this is observed for fuming of anthracene, camphor and naphthalene.

2.6 Selective etching/attack of ridges (or background) by fumes of acid or other substance – this can be seen for hydrogen fluoride on glass, nitric acid on brass, and chloroform on polystyrene.

3. Reasons technique is not recommended by CAST

3.1 CAST does recommend two fuming processes, iodine and superglue (Chapters 5.10 and 3.10 respectively in this book), in the *Manual of Fingerprint Development Techniques* [10]. Other fuming techniques are not recommended because they are either less effective than other techniques (e.g. anthracene) or there are health and safety issues associated with their use. In particular, there are concerns about the fuming of concentrated acids because they are highly corrosive. In general, all fuming processes need to be well-contained and carried out in areas with good ventilation.

4. References

1. **Micik, W.** (1974) ‘Latent Print Techniques’, *Ident.*, October, pp 3–9.

2. **Almog, J. and Gabay, A.** (1980) ‘Chemical Reagents for the Development of Latent Fingerprints. III: Visualisation of Latent Fingerprints by Fluorescent Reagents in Vapor Phase’, *J. Forens. Sci*, vol. 25 (2), pp 408–410.

3. **Peacock, P. M.** (1982) *The Development of Latent Fingerprints by the Evaporation of Anthracene*, February. Polytechnic of the South Bank.

4. **Haque, F.** (1982) *Organic Vapours for Developing Latent Fingerprints on Non-porous Surfaces*, Fingerprint Report No. 7, Chemistry Department, March. University of Ottawa, Canada.

5. **Meylan, N., Lennard, C. and Margot, P.** (1990) 'Use of a Gaseous Electrical Discharge to Induce Luminescence in Latent Fingerprints', *Forens. Sci. Int*., vol. 45, pp 73–83.

6. **Davies, L. M., Jones, N. E., Brennan, J. S. and Bramble, S. K.** (2000) 'A New Visibly-Excited Fluorescent Component in Latent Fingerprint Residue Induced by Gaseous Electrical Discharge', *J. Forens. Sci*, vol. 45 (6), pp 1294–1298.

7. **Bentsen, R. K., Brown, J. K., Dinsmore, A., Harvey, K. K. and Kee, T. G.** (1996) 'Post Firing Visualisation of Fingerprints on Spent Cartridge Cases', *Sci. and Jus.*, vol. 36 (1), pp 3–8.

8. **Broniek, B. and Knaap, W.** (2002) 'Latent Fingerprint Development on Thermal Paper using Muriatic (Hydrochloric) Acid', *J. Forens. Ident*., vol. 54 (4), pp 427–432.

9. **Kelly, P. F., King, R. S. P. and Mortimer, R. J.** (2008) 'Fingerprint and inkjet trace imaging using disulfur dinitride'*, Chem. Comm*., pp 6111–6113.

10. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

5.6 Gelatine lifting

1. History

1.1 Gelatine lifting has been used for the recovery of fingerprints from the early 20th century. The concept was first proposed in 1913 for lifting of marks powdered with lead acetate and subsequently treated with hydrogen sulphide. The lifting medium used in this case was a paper coated with a gelatine/glycerol mix [1]. Gelatine lifting was not widely adopted for fingerprint lifting at that time, but the lifting concept was further investigated for the recovery of footwear marks [2]. By the late 1970s several rubber- and gelatine-based lifters were commercially available for the lifting of footwear marks, including latent marks in dust and dried contaminant, and marks developed by powdering. Experiments conducted by the Police Scientific Development Branch (PSDB) in the early 1970s utilised gelatine films to lift marks developed using vacuum metal deposition from patterned surfaces [3]. Physical developer was then used to intensify the images of the lifted marks. This was reasonably successful, but a high contrast mark developed using vacuum metal deposition was required as a starting point.

1.2 There has been subsequent research into the broader forensic applications of gelatine lifts. The mildly adhesive nature of the gelatine lift combined with a degree of flexibility and compressibility makes them well suited for the lifting of trace evidence from a range of surfaces, without causing significant damage to the surface itself.

1.3 As a consequence of these studies, gelatine lifts are now marketed for the lifting of footwear marks [4,5,6], the lifting of paint and other micro traces [4], recording patterns around bullet holes [4] and the lifting of blood traces from surfaces [4]. They have also been shown to be effective in detecting indented writing, and in comparisons with the electrostatic detection apparatus (ESDA) technique (see Chapter 5.4) have shown better performance than ESDA on thick, glossy paper types, and to be capable of being used sequentially after ESDA on documents [7].

1.4 The principal application of gelatine lifts has remained the lifting of fingerprint and footwear evidence, both latent marks and marks developed using processes such as powders and superglue. Gelatine lifts are currently (2011) available in black, white and clear forms, and because they are flexible and can be compressed against a surface on application, they are better suited to lifting of powdered marks from textured surfaces than some types of tape. The colour of the lift can be selected to give optimum contrast with the powder used, and the lifts are better suited for lifting and subsequent imaging of marks powdered with granular and magnetic powders [4].

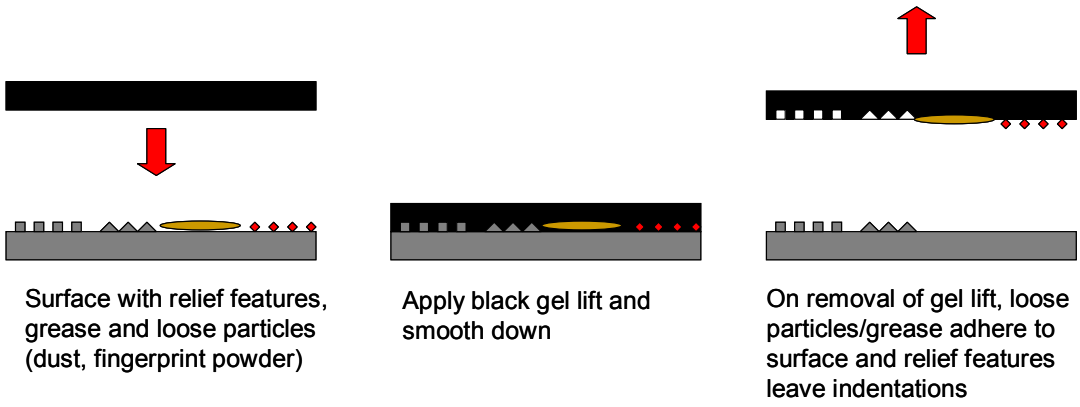
1.5 The gelatine lifting process has also been shown to be a potentially useful technique in recovering marks for subsequent chemical analysis.

The gelatine lift acts as a transfer medium for marks lifted from a scene to be transported back to a laboratory for subsequent compositional analysis [8].

1.6 The recent (circa 2005) development of specialist imaging equipment for the enhancement of marks lifted on black gelatine lifts (GLScan, produced by BVDA, Haarlem, Netherlands) has increased interest in the use of gelatine lifts for the recovery of latent fingerprints prior to chemical development. Several police forces in the UK have proposed the use of the technique as an alternative to powdering. This chapter deals with the application of gelatine lifting as the sole recovery process for latent fingerprints, as opposed to a lifting process for marks developed using other processes, such as powdering or superglue.

2. Theory

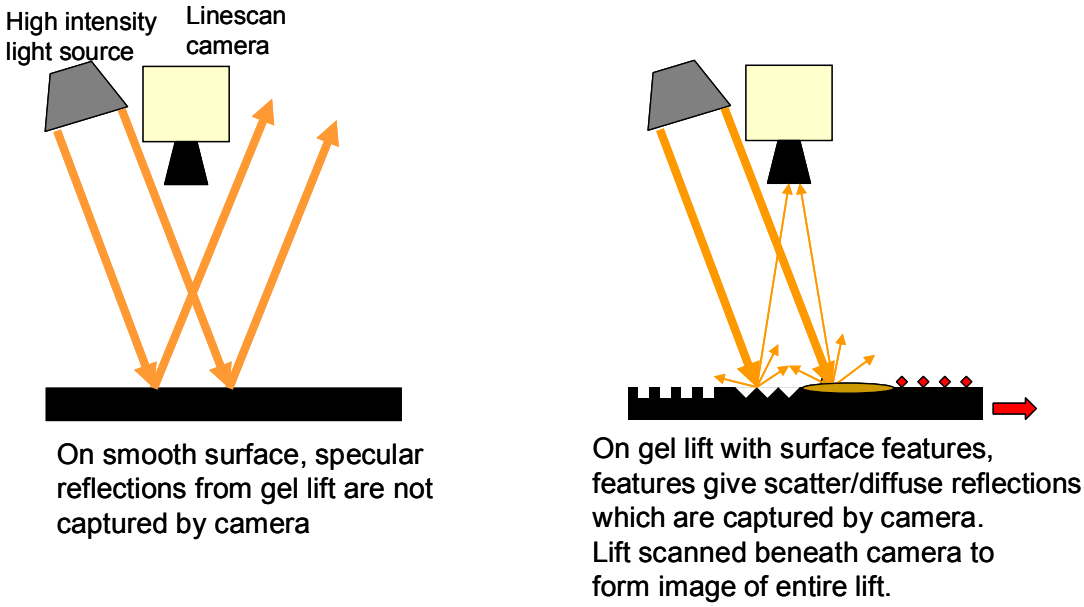
2.1 The theory behind the gelatine-lifting technique is that the gelatine is able to deform to the surface contours during application and smoothing in place. The slight adhesive nature of the surface also means that on removal of the gelatine lift, some of the loose particulate matter and any grease on the surface will be transferred to the surface of the gel. The gel may also retain some impression of the contours of the surface it has been applied to.



Schematic diagram showing how gelatine lifts can lift and reproduce surface features.

2.2 The surface features retained on the lift are then imaged in a way that maximises the contrast between the surface feature and the black background of the lift. This can be carried out using photography in a dark room with the light source perpendicular to the surface and close to the imaging system. Alternatively a specialist imaging system such as the BVDA GLScan may be used. The GLScan system consists of a line scan camera combined with high intensity white light illuminating the gel at an angle close to perpendicular to the surface. The gel itself is mounted on a vacuum stage drawing it flat, and then scanned slowly by moving the vacuum stage underneath the fixed focus position of the line

scan camera. The principle common to both imaging methods is that with nothing on the gel, the specular reflection from the surface means that no light is reflected into the camera and the background appears black. The particulates and grease on the surface scatter light and produce diffuse reflections, meaning that some light reaches the camera and those regions appear lighter.



Schematic diagram showing the way in which images are produced in the GLScan system.

2.3 An example of a section of a gelatine lift taken from a door handle and scanned on GLScan equipment is illustrated below.

Example of a series of latent marks lifted from a door handle using a gelatine lift and imaged on a GLScan system (greyscale inverted).

3. CAST processes

3.1 The Home Office Centre for Applied Science and Technology (CAST) recommends using the process in accord with the gel manufacturer's instructions, peeling the acetate from the gelatine lift and applying it to the surface being treated. The gel is then smoothed in place to remove air bubbles. It may be beneficial to leave the gel in place for several minutes or to warm it slightly, but CAST has no data to conclusively demonstrate the benefit of either of these approaches.

3.2 'Gelatine' lifts can be obtained from more than one manufacturer, the principal supplier being BVDA (Haarlem, Netherlands). A rubber-based lifter is available from Dycem (Bristol, UK) for the same applications and there are other producers of similar products worldwide. It is not possible to recommend a single type of lifter for all applications. In general the BVDA lifts have been found to have higher tack and be more effective than the Dycem lifts, but in some cases the higher tack of the BVDA lift may cause damage to the surface. The ultimate selection of lifter by the user must take these factors into account.

3.3 Once lifted, the gelatine lift should be stored without a cover material, if at all possible, and imaged as soon as it is retrieved to the laboratory. This is because any lifted latent marks will progressively degrade and the reapplication of a cover material exacerbates this.

3.4 Imaging of the lift should be carried out under the conditions outlined in the 'Theory' section above. However, they should also be examined under oblique lighting. This is because any lifted marks in dust are best visualised under oblique light but may not be so prominent under the specular lighting conditions used to capture greasy deposits.

4. Critical issues

4.1 The temperature of the surface to be lifted must be below 40°C otherwise the gelatine lift may melt on the surface.

4.2 The gelatine lift must be smoothed in place to eliminate air bubbles, enabling all parts of the surface to come into contact with the lifting material.

4.3 The lift should ideally be stored without a cover and imaged as soon as possible after lifting, to reduce degradation in the quality of the lifted marks.

5. Application

5.1 Suitable surfaces: Gelatine lifts are suitable for use on all smooth non-porous surfaces where they can be readily formed to the shape of the surface. They can also be used on surfaces where a layer of contaminant is present. It is possible to use gelatine lifts on textured, semi-porous and porous surfaces, but their effectiveness is considerably reduced.

5.2 There are no special application methods for the gelatine lifts other than those recommended by the manufacturer [4]. The lifts may be cut to shape to suit the article or surface they are being applied to.

5.3 Gelatine lifting is recommended for situations where the primary processes in the *Manual of Fingerprint Development Techniques* [9] may not be applicable, primarily as an alternative to powdering. Such circumstances may include the following.

- Heavily contaminated surfaces where marks are visible in the contaminant, but cannot be imaged in situ and chemical development is not feasible.
- Articles that cannot be chemically treated and/or the application of powders may leave permanent traces or have a risk of damage (e.g. electrical equipment such as laptops, valuable antiques, etc.).
- Areas that are not easy to reach using powdering and where any developed marks would be difficult to see (e.g. on the inside of door handles).

6. Alternative formulations and processes

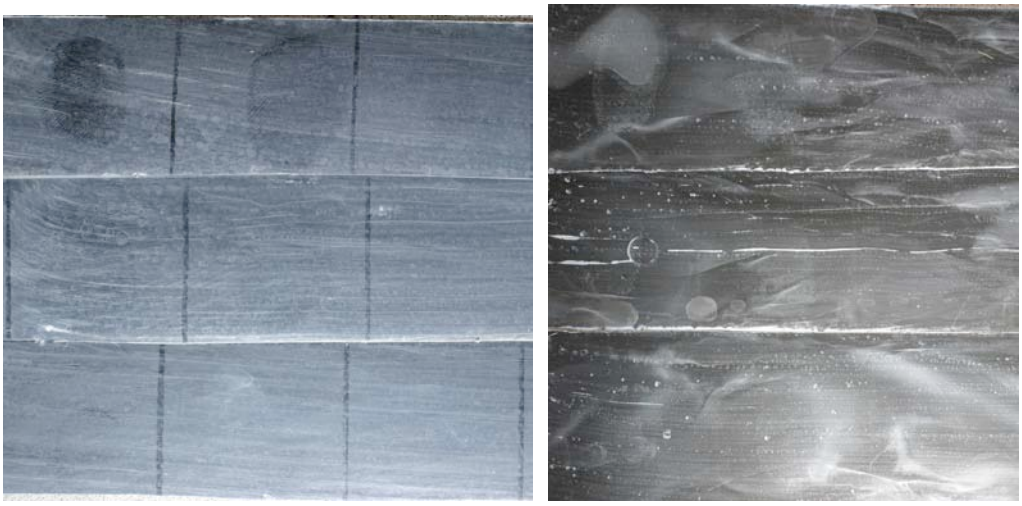
6.1 As alluded to above, there are several different types of gelatine lifter on the market. The only ones evaluated by HOSDB are the BVDA Black Gelatin Lift and the Dycem High Performance Evidence Lifter. Both of these have advantages and disadvantages and the user is encouraged to make a judgement on which lift to use according to the individual circumstances of the scene.

6.2 Silicone casting compounds have also been used to lift latent marks from surfaces, but in this case the lifted marks are not imaged directly on the surface, but are first developed using another enhancement process such as superglue [10].

7. Post-treatments

7.1 In some circumstances it may be able to enhance the latent marks lifted by a secondary chemical process. Attempts have been made to enhance marks lifted on both Dycem and BVDA lifters using white powder suspensions and superglue, which were selected to give maximum contrast with the black lift.

7.2 The results obtained for some donors on post-treated lifts are shown below.



a) b)

c)

d)

Post-recovery enhancement of marks on gelatine lifts a) white powder suspension on BVDA lift b) white powder suspension on Dycem lift c) superglue on BVDA lift and d) superglue on Dycem lift.

7.3 The results suggest that although there is little benefit in applying subsequent chemical treatments to BVDA gels, chemical treatment (superglue in particular) of Dycem gel lifts may improve marks in some cases or even develop additional marks, in particular for superglue treatment. This is in accordance with observations made by other researchers using silicone rubber-based casting compounds [10].

8. Validation and operational experience

8.1 Laboratory trials

8.1.1 CAST has carried out a direct comparison of the effectiveness of gelatine lifting with powdering [11]. This study compared gelatine lifting using black gelatine lifts produced by BVDA with the powdering process found to be most appropriate to the particular surface type being studied, according to guidelines published by CAST [12]. In this study six surfaces, representative of those found at crime scenes, were used:

- glass;
- u-PVC;
- painted metal;
- laminate (fake textured granite);
- laminate (fake ash);
- silk painted plasterboard.

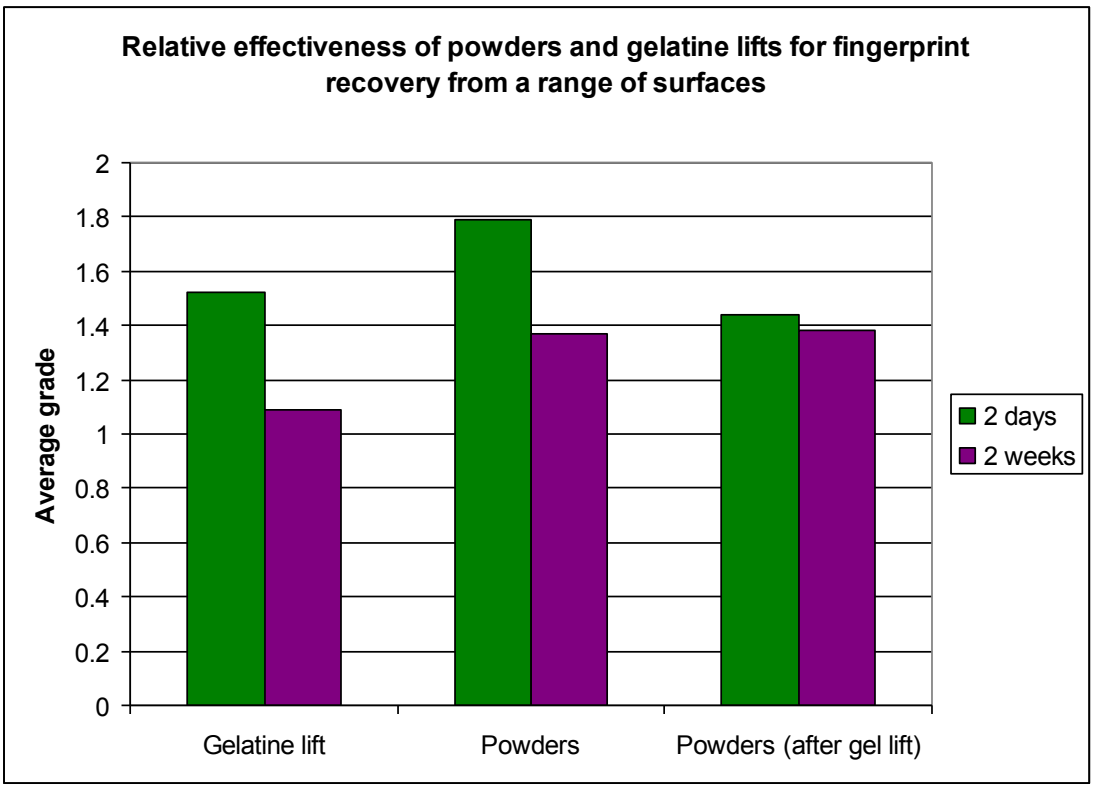
8.1.2 In this trial 70 separate donors were used, each depositing three fingerprints on each of the six surfaces. Donors were asked to wait at least 30 minutes after washing their hands before deposition of the marks, rubbing their hands together before deposition to evenly

distribute the sweat over the entire surface. No 'grooming' of marks (i.e. rubbing fingers on nose or forehead) was permitted.

8.1.3 The surfaces were then aged for periods of two days and two weeks. The marks were processed by three different routes:

- gelatine lift of the latent mark and subsequent imaging on the GLScan;
- application of fingerprint powder according to recommendations of the CAST *Fingerprint Powders Guidelines* [12];
- application of fingerprint powder as above, but on the same surface previously treated with the gelatine lift.

8.1.4 The results of the experiment to compare relative process effectiveness are shown below, with the average grade of developed mark across all 70 donors being compared for powdering and gel lifting.



Results of the experiment to compare the effectiveness of gelatine lifting and powdering for fingerprint recovery.

8.1.5 The information depicted in the graph shows that for both powdering and gelatine lifting used as a single process there is a drop in the average grade of marks developed as the age of the mark increases from two days to two weeks. This is consistent with trends seen in previous studies of the powdering process.

8.1.6 It can also be seen that for both ages of fingerprint, powdering gives superior results to gelatine lifting.

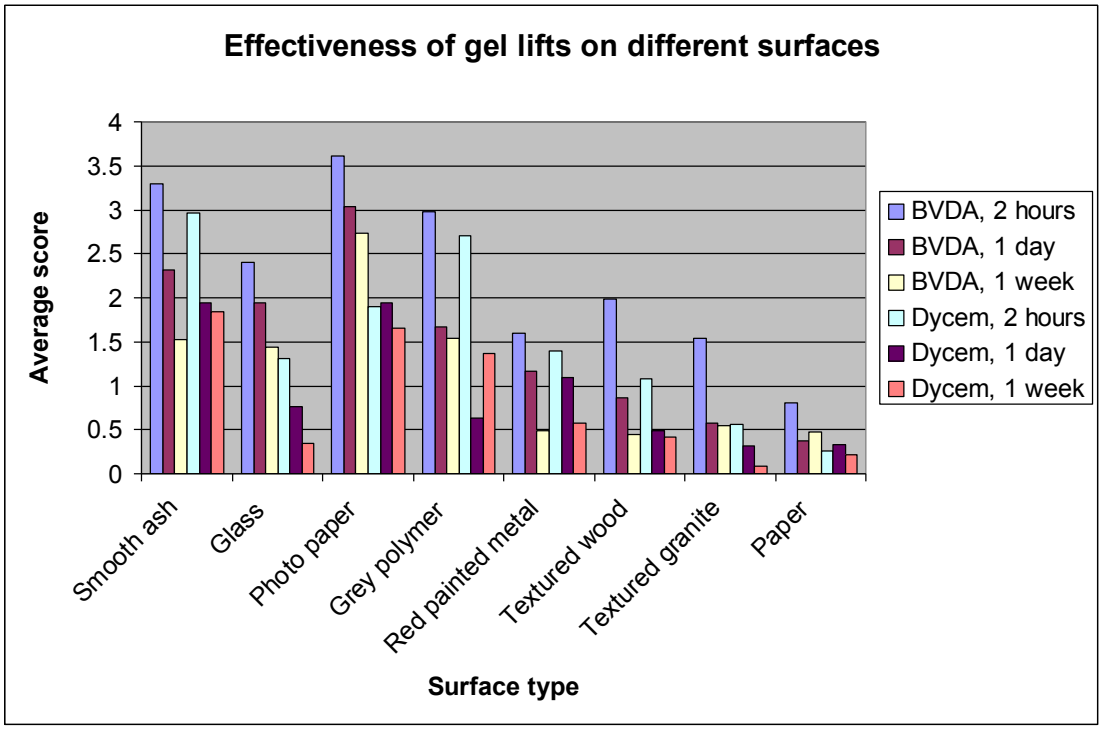
8.1.7 For two-day-old marks, the average grade of powdered marks is reduced by 20% if the gelatine lift is applied prior to powdering. Gelatine lifting transfers some of the residue to the lift, hence reducing the amount of material left on the surface for powders to adhere to. For two-week-old marks, application of the gelatine lift prior to powdering is far less detrimental because the mark has hardened and less residue is transferred.

8.1.8 A second study [13] was carried out to establish the relative effectiveness of two types of lifter (BVDA and Dycem) across surfaces ranging from smooth non-porous through rough non-porous to porous surfaces. The following surfaces were used in the study:

- glass;
- glossy photographic paper;
- laminate (fake smooth ash);
- grey polypropylene polymer;
- red painted metal (car paint scheme);
- laminate (fake textured oak);
- laminate (fake textured granite);
- printer paper.

8.1.9 Depletion series of nine marks were deposited by a range of six to seven donors (depending on the size of the surface used for deposition) using the same process described above. Marks were aged for two hours, one day and one week prior to gel lifting.

8.1.10 The results of this study are summarised in the graph below, which shows the average score across all marks deposited.



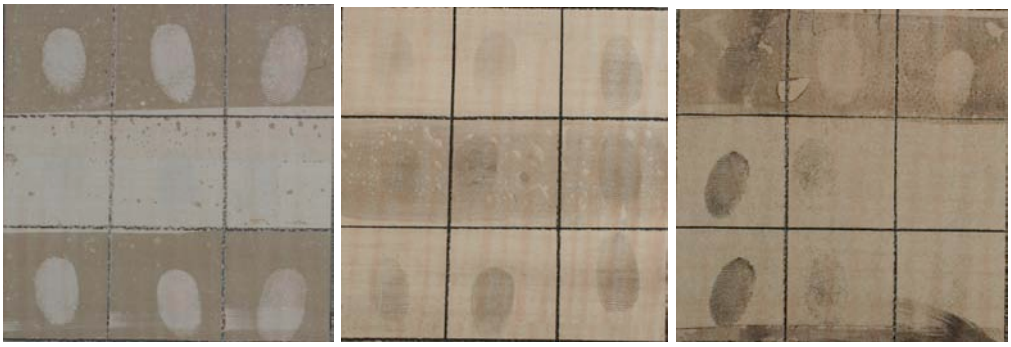
Graph showing the effectiveness of gelatine lifting on different surfaces and on marks of different age.

8.1.11 The BVDA gel lifts were found to be more effective than Dycem lifters on all the surfaces studied to date (2008), and this is consistent with the greater surface tack of the BVDA gel when the protective acetate sheet is removed. However, the potential of the higher tack BVDA lifts to cause surface damage should be recognised.

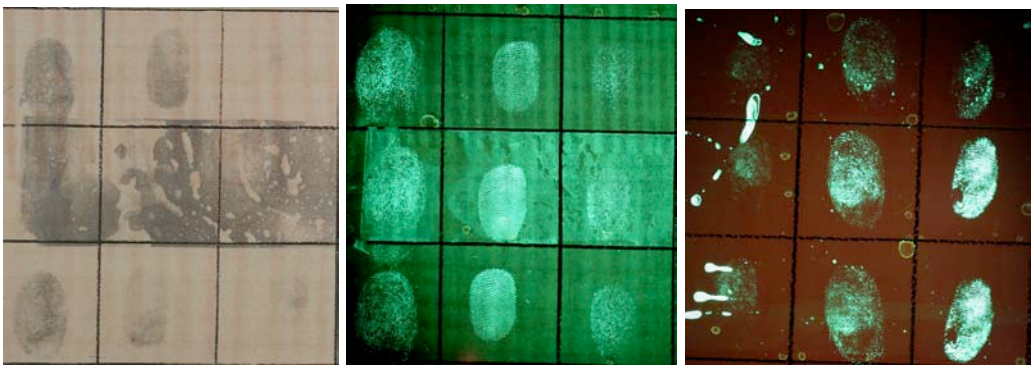
8.1.12 The effectiveness of gel lifts was seen to decrease as surface roughness and porosity increases.

8.1.13 In accordance with the initial study above, it can be seen that the effectiveness of gelatine lifting decreases as the age of the mark increases, and that significant degradation in the average score of lifted marks actually occurs in the period between two hours and one day after deposition.

8.1.14 Further experiments were carried out to establish if gelatine lifting could be used in sequence with other processes. It was shown that both types of lifter could be detrimental to subsequent treatment, but that it was not always possible to tell which combination of lifting material, surface and subsequent development technique would cause problems. The use of gelatine lifting as a development process is therefore not recommended if further treatments are likely to be carried out to the surface.



a) b) c)



d) e) f)

The effects of gelatine lifting on subsequent fingerprint development processes a) vacuum metal deposition on fake ash laminate b) black magnetic powder on fake ash laminate c) black powder suspensions on fake ash laminate d) Magneta flake powder on fake ash laminate e) superglue/basic yellow 40 on fake ash laminate and f) superglue/basic yellow 40 on painted metal. Top row = lifted with BVDA lift, middle row = lifted with Dycem lift and bottom row = control (no lifter applied).

8.2 Pseudo-operational trials and operational experience

8.2.1 No fully recorded pseudo-operational trials have been conducted on gelatine lifting, although small-scale exercises have been conducted on 'real' surfaces by HOSDB to see what types of item the technique can recover marks from. These were articles and surfaces tested during a tour around the laboratories and common areas at HOSDB, without any pre-planting of marks. Surfaces that marks were successfully lifted from included: soft drinks cans, coffee mugs, door handles and push plates, glass windows, wooden pool cue handle, bench top, credit card, pens, guns and glossy magazine covers.

8.2.2 Operationally there are few situations where the process should be used in preference to powdering using the optimum brush/powder combination, but there are some police forces using the technique routinely for lifting of latent marks. One widely publicised success was obtained from gelatine lifting a mark in grease from the ceiling of an

abattoir. This mark could not be powdered because of contamination, could not be chemically treated because of the difficulties in washing chemicals over the ceiling, and was difficult to photograph in situ because of it being on a white background.

9. References

1) **Crispo, D.** (1913) *Bull. Soc. Chim. Belg*., vol. 26, pp 190–193.

2) **Bodziak, W. J.** (2000) *Footwear Impression Evidence – Detection, Recovery and Examination*, 2nd edition, pp 116–122. Boca Raton: CRC Press.

3) **Newton, R.** (1974) *Detection of Latent Fingerprints on Banknotes by Metal Deposition,* Student placement report, September. Polytechnic of the South Bank

4) **BVDA International** (2009) *Gellifter brochure*, BVDA International. Available from: <http://www.bvda.com/EN/download/Gellifter_brochure.pdf >, accessed 3 April, 2009.

5) **Theeuwen, A. B. E., van Barneveld, S., Drok, J. W., Keereweer, I., Limborgh, J. C.M., Naber, W. M. and Velders, T.** (2001) 'Enhancement of Muddy Footwear Impressions', *Forens. Sci. Int*., vol. 119 (1), pp 57–67.

6) **Shor, Y., Tsach, T., Vinokurov, A., Glattstein, B., Landau, E. and Levin, N.** (2003) 'Lifting Shoeprints using Gelatin Lifters and a Hydraulic Press', *J. Forens. Sci*, vol. 48 (2), pp 368–372.

7) **De Koeijer, J. A., Berger, C. E. H., Glas, W. and Madhuizen, H. T.**(2006) 'Gelatine Lifting, a Novel Technique for the Examination of Indented Writing', *J. Forens. Sci*, vol. 51 (4), pp 908–914.

8) **Ricci, C., Bleay, S. and Kazarian, S. G.** (2007) 'Spectroscopic Imaging of latent fingermarks collected with the aid of a gelatin tape', *Anal. Chem.*, vol. 79 (15), pp 5771–5775.

9) **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

10) **Shalhoub, R., Quinones, I., Ames, C. Multaney, B., Curtis, S., Seeboruth, H., Moore, S. and Daniel, B.** (2008) 'The Recovery of Latent Fingerprints and DNA using a Silicone-based Casting Material', *Forens. Sci. Int*., vol. 178 (2), pp 199–203.

11) **HOSDB** (2007) *Fingerprint Development and Imaging Newsletter*, HOSDB Publication No. 17/07, April. London: Home Office.

12) **Bandey, H. L.** (2007) *Fingerprint Powders Guidelines*, HOSDB Publication No. 09/07, March. London: Home Office.

13) **Bleay, S.M., Bandey, H. L., Black, M., and Sears, V. G.** (2011) 'The Gelatin Lifting Process: An Evaluation of its Effectiveness in the Recovery of Latent Fingerprints', *J. Forens. Ident.,* vol. 61 (6), pp 549–574.

5.7 1,2 Indandione

1. History

1.1 1,2 Indandione was first proposed as a fingerprint development reagent in 1997 [1,2], following observations by researchers at the University of Pennsylvania that it reacted with amino acids to give products that were both coloured (pink) and fluorescent. A range of analogues were also developed in this study, but only 1,2 indandione has been extensively researched since.

1.2 1,2 Indandione is applied in a very similar way to 1,8-diazafluoren-9-one (DFO) and ninhydrin, drawing the exhibit through a bath of solution, allowing it to dry and then placing it in an oven to develop the marks. The initial observations of both coloured and fluorescent reaction products prompted more detailed investigations of the reagent in comparison to the ninhydrin and DFO formulations then in common use [3,4]. Both these studies indicated that 1,2 indandione merited further study, with results equivalent to DFO being obtained in laboratory tests. However, it was also observed that sequential treatments using combinations of ninhydrin and 1,2 indandione were not particularly effective [3].

1.3 Further studies were carried out in both Israel [5] and by the Police Scientific Development Branch (PSDB) in the UK [6] to establish the optimum processing conditions for 1,2 indandione, although these arrived at different conclusions. The Israeli researchers found that a formulation free of acetic acid gave the best results, and suggested processing conditions of 20 minutes at 100ºC and 60% relative humidity, whereas the UK research identified an optimal level of acetic acid to promote fluorescence and suggested processing for 10 minutes at 100ºC and 0% relative humidity. Variable results have since been obtained from 1,2 indandione at different laboratories worldwide and it has been concluded that humidity is very important in the development process and variations in local humidity conditions affect the results obtained.

1.4 However, both the Israeli and UK research provided further evidence that 1,2 indandione justified operational trials, PSDB [6] finding it giving equivalent results to DFO on batches of 75 cheques and a range of representative porous items, and the Israelis [5] reporting an improved performance over DFO on a pseudo-operational trial conducted over batches of 500 cheques per process. Once again it was found that using ninhydrin in sequence after 1,2 indandione developed few, if any, additional marks.

1.5 Based on these results, the process was adopted for operational use in Israel and taken forward into a full operational trial in the UK [7]. In the UK operational trials the performance of 1,2 indandione was the least effective of the formulations under test and was consequently not recommended for operational use. A similar operational trial in Canada

[8] arrived at similar conclusions and in both countries DFO remained the technique of choice.

1.6 1,2 Indandione has become more widely used in Australia and Israel and to some extent in the USA, and further research into the reagent has been conducted in all three countries. Stimac [9] has proposed a formulation of 1,2 indandione for use with thermal papers and Azoury *et al.* [10] have reported that the treatment of exhibits with 1,2 indandione is not detrimental to subsequent DNA profiling. However, a survey conducted into the usage of chemical treatments worldwide demonstrated that 1,2 indandione was still not in widespread use in many countries and in some cases the respondents were not aware of it at all [11].

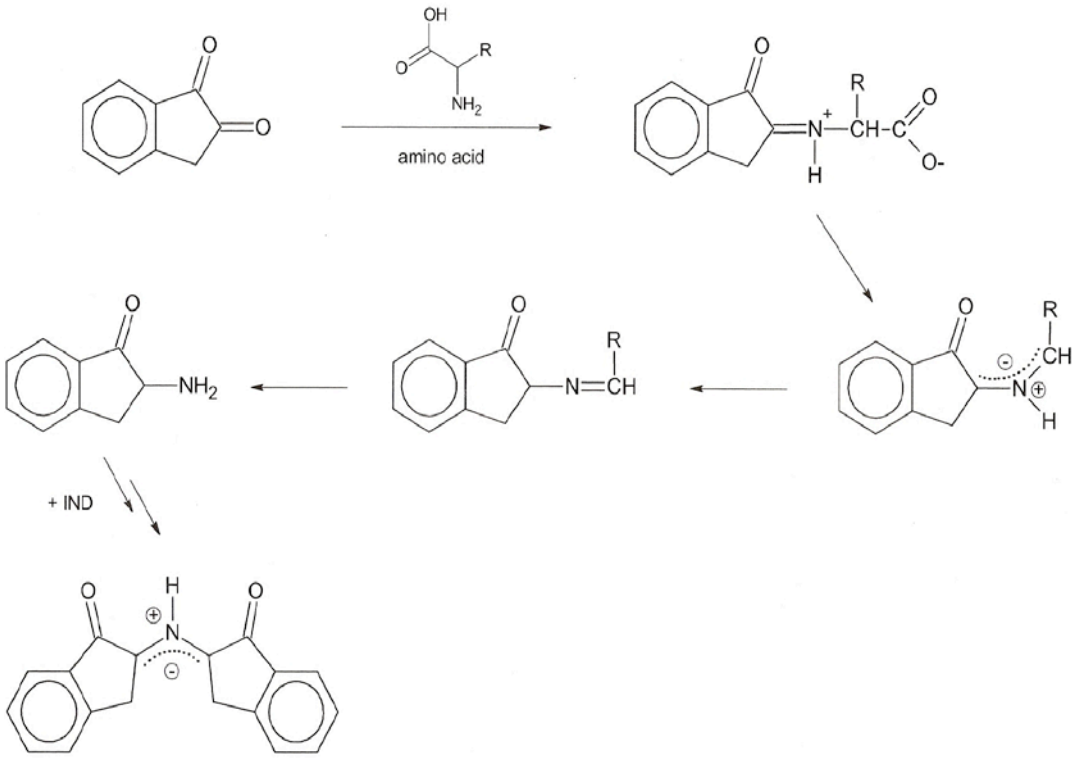
1.7 The lack of widespread use may partly be attributed to the variable results that were obtained worldwide, in some cases different cities in the same country giving very different results according to local weather conditions. As a consequence optimised formulations differed according to local humidity and environment. More recently, a potential solution to this problem has been identified. In early assessments of 1,2 indandione it was noted that the fluorescence may be enhanced by toning with metal salts in a similar manner to ninhydrin [3]. It has recently been shown that by adding zinc salts to the treatment solution the fluorescence of the mark can be enhanced without the need for a post-treatment and the resultant formulation is considerably more resilient to local fluctuations in humidity and environment [12,13]. Optimised formulations were developed for use under Australian conditions, with the best results claimed after hot-pressing at 165ºC for 10 seconds. The development of this formulation prompted further studies by HOSDB to see whether this offered a credible alternative to or replacement for DFO in the UK [14,15]. Comparative studies were carried out of the formulations recommended by Australian and US researchers in 2007 alongside the PSDB formula used in the late 1999 comparison with DFO, with zinc salts added into the formula. In this study the modified HOSDB formulation was found to be most effective and was compared with DFO in a further comparison. The results of this experiment are reported below, and showed the performance of 1,2 indandione-zinc to be closely equivalent to DFO.

1.8 Further work has since been carried out by the Australian and US research teams in further optimising formulations. Bicknell and Ramotowski in the US further refined the reagent formulation and found it to out-perform DFO [16]. They also observed that although the stability of the 1,2 indandione-zinc system to humidity fluctuations was much improved, the humidity level in the paper after dipping did have an influence on the subsequent development route. For papers below a critical humidity level (approximately 70%), treatment in a humidity oven using ninhydrin processing conditions was recommended, whereas for papers with humidity content above this level a dry oven and DFO processing conditions gave best results.

1.9 CAST is aware that the US researchers have produced a further modification to the 1,2 indandione-zinc formulation and have directly observed it to perform better than the HOSDB DFO formulation in small-scale comparative trials. This formulation is based on petroleum ether and would not be recommended for operational use in the UK, but could form the starting point for more reformulation studies. There has also been a further comparative trial between 1,2 indandione-zinc and DFO in Australia, which again reinforces the fact that 1,2 indandione-zinc may need further investigation to see if it could replace DFO. One element of these studies will be to assess whether the DFO-ninhydrin sequence gives more marks than 1,2 indandione-ninhydrin, because sequential processing must be considered in addition to the single most effective process.

2. Theory

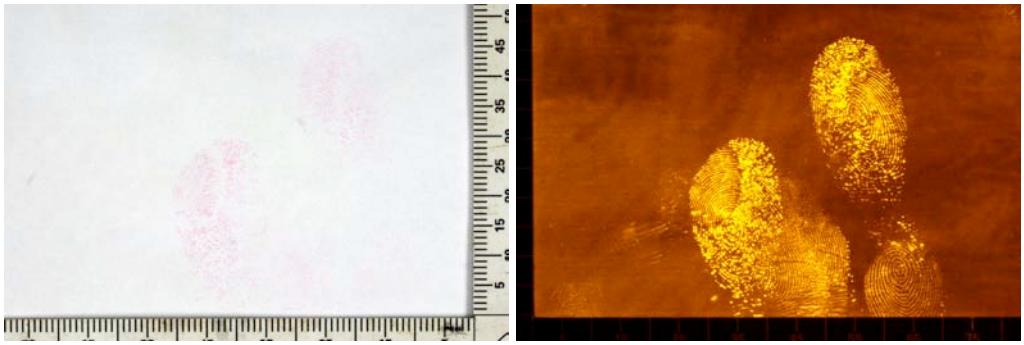
2.1 1,2 Indandione is closely related to ninhydrin and it has been proposed that its reaction with amino acids follows a very similar pathway, one suggested reaction path being illustrated below.



Proposed reaction pathway for 1,2 indandione with amino acids.

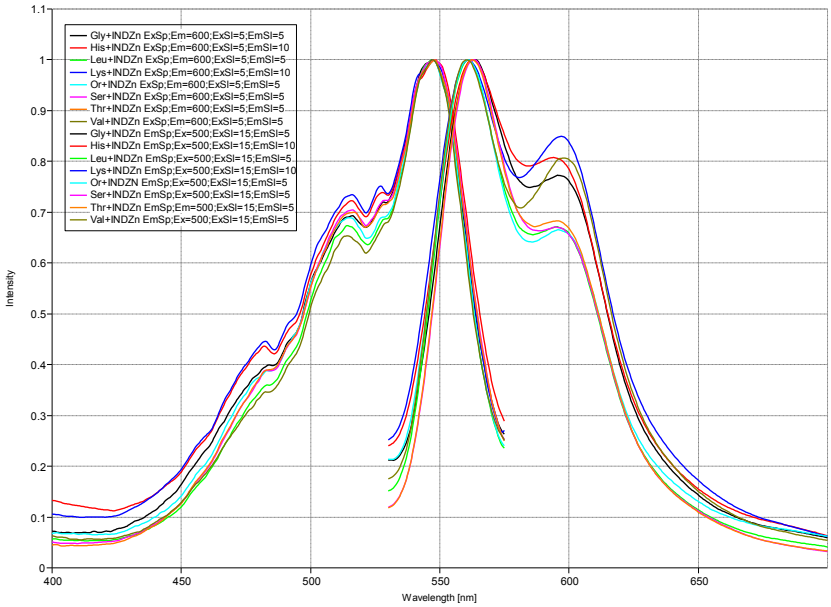
2.2 The proposed formation of a Ruhemann's purple analogue as shown above may account for the pink coloration seen for prints developed from formulations using high 1,2 indandione contents. However the product responsible for the fluorescence has not been conclusively identified.

2.3 Unlike DFO, methanol is not necessary for the reaction to proceed and may in fact inhibit it. This is because 1,2 indandione forms a stable hemiketal with methanol and this prevents the reaction with amino acids taking place.



a) *b)*

Fingerprints on paper developed using 1,2 indandione and imaged in a) reflected light and b) fluorescence mode.



Absorption and emission spectra measured for 1,2 indandione-zinc [17]

2.4 The addition of zinc to the formulation has been shown to give reaction products that are consistent in their excitation and emission spectra across a wide range of amino acids. This was not true of 1,2 indandione formulations without zinc salt additions [17] and it was proposed that the Zn^{2+} present in the solution has a catalytic effect in driving the formation of the fluorescent reaction product.

3. Reasons technique is not recommended by CAST

3.1 CAST does not currently (2011) recommend the use of 1,2 indandione because an extensive research programme including operational trials

has not demonstrated that it gives any improvements in effectiveness over the currently recommended DFO formulation. There have also been suggestions in previous published work that 1,2 indandione may not be as effective as DFO when used as part of sequential treatments [3,5] although other studies indicated that 1,2 indandione may develop more fingerprints than the DFO-ninhydrin sequence [5,12]. However, recent formulations incorporating zinc salts give improved performance and may need further evaluation.

3.2 A summary of the experiments performed and the results on which these conclusions are based is given below.

3.3 In the late 1990s/early 2000s, PSDB began a programme of work to optimise the 1,2 indandione formulation for use in UK conditions [6]. Observations from this work included:

- 0.25g 1,2 indandione per litre of solution gives the optimum fluorescence level in the developed mark. Higher concentrations can give a more intense pink colour, but in common with the DFO formulation, CAST regards fluorescence as the most important characteristic;
- 10 mL of acetic acid per litre of solution gives the optimum fluorescence of fingerprints without increasing undesirable background fluorescence to a level where it begins to obscure marks;
- 90 mL of ethyl acetate per litre of solution is added as a co-solvent;
- the solution is made up to 1 litre with 1-methoxynonafluorobutane (HFE7100), selected as a proven non-flammable, non-toxic solvent for fingerprint formulations.

3.4 It was also determined that the optimum processing conditions for maximum fluorescence were heating for 10 minutes at 100ºC without humidity, and that processed exhibits should be stored in the dark to maximise subsequent development of marks and to retain fluorescence. More recently, other researchers have suggested that equivalent (if not better) performance can be obtained by heating at higher temperatures (~160°C) for shorter times [12], but this has not yet been investigated by CAST.

3.5 Pseudo-operational trials were then conducted on batches of 75 cheques, comparing the optimised 1,2 indandione formulation with DFO. The results are shown in the graphs below.

Chart 1

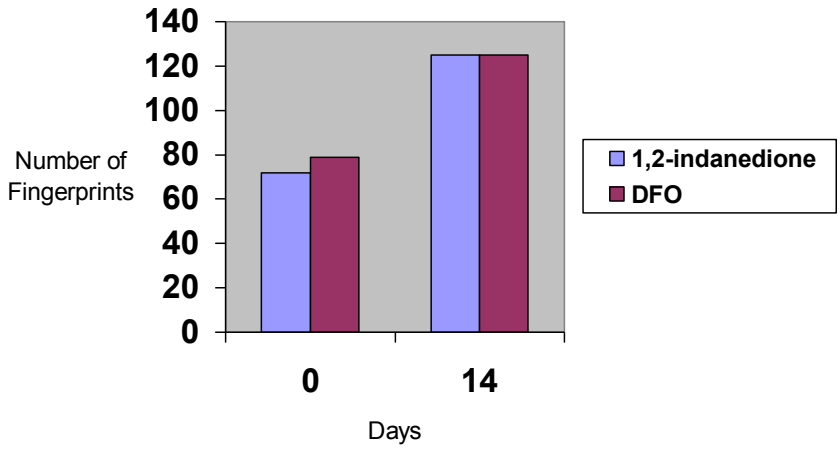
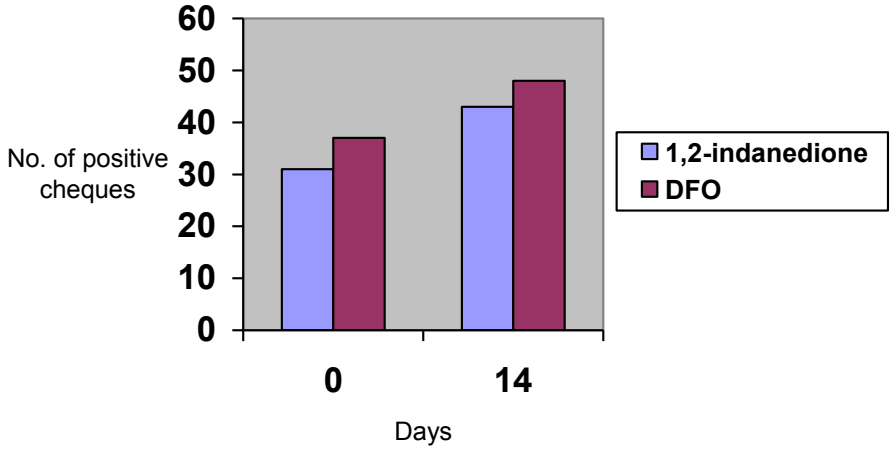


Chart 2



Number of fingerprints detected on cheques developed using 1,2 indandione and 1,8-diazafluoren-9-one processes and number of cheques yielding positive results.

3.6 These results indicated that the two systems were closely equivalent in performance, with DFO developing slightly more marks. A further pseudo-operational trial was conducted on a range of different porous exhibits, the results being tabulated below.

Article	Number of articles	Number of developed fingerprints	
		DFO	1,2 Indandione
White envelopes	20	41	44
Brown envelopes	15	17	33
Photocopy paper	20	92	82
Newspaper	20	2	2
Receipts	20	5	7
Train tickets	19	10	6
Total	**114**	**167**	**174**

Results of pseudo-operational trial on samples typical of porous exhibits encountered in casework.

3.7 These results were sufficiently encouraging to justify inclusion of 1,2 indandione in an operational trial of two ozone-friendly DFO formulations against the chlorofluorocarbon (CFC) 1,1,2-trifluorotrichloroethane (CFC113)-based DFO formulation [7]. In these trials the 1,2 indandione formulation proved least effective on operational work and was not pursued further. Similar results were obtained from an operational trial in Canada [8].

3.8 However, more recent publications from Australia [12,13] suggest stabilisation of the 1,2 indandione system to humidity by additions of zinc salts in solution. This necessitated a re-evaluation of the process and HOSDB carried out a further work programme with the objectives of identifying the optimum formulation with zinc salt additions and carrying out a comparative trial with DFO. In this trial18 different porous substrates were used, covering a range of different paper types.

Substrate	Brand	Size	Weight	Description
1	Tesco value	A4	75gsm	White copier paper
2	Woolworths	A4	80gsm	Multipurpose paper
3	WH Smith	A4	100gsm	Premium inkjet paper
4	XEROX	A4	80gsm	Laser copier paper
5	HP	A4	80gsm	Everyday inkjet paper
6	WH Smith	A5	–	Writing paper
7	PUKKA	A4	80gsm	Premium quality lined writing paper
8	Woolworths	A4	–	Premium pad lined
9	Tesco value	A4	–	Refill pad lined
10	Tesco	C4	–	White envelopes
11	Tesco value	C4	–	Brown envelopes
12	Woolworths	50cm x 5m	–	Brown paper

13	*TV Choice*	–	–	TV magazine
14	*Heat*	–	–	Magazine
15	*Sun*	–	–	Newspaper
16	Ryman	A4	–	Silk finish paper laser printers
17	Ryman	C4	130gsm	White envelopes
18	Ryman	A4	90gsm	Ivory parchment paper

Summary of porous surfaces used in comparative studies.

3.9 This study consisted of over 180 deposited marks per substrate, per condition examined. For experiments using all 18 substrates, greater than 180 x 18 prints were examined. The intensity of the fluorescence for developed marks was measured using a Minolta 100LS spot meter, and marks graded using a 0–4 grading scheme [18]. The fluorescence conditions used were illumination with the 473–548nm excitation band of a Quaser 40, viewed through a 549nm cut-on long-pass filter (Schott glass OG570).

3.10 Initial studies compared the effectiveness of 'optimised' formulations, including zinc salts developed by the Australian Federal Police, HOSDB in the UK, and the US Secret Service in the USA.

UK 1,2 indandione-zinc formulation

0.125g 1,2 indandione

45mL ethyl acetate

5mL acetic acid

0.25mL $ZnCl_2$ stock solution (0.2g $ZnCl_2$ in 5mL absolute ethanol)

500mL HFE7100.

USA 1,2 indandione-zinc formulation

0.5g 1,2 indandione

15mL dichloromethane

30mL ethyl acetate

5mL acetic acid

2mL $ZnCl_2$ stock solution

448mL petroleum ether.

Australia 1,2 indandione-zinc formulation

0.5g 1,2 indandione

15mL dichloromethane

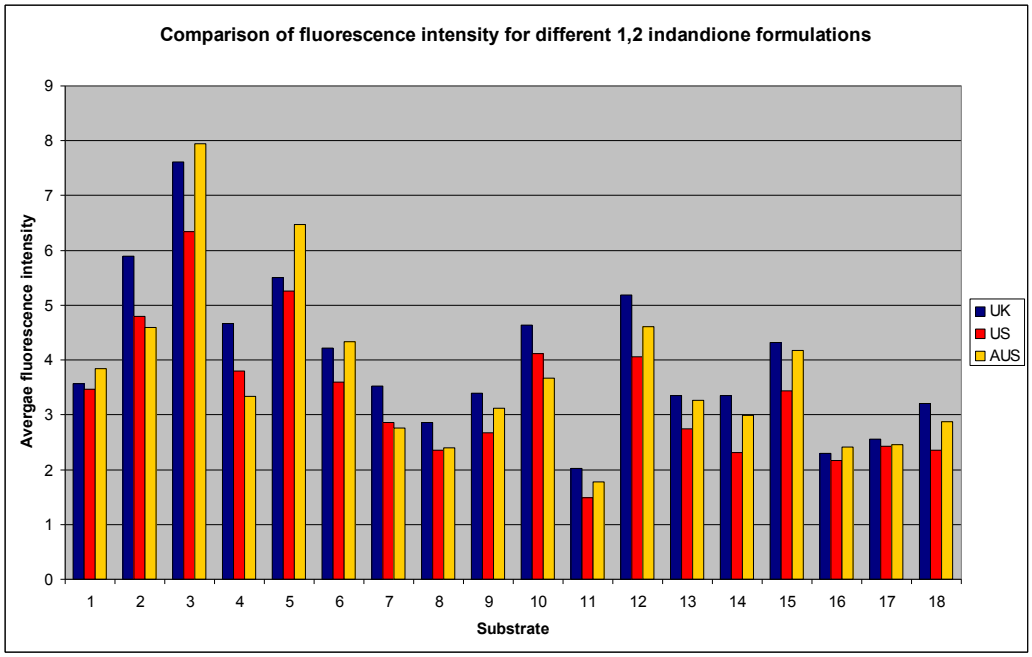
30mL ethyl acetate

5mL acetic acid

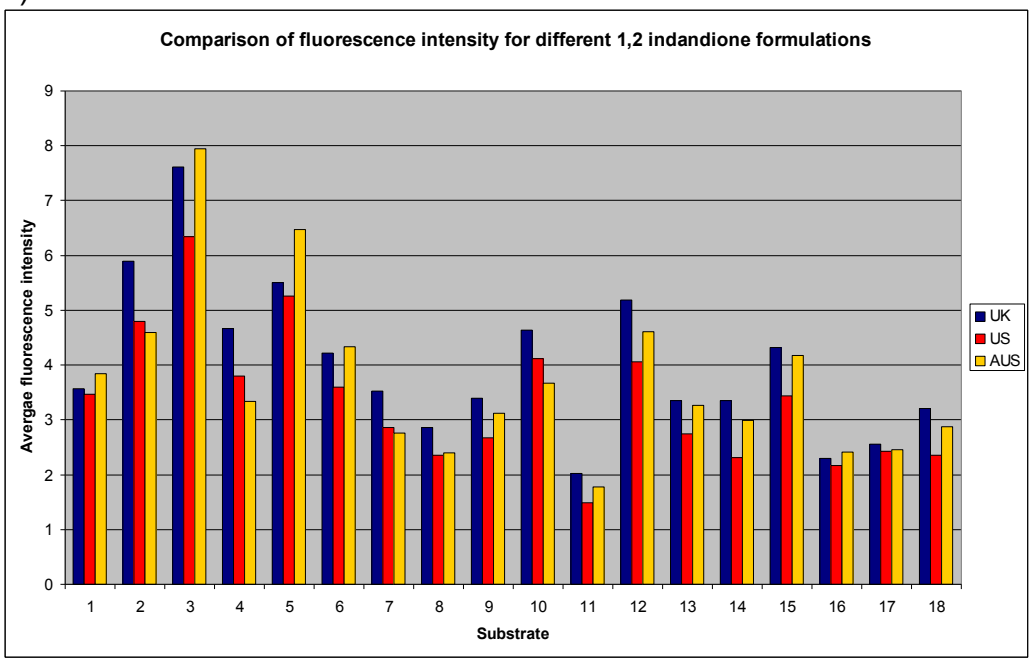
0.5mL $ZnCl_2$ stock solution

450mL HFE-7100.

3.11 The results of this pre-selection exercise are depicted graphically below.



a)



b)

Results of comparative trials between different 1,2 indandione formulations a) fingerprint quality and b) intensity of fluorescence.

3.12 From these trials it appeared that under UK conditions the HOSDB formulation gave the best performance in terms of both quality of fingerprints developed and intensity of fluorescence from the developed mark. The HOSDB formulation with zinc salt additions was therefore compared with the existing HOSDB DFO formulation, both formulations being given below.

UK 1,2 indandione-zinc formulation

0.125g 1,2 indandione

45mL ethyl acetate

5mL acetic acid

0.25mL $ZnCl_2$ stock solution

500mL HFE7100.

DFO formulation

0.25g DFO

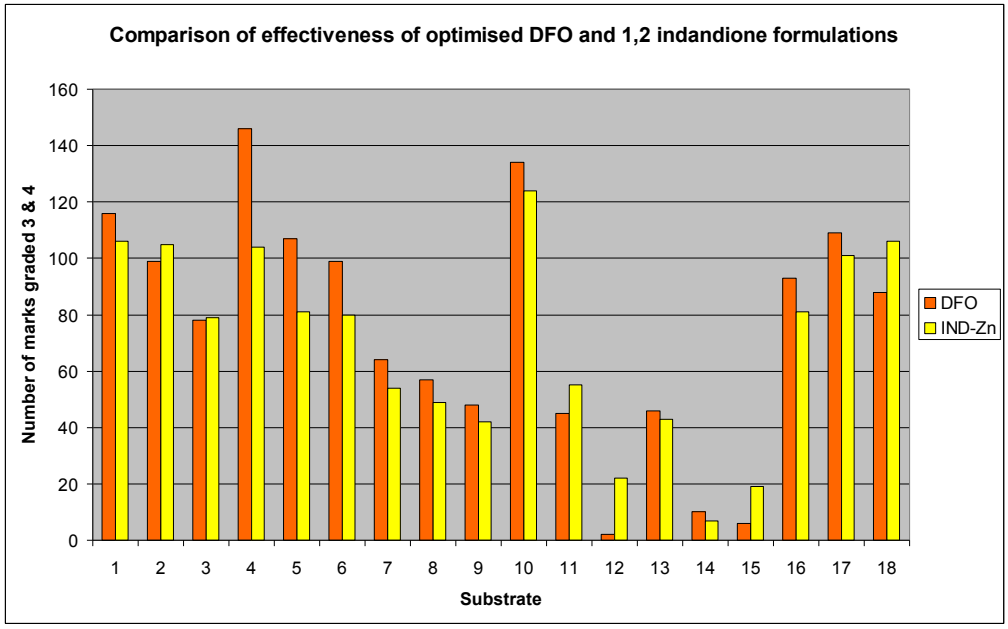
30mL methanol

20mL acetic acid

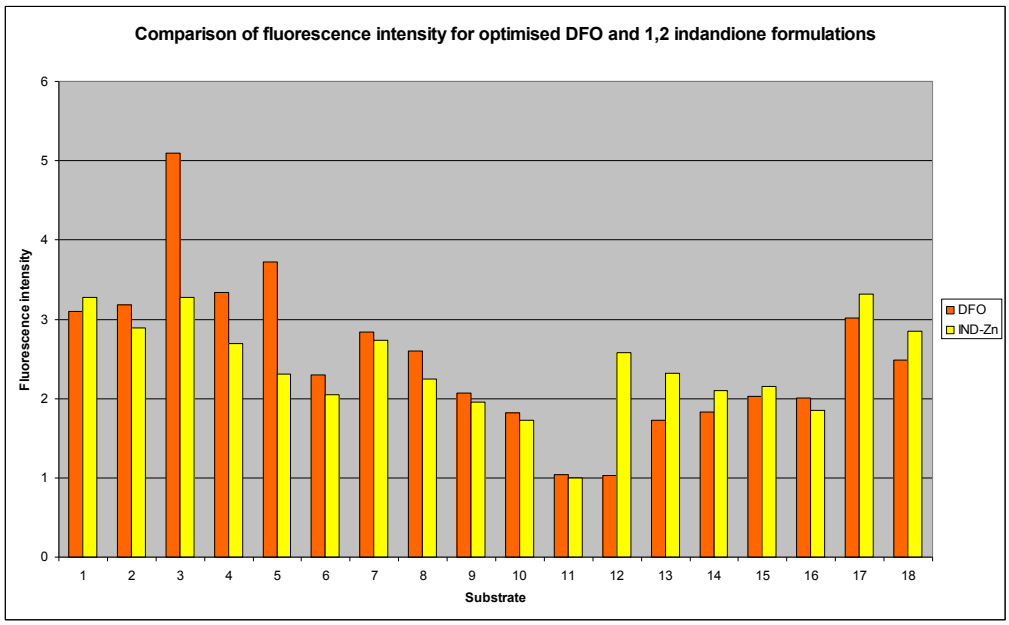
275mL HFE71DE (trans-1,2-dichloroethylene)

725mL HFE7100.

3.13 Once again, comparisons were made between the quality of the developed mark and intensity of fluorescence. Results from this comparison on two-day-old marks are shown in the graphs below.



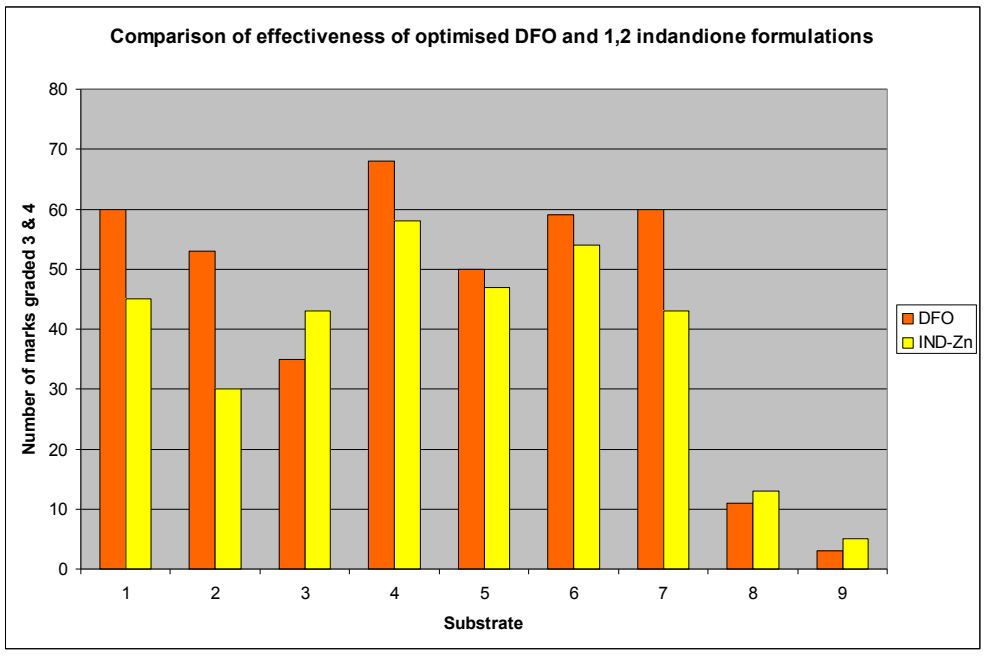
a)



b)

Results of comparative trials between 1,8-diazafluoren-9-one and 1,2 indandione formulations on two-day-old marks a) fingerprint quality and b) intensity of fluorescence.

3.14 In these trials the formulations give closely equivalent performance, with DFO giving marginally better results. An assessment on 14-day-old marks was commenced but it was not possible to complete the study in the time available. However, initial results (illustrated below) suggested a similar trend.



Results of comparative trials between 1,8-diazafluoren-9-one and 1,2 indandione formulations on 14–day-old marks, assessing fingerprint quality alone.

3.15 The results obtained showed no improvement in performance from 1,2 indandione over DFO and this, combined with reports that ninhydrin develops no additional marks after 1,2 indandione, but is known to do so after DFO, resulted in HOSDB recommending no further evaluation of 1,2 indandione. DFO was therefore retained as the recommended HOSDB process and the sequential treatment of DFO-ninhydrin-physical developer remained unchanged.

3.16 However, as outlined in the sections above subsequent studies have been carried out in Australia, which again found that 1,2 indandione-zinc outperformed DFO (in this case the HOSDB DFO formulation) under Australian conditions. Similarly, the US Secret Service has developed a revised 1,2 indandione-zinc formulation, which appears to give improved performance over the HOSDB DFO formulation in a limited study under UK conditions. Clearly, further work is required to see if 1,2 indandione-zinc has potential for replacing DFO in operational use.

4. References

1. **Ramotowski, R., Cantu, A. A., Joullie, M. M. and Petrovskaia, O.** (1997) '1,2-Indanediones: A Preliminary Evaluation of a New Class of Amino Acid Visualising Reagents', *Fingerprint Whorld*, vol. 23 (90), pp 131–140.

2. **Hauze, D. B., Petrovskaia, O., Taylor, B., Joullie, M. M., Ramotowski, R. and Cantu, A. A.** (1998) '1,2-indandiones: new reagents for visualising the amino acid components of latent prints', *J. Forens. Sci*, vol. 43 (4), pp 744–747.

3. **Roux, C., Jones, N., Lennard, C. and Stoilovic, M.** (2002) 'Evaluation of 1,2-Indanedione and 5,6-Dimethoxy-1,2-Indanedione for the detection of latent fingerprints on porous surfaces', *J. Forens. Sci.*, vol. 45 (4), pp 761–769.

4. **Almog, J., Springer, E., Wiesener, S., Frank, A., Khodzhaev, O. Lidor, R., Bahar, E., Varkony, H., Dayan, S. and Rozen, S.** (1999) 'Latent Fingerprint Visualisation by 1,2-indanedione and Related Compounds: Preliminary Results', *J. Forens, Sci*, vol. 44 (1), pp 114–118.

5. **Wiesener, S., Springer, E., Sasson, Y. and Almog, J.** (2001) 'Chemical Development of Latent Fingerprints: 1,2 Indandione Has Come of Age', *J. Forens. Sci*, vol. 46 (5), pp 1082–1084.

6. **Gardener, S. J. and Hewlett, D. F.** (2003) 'Optimisation and Initial Evaluation of 1,2 Indandione as a reagent for Fingerprint Detection', *J. Forens. Sci*, vol. 48 (6), pp 1288–1292.

7. **Merrick, S., Gardner, S., Sears, V. and Hewlett, D.** (2002) 'An Operational Trial of Ozone-friendly DFO and 1,2 Indandione Formulations for Latent Fingerprint Detection', *J. Forens. Ident*., vol. 52 (5), pp 595–605.

8. **Wilkinson, D., McKenzie, E., Leech, C., Mayowski, D., Bertrand, S. and Walker, T.** (2003) 'The Results from a Canadian National Field Trial Comparing Two Formulations of 1,8-Diazafluoren-9-one (DFO) with 1,2 Indandione', *Ident. Canada*, vol. 26 (2), pp 8–18.

9. **Stimac, J. T.** (2003) 'Thermal Paper: Latent Friction Ridge Development via 1,2-Indanedione', *J. Forens. Ident*., vol. 53 (3), pp 265–271.

10. **Azoury, M., Zamir, A., Oz, C. and Wiesner, S.** (2002) 'The Effect of 1,2-Indanedione, a Latent Fingerprint Reagent on Subsequent DNA Profiling', *J. Forens, Sci*, vol. 47 (3), pp 586–588.

11. **Wallace-Kunkel, C., Roux, C., Lennard, C. and Stoilovic, M.** (2004) 'The Detection and Enhancement of Latent Fingermarks on Porous Surfaces – A Survey', *J. Forens. Ident*., vol. 54 (6), pp 687–705.

12. **Wallace-Kunkel, C., Lennard, C., Stoilovic, M. and Roux, C.** (2007) 'Optimisation and Evaluation of 1,2-Indanedione for use as a Fingerprint Reagent and its Application to Real Samples', *Forens. Sci. Int*., vol. 168 (1), pp 14–26.

13. **Stoilovic, M., Lennard, C., Wallace-Kunkel, C. and Roux, C.** (2007) 'Evaluation of a 1,2-indanedione Formulation Containing Zinc Chloride for Improved Fingermark Detection on Paper', *J. Forens. Ident*., vol. 57 (1), pp 4–18

14. **Batham, R.** (2007) *Comparison of DFO with 1,2 Indandione Formulations containing Zinc*, MSc Thesis, September. University of Strathclyde.

15. **Sears, V., Batham, R. and Bleay, S.** (2009) 'The Effectiveness of 1,2 Indandione – Zinc Formulations and Comparison with HFE-Based 1,8-diazafluoren-9-one for Fingerprint Development', *J. Forens. Ident*., vol. 59 (6), pp 654–678.

16. **Bicknell, D. E. and Ramotowski, R. S.** (2008) 'Use of an Optimised 1,2 Indanedione Process for the Development of Latent Prints', *J. Forens. Sci*, vol. 53 (5), pp 1108–1116.

17. **Spindler, X., Stoilovic, M., Lennard, C. and Lennard, A.** (2009) 'Spectral Variations for Reaction Products Formed Between Different Amino Acids and Latent Fingermark Detection Reagents on a Range of Cellulose-Based Substrates', *J. Forens. Ident*., vol. 59 (3), pp 308–324.

18. **Sears, V. G., Bleay, S. M., Bandey, H. L. and Bowman, V. J.** (2011) 'A Methjodology for Finger Mark Research', accepted for publication in *Science and Justice*

5.8 Ninhydrin analogues

1. History

1.1 Many ninhydrin analogues have been synthesised, but the first concerted synthesis of such analogues for assessment as fingerprint reagents was carried out by Almog *et al.*[1] in the early 1980s. These studies identified benzo[f]ninhydrin as a reagent with potential for operational use, the reaction product being a dark green in colour.

1.2 Benzo[f]ninhydrin was first assessed in the UK by Jones and Pounds [2], who conducted a comparison of the new reagent with ninhydrin. These studies found that there was little difference in sensitivity between the two reagents, but benzo[f]ninhydrin was less soluble and the increased solvent levels required in the formulation caused ink to run. However, it was noted that benzo[f]ninhydrin may allow better distinction of marks on coloured backgrounds, because of the darker colour of the developed marks.

1.3 It was later found that benzo[f]ninhydrin could be treated with metal salts in a similar manner to ninhydrin to produce a fluorescent reaction product. An examination of zinc chloride ($ZnCl_2$)-toned benzo[f]ninhydrin marks was conducted using a neodymium:yttrium aluminium garnet (Nd:YAG) laser (green, 532nm), and these were found to be well-matched to the absorption spectrum of the toned mark [3].

1.4 In the mid-1980s, a wider range of ninhydrin analogues were synthesised including 5-methoxyninhydrin. These studies included an extensive investigation of the reactions between these analogues and metal salts, and the fluorescence characteristics of the reaction products [4]. The same researchers carried out further studies of the fluorescence produced from metal toning [5] and found that in this respect benzo[f]ninhydrin and 5-methoxyninhydrin were particularly useful. Both these compounds gave reaction products with more intense fluorescence than ninhydrin and fluorescence occurred at longer wavelengths, thus reducing problems associated with background fluorescence.

1.5 A further investigation into reactions of both ninhydrin analogues and related compounds with amino acids was carried out by Almog [6]. It was observed in these studies that only cyclic triketones gave coloured reaction products with amino acids, whereas open chained triketones did not.

1.6 The intense fluorescence from metal-toned 5-methoxyninhydrin was investigated using a copper-vapour laser [7,8]. Marks developed using this reagent had the same visible appearance as ninhydrin but were considerably more fluorescent when illuminated with the copper-vapour laser at 510.6nm. The laser was found to be the most appropriate light source for excitation of this fluorescence.

1.7 An extensive review of ninhydrin, its analogues and reactions was published by Joullie *et al.*[9]. However, the interest in developing ninhydrin analogues for the fluorescent properties of their reaction products did fall off with the introduction of 1,8-diazafluoren-9-one (DFO), which did not require a post-treatment to give fluorescent marks. One final class of ninhydrin analogues that were investigated were the thioninhydrins [10], which were found to give the most intense fluorescence from marks after metal toning than any other ninhydrin analogue.

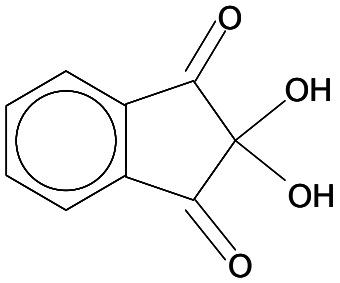
1.8 The Police Scientific Development Branch (PSDB) carried out limited evaluations on some ninhydrin analogues, including 5-methoxyninhydrin and 5-(2-thienyl) ninhydrin. The most comprehensive study was carried out on benzo[f]ninhydrin in collaboration with the Israeli National Police, comparing the effectiveness of the two reagents on bundles of cheques in a pseudo-operational trial [11]. It was found that ninhydrin gave significantly better results and therefore benzo[f]ninhydrin was not recommended for operational use in the UK.

1.9 Recently Israeli researchers have revisited the toning of ninhydrin analogues with metal salts, most notably by incorporating the metal salts into the formulation and eliminating the need for a post-treatment stage [12]. The analogues used in this study were 5-methoxyninhydrin (5-MN) and 5-methylthioninhydrin (5-MTN). It was reported that these 'dual action' reagents gave a more intense colorimetric reaction than ninhydrin, and the zinc toned marks of 5-MTN produced a fluorescent product of intensity equivalent to DFO.

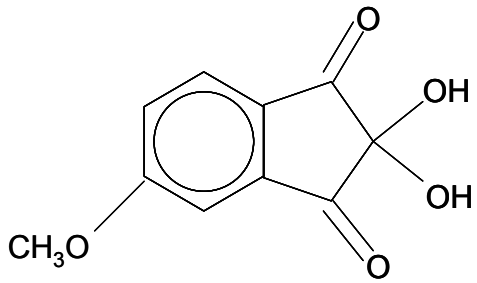
2. Theory

2.1 All ninhydrin analogues essentially follow a similar reaction path with amino acids to ninhydrin itself, and for those analogues that do form complexes with metal salts, the structures of these complexes are similar to those observed for ninhydrin.

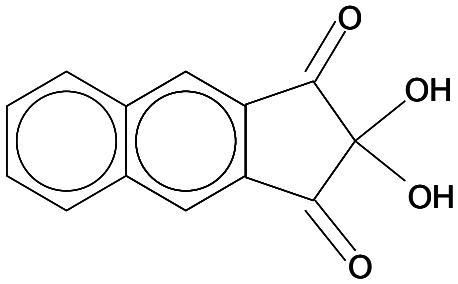
2.2 The structures of ninhydrin and the principal analogues that have been considered for fingerprint development are illustrated below.



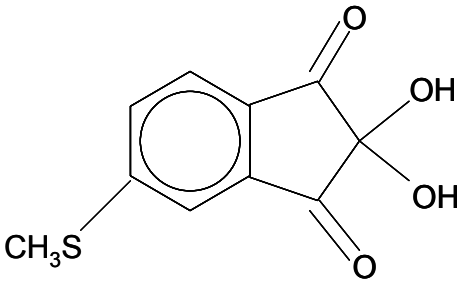
Ninhydrin



5-methoxyninhydrin



Benzo[f]ninhydrin



5-methylthioninhydrin

Structures of some of the principal ninhydrin analogues.

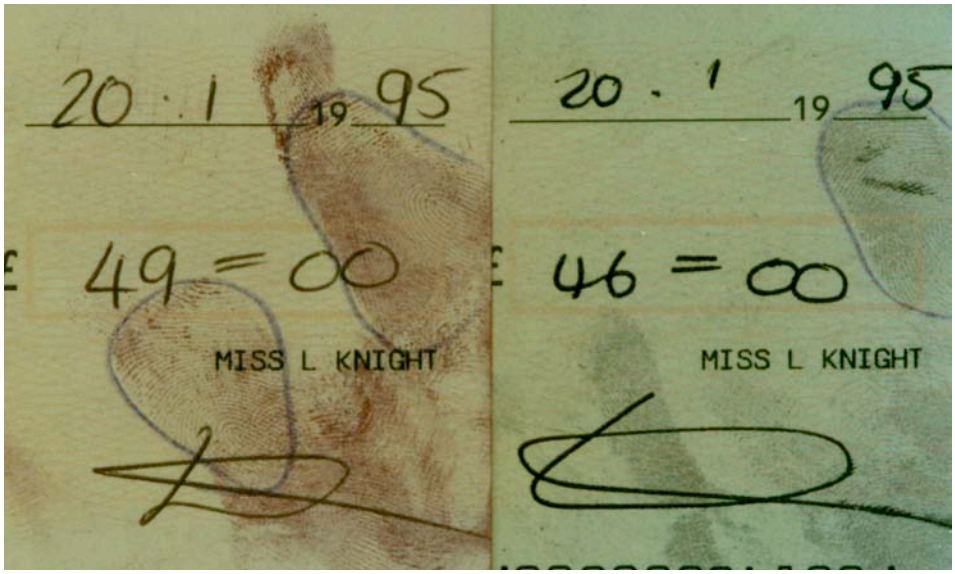
3. Reasons technique is not recommended by CAST

3.1 CAST does not currently (2011) recommend the use of ninhydrin analogues because those studied to date offer no performance benefits over ninhydrin itself. The two analogues that have generated the most interest, benzo[f]ninhydrin and 5-MN, may have niche applications, but in routine use are no more effective. However, the recent development of 5-MN and 5-MTN formulations incorporating metal salts [12] merits further study and may lead to one or both of these analogues being preferred over ninhydrin.

3.2 Both 5-MN (and 5-MTN) are of interest because they produce a more intensely fluorescent reaction product than ninhydrin when post-treated with metal salts. However, they are no more sensitive than ninhydrin and the visible reaction product is almost identical in colour. The requirement for intense fluorescence after metal toning reduced significantly with the introduction of reagents producing fluorescent products such as DFO and therefore it was not considered necessary to change from the currently recommended ninhydrin formulation. As stated above, the recent observation that metal salts can be incorporated into 5-MN and 5-MTN formulations rather than being used as an additional, post-treatment step may revive interest in these compounds. The development of intensely coloured, inherently fluorescent marks may

offer operational advantages and will be studied by CAST in the near future.

3.3 Benzo[f]ninhydrin has been of interest because it produces a grey-green reaction product, which may be easier to distinguish on coloured papers than the purple colour produced by ninhydrin. It also fluoresces at a longer wavelength after metal toning than ninhydrin, which again may be useful in distinguishing developed marks against background fluorescence.



a) b)

Comparison of reaction products produced with a) ninhydrin and b) benzo[f]ninhydrin.

3.4 However, in comparative trials between ninhydrin and benzo[f]ninhydrin, ninhydrin was found to be significantly more effective in terms of the numbers of fingerprints developed on batches of cheques from different banks [11]. A brief summary of this trial is given below.

3.5 The formulations used were as follows:

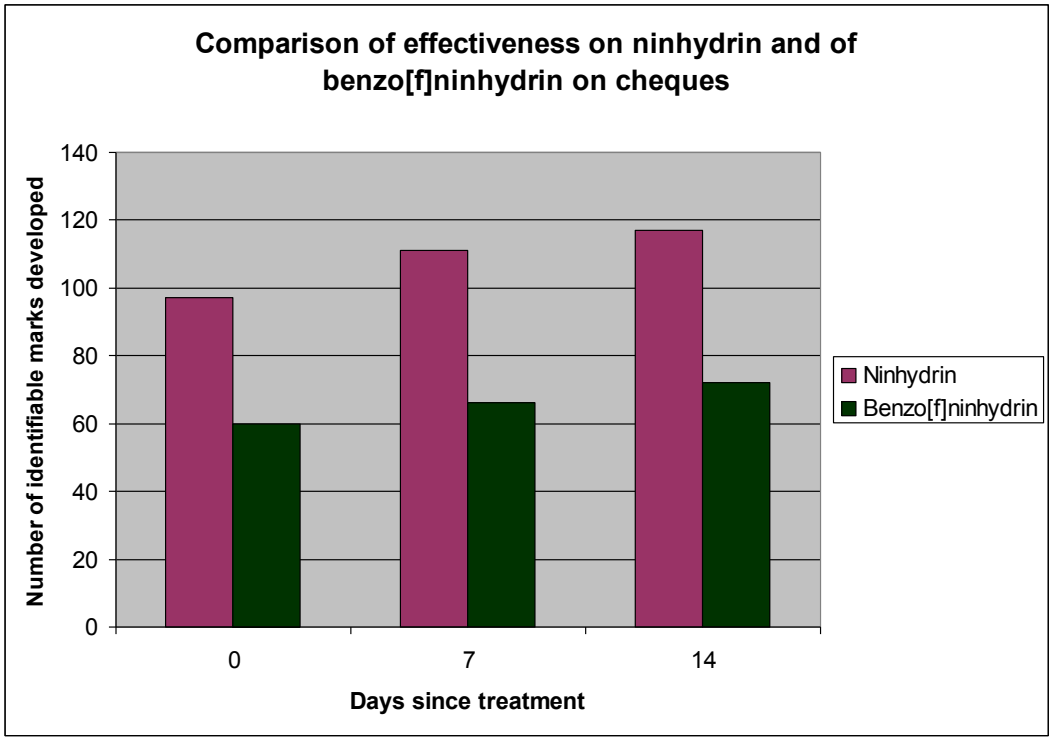
ninhydrin: 5g ninhydrin, 45mL ethanol, 5mL acetic acid, 2mL ethyl acetate, 1 litre 1-methoxynonafluorobutane (HFE7100);

benzo[f]ninhydrin: 6g benzo[f]ninhydrin, 60mL methanol, 30mL acetic acid, 60mL methyl acetate, 850mL 1,1,2-trifluorotrichloroethane (CFC113).

3.6 The numbers of fingerprints containing more than eight points developed using each process is recorded in the table below, and also shown graphically.

Days since treatment	Number of fingerprints							
	Ninhydrin (HFE7100)				Benzo[f]ninhydrin (CFC113)			
	B	M	N	Total	B	M	N	Total
0	22	41	34	97	12	21	27	60
7	28	46	37	111	13	23	30	66
14	30	50	37	117	15	26	31	72

Number of fingerprints developed on bundles of fraudulently passed cheques (B = Barclays, M = Midland, N = Natwest).



Total number of fingerprints developed on bundles of 75 fraudulently passed cheques.

3.7 As can be seen, the results do not justify the operational use of benzo[f]ninhydrin on grounds of effectiveness, although there may be niche applications, such as the development of marks on coloured surfaces.

4. References

1. **Almog, J., Hirshfield, A. and Klug, J. T.** (1982) 'Reagents for the Chemical Development of Latent Fingerprints: Synthesis and Properties of Some Ninhydrin Analogues', *J. Forens. Sci*, vol. 27 (4), pp 912–917.

2. **Jones, R. J. and Pounds, C. A.** (1982) *Comparison of Ninhydrin and Benzoninhydrin for Developing Latent Fingerprints on Paper Surfaces*, HO CRE Fingerprint Report No. 10, December. London: Home Office.

3. **Menzel, E. R. and Almog, J.** (1985) 'Latent Fingerprint Development by Frequency Doubled Neodymium: Yttrium Aluminium Garnet (Nd:YAG) Laser: Benzo[f]ninhydrin', *J. Forens. Sci*, vol. 30 (2), (1985) pp 371–382.

4. **Lennard, C. J., Margot, P. A., Stoilovic, M. and Warrener, R. N.** (1986) 'Synthesis of Ninhydrin Analogues and Their Application to Fingerprint Development: Preliminary Results', *J. Forens. Sci. Soc*., vol. 26, pp 323–328.

5. **Lennard, C. J., Margot, P. A., Stoilovic, M. and Warrener, R. N.** (1988) 'Synthesis and Evaluation of Ninhydrin Analogues as Reagents for the Development of Latent Fingerprints on Paper Surfaces', *J. Forens. Sci. Soc*., vol. 28, pp 3–23.

6. **Almog, J.** (1987) 'Reagents for Chemical Development of Latent Fingerprints: Vicinal Triketones – Their Reaction with Amino Acids and with Latent Fingerprints on Paper', *J. Forens. Sci*, vol. 32 (6), pp 1565–1573.

7. **Almog, J. and Hirshfield, A.** (1988) '5-Methoxyninhydrin: A Reagent for the Chemical Development of Latent Fingerprints That is Compatible with the Copper-Laser Laser', *J. Forens. Sci*, vol. 33 (4), pp 1027–1030.

8. **Menzel, E. R.** (1989) 'Detection of Latent Fingerprints by Laser-excited Luminescence', *Anal. Chem*., vol. 61 (8), pp 557–561.

9. **Joullie, M. M., Thompson, T. R. and Nemeroff, N. H.** (1991) 'Ninhydrin and Ninhydrin Analogs. Syntheses and Applications', *Tetra.*, vol. 47 (42), pp 8791–8830.

10. **Almog, J., Hirshfield, A., Frank, A., Grant, H., Harel, Z. and Ittah, Y.** (1992) '5-Methylthio Ninhydrin and Related Compounds: A Novel Class of Fluorogenic Fingerprint Reagents', *J. Forens. Sci*, vol. 37 (3), pp 688–694.

11. **Almog, J., Sears, V. G., Springer, E., Hewlett, D. F., Walker, S., Wisener, S., Lidor, R. and Bahar, E.** (2000) 'Reagents for the Chemical Development of Latent Fingerprints: Scope and Limitations of Benzo[f]ninhydrin in Comparison to Ninhydrin', *J. Forens. Sci*, vol. 45 (3), pp 538–544.

12. **Almog, J., Klein, A., Davidi, I., Cohen, Y., Azoury, M. and Levin-Elad, M.** (2008) 'Dual Fingerprint Reagents with Enhanced Sensitivity: 5-Methoxy- and 5-Methylthioninhydrin', *J. Forens. Sci*, vol. 53 (2), pp 364–367.

5.9 Miscellaneous amino acid reagents:

5.9.1 Fluorescamine

1. History

1.1 Fluorescamine (4-phenylspiro[furan-2(3H), 1'-phthalan]-3,3'-dione) was developed in the early 1970s as a fluorescent reagent for automated assay of amino acids [1,2]. This was based on earlier work showing that fluorescent products could be obtained when treating phenylalanine with ninhydrin and a peptide. By deducing the structure of these fluorescent products, it was possible to identify a novel reagent (fluorescamine) that would react directly with primary amines to give the same fluorescent reaction products. Tests demonstrated that fluorescamine was capable of detecting both amino acids and peptides and had a high level of sensitivity.

1.2 Several studies were carried out to compare the sensitivity of fluorescamine, ninhydrin and o-phthaldialdehyde (another reagent proposed for assay of amino acids). These concluded that for detection of most free amino acids, fluorescamine offered no advantages over ninhydrin. However, for recovery of peptides, fluorescamine did appear to work over a wider range of substances than ninhydrin [3].

1.3 The reagent also became considered as an alternative to ninhydrin for the development of fingerprints on porous surfaces. However, initial tests indicated that the aqueous buffer required to provide the optimal pH environment washed out the fingerprint ridge detail and therefore organic bases were investigated as alternative ways of providing an alkaline environment. A suitable formulation was developed based on fluorescamine dissolved in acetone with addition of triethylamine [4].

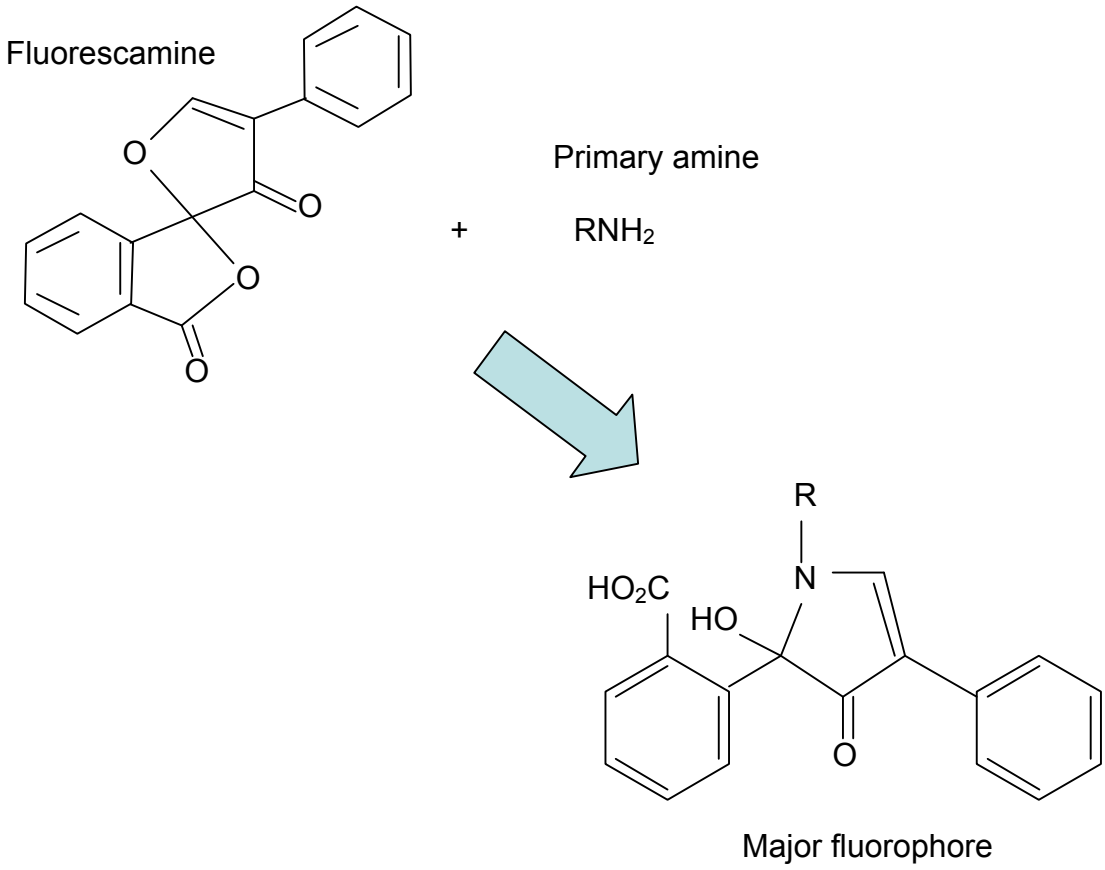
1.4 This formulation was then compared with ninhydrin and an optimised formulation of o-phthaldialdehyde for the detection of fingerprints was deposited on a range of surfaces, all reagents being applied as sprays [5]. These studies indicated that fluorescamine had some advantages over ninhydrin, including greater sensitivity, ability for mark detection on dark and multicoloured surfaces, and the fact that heat is not required for the reaction to occur. However, there were also some disadvantages: the solution does not have long-term stability and water will hydrolyse fluorescamine to a non-fluorescent product; in addition, ultraviolet (UV) light is required to visualise developed marks.

1.5 The reagent does not appear to have become widely used for fingerprint detection, possibly because of the greater ease in visualising the purple marks produced by ninhydrin and the fact that ninhydrin solution is more stable for long-term storage. The increasing use of optical brighteners in papers also means that many such surfaces now fluoresce a bright blue when illuminated with UV light, and this will swamp the weaker, pale blue fluorescence of any marks developed using fluorescamine. The

technique is therefore no longer appropriate for the types of surface that it was originally intended for.

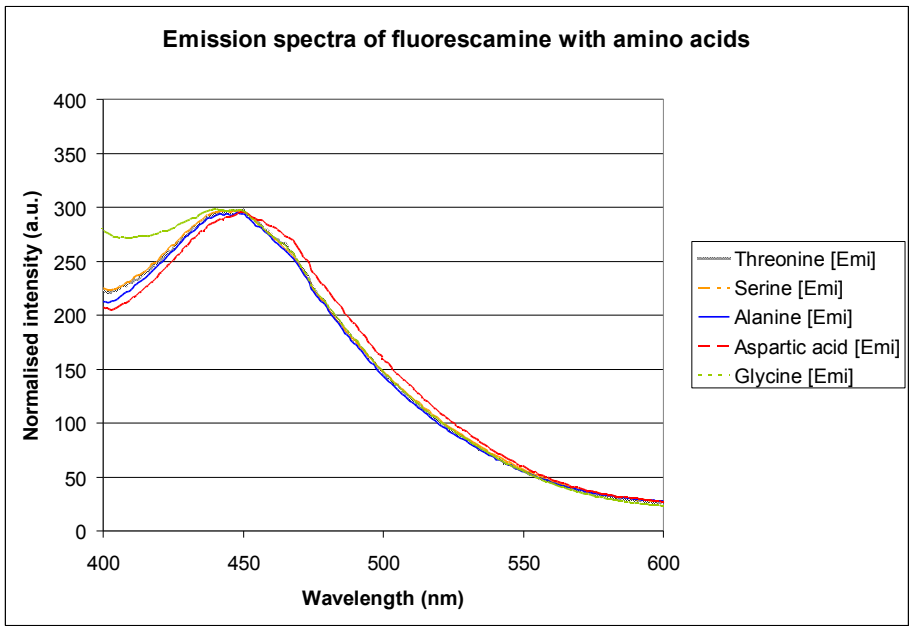
2. Theory

2.1 The way in which fluorescamine works is by a chemical reaction between the fluorescamine molecule and the amine groups present in amino acids and peptides to give a fluorescent reaction product. This is illustrated below:



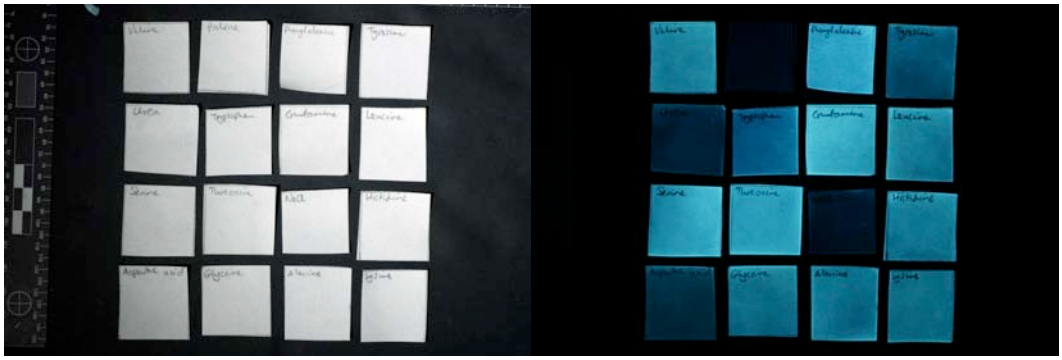
Reaction of fluorescamine with amines to form fluorescent products,

2.2 The major fluorophore produced by this reaction can be best visualised using an excitation wavelength ~390nm (long-wave UV) that produces a visible emission in the region 475–495nm.



Emission spectra of reaction products of fluorescamine with amino acids.

Palm print developed on painted wall using fluorescamine.



a) b)

Reaction products formed between fluorescamine and 0.1M solutions of amino acids and other fingerprint constituents a) visible and b) fluorescence.

2.3 The formulations proposed for use in the late 1970s utilised acetone as the principal solvent, small additions of triethylamine as an organic base, and fluorescamine. One such formulation is given below [6].

15mg fluorescamine
100mL acetone
0.1mL triethylamine.

2.4 These constituents were mixed together and then sprayed using an atomiser onto the surface being treated.

3. Reasons technique is not recommended by CAST

3.1 Although the technique was evaluated in the late 1970s, the Home Office Centre for Applied Science and Technology (CAST) has not recently conducted any extensive trials to compare fluorescamine with ninhydrin and/or 1,8-diazafluoren-9-one (DFO) and therefore it is not known whether there are any operational benefits in its use. However, reformulation work would be required to change the base solvent for fluorescamine from acetone to another less flammable substance less likely to cause ink to run and affect any subsequent document analysis, e.g. 1-methoxynonafluorobutane (HFE7100). Some components of the original formulation (such as dichloromethane) also have health and safety issues associated with them and alternatives would need to be identified. The solution is also unstable in contact with water and is more difficult to store than ninhydrin.

3.2 The fact that long-wave UV is required to visualise the developed fingerprints also makes fluorescamine less attractive for operational use. Extended usage of long-wave UV light sources does have health and safety implications for the operator and many modern papers also contain optical brighteners that are excited by long-wave UV, making developed marks more difficult to see against the fluorescing background.

4. References

1. **Udenfriend, S.** (1972) 'Development of a New Fluorescent Reagent and Its Application to the Automated Assay of Amino Acids and Peptides at the Picomole Level', *J. Res. Nat. Bur. Stan., – A. Phys. & Chem*., vol. 76A (6), pp 637–640.

2. **Udenfriend, S., Stein, S., Bohlen, P., Dairman, W., Leimgruber, W. and Weigele, M.** (1972) 'Fluorescamine: A Reagent for Assay of Amino Acids, Peptides, Proteins, and Primary Amines in the Picomole Range', *Sci.*, vol. 178, pp 871–872.

3. **Schiltz, E., Schnackerz, K. D. and Gracy, R. W.** (1977) 'Comparison of Ninhydrin, Fluorescamine, and o-Phthaldialdehyde for the Detection of Amino Acids and peptides and Their Effects on the Recovery and Composition of Peptides from Thin-Layer Fingerprints', *Anal. Biochem*., vol. 79, pp 33–41.

4. **Attard, A. E. and Lee, H. C.** (undated) *Fluorescamine: A New Reagent For Detection of Latent Fingerprints*, Undated Report, Forensic Science Laboratory. University of New Haven, Connecticut, USA

5. **Lee, H. C. and Attard, A. E.** (1979) 'Comparison of Fluorescamine, O-phthalaldehyde, and Ninhydrin for the Detection and Visualization of Latent Fingerprints', *J. Police Sci. & Admin*., vol. 7 (3), pp 333–335.

6. **Ohki, H.** (1976) 'A New Detection Method for Latent Fingerprints with Fluorescamine', *Natl. Res. Inst. of Police Sci*., vol. 29, pp 46–47.

5.9.2 O-phthaldialdehyde

1. History

1.1 O-phthaldialdehyde is another reagent originally developed for assay of amino acids in the early 1970s. Initially, o-phthaldialdehyde was not found to be as effective as ninhydrin or fluorescamine for detection of peptides, but by the mid-1970s revised formulations were published that were stated to overcome these issues [1]. The authors suggested that o-phthaldialdehyde was actually preferable to fluorescamine for fingerprint development because it exhibited greater fluorescent quantum yields, was stable in aqueous buffers, and was cheaper.

1.2 Similarly to fluorescamine, work was carried out to adapt the assay formulations for the development of fingerprints. One reported study investigated the use of a Babington nebuliser to provide a means of delivering o-phthaldialdehyde to large areas with saturating the surface [2]. In this formulation boric acid and potassium hydroxide were used as

a buffer solution, with additions of a detergent (Brij 35) and 2-mercaptoethanol.

1.3 O-phthaldialdehyde was also compared with ninhydrin and fluorescamine in spray reagent form. No single reagent out-performed the others in all respects, with o-phthaldialdehyde performing well in terms of sensitivity but suffering from a complex formulation and application procedure coupled with lack of stability in air [3].

1.4 Alternatives to the boric acid/potassium hydroxide buffer solution were investigated, this being found to cause diffusion of ridge detail. Ohki reported a formulation based on chloroform, triethylamine and 2-mercaptoethanol that overcame this problem [4].

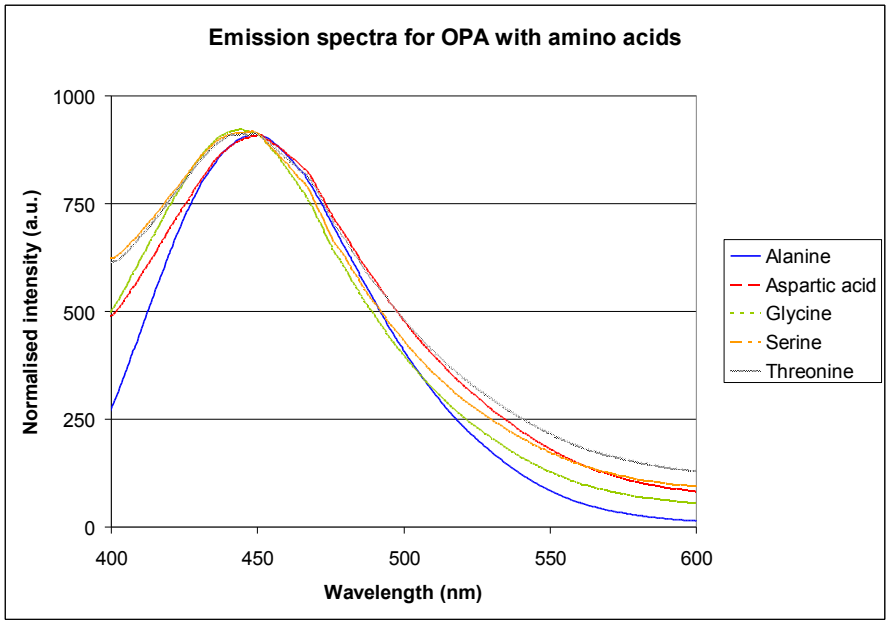
1.5 Subsequently Fischer [5] investigated a simpler and less hazardous formulation that involved dissolving o-phthaldialdehyde in acetone, dipping the exhibit and then lightly spraying with a 1% nitric acid solution in acetone. The fluorescent products produced in this way were excited with blue/green light rather than ultraviolet (UV).

1.6 The reagent does not appear to have become widely used for fingerprint detection, possibly because of the greater ease in visualising the purple marks produced by ninhydrin. The increasing use of optical brighteners in papers also mean that many such surfaces now fluoresce a bright blue when illuminated with UV light, and this will swamp the weaker, pale blue fluorescence of any marks developed using o-phthaldialdehyde. The technique is therefore no longer appropriate for the types of surface it was originally intended for.

2. Theory

2.1 O-phthaldialdehyde undergoes a chemical reaction with primary amines that may be present in fingerprint deposits to form fluorescent reaction products. The reaction products have an optimum excitation wavelength of ~340nm and an emission ~455nm.

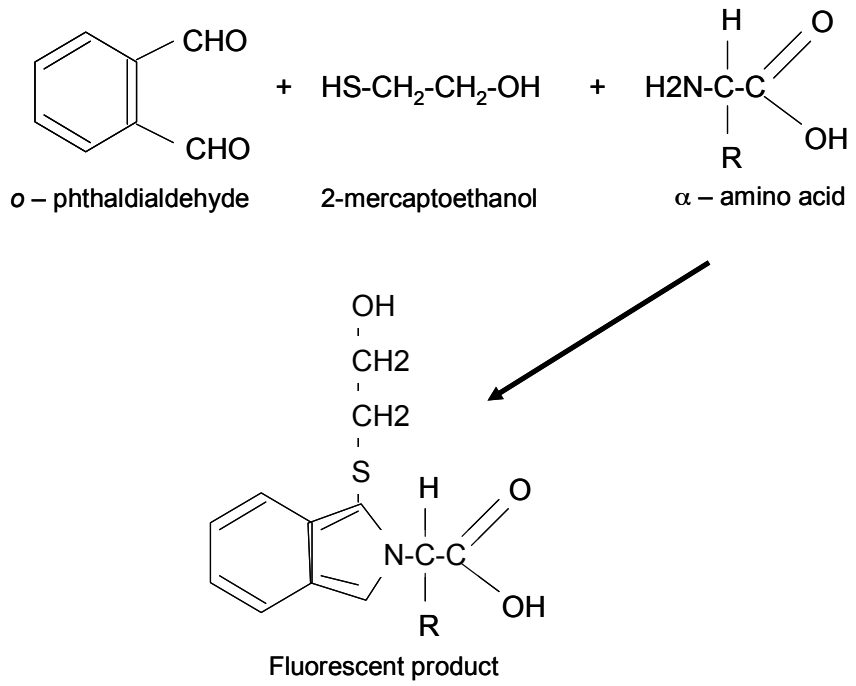
Fingerprint developed on filter paper using o-phthaldialdehyde.



Emission spectra for reaction products of o-phthaldialdehyde (OPA) with amino acids.

2.2 Research has indicated that the fluorescent reaction products are 1-alkylthio-2-alkyl-substituted isoindoles [6,7,8].

2.3 Some of the reactions proposed for o-phthaldialdehyde are given below.



Proposed reaction between o-phthaldialdehyde, 2-mercaptoethanol and α-amino acids [8]

2.4 Lee and Attard [3] proposed a two-part formulation with an aqueous base, where solution A comprised:

2.5g boric acid
95mL distilled water
pH adjusted to 10.40 with additions of 6M potassium hydroxide
0.3mL Brij 35 detergent
0.2mL 2-mercaptoethanol;

and solution B comprised:

0.5g o-phthaldialdehyde
1mL methanol.

The solutions were mixed together and then sprayed.

2.5 Ohki [4] proposed an alternative, one-part solution with an organic base:

40mg o-phthaldialdehyde
1mL 95% ethanol
50mL chloroform
0.5mL triethylamine
0.1mL 2-mercaptoethanol.

Again, the solution was sprayed onto the surface being treated.

3. Reasons technique is not recommended by CAST

3.1 CAST has not recently (since the late 1970s) conducted any extensive trials to compare o-phthaldialdehyde with ninhydrin and/or 1,8-diazafluoren-9-one (DFO) and therefore it is not known whether there are any operational benefits in its use. However, most o-phthaldialdehyde formulations are based on 2-mercaptoethanol, which is toxic, corrosive and dangerous for the environment and therefore it is unlikely that any formulation based on this substance would be recommended for operational use for health and safety reasons. Alternatives are available, but this would require extensive reformulation work for little operational benefit.

3.2 In common with fluorescamine, there is the problem that long-wave UV is required to visualise the developed marks and this brings with it health and safety issues associated with long exposures, and also interference with the developed mark from background paper fluorescence. The solution is also unstable in contact with air and may need to be stored under an inert gas, making it impractical for routine use.

4. References

1. **Benson, J. R. and Hare, P. E.** (1975) ‘o-Phthalaldehyde: Fluorogenic Detection of Primary Amines in the Picomole Range. Comparison with Fluorescamine and Ninhydrin’, *Proc. Nat. Acad. Sci. USA*, vol. 72 (2), pp 619–622.

2. **Mayer, S. W., Meilleur, C. P. and Jones, P. F.** (1978) ‘The Use of Ortho-phthalaldehyde for Superior Fluorescent Visualization of Latent Fingerprints’, *J. Forens. Sci. Soc*., vol. 18, pp 233–235.

3. **Lee, H. C. and Attard, A. E.** (1979) ‘Comparison of Fluorescamine, O-phthalaldehyde, and Ninhydrin for the Detection and Visualization of Latent Fingerprints’, *J. Police Sci. & Admin*., vol. 7 (3), pp 333–335.

4. **Ohki, H.** (1978) ‘A New Detection Method for Latent Fingerprints with O-Phthalaldehyde’, *Rep. Nat. Res. Inst. Police Sci.*, vol. 31 (4), pp 295–300.

5. **Fischer, J. F.** (1990) ‘A Modified o-Phthalaldehyde Technique Utilizing Blue-Green Light Excitation for Developing Luminescent Latent Prints’, *J. Forens. Ident.*, vol. 40 (6), pp 327–333.

6. **Simons, S. S. and Johnson, D. F.** (1976) ‘The Structure of the Fluorescent Adduct Formed in the Reaction of Ortho-Phthalaldehyde and Thiols with Amines’, *J. Am. Chem. Soc*., vol. 98, pp 7098–7099.

7. **Svedas, V. J. K., Galaev, I. J., Borisov, I. L. and Berezin, I. V.** (1980) 'The Interaction of Amino Acids with O-Phthaldialdehyde; A Kinetic Study and Spectrophotometric Assay of the Reaction Product', *Anal. Biochem*., vol. 101, pp 188–195.

8. **Lee, K. S. and Drescher, D. G.** (1978) 'Fluorometric Amino-Acid Analysis with o-Phthadialdehyde (OPA)', *Int. J. Biochem*., vol. 9, pp 457–467.

5.9.3 Genipin and lawsone

1. History

1.1 Genipin is a natural product that can be extracted from the fruit of Gardenia jasminoides. Since the 1960s it has been recognised that genipin brought into contact with skin produces an indelible blue-violet colour and that the same reaction readily occurs with amino acids [1]. However, the potential of such systems for the development of latent fingerprints was only explored in the mid-2000s when Almog *et. al*. [1] demonstrated that genipin could be mixed into solution with solvents such as 1-methoxynonafluorobutane (HFE7100) and petroleum ether, and the resultant formulations used to develop fingerprints on porous surfaces. It was also noted that in addition to the colorimetric reaction giving developed fingerprints a blue/black colour, the reaction products were also fluorescent with maximum emission at the red end of the spectrum.

1.2 Further experiments were carried out to establish optimum processing conditions for genipin, to compare its sensitivity with both ninhydrin and 1,8-diazafluoren-9-one (DFO) and to look at the reaction products formed between genipin and a range of amino acids [2]. The studies of these exercises identified a formulation based on genipin dissolved in ethanol/ethyl acetate and diluted using HFE7100, which could be used on documents without causing inks to run. It was found that genipin was slightly less sensitive than ninhydrin when considering the coloured reaction product, and less sensitive than DFO when considering the fluorescent reaction product. However, unlike DFO the emission spectra obtained from reaction products with a range of amino acids differed slightly from each other. On some types of paper genipin gave advantages over DFO because the longer wavelength fluorescent product reduced interference from background fluorescence of the paper and/or inks.

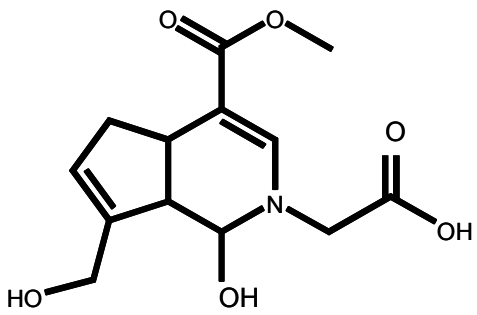
1.3 More rigorous comparative testing against other reagents with dual colorimetric and fluorescent reaction products, e.g. ninhydrin combined with metal salts [3] confirmed that genipin was, on the whole, less sensitive than such reagents, but that this longer wavelength fluorescence could make genipin the reagent of choice on brown paper

articles where background fluorescence may cause problems in imaging developed marks.

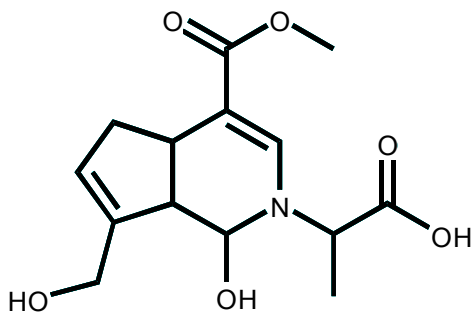
1.4 The research into genipin has since prompted research into other naturally occurring products that could be used as fingerprint development reagents, and information has recently been published on lawsone (2-hydroxy-1,4-napthoquinone), a component of henna [4]. This gives purple-brown marks with a red fluorescence when reacting with the amino acids in fingerprints. Further developments based on naturally occurring products are anticipated.

2. Theory

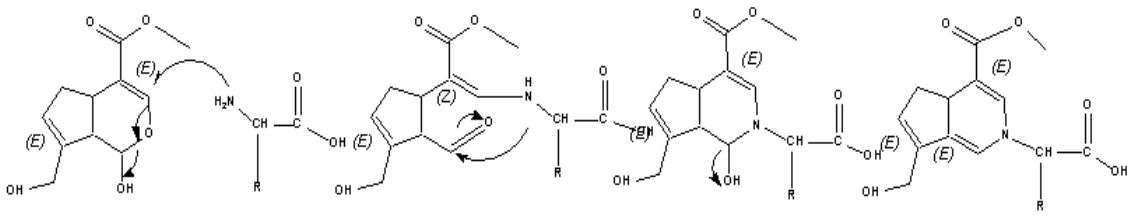
2.1 The mechanism of the reaction between genipin and amino acids and the nature of the coloured and fluorescent reaction products has not yet been established. The studies above [2] have established that slightly different reactions will occur between genipin and the different amino acids present in the fingerprint. Some of the blue reaction products produced with amino acids have been identified and the proposed reaction and structures are shown below.



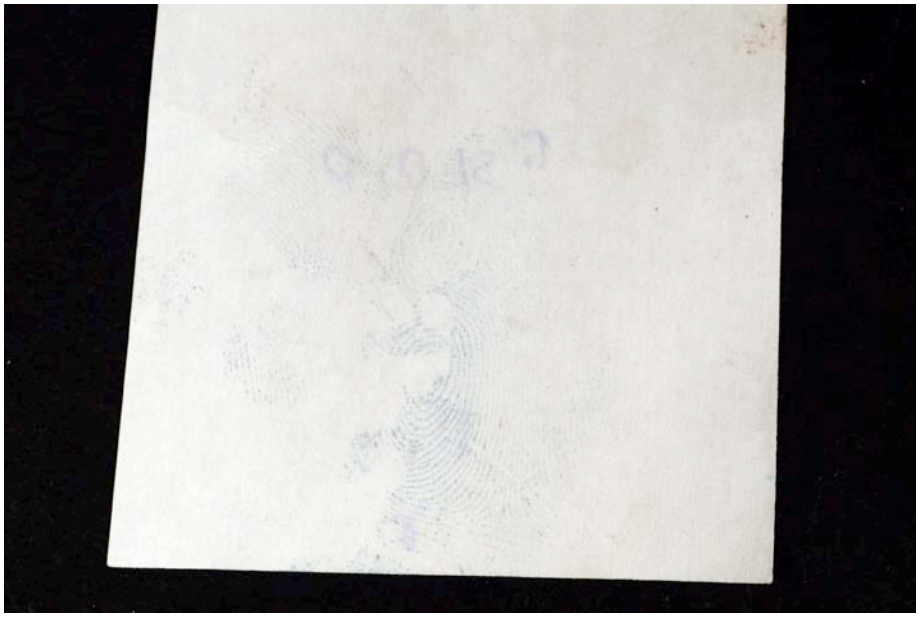
Genipin/Glycine



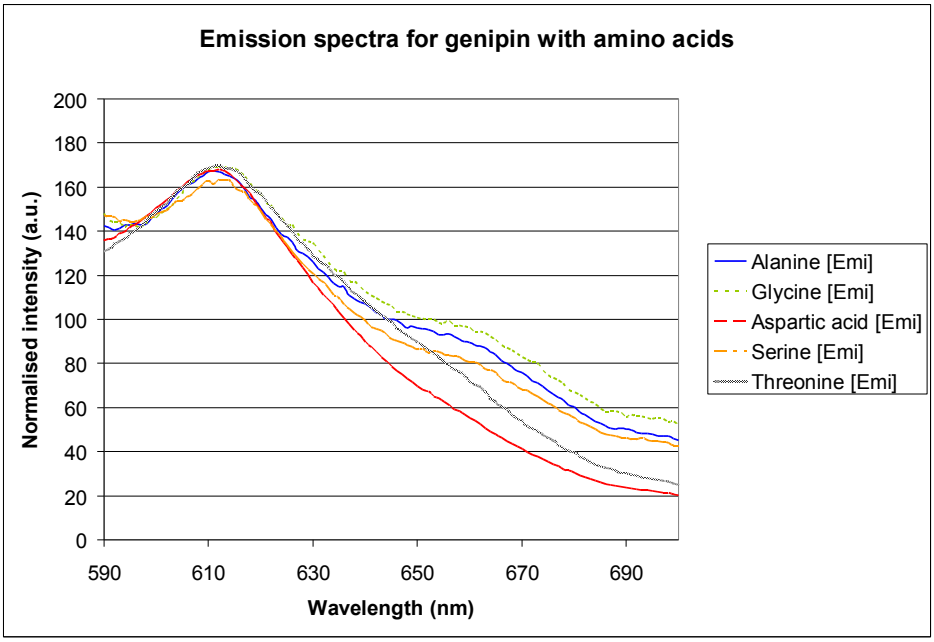
Genipin/L -Alanine



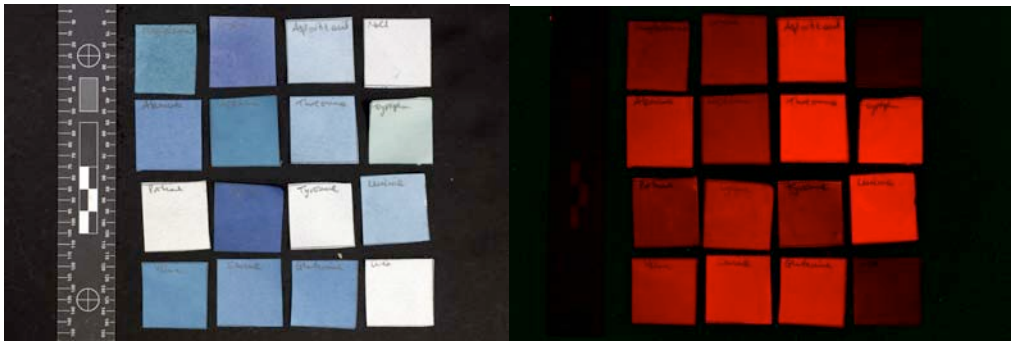
Proposed reaction mechanism for genipin with amino acids and structures of some reaction products [5].



Blue reaction product obtained from genipin.



Emission spectra for reaction products of genipin with amino acids.



a) *b)*

Reaction products formed between genipin and 0.1M solutions of amino acids and other fingerprint constituents a) visible and b) fluorescence.

2.2 The formulation proposed for use by the Israeli National Police (and used in the comparative studies below) comprises:

1.7g genipin
57mL absolute ethanol
86mL ethyl acetate
587mL HFE7100.

3. Reasons technique is not recommended by CAST

3.1 Genipin is not recommended for operational use because it is not as effective as ninhydrin in colorimetric mode, and not as effective as DFO in fluorescence mode.

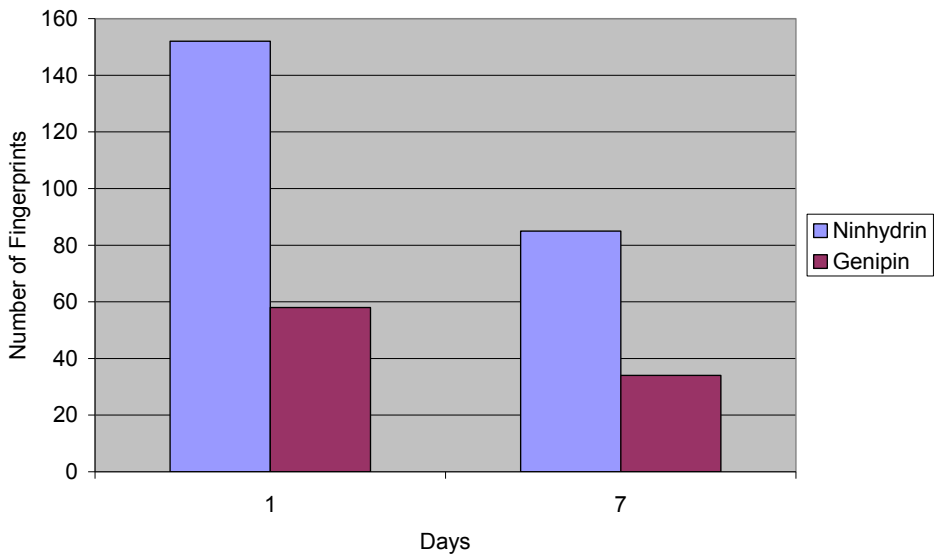
3.2 On certain paper types genipin may give better results than ninhydrin or DFO, but unless a detailed analysis of paper type is carried out prior to chemical treatment it will not be possible to identify when genipin should be used. This is clearly not practical for routine operational work.

3.3 These observations are based on the results of a short study of the effectiveness of genipin conducted by CAST in 2005 [6]. These studies utilised six donors leaving depletion series of six fingerprints on ten different types of paper found in the UK, namely:

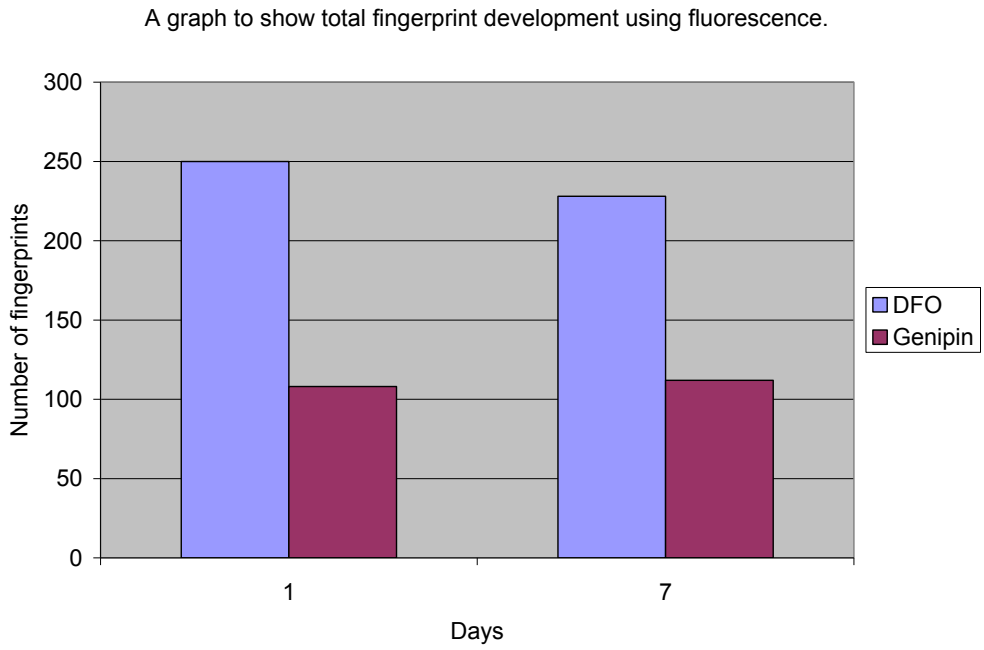
business paper (wove);
parchment paper;
photocopier paper;
writing paper;
white envelope;
brown envelope;
yellow card;
laser printer paper;
newspaper;
magazine.

3.4 The depletion series were split down the middle, one-half being treated with genipin and the other with ninhydrin or DFO. Prints were aged for one day and seven days before processing. The results are illustrated below.

A graph to show fingerprint development using visual examination.



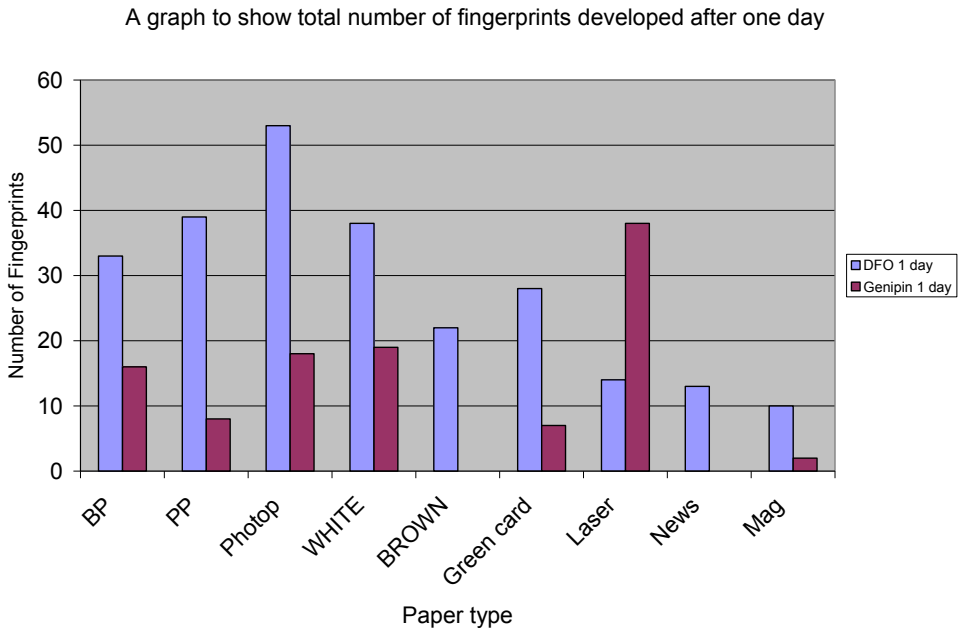
a)



b)

Comparison of the effectiveness of genipin with existing techniques for porous surfaces a) with ninhydrin in colorimetric mode and b) with DFO in fluorescence mode.

3.5 It can be seen that when overall numbers of marks developed on all types of paper are considered, genipin is clearly not as effective as either DFO or ninhydrin. However, a more detailed breakdown by paper type (an example is given below) actually showed that on laser printer paper genipin was the most effective reagent. However, this observation is of no operational benefit unless it is positively known that a particular exhibit is laser printer paper and genipin can be recommended as an alternative treatment.



More detailed comparison of results obtained comparing genipin to DFO on individual paper types

4. References

1. **Almog, J., Cohen, Y., Azoury, M. and Hahn, T. - R.** (2004) 'Genipin – A Novel Fingerprint Reagent with Colorimetric and Fluorogenic Activity', *J. Forens. Sci.*, vol. 49 (2), pp 255–257.

2. **Levinton-Shamuilov, G., Cohen, Y., Azoury, M., Chaikovsky, A. and Almog, J.** (2005) 'Genipin, a Novel Fingerprint Reagent With Colorimetric and Fluorogenic Activity, Part II: Optimization, Scope and Limitations', *J. Forens. Sci.*, vol. 50 (6), pp 1367–1371.

3. **Almog, J., Levinton-Shamuilov, G., Cohen, Y. and Azoury, M.** (2007) 'Fingerprint Reagents with Dual Action: Color and Fluorescence', *J. Forens. Sci.*, vol. 52 (2), pp 330–334.

4. **Jelly, R., Lewis, S. W., Lennard, C., Lim, K. F. and Almog, J.** (2008) 'Lawsone: a novel reagent for the detection of latent fingermarks on paper surfaces', *Chem. Comm*., pp 3513–3515.

5. **Lewis, S. W.,** (2007) 'Natural products as novel reagents for the detection of latent fingermarks'**,** *Presentation at International Fingerprint Research Group*, 26–30 March, 2007. Canberra: Australian Federal Police.

6. **Fitzgerald, L.** (2005) *Feasibility Report on Genipin*, Internal HOSDB Report, October. London: Home Office.

5.9.4 Alloxan

1. History

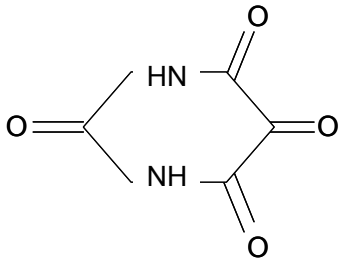
1.1 Reactions of the substance alloxan with amino acids had been observed in the 1860s, and the formation of red reaction products between alloxan and the amino acid glycine were noted at the beginning of the 20th century. After his synthesis of ninhydrin in 1910, Ruhemann conducted a series of experiments that demonstrated that alloxan and ninhydrin must be closely related compounds [1] because of the similar nature of their reactions.

1.2 Alloxan was not investigated as a fingerprint reagent until the discovery that ninhydrin could develop fingerprints on paper in 1954. This led to the re-evaluation of several related compounds in the same role and alloxan formulations for fingerprint development were reported in Japan in the late 1950s [2], the fingerprints thus developed being orange-yellow in colour. However, it was noted that for the majority of surfaces studied ninhydrin gave superior performance.

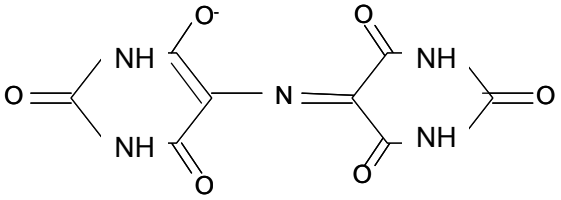
1.3 The use of alloxan for fingerprint development was mentioned in the 1970s [3], although it was still regarded as inferior to ninhydrin, developing fewer fingerprints with lower contrast and higher levels of background staining. The most recent comparative study of alloxan was carried out by Almog [4] in 1987, in an assessment of the reactivity and colour intensity of a range on ninhydrin analogues. It was concluded that alloxan was inferior to ninhydrin as a fingerprint development reagent in all respects.

2. Theory

2.1 The reaction between alloxan and amino acids is directly analogous to that with ninhydrin, and a Ruhemann's purple analogue is formed as a result. The structure of alloxan and the corresponding coloured product is shown below.



Alloxan



Ruhemann's Purple Analogue

Structures of alloxan and the corresponding Ruhemann's purple analogue.

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend alloxan because it is significantly less sensitive than the currently (2011) available 1,8-diazafluoren-9-one (DFO) and ninhydrin processes.

4. References

1. **Ruhemann, S.** (1911) 'Triketohydrindene Hydrate. Part III. Its Relation to Alloxan', *J. Chem. Soc*., vol. 23, pp 792–800.

2. **Morris, J. R.** (1974) *An Examination of the Chemical Literature on Fingerprint Technology for the Period 1890 to August 1974*, AWRE SSCD Memorandum 359, October. Aldermaston: Atomic Weapons Research Establishment.

3. **Caton, H. E.** (1974) 'Physical and Chemical Aspects of Latent Print Development', *Proceedings of the Conference on the Science of Fingerprints*, 24–25 September 1974, London, UK.

4. **Almog, J.** (1987) 'Reagents for Chemical Development of Latent Fingerprints: Vicinal Triketones – Their Reaction with Amino Acids and with Latent Fingerprints on Paper', *J. Forens. Sci*, vol. 32 (6), pp 1565–1573.

5.9.5 4-Chloro-7-nitrobenzofurazan (NBD chloride)

1. History

1.1 In the late 1960s and early 1970s, a series of chemicals were developed that gave fluorescent reaction products with amino acids. The primary application of these compounds was in amino acid detection in thin layer chromatography, although it was soon recognised that these could also be applied to fingerprint detection in the same way as ninhydrin. 4-Chloro-7-nitrobenzofurazan (NBD chloride) was one such compound, introduced in the late 1960s with investigations of its effectiveness in amino acid detection under way in the early 1970s [1].

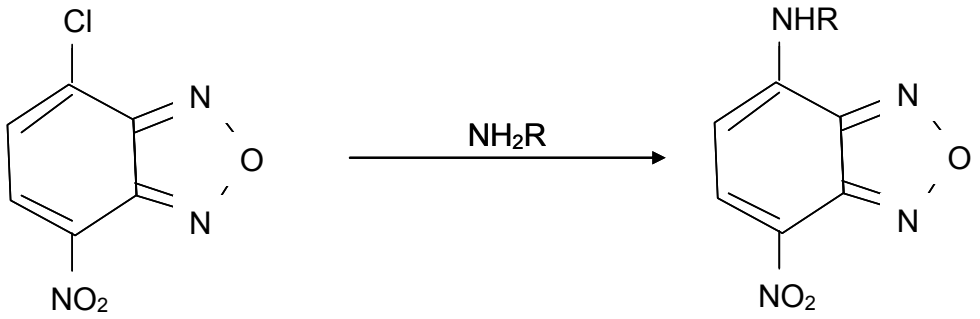
1.2 Initial studies into the use of NBD chloride as a fingerprint reagent were conducted in the late 1970s [2], the results of which suggested that the technique may give improved sensitivity over ninhydrin when developed marks were excited using a laser.

1.3 By the early 1980s, NBD chloride was still being evaluated as a fingerprint reagent for the development of fingerprints on porous surfaces, using blue light (~475nm) from a filtered xenon arc lamp to promote fluorescence in the developed marks [3]. A further, more extensive comparative study with ninhydrin demonstrated similar sensitivity between the two techniques [4]. In some cases the background fluorescence of the paper caused issues and it was recommended that an area of paper be tested to assess the level of background fluorescence prior to treatment of the entire exhibit.

1.4 The process was introduced into operational use in several police forces, including the Metropolitan Police [5] where it was used as part of a sequential treatment routine in serious cases. However, by the late 1980s, concerns were being raised about the fact that NBD chloride was a potential mutagen and its use began to decline. Almog *et al.* investigated the synthesis and properties of a range of NBD chloride derivatives [6] and identified several with potential for further study, but with the introduction of 1,8-diazafluoren-9-one (DFO) this class of compounds does not appear to have been developed further.

2. Theory

2.1 NBD chloride is a non-fluorescent compound that reacts with amino acids to produce a fluorescent reaction product, shown in outline below.



NBD Chloride

Fluorescent product

Fluorescent product formed by reaction between 4-chloro-7-nitrobenzofurazan and amino acids.

2.2 Published NBD chloride formulations utilised chlorofluorocarbon (CFC) 1,1,2-trifluorotrichloroethane (CFC113) as the carrier solvent and either ethanol or acetonitrile as the principal solvent. The formulation used by Salares [2] consisted of:

20mg NBD chloride
2mL absolute ethanol
20mL CFC113.

2.3 The resultant solution was sprayed, the treated article allowed to dry and then heated for 10 minutes at 90°C. Other researchers [3] used the solution as a dip bath, and suggested heating for the same time at the slightly higher temperature of 110°C.

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend the use of NBD chloride because it is not as effective as DFO, and there are concerns about it being a suspected mutagenic compound.

4. References

1. **Fager, R. S., Kutina, C. B. and Abrahamson, E. W.** (1973) 'The Use of NBD Chloride (4-chloro-7-nitrobenzo-2-oxa-1,3-diazole) in Detecting Amino Acids and as an N-Terminal Reagent', *Anal. Biochem*., vol. 53, pp 290–294.

2. **Salares, V., Eves, C. and Carey, P.** (1979) 'On the Detection of Fingerprints by Laser Excited Luminescence', *Forens. Sci. Int*., vol. 14, pp 229–238.

3. **Warrener, R. N., Kobus, H. J. and Stoilovic, M.** (1983) 'An Evaluation of the Reagent NBD Chloride for the Production of Luminescent Fingerprints on Paper: I. Support for a Xenon Arc Lamp Being a Cheaper and Valuable Alternative to an Argon Ion Laser as an Excitation Source', *Forens. Sci. Int*., vol. 23, pp 179–188.

4. **Stoilovic, M. Warrener, R. N. and Kobus, H. J.** (1984) 'An Evaluation of the Reagent NBD Chloride for the Production of Luminescent Fingerprints on Paper: II. A Comparison with Ninhydrin', *Forens. Sci. Int*., vol. 24, pp 279–284.

5. **Creer, K. E. and Brennan, J. S.** (1987) 'The work of the serious crime unit', *Proceedings of the International Forensic Symposium on Latent Prints,* 7–10 July 1987, pp 91–99. Virginia USA: FBI Academy, Quantico.

6. **Almog, J., Zeichner, A., Shifrina, S. and Scharf, G.** (1987) 'Nitrofurazanyl Ethers – A New Series of Fluorigenic Fingerprint Reagents', *J. Forens. Sci*, vol. 32 (3), pp 585–596.

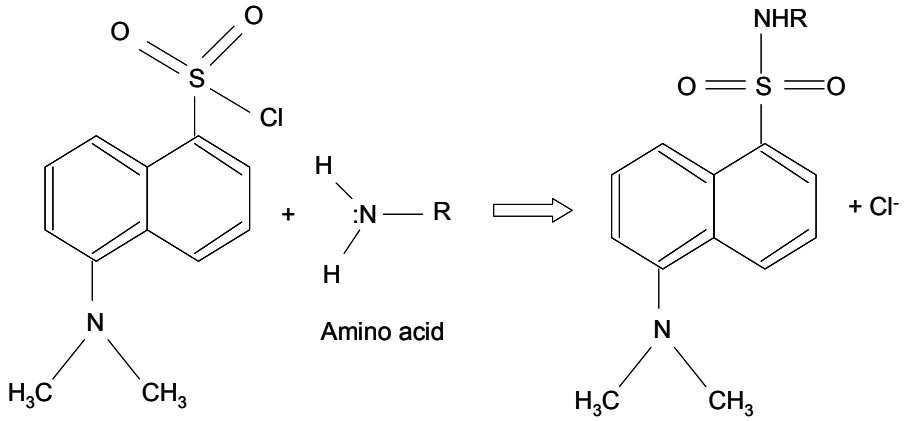
5.9.6 Dansyl chloride

1. History

1.1 Dansyl chloride is another reagent originally developed for the analysis of amino acids [1-3], producing a fluorescent reaction product that is excited by ultraviolet (UV) light. In common with other amino acid detection compounds, it has been investigated as a fingerprint development reagent [4]. In tests where ninhydrin and dansyl chloride were used as spray reagents on brown paper and cardboard, dansyl chloride appeared to give higher sensitivity on weaker marks. However, the process has not been extensively pursued as a practical technique since the mid-1980s.

2. Theory

2.1 The dansylation reaction of amino acids is described in detail elsewhere [1]. The reaction product formed by the reaction of dansyl chloride with fingerprint residues has been shown to absorb at 360nm (UV) and an emission maximum at around 475nm.



Reaction between dansyl chloride and amino acids.

2.2 A formulation given for dansyl chloride is:

0.2g dansyl chloride
100mL acetone
adjust pH to 10 using additions of 8M potassium hydroxide.

The resultant solution was applied by spraying.

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend the process because no extensive comparative studies have been carried out on its effectiveness. Dansyl chloride is also corrosive and potentially explosive under certain conditions and is therefore not recommended for health and safety reasons.

4. References

1. **Sieiler, N.** (1970) 'Use of the Dansyl Chloride in Biochemical Analysis'. In *Methods of Biochemical Analysis*, Glick, E. J. (ed). J Wiley.

2. **Lee, M. - L. and Safille, A.** (1976) 'Improved Solvent System for Thin-layer Chromatography of Dns-amino acids', *J. Chromatog.*, vol. 116, pp 462–464.

3. **Tapuhi, Y., Schmidt, D. E., Lindner, W. and Karger, B. L.** (1981) 'Dansylation of Amino Acids for High-Performance Liquid Chromatography Analysis', *Anal. Biochem*., vol. 115, pp 123–129.

4. **Burt, J. A. and Menzel, E. R.** (1985) 'Laser Detection of Latent Fingerprints: Difficult Surfaces', *J. Forens. Sci*, vol. 13 (2), pp 364–370.

5.10 Iodine

1. History

1.1 Iodine is one of the earliest chemical processes proposed for the development of latent fingerprints and is still in operational use today. The observation that iodine fumes could be used both to detect handwriting alterations and to develop latent fingerprints was reported by Coulier in 1863 [1]. In a review of early literature relating to fingerprint development conducted by Morris [2] references to the use of iodine fuming are made in 1891 and a procedure for its application given in 1912. It was noted that fumes of iodine directed onto paper produced a yellow colour where the iodine was absorbed by the fingerprint residues. However, this staining was only transitory, fading in minutes, and further experiments were conducted to identify a method of fixing the mark.

1.2 Iodine fuming was in operational use in the UK by 1931 [3] and a method of transferring and fixing developed marks using moist paper carrying rice starch was proposed in 1935 [2]. An alternative means of 'lifting' developed marks by means of a silver foil was being used by the 1960s [4], the iodine selectively reacting with the surface to form silver iodide, which then darkened when exposed to strong light. A refinement to the starch fixing process was proposed at about the same time [5], the proposed method being to brush the mark with finely ground starch powder, blow to remove the excess and then expose the mark to gentle steam for 1–2 seconds. In a summary of methods used to develop fingerprints produced by Scotland Yard in 1970 [6] iodine fuming was among the recommended development techniques, in this case utilising the starch powder fixing method. Iodine fuming was either applied within an enclosure, or could be applied to surfaces using a fuming pipe, the latter approach not now recommended because of health and safety concerns.

1.3 The lifting of fingerprints developed using iodine with silver or tin plates was further investigated as a means of recovering fingerprints from skin [7,8,9]. Experiments demonstrated that marks could be recovered from both live and dead skin using this technique and although marks could be recovered up to 72 hours after deposition on dead skin, the retention time on live skin was significantly shorter. It was also observed that only oil-rich, sebaceous marks were developed in this way, no development being observed for eccrine marks.

1.4 Further work was carried out on iodine fixatives in the 1970s. 'Tetrabase'(4,4-Bis(dimethylamino)diphenylmethane) was investigated as a fixing solution and also as an additive in uncured silicone rubber mixes, which could be moulded over a developed mark to fix it without recourse to solvent dipping or spraying [10]. Other researchers proposed α-naphthoflavone [11], with this method of fixing ultimately being favoured in the UK for operational use [12,13]. Simultaneous fuming of iodine and steam was studied as a means of improving the sensitivity of

iodine fuming on paper and also improving the performance of the reagent on older marks [14]. Iodine fuming was also applied to non-porous surfaces, with successful results on brass being claimed [6].

1.5 It was subsequently proposed that the sensitivity of the technique could be improved by applying the iodine in solution, combined with the a-naphthoflavone fixative [15]. This formulation used cyclohexane as the solvent for iodine, which is highly flammable and not considered appropriate for use at scenes of crime. The Metropolitan Police and Home Office Central Research Establishment (HO CRE), Aldermaston developed a non-flammable, two-part formulation with the objective of treating large areas such as painted and papered walls at scenes [16]. This formulation was based on iodine dissolved in Fluorisol (1,1,2-trichlorotrifluoroethane, also known as CFC113 or Arklone), with the α-naphthoflavone fixative dissolved in dichloromethane applied as a separate solution. Comparative trials were carried out with the chlorofluorocarbon (CFC)-based ninhydrin formulation – known as non-flammable ninhydrin (NFN) – then in operational use, recording the marks developed under ambient conditions [16]. It was shown that the iodine solution was more effective in these conditions, although on paper and paper-based wallpaper ninhydrin gave superior performance if it was exposed to elevated temperature and humidity. Iodine solution was introduced into operational use by some organisations with marks being developed at around one-third of scenes treated [16].

1.6 The iodine solution formulation developed by HO CRE was considered for inclusion in the first edition of the *Manual of Fingerprint Development Techniques*, primarily as a process for application to wall surfaces at crime scenes. However, further comparative testing carried out by the Home Office Scientific Research and Development Branch (HO SRDB) between iodine solution and the CFC-based ninhydrin formulation indicated that ninhydrin was in fact the more effective process and that the sequence of iodine solution and fixative followed by ninhydrin may produce fewer marks than ninhydrin as a single treatment [17]. The principal advantage of iodine solution was that it developed marks instantly, compared with the period of up to ten days required for the development of marks treated with ninhydrin at a scene. Because of the potentially detrimental (albeit slight) effect of iodine solution and fixative on subsequent ninhydrin development, the process was ultimately omitted from the manual. The possibility of applying the reagent as a spray was also investigated [18], and was claimed to be more effective than both ninhydrin and the brush application of iodine solution.

1.7 With the introduction of the Montreal Protocols in 1987 and the banning of the use of CFCs, it became necessary to look at alternative formulations of iodine solution. PSDB initiated a programme of work to revisit the iodine solution formulation and assess alternatives to the solvent, fixing agent and to iodine itself [19]. These studies identified heptane and methyl cyclohexane as possible alternative solvents to CFC113. However, both these solvents are flammable and not suitable

for crime scene use without significant precautions. Alternative non-flammable solvents gave inferior performance in the development of fingerprints. Of the range of fixing agents studied, α-naphthoflavone proved to be the most effective in terms of colour and longevity of the fixed mark.

1.8 Australian researchers also studied the use of formulations based on the non-CFC solvents 1-methoxynonafluorobutane (HFE7100) and 2,3-dihydrodecafluoropentane (HFC4310mee) [20]. Although not as effective as the CFC-based formulation, the HFC4310mee formulation was investigated as a spray reagent on a range of surfaces, including treated wood, glass, wallpaper, vinyl, paint, brick and raw wood, and its performance compared with powdering and a ruthenium tetroxide spray reagent. In these trials, iodine was found to be the most effective treatment of those evaluated for vinyl, wallpaper and brick. Other studies using iodine have looked at α-naphthoflavone as a fixative for marks developed on skin using fuming [21] and fuming as a technique for developing marks on adhesive tape [22].

1.9 More recently, in 2004-2005, HOSDB included a flammable (heptane-based) iodine solution in comparative studies of techniques for developing marks on contaminated surfaces, where it was compared with solvent black 3 and basic violet 3. On certain surfaces iodine did appear to give superior results and it will be necessary to explore this in more detail to see if iodine has a place in some sequential treatment charts.

1.10 A further study has been conducted by HOSDB to compare the flammable, heptane-based iodine solution to ninhydrin on a range of wall coverings representative of those commercially available in 2009 [23]. The materials used to manufacture wallcoverings have changed in the years since the previous studies, and the results demonstrated that the flammable iodine formulation was far more effective than ninhydrin on the surfaces studied, in contrast to previously observed trends. This does raise some operational issues because the flammable formulation should not be applied at scenes and the effectiveness of ninhydrin has evidently declined. Subsequent treatment of these surfaces with powder suspensions has indicated that this process is potentially far more effective than either iodine or ninhydrin and further work is required to optimise advice given for treatment of such surfaces.

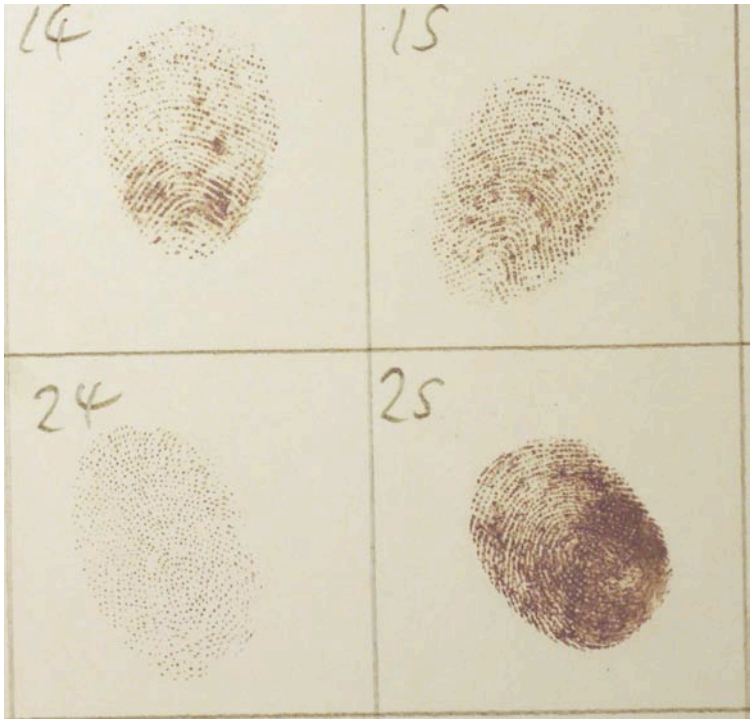
2. Theory

2.1 It has been suggested that the development of fingerprints using iodine occurs by an addition reaction across the carbon double bonds in the unsaturated fatty acid components of the fingerprint residue [24]. The readily reversible nature of this reaction is used to explain the rapid fading of prints developed using iodine.

2.2 However, observations by subsequent researchers indicate that this may not be the sole reaction mechanism [14]. The following reasons for this are cited.

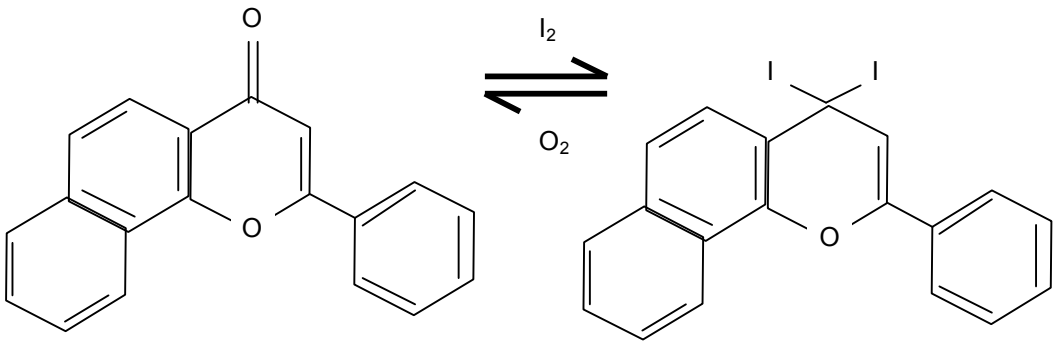
- The addition reaction across the double bonds of unsaturated compounds is known to be slow, whereas the development of prints using iodine is instantaneous and still occurs at sub-zero temperatures.
- The reactions that occur to fix the developed marks would not occur unless free iodine was present; the iodine compounds formed by the saturation reaction would not react in the same way.
- Laboratory trials with chemical compounds representative of other fingerprint constituents, including saturated hydrocarbons, amino acids, inorganic salts and water, also gave visible reaction products on exposure to iodine fumes.

2.3 It was proposed that the principal mechanism binding iodine into the fingerprint deposit and causing its yellow/brown coloration is the attractive interaction between the constant dipole of water molecules in the fingerprint and a dipole induced on the iodine molecule. It is proposed that this effect is enhanced by the presence of inorganic salts in the fingerprint residue [14]. Because the presence of water is necessary, this would account for the observed poor performance of iodine on older marks where water has evaporated.



Fingerprints from different donors developed on glossy paper by iodine fuming.

2.4 The mechanism of the fixing reaction has not been conclusively identified, but Sears [17] suggests a reversible reaction between iodine and α-naphthoflavone, which would account for the fading of the fixed marks with time.

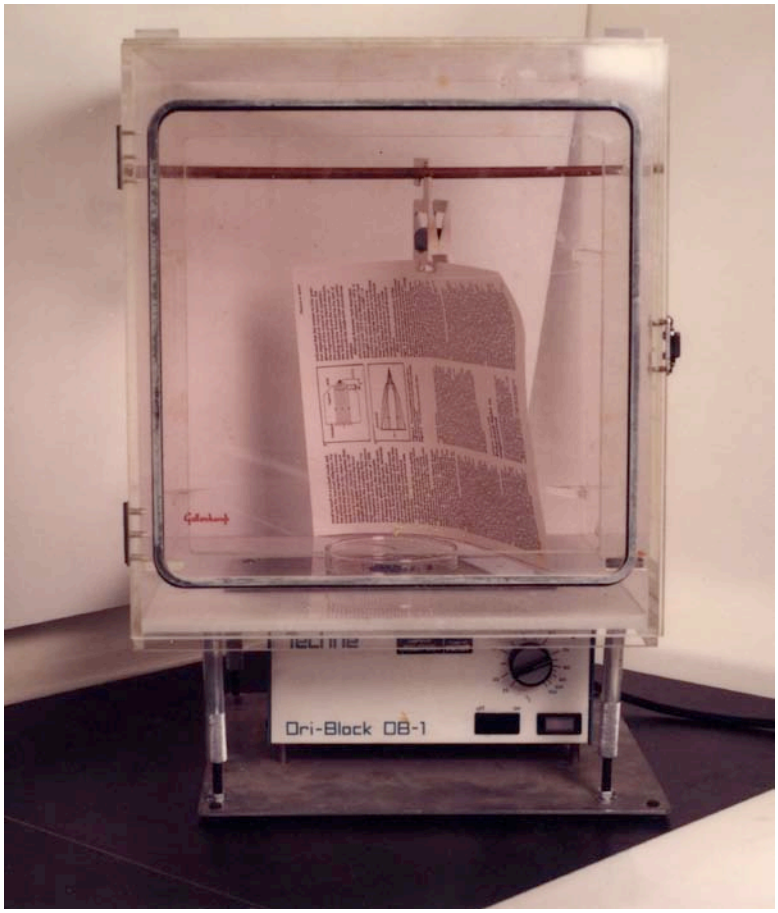


α-Naphthoflavone

Proposed fixing mechanism for iodine using α-naphthoflavone.

3. CAST processes

3.1 The currently (2011) recommended CAST process is iodine fuming rather than solution treatment, although this will be withdrawn from the principal processes recommended in the forthcoming 3rd edition of the *Manual of Fingerprint Development Techniques*. In the fuming process, the article to be treated is supported or suspended within a small chamber, 1g of iodine placed onto a glass dish at the base of the chamber, and the chamber sealed. The iodine is then allowed to sublime (or can be gently heated to 50ºC), producing a violet/brown vapour.



Article being treated by iodine fuming.

3.2 Development of fingerprints is monitored and when the maximum contrast is reached between ridges and the background, the excess iodine vapour is removed from the chamber and the article removed and photographed.

3.3 Fingerprints may then be fixed using a solution based on α-naphthoflavone, but the formulation given in the 2nd edition of the *Manual of Fingerprint Development Techniques* [13] requires review because it is based on CFC113.

4. Critical issues

4.1 Iodine fumes are corrosive and harmful, and iodine solution is harmful and flammable. Neither process should be used outside the controlled environment of a laboratory.

4.2 Marks developed using iodine may fade rapidly and require fixing to make them more visible and more permanent for subsequent imaging.

4.3 Iodine is not effective on marks more than a few days old, and should not be used if older marks are being targeted.

5. Application

5.1 Suitable surfaces: Iodine fuming is suitable for porous surfaces, in particular paper. Performance is best on glossy paper types. It can be used on non-porous surfaces, but is most suited for those where greasy contamination is present. Iodine solution is suitable for all surfaces where iodine fuming is successful, and is also effective in developing fingerprints on painted wall surfaces. However, it is not recommended for use on painted walls because of the flammability of the solution.

5.2 The iodine process does not appear in any of the sequential treatment flow charts in the *Manual of Fingerprint Development Techniques* [13]. This is because there is no surface for which iodine is more effective in developing fingerprints than any of the other recommended processes. Iodine fuming is retained in the manual because it is the only chemical treatment that can be used without leaving visible traces on the treated article. There are specialist applications where the lack of a visible, developed mark is important and iodine fuming is an option in these cases. Because it is not often possible to determine what substances may be present in an article before treatment, iodine should be used with caution because some substances may be capable of temporarily fixing the mark and inhibiting the normally rapid fading process.

5.3 Iodine fuming (and iodine solution) are also capable of detecting fingerprints on contaminated surfaces. Because iodine, basic violet 3 and solvent black 3 all develop marks in slightly different ways and may not target the same constituents, iodine may develop marks where other processes are ineffective. At least one practical case of this is known (see section 8 on validation and operational experience, below). Because it has proved difficult to generate consistent ‘contaminated’ surfaces for laboratory trials, it is not currently (2011) possible to give comprehensive guidance for when (or if) iodine should be considered in either fuming or solution form, or to propose sequences with other reagents for contaminated surfaces.

5.4 In the laboratory, iodine fuming should be carried out in a chamber sited within a fume cupboard. Fuming can be also be carried out on larger items or at scenes of crime using portable glass pipes with heated compartments to start iodine fuming, and desiccant crystals to dry the fumes. Because of the toxic and corrosive nature of iodine vapour, this should only be carried out in well ventilated and/or extracted areas by operators with the appropriate protective equipment.

5.5 Iodine solution can also be applied in a laboratory or at a crime scene by brushing or spraying. The solvents used as carriers for iodine are either flammable or capable of displacing air and should therefore be used with appropriate health and safety measures. CAST does not currently (2011) recommend the use of iodine solution.

6. Alternative formulations and processes

6.1 Several processes have been proposed as alternatives to the fuming technique outlined in the manual [13]. The principal one of these is the iodine solution treatment, until recently (up to around 2008) in regular use in the UK by the Forensic Science Service (FSS). CAST does not recommend this process for a variety of reasons, including effectiveness (although this may need to be reassessed), impact on subsequent treatments, health and safety, and scene clean up considerations. However, it is recognised that the technique does have some potential advantages and may warrant more evaluation.

6.2 Previous assessments of the iodine solution process carried out by CAST in the late 1980s and late 1990s [17,19] have included investigations into alternatives to the solvent, fixing agent and iodine itself.

6.3 In the late 1980s, replacements to cyclohexane as the solvent for iodine were considered [17], with the main consideration being to identify a less or non-flammable formulation. Several candidate systems were rejected on the basis of cost (dichlorocyclohexane, dibromocyclohexane, 1,9-dichlorononane, 1,10-dichlorodecane and 1,7-dibromoheptane). Decahydronaphthalene ('Decalin') was tested as an alternative solvent and found to give fingerprint development equivalent to the cyclohexane formulation. However, evaporation time of the solvent from the treated surface increased from seconds to 20–40 minutes and this was not deemed practical for operational use. Ultimately, the CFC113-based formulation developed by HO CRE provided a non-flammable system that could be used at scenes of crime.

6.4 Replacements for dichloromethane in the α-naphthoflavone fixing solution were also investigated. Ethanol, ether, 2-ethoxyethanol, 1,1,1 trichloroethane did not dissolve α-naphthoflavone and were therefore unsuitable. α-Naphthoflavone did dissolve in acetone and glacial acetic acid, but in both cases the quantity of solvent required was far greater than the amount of dichloromethane and no change to the existing formulation was made.

6.5 After CFCs were withdrawn from regular use, HOSDB reassessed several formulations that included CFC113, including iodine solution. The objective was to produce an all-in-one formulation containing iodine and fixing agent. The CFC formulation was compared with a range of different solvent types, including:

- hydrofluorocarbons (HFCs) – 2,3-dihydrodecafluoropentane (HFC4310mee), 1,1,1,3,3-pentafluorobutane (HFC365mfc), 1-methoxynonafluorobutane (HFE7100) and 1-ethoxynonafluorobutane (HFE7200);

- siloxanes –octylmethylcyclotetrasiloxane (Volasil 244), decamethylcyclopentasiloxane (Volasil 245);
- hydrocarbons – cyclohexane, heptane, methyl cyclohexane.

6.6 Of these, iodine had only limited solubility in the hydrofluorocarbons, as did α-naphthoflavone. This resulted in rapid precipitation of the fixative unless excess dichloromethane was added. Solutions based on the siloxane solvents were more stable, but often took in excess of one hour to evaporate from the surface being treated. Solutions based on siloxanes also developed fewer fingerprints. All hydrocarbon solvents produced solutions that were effective in fingerprint development. However, all are flammable.

6.7 The opportunity was also taken to review alternatives to α-naphthoflavone. The alternatives considered were starch, β-cyclodextrin and the leuco-dyes leuco crystal violet, leuco malachite green, leuco patent blue and leuco berbelin blue.

6.8 The leuco dyes were effective fixing agents but for a variety of reasons, including the cost of the reagent and background staining, were not proposed as replacements for α-naphthoflavone. Starch was the least effective of the fixing agents examined and although β-cyclodextrin did fix marks, the colour contrast was poor and ninhydrin could not be used sequentially after its use.

6.9 The interhalides iodine monobromide and iodine monochloride were considered as replacements for iodine. Solutions formed with these compounds were less stable and the colours of the fixed marks were less strong. As a consequence, these compounds were not pursued further.

6.10 The most effective all-in-one iodine solution was identified as:

part A: 0.4g iodine dissolved in 194mL of heptane or methyl cyclohexane;
part B: 0.6g α-naphthoflavone dissolved in 6mL of dichloromethane.

Part B is added to part A, the resultant solution is filtered and applied with a brush.

6.11 However, there were disadvantages with the formulation (such as flammability) making it difficult to recommend for widespread use, especially at crime scenes. This formulation is used internally by CAST as the standard formulation for comparative laboratory studies of technique effectiveness.

7. Post-treatments

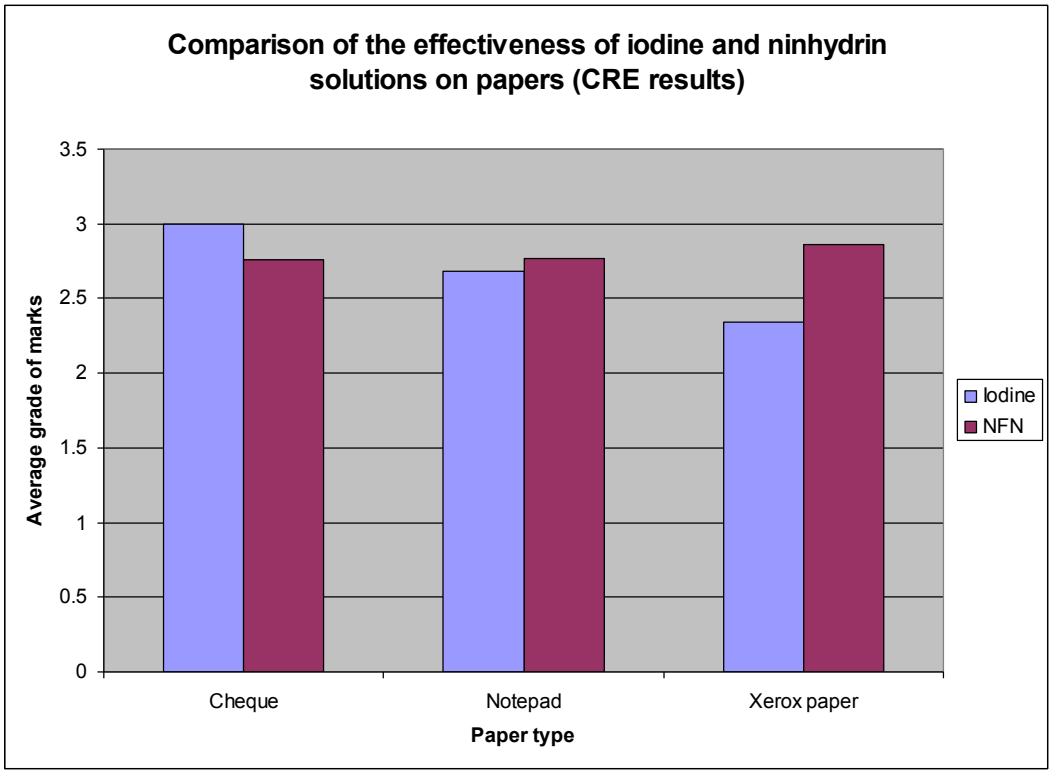
7.1 The principal post-treatment used for iodine is fixing solution, which can be applied to marks developed using both fuming and solution treatments (although the CAST solution contains the fixative in the solution itself). The fixing solution converts the yellow/brown marks into a product with a more highly contrasting colour, and prevents them from rapidly fading. The most commonly used fixing agent for iodine is α-naphthoflavone, which gives a deep blue coloration.

7.2 For fingerprints developed using iodine on skin, lifting using tin or silver plates has been proposed, which involves placing the metal plate in contact with the developed mark. The reactive iodine will form a metal iodide on regions of the metal in contact with the fingerprint ridges, which can then be darkened by illumination with strong light to reveal the ridges.

8. Validation and operational experience

8.1 Laboratory trials

8.1.1 Iodine solution was proposed as one possible treatment for paper samples in a laboratory during the early/mid-1980s. To explore this potential application HO SRDB and HO CRE both carried out laboratory trials on paper exhibits, comparing the effectiveness of iodine solution and the non-flammable ninhydrin formulation then in operational use. The results obtained by HO CRE are tabulated below, being based on the results of developing and assessing single fingerprints deposited by 100 different donors.



Comparative results obtained for iodine and ninhydrin (NFN) on porous surfaces.

8.1.2 The HO SRDB studies consisted of trials using 40 split prints on white card and cheques. Three comparisons were made: iodine versus ninhydrin; the effect of subsequent ninhydrin treatment after iodine; and the iodine/ninhydrin sequence versus ninhydrin. The results are summarised below.

Grading	Iodine = ninhydrin	Iodine > ninhydrin	Iodine < ninhydrin
Percentage	75	10	15

Grading	Iodine = iodine/NFN	Iodine > iodine/NFN	Iodine < iodine/NFN
Percentage	70	25	5

Grading	Iodine/NFN = ninhydrin	Iodine/NFN > ninhydrin	Iodine/NFN < ninhydrin
Percentage	60	10	30

Results of comparative tests between iodine, ninhydrin and iodine/ninhydrin sequences.

8.1.3 The PSDB results were in accordance with the HO CRE results. Iodine solution was, in general, a less effective treatment than ninhydrin for paper samples and the sequential use of ninhydrin after iodine did not yield as many marks as ninhydrin alone. Iodine solution was therefore not recommended for use on paper articles by HO SRDB.

8.1.4 To assess the effectiveness of iodine solution on wall surfaces, Pounds [16] carried out a series of laboratory trials at HO CRE comparing iodine solution and the CFC-based ninhydrin formulation on surfaces representative of wall coverings. Initial tests looked at sheets of substrate stored in a laboratory, and consistently showed the iodine solution to be more effective when marks were developed under ambient conditions. Subsequent tests utilised actual sections of painted wall and added powders to the techniques used in comparative studies. The results of these studies are tabulated below.

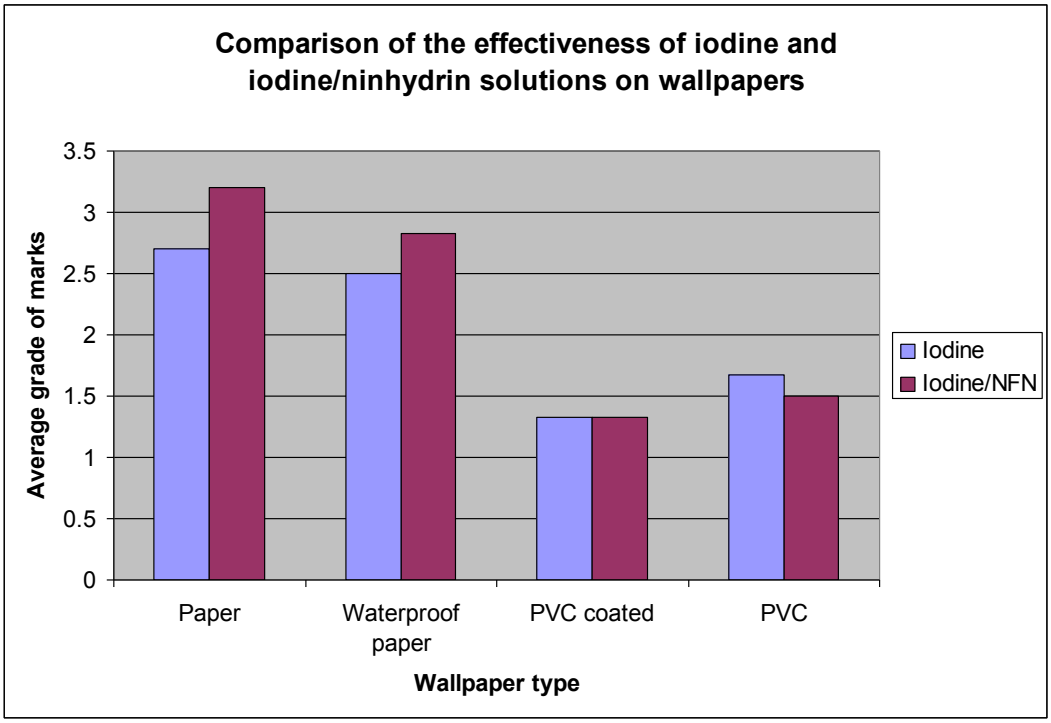
Storage condition	**Development method**	**Days treated and assessed**					
		0	**5**	**12**	**20**	**32**	**53**
Wall by window	Iodine solution	3.1	1.5	1.7	1.5	1.5	1.1
	Ninhydrin (CFC)	2.2	–	–	–	–	1.6
Wall in shade	Iodine solution	2.9	1.7	2.1	1.4	1.4	1.6
	Ninhydrin (CFC)	2.0	–	–	–	–	1.9

Average quality score for fingerprints developed on emulsion painted wall.

Development method	**Days treated and assessed**			
	1	**5**	**12**	**27**
Iodine solution	3.9	3.3	2.9	1.9
Ninhydrin (CFC)	1.0	–	–	–
Magnetic powder	2.9	–	–	–
Aluminium powder	1.0	–	–	–

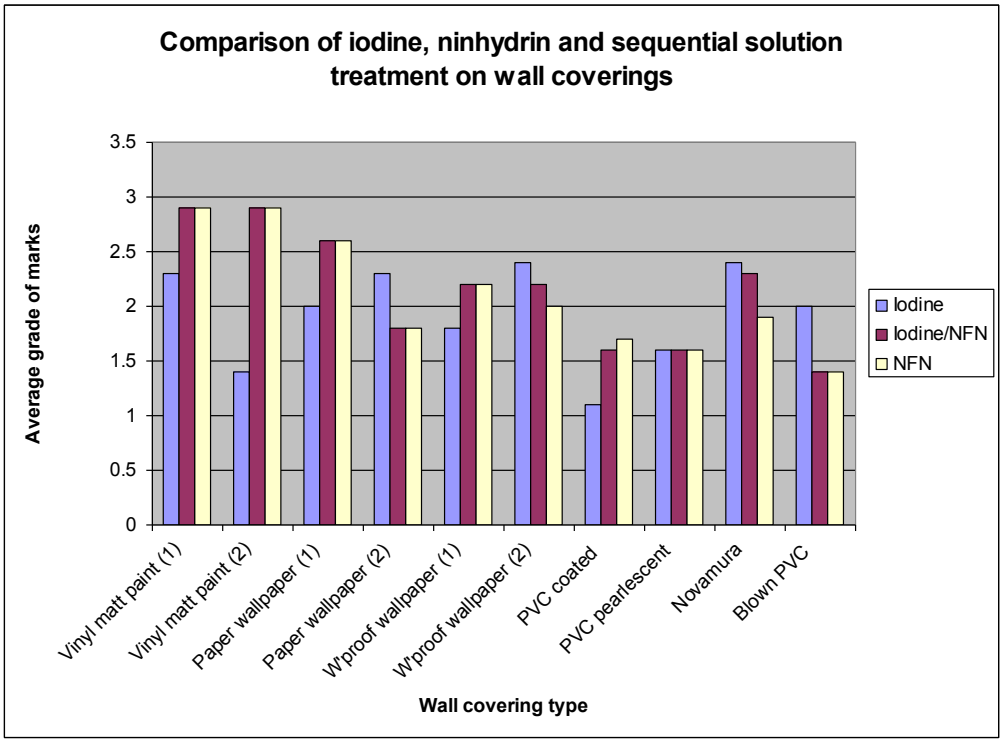
Average quality score for fingerprints developed on emulsion painted wall.

8.1.5 HO SRDB carried out similar studies [17] looking at a wider, more representative range of wall coverings, including painted walls and different types of wallpaper. Two trials were conducted, both involving the grading of over 200 prints. The first looked at aluminium powder, black granular powder, iodine and iodine followed by ninhydrin on the full range of surfaces. In this trial, aluminium powder was found to out-perform iodine solution on vinyl silk painted walls, but apart from this surface both powders produced significant clogging on the surface and were not recommended for use. On matt paint iodine worked well, but additional marks were developed by subsequent ninhydrin treatment. A summary of the results for the wallpaper surfaces is given below.



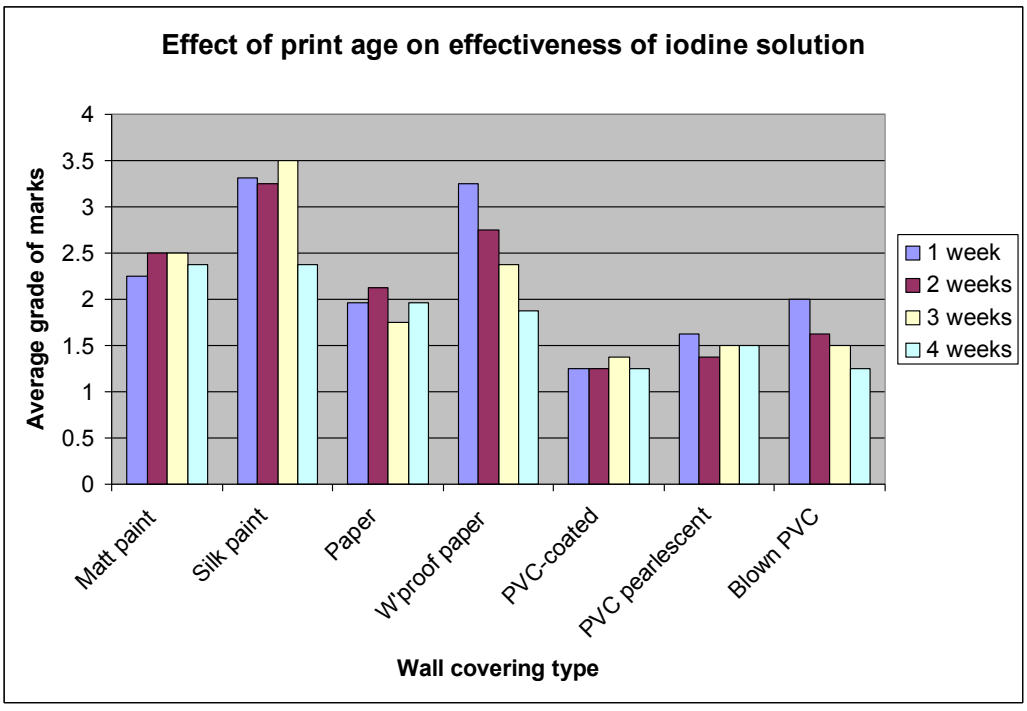
Comparative results obtained for iodine and iodine/ninhydrin on wallpapers.

8.1.6 The results again showed a general trend that ninhydrin developed more marks after iodine solution, but this was not true on every surface examined. This trial was repeated, but now including marks that were treated with ninhydrin alone. The results were similar to those above, showing that in general the iodine solution was less effective than the iodine/ninhydrin sequence and ninhydrin alone. However, there were certain surfaces where iodine solution was the single most effective treatment, although it was not always possible to determine which type of surface was present before commencing treatment.



Comparison of effectiveness of different processes and sequences on different wall coverings.

8.1.7 The effect of the age of the print on development using iodine solution was also studied by HO SRDB, 96 marks being deposited and graded for each age (from one to four weeks). Results are summarised below.



Effect of age of mark on effectiveness of iodine solution.

8.1.8 There is again agreement between the HO SRDB results and those obtained by HO CRE. In general the effectiveness of iodine solution falls with time but on certain surfaces there is a less obvious fall off.

8.1.9 Based on laboratory trials, HO CRE introduced iodine solution into operational use in the late 1980s. The performance of the Fluorisol-(CFC113)-based iodine formulation was found to be equivalent to the cyclohexane-based formulation in laboratory tests, and superseded it in operational use until CFCs were banned by the Montreal Protocols and the formulation reverted to one based on a flammable solvent. Operational performance figures recorded for iodine solution (including results obtained using both formulations) are given below.

Type of case	**Number of scenes**		**Number of iodine marks recorded**
	Examined	**Marks found**	
Murder	71	28	56
Rape	12	3	4
Burglary	9	4	6
Other major crime	11	2	3
Total	**103**	**37**	**69**

Operational results obtained by the use of iodine solution at scenes of crime.

8.1.10 The results presented above were criticised by HO SRDB at the time in that they did not provide a detailed assessment of the types of surface the iodine solution had been applied to (only the crime type), nor did they record the effectiveness of subsequent ninhydrin treatment [25]. In several of these cases, it was known that ninhydrin had developed significant numbers of additional marks. Subsequent development of a spray formulation [18] resulted in further operational trials by HO CRE, the initial results of which are given below.

Surface type	**Number of scenes**		**Number of iodine marks recorded**
	Examined	**Marks found**	
Wallpaper	5	2	15
Emulsion paint	11	6	24
Total	**16**	**8**	**39**

Operational results obtained by the use of iodine spray at scenes of crime.

8.1.11 Subsequent testing of the different application routes for iodine solution by PSDB [19] found that solution dipping was the most effective, followed by brush application, with spray being the least effective. Brush application is the technique that was used at crime scenes up until 2008.

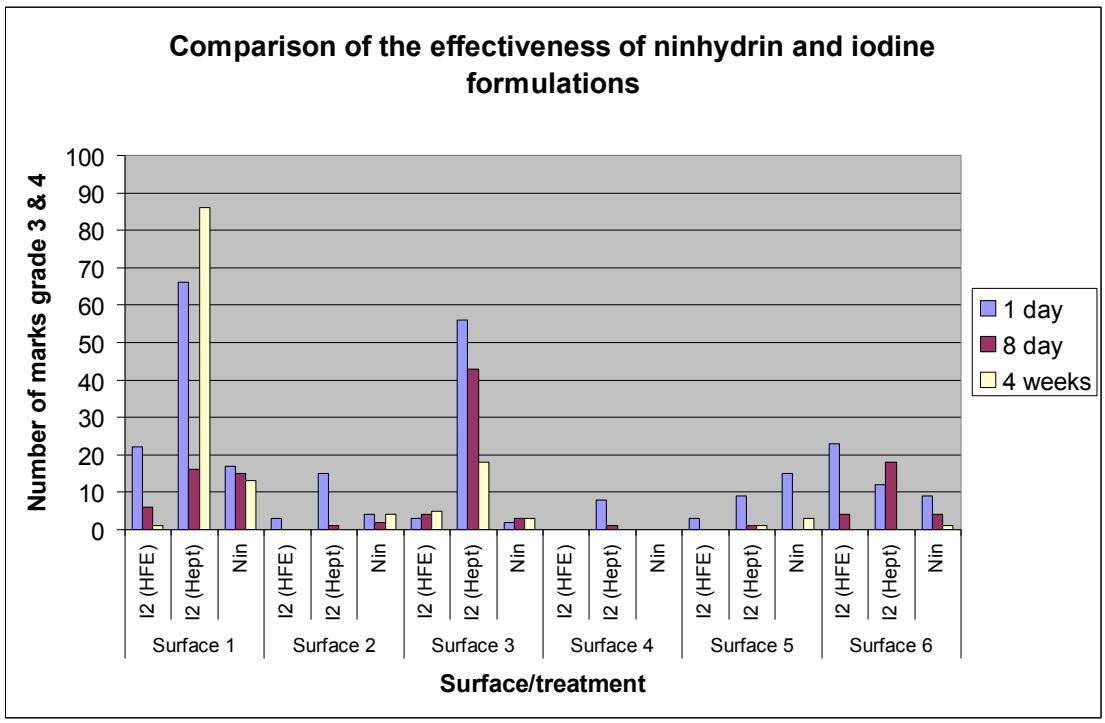
8.1.12 One of the other potential applications of iodine solution (and fuming) is in the development of marks on contaminated surfaces. Comparative laboratory trials were carried out between the CAST iodine solution formulation and solvent black 3. These are more fully reported in Chapter 3.9 Solvent black 3 and demonstrated that in general solvent black 3 was more effective, although there were some surfaces, such as gloss painted wood, where iodine solution was more effective.

8.1.13 In the repeat trials on wallcoverings conducted in 2009 [23], the following surfaces were examined.

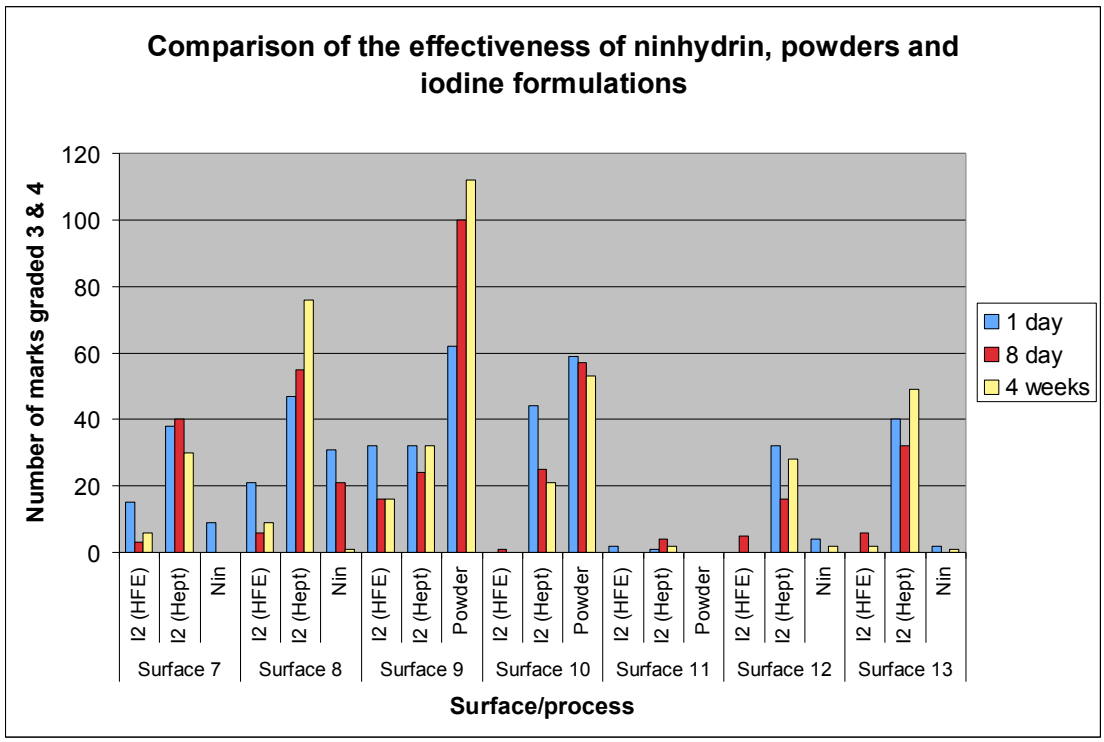
Surface	**Type**	**Porosity**
1	Wickes master washable matt paint	Porous
2	Dulux interior matt paint	Porous
3	Wallpaper – pulp	Porous
4	Wallpaper – vinyl	Non-porous
5	Wickes interior matt emulsion paint	Porous
6	Crown silk emulsion	Non-porous
7	Wallpaper – foamed polyethylene	Semi-porous
8	Wallpaper – washable vinyl coated	Semi-porous
9	Wickes liquid gloss paint	Non-porous
10	Dulux liquid gloss paint	Non-porous
11	Crown non-drip satin paint	Semi-porous
12	Dulux grease and stain resistant, tough matt paint	Porous
13	Wallpaper – vinyl coated	Semi-porous

Description of the surfaces examined in the 2009 study.

8.1.14 For all surfaces except gloss paint, the effectiveness of the heptane-based iodine solution was compared with a low-flammability, HFE71DE-based iodine solution and ninhydrin, whereas on gloss painted surfaces powders were substituted for ninhydrin. Over 4,500 marks were graded in this study.



Results of the 2009 comparative study on surfaces 1–6.



Results of the 2009 comparative study on surfaces 7–13.

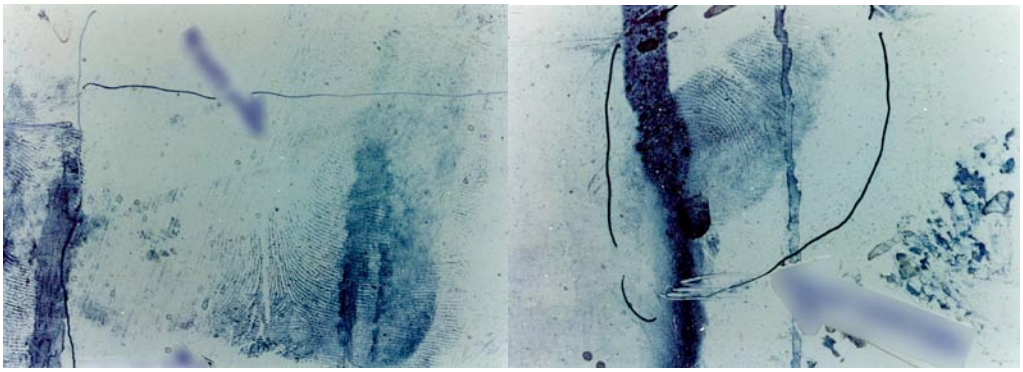
8.1.15 It can be seen that the heptane-based iodine solution out-performed the HFE71DE-based iodine solution and ninhydrin on all almost surfaces, except for 1 day old marks on surface 6. The heptane-based solution gave far less background staining than the HFE71DE-based solution, and marks of greater contrast than ninhydrin. On gloss

surfaces, powdering gave superior performance to both of the iodine solutions. Very few additional marks were found to be developed by leaving surfaces treated with ninhydrin for a further two-week period. Subsequent treatment of the surfaces with powder suspensions produced a significant improvement in the number and quality of developed marks over and above all of the initial treatments. The results of this further study are still (2011) being analysed.

8.2 Pseudo-operational trials and operational experience

8.2.1 The use of iodine fuming on operational work is rare, because it is only recommended in special circumstances, such as where the surface is contaminated or the treatment should ideally leave no trace on the article being examined. However, there are recorded cases where iodine has produced marks of value and the other processes recommended for contaminated surfaces (basic violet 3, solvent black 3) have not. PSDB was involved in the treatment of a contaminated fridge from a fast food outlet in the 1990s where iodine fuming yielded identifiable marks.

Contaminated fridge treated using iodine fuming followed by fixing with α-naphthoflavone.



Marks developed on contaminated fridge using iodine fuming followed by fixing with α-naphthoflavone.

8.2.2 The operational use of iodine solution has been more contentious, with CAST not recommending the process and the FSS until recently (2008) often using it on serious operational cases. One of the reasons CAST has not recommended iodine solution is that previous studies conducted in the late 1980s and again in the late 1990s indicated that it was less effective than ninhydrin across the range of surfaces it was likely to be applied to, and the use of fixative may inhibit subsequent ninhydrin development. However, the most recent studies (2009) have shown that this may no longer be true, with considerably more marks being found by iodine solution than were developed by ninhydrin. In addition, marks are revealed instantly with iodine and may take several days to develop fully with ninhydrin. Some of the background data behind these original recommendations are presented here, although it should be noted that all the early comparisons were between iodine solution and the CFC-based ninhydrin formulation (NFN). The more recently developed HFE7100-based ninhydrin formulation was more effective than the CFC-based formulation on paper (see Chapter 3.4, Ninhydrin) but until the recent study [23] no tests had been carried out on surfaces representative of wall coverings.

9. References

1. **Quinche, N. and Margot, P.** (2010) 'Coulier, Paul-Jean (1824–1890): A Precursor in the History of Fingermark Detection and Their Potential Use for Identifying Their Source (1863)', *J. Forens. Ident.*, vol. 60 (2), pp 129–134.

2. **Morris, J. R.** (1974) *An Examination of the Chemical Literature on Fingerprint Technology for the Period 1890 to August 1974*, SSCD Memo 359, October. Aldermaston: Atomic Weapons Research Establishment.

3. **Rhodes, H. T. F.** (1931) *Some Persons Unknown*. London: John Murray.

4. **McLaughlin, A. R.** (1961) 'Chemicals and Their Application For Developing Latent Prints', *Ident.*, July, pp 3–7.

5. **Larsen, J. K.** (1962) 'The Starch Powder-Steam Method of Fixing Iodine Fumed Latent Prints', *Ident.* July, pp 3–5.

6. **New Scotland Yard** (1970) *Chemical Development of Latent and Other Marks*, Fingerprint Branch. London: New Scotland Yard.

7. **Shin, D. H. and Argue, D. G.** (1976) 'Identification of Fingerprints Left on Human Skin', *Can. Soc. Forens. Sci. J.*, vol. 9 (2), pp 81–84.

8. **Adcock, J. M.** (1977) 'The Development of Latent Fingerprints on Human Skin: The Iodine-Silver Plate Transfer Method', *J. Forens. Sci* vol. 22 (3), pp 599–605.

9. **Gray, C.** (1978) 'The Detection and Persistence of Latent Fingerprints on Human Skin: An Assessment of the Iodine-Silver Plate Method', *J. Forens. Sci. Soc.*, vol. 18 (1/2), pp 47–52.

10. **Trowell, F.** (1975) 'A Method for Fixing Latent Fingerprints Developed with Iodine', *J. Forens. Sci. Soc.*, vol. 15, pp 189–195.

11. **Mashiko, K. and Ishizaki, M.** (1977) 'Latent Fingerprint Processing: Iodine-7,8-Benzoflavone Method', *Ident. News*, vol. 27 (11), pp 3–5.

12. **Goode, G. C. and Morris, J. R.** (1983) *Latent Fingerprints: A Review of Their Origin, Composition and Method for Detection*, AWRE Report O 22/83. Aldermaston: Atomic Weapons Research Establishment.

13. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

14. **Almog, J., Sasson, Y. and Anati, A.** (1979) 'Chemical Reagents for the Development of Latent Fingerprints II: Controlled Addition of Water Vapor to Iodine Fumes – A Solution to the Ageing Problem', *J. Forens. Sci,* vol. 24 (2), pp 431–36.

15. **Haque, F., Westland, A. and Kerr, M. F.** (1983) 'An Improved Non-Destructive Method for Detection of Latent Fingerprints on Documents with Iodine-7,8-Benzoflavone', *Forens. Sci. Int.*, vol. 21, pp 78–83.

16. **Pounds, C. A.** (1989) *The Use of Iodine Solution to Reveal Latent Fingerprints on Wallpaper and Emulsion Painted Walls*, Home Office Forensic Science Service CRSE Report No. 694. London: Home Office.

17. **Sears, V. G.** (1987) *I_2 Experiments and Notes 1987*. Unpublished PSDB Project File. London: Home Office.

18. **Pounds, C. A., Allman, D. S. and Wild, F. M.** (1992) 'The Development of Latent Fingerprints Using an Iodine Spray Technique', *Forensic Science Service CRSE Report No. 746*, March. London: Home Office.

19. **Sears, V. G.** (1999) 'Iodine Solution for the Development of Latent Fingerprints', unsubmitted journal paper, 1999. Presented at the *International Symposium of Fingerprint Detection Chemistry*, Ottawa, May 25–28. Canada: Ottawa.

20. **Flynn, K., Maynard, P., Du Pasquier, E., Lennard, C., Stoilovic, M. and Roux, C.** (2004) 'Evaluation of Iodine-Benzoflavone and Ruthenium Tetroxide Spray Reagents for the Detection of Latent Fingermarks at the Crime Scene', *J. Forens. Sci*, vol. 49 (4), pp 707–715.

21. **Wilkinson, D. A., Watkin, J. E. and Misner, A. H.** (1996) 'A Comparison of Techniques for the Visualisation of Fingerprints on Human Skin including the Application of Iodine and a-Naphthoflavone', *J. Forens. Ident*., vol. 46 (4), pp 432–451.

22. **Midkiff, C. R.** (1997) 'Development of Prints on Tape – Part III', *Fingerprint Whorld*, vol. 23, pp 83–86.

23. **Fletcher, G. W.** (2009) 'The Effectiveness of Iodine Solutions in the Visualisation of Latent Fingerprints', journal article submitted in part fulfilment of an MSc degree, September. King's College, University of London.

24. **Olsen, R. D.** (1975) 'The Oils of Latent Fingerprints', *Fingerprint and Identification Magazine* vol. 56 (7), pp 3–12.

25. **PSDB** (1990) 'T. Kent to Dr G. Turnbull', Internal Memorandum, 23 April, PSDB. London: Home Office.

5.11 Multimetal deposition

1. History

1.1 The multimetal deposition (MMD) system for developing fingerprints was first proposed by Saunders [1] in the late 1980s. The system incorporated principles of both small particle reagent and physical developer and provided a universal developing agent capable of producing marks on porous, semi-porous and non-porous surfaces.

1.2 To carry out the MMD process, porous items were immersed in distilled water for 20–30 minutes (treatment of other types of article omitted this immersion stage). Items were then immersed in colloidal gold solution for 30–120 minutes, rinsed in distilled water (for up to 15 minutes in the case of porous items) and then immersed in a silver physical developer solution for 5–15 minutes. After a final rinse in distilled water items were air dried and photographed.

1.3 After the publication of this technique, researchers in the UK and elsewhere began to investigate the capabilities of MMD. In the UK, the Central Research and Support Establishment (CRSE) of the Home Office Forensic Science Service (FSS) carried out a trial comparing MMD with superglue fuming and vacuum metal deposition on a range of surfaces known to be difficult to treat, including clingfilm, plastic shotgun cartridges, masking tape and expanded polystyrene [2]. These results suggested that for some of these surfaces MMD did produce superior results, although it could not be used sequentially after superglue.

1.4 The Police Scientific Development Branch (PSDB) also carried out an assessment of the process and confirmed that it worked on a wide range of substrates, including polythene bags, metal, fabric tape, coated cardboard, masking tape, wax candles, leather and cling film [3]. Tests were carried out on paper, but no results were obtained because the paper blackened. Development of fingerprints on fabrics was also attempted, as was subsequent radioactive toning of any marks developed. Faint ridges were seen during drying but these were not visible when fully dry, although some detail could be seen after radioactive toning and autoradiography. The microstructure of the marks developed was also studied by scanning electron microscopy. HOSDB concluded that MMD was a versatile technique, but gave no better results for any given surface than other techniques already available, and therefore it was not pursued further.

1.5 The process was later extensively re-evaluated by Schnetz and Margot [4]; they proposed an improved formulation offering increased reactivity, improved resolution and greater amplification selectivity (and therefore reduced background interference). Important elements in the revised formulation were the use of an alternative means of producing colloidal gold, giving smaller particle sizes, and the replacement of the silver

nitrate/iron(II), iron(III) redox system in the physical developer stage with silver acetate/hydroquinone.

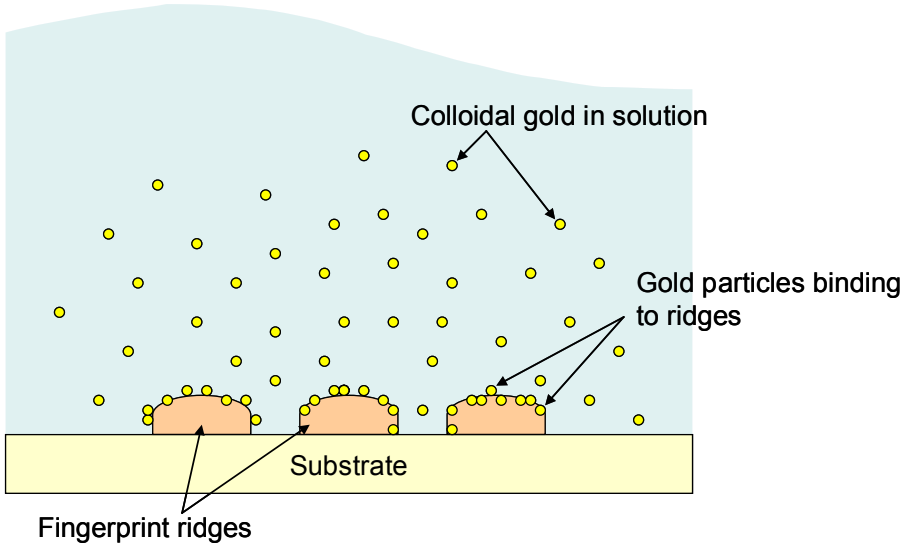
1.6 Jones [5] used the revised MMD formulation in an extensive study of processes for developing fingerprints on semi-porous surfaces. It was found that although not particularly effective on the polymer banknotes used in Australia, MMD did have potential applications for other semi-porous surfaces, including expanded polystyrene, latex and nitrile gloves, and waxed paper.

1.7 More recently Becue *et al.* [6,7] have considered further revisions to the MMD process, trying to simplify the process and to investigate the possibility of functionalising the gold nanoparticles with colorimetric or fluorescent tags. These studies are ongoing and may yield further revised formulations in future.

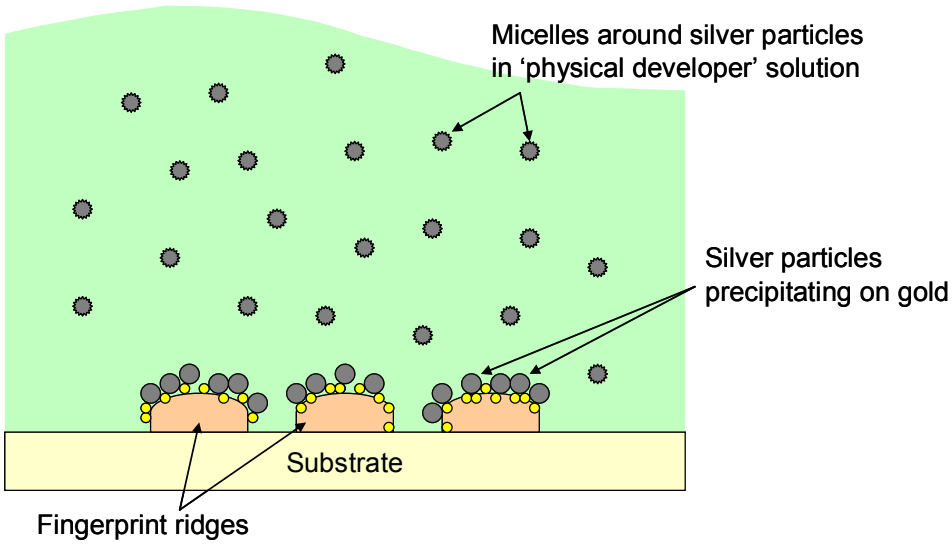
1.8 Other recent refinements have included the development of formulations for single metal deposition (SMD) [8,9] where the two-stage silver and gold deposition is replaced by a single-stage gold deposition process. This is claimed to have the advantages of reducing the number of treatment stages, reducing the number of different reagents and associated costs, and utilising reagents with a longer shelf life.

2. Theory

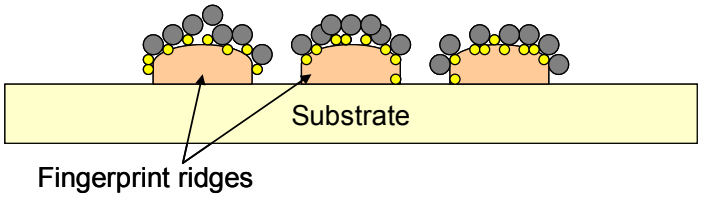
2.1 MMD is essentially a two-phase development process, illustrated schematically in the diagrams below. The exhibit to be treated is immersed in an acidified solution containing colloidal gold particles, which bind preferentially to the amino acid, protein and peptide constituents of the fingerprint. This stage alone generally gives poor contrast of the ridges and therefore a second amplification stage is used. This involves the use of a modified physical developer solution, where surfactant stabilised silver particles preferentially deposit on the colloidal gold, thus turning the ridges dark grey to black in colour.



a)



b)



c)

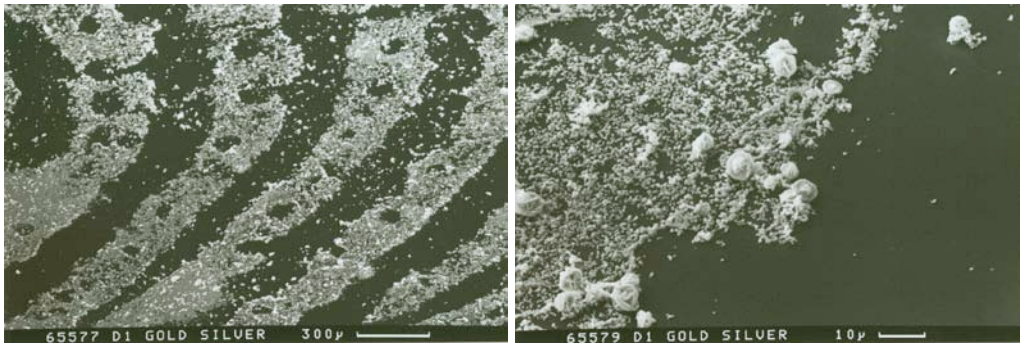
Schematic diagrams illustrating the stages in the multimetal deposition process a) colloidal gold binding to ridges b) preferential deposition of silver particles on pre-existing gold and c) dried mark with contrast provided by silver particles.

2.2 The reason that colloidal gold particles (in the case of MMD formed by the chemical reduction of tetrachloroauric acid) are used is that they are both negatively charged and hydrophobic. Binding between organic compounds and colloidal gold particles can occur by both electrostatic and hydrophobic reactions. The dominant binding mechanism varies with pH, hydrophobic interactions dominating at high pH and electrostatic interactions dominating at low pH. Schnetz and Margot [4] have suggested that it is the electrostatic interactions that are responsible for the reaction with fingerprint deposits and the pH of the treatment solution is kept low (pH 2.5–3) to facilitate this. Mildly acidic compounds such as amino acids, fatty acids and proteins carry a positive charge under these conditions and attract and bind to gold particles from the solution.

2.3 The size of the gold particles is also regarded as important, with smaller particles claimed to result in higher specificity. A size of 5–15nm is recommended, although some researchers claim to have obtained equivalent results with 30nm particles.

2.4 The physical developer solution is effectively a modification of the system used to develop fingerprints on paper, containing silver ions in the presence of a reducing system, the solution being stabilised by surfactants. The silver ions are reduced to silver metal and the gold particles bound to the ridges act as a nucleation site for this to occur. The gold particles also act to catalyse the reduction of the silver.

2.5 Scanning electron micrographs of a mark developed using MMD are shown below [3].



a) b)

Scanning electron micrographs of marks developed using multimetal deposition a) low magnification showing fingerprint ridges and b) higher magnification showing precipitated particles.

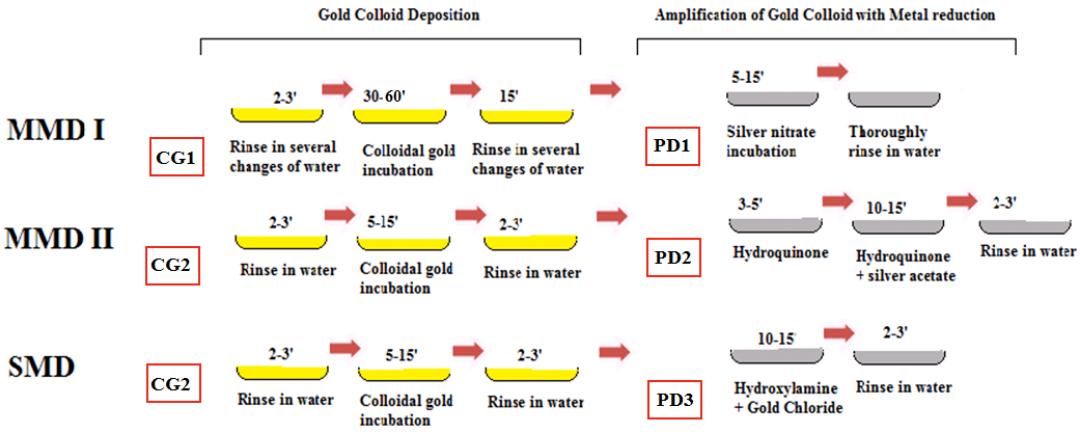
3. Reasons technique is not recommended by CAST

3.1 CAST does not currently (2011) recommend MMD because it has not been shown to give better performance than any other technique currently recommended. MMD does give reasonable results on a wide range of surfaces, but in tests carried out by PSDB in 1992 there was no single surface on which MMD gave better results than any other recommended process.

3.2 In addition to this, the technique is difficult to carry out effectively compared with many other existing processes. It requires siliconised glassware, all items used in the process must be kept scrupulously clean and it is necessary to constantly monitor pH while carrying out initial colloidal gold deposition. There are also many stages to the process and some of these may be time-consuming, even more so than the physical developer process. It has proven difficult to obtain good, reliable results and therefore the process is not recommended for routine use. The more recent SMD process [8,9] utilises fewer stages and offers potential for further study.

3.3 More recently, a further study has been carried out at HOSDB [10] to assess the relative effectiveness of MMD I, MMD II and SMD and to compare an optimised MMD process with currently recommended

processes in the *Manual of Fingerprint Development Techniques* [11]. The essential elements of the two MMD and one SMD processes are shown schematically below.



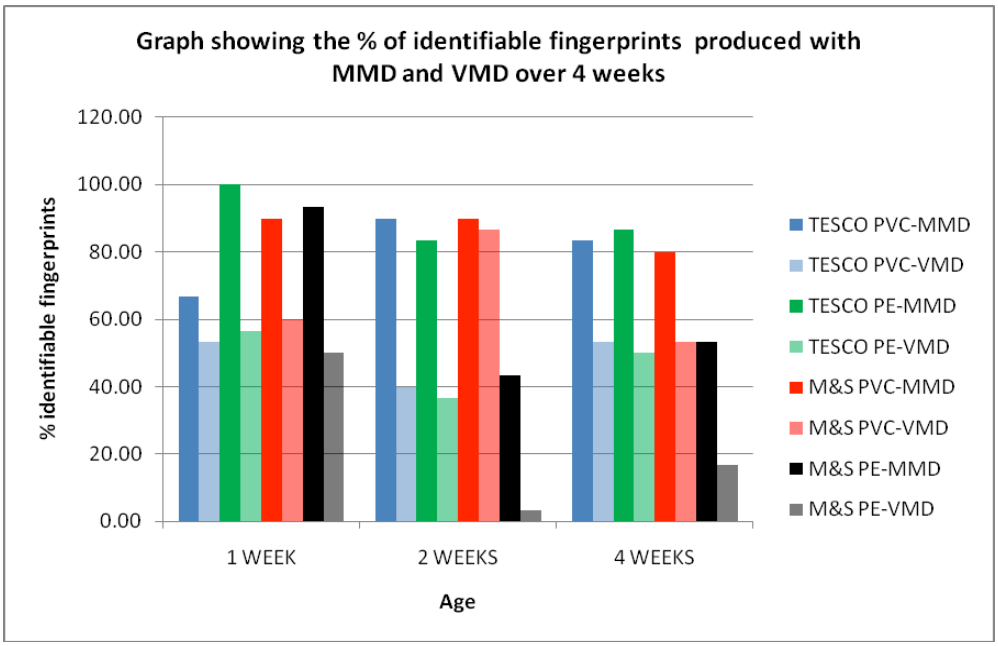
Schematic diagram showing the stages in the multimetal deposition and single metal deposition processes, and their duration.

3.4 Comparative tests confirmed that the MMD II process was the most effective. However, it was felt that this was impractical for routine use and the MMD I technique, with the pH of the colloidal gold solution reduced to pH 2.5–2.8, was chosen as the preferred method for comparative trials. This was compared with the techniques currently (2011) recommended in the CAST manual [11] for the various surfaces, summarised in the table below.

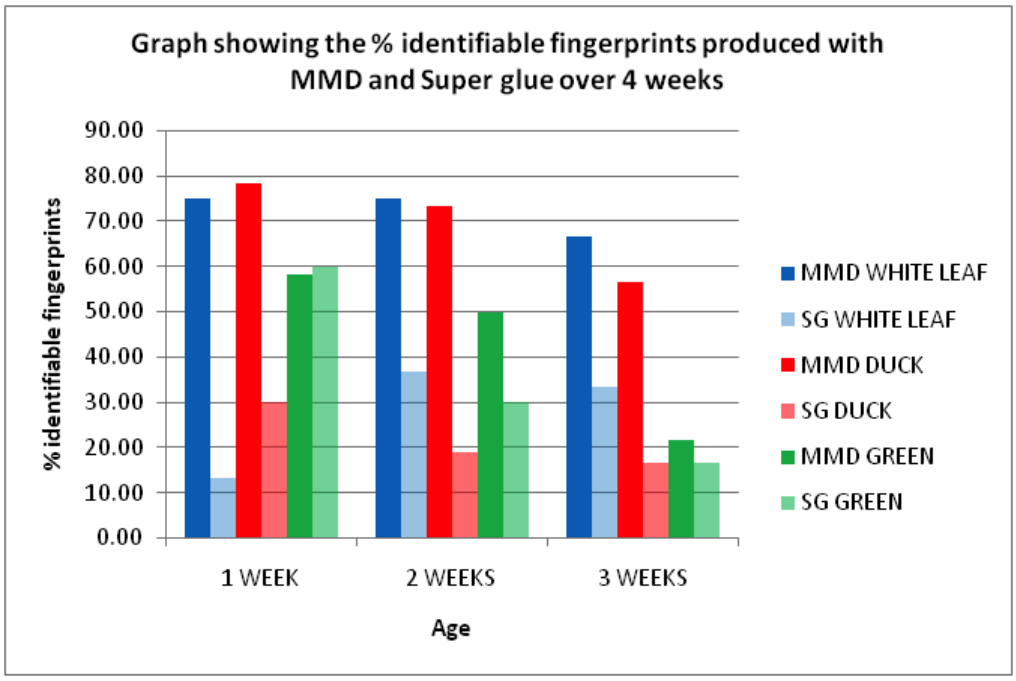
Surface	Current recommendation
Cling film – PVC/PE-based	Silver vacuum metal deposition (VMD)
Shower curtains – vinyl-based	VMD and cyanoacrylate fuming
Leatherette – PVC-based	Powder suspensions – Wet Powder Black/White ™
Leather	Powder suspensions – Wet Powder Black/White ™

Surfaces for which comparative experiments were carried out, and the processes used in the comparisons.

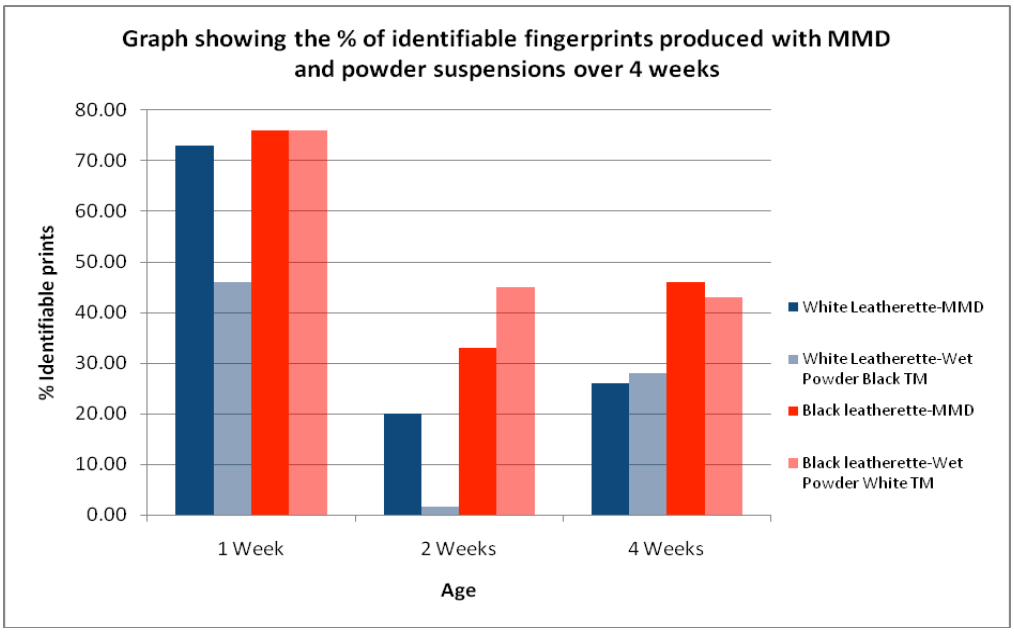
3.5 The results of these comparisons are summarised in the series of graphs and tables below.



Graph showing the proportion of potentially identifiable fingerprints developed on clingfilm over four weeks.



Graph showing the proportion of potentially identifiable fingerprints developed on shower curtains over four weeks.



Graph showing the proportion of potentially identifiable fingerprints developed on leatherette over four weeks.

	Leather white textured	
	MMD	**Wet Powder Black™**
% Identifiable prints	5.00	18.00
Average score	0.57	0.97
Standard deviation	0.89	1.37

The average score, proportion of identifiable prints and standard deviation for marks developed using multimetal deposition and powder suspension on white leather – whole prints.

3.6 MMD did show improved performance over existing techniques for vinyl-based polymer surfaces in general, and on clingfilm in particular. These results suggest there may be operational merit in using MMD on such surfaces and that further research is desirable to see if the process could be incorporated into processing sequences.

4. References

1. **Saunders, G. C.** (1989) 'Multimetal Deposition Method For Latent Fingerprint Development', *Los Alamos National Laboratory Guidelines*. Los Alamos National Laboratory, USA.

2. **Allman, D. S., Maggs, S. J. and Pounds, C. A.** (1992) *The Use of Colloidal Gold/Multi-metal Deposition for the Detection of Latent Fingerprints – A Preliminary Study*, HO FSS CSRE Report No. 747. London: Home Office.

3. **PSDB** (1992) 'Multimetal Deposition Mar/Apr 1992', PSDB file, unpublished work. London: Home Office.

4. **Schnetz, B. and Margot, P.** (2001) 'Technical Note: Latent Fingermarks, Colloidal Gold and Multimetal Deposition (MMD). Optimisation of the Method', *Forens. Sci. Int*., vol. 118, pp 21–28.

5. **Jones, N.** (2002) *Metal Deposition Techniques for the Detection and Enhancement of Latent Fingerprints on Semi-porous Surfaces*, PhD Thesis. Centre for Forensic Science, University of Sydney.

6. **Becue, A., Champod, C. and Margot, P,** (2007) 'Use of Gold Nanoparticles as Molecular Intermediates for the Detection of Fingermarks', *Forens. Sci. Int*., vol. 168 (2–3), pp 169–176.

7. **Becue, A., Scoundrianos, A., Champod, C. and Margot, P.** (2008) 'Fingermark detection based on the in-situ growth of luminescent nanoparticles: towards a new generation of multimetal deposition', *Forens. Sci. Int*., vol. 179 (1), pp 39–43.

8. **Stauffer, E., Becue, A., Singh, K. V., Champod, C. and Margot, P.** (2007) 'Single Metal Deposition (SMD) as a latent fingermark enhancement technique: An alternative to multimetal deposition (MMD)', *Forens. Sci. Int*., vol. 168 (1), pp 5–9.

9. **Durussel, P., Stauffer, E., Becue, A., Champod, C. and Margot, P.** (2009) 'Single Metal Deposition: Optimisation of this fingermark enhancement technique', *J. Forens. Ident*., vol. 59 (1), pp 88–96.

10. **Fairley, C.** (2010) *Investigation of the most effective formulation and practical use of multimetal deposition*, MSc Thesis, September. Strathclyde University.

11. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

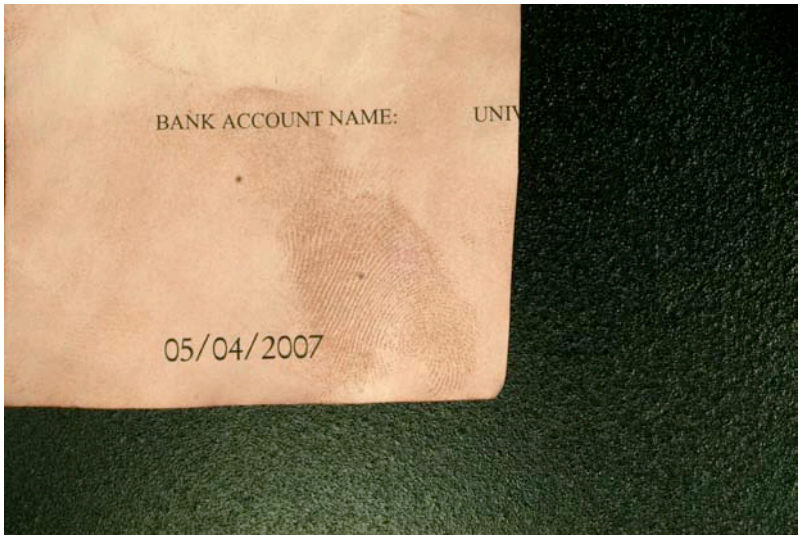
5.12 Oil Red O

1. History

1.1 Oil Red O (also known by its Colour Index name solvent red 27) is a superlipophilic diazo dye and is closely chemically related to Solvent Black 3. It has been used as a fat stain for biological samples and also industrially as a colorant for oils, fats and waxes. As the name suggests, the dye is red in colour and selectively stains lipid components. The Police Scientific Development Branch (PSDB) initially investigated Oil Red O, amongst other lipid dyes, as an alternative to solvent black 3 on non-porous surfaces [1]. These studies indicated that solvent black 3 was a superior dye for the particular range of surfaces being investigated (i.e. non-porous surfaces) and no further work was carried out on Oil Red O at this time.

1.2 The next reported forensic application of Oil Red O was for the development of lip prints [2], with a range of similar dyes including Oil Red O, solvent black 3, solvent red 23 (Sudan III) and solvent red 24 (Sudan IV) being applied, both in powder form and in solution for the staining of lip prints deposited on tissue paper.

1.3 In 2004, Beaudoin [3] reported an Oil Red O formulation for the development of fingerprints on wetted papers. The work was carried out to identify alternatives to the complex and time-consuming physical developer process, and resulted in a two-stage method consisting of a dip bath of Oil Red O in a methanol/sodium hydroxide solvent, followed by immersion of the exhibit in a sodium carbonate/nitric acid buffer solution. Initial tests on wetted surfaces ranging from porous to non-porous in nature indicated that Oil Red O was effective on porous and semi-porous surfaces, but that developed marks were difficult to visualise on non-porous surfaces.



Photograph of fingerprint developed on paper using Oil Red O.

1.4 This was followed by a comparative study looking at the relative effectiveness of Oil Red O and physical developer on thermal papers, white printer paper and brown Kraft paper [4]. In these studies sebum-rich fingerprints were deposited on paper that was wetted, divided in two and then treated using the selected process. For the range of surfaces examined, Oil Red O gave superior results on both thermal papers and white printer paper. On the brown Kraft paper, average scores were similar, but physical developer gave more marks of high quality.

1.5 A further study was conducted to look at the insertion of Oil Red O into sequential treatments on porous surfaces [5]. Again sebum-rich fingerprints were used, and comparisons made between the quality of fingerprints developed in the sequences including Oil Red O and those omitting it. Both wetted and dry papers were considered in these studies. For white paper, results indicated that improved fingerprint quality could be achieved by inserting Oil Red O into standard sequential treatments as the stage before physical developer. For brown papers Oil Red O was found to be detrimental, primarily because of the pink background staining caused by Oil Red O making marks subsequently developed using physical developer more difficult to visualise.

1.6 The promise of these studies has resulted in more detailed studies being carried out in several countries, including Australia, the UK and the USA [6]. These studies have generally used ‘standard’ fingerprints rather than deliberately sebum-rich marks and have tended to indicate that the effectiveness of Oil Red O begins to fall with the increasing age of the mark, and for marks much older than four weeks, the marks are very diffuse with little ridge detail being developed. The same effect is observed for longer immersion times in water. In both these cases, physical developer continues to develop marks with good clarity of ridge detail.

1.7 Further studies have been carried out at universities within the UK [8,9]. These again demonstrated that on groomed, sebum-rich prints Oil Red O gave superior performance to physical developer, but when normally deposited marks were used, the performance was closely equivalent. It was shown that exposing porous surfaces to accelerant was detrimental for both processes, no marks being developed by Oil Red O or physical developer after exposure.

2. Theory

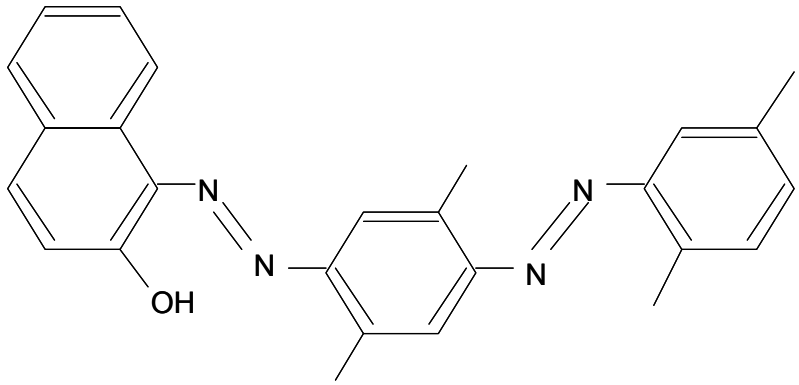
2.1 Oil Red O is a lysochrome, more commonly known as a fat stain. Most lysochromes are azo dyes that, because of their structure, have undergone molecular rearrangement making them incapable of ionising.

2.2 The basis for these dyes colouring fats is that they dissolve into it. From another perspective, the fat is the solvent for the dye. Lysochromes are

mostly insoluble in strongly polar solvents, such as water, and somewhat more so in less polar solvents, such as ethanol. They are quite strongly soluble in non-polar solvents, such as xylene. Triglycerides, being non-polar compounds, dissolve them quite well. Other lipids, having fatty components, may also dissolve them.

2.3 Lysochromes such as Oil Red O are applied from solvents in which they are sparingly soluble. As they come into contact with materials in which they are strongly soluble, they transfer to them significantly, often colouring them more strongly than the original solvent. This process is known as preferential solubility.

2.4 Oil Red O is more strongly hydrophobic than some earlier dyes used for staining lipids, and it is thought that this makes it more effective in staining applications [7]. The structure of Oil Red O is shown below.



Structure of Oil Red O (solvent red 27).

2.5 The formulation proposed by Beaudoin [3] consists of three separate baths, a staining bath to stain the lipid components of the fingerprint, a buffer solution to neutralise the base side of the staining solution and stabilise the developed marks, and finally a water wash. The formulations used are as follows:

stain bath – dissolve 1.54g Oil Red O in 770mL methanol;
dissolve 9.2g of NaOH in 230mL water;
add the two solutions, mix together, filter and store in a brown bottle.

buffer solution – add 26.5g of Na_2CO_3 to 2 litres of water and stir to dissolve;
add 18.3 mL of concentrated HNO_3;
increase volume of solution to 2.5 litres with water.

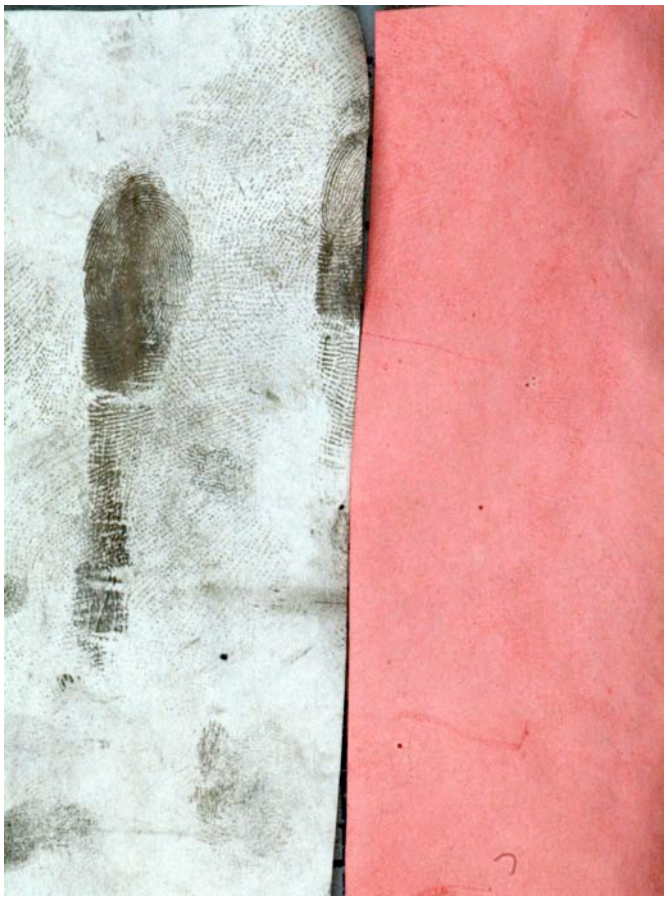
2.6 Articles to be treated are immersed in the stain bath for up to 90 minutes, then removed, drained and placed in the buffer solution. Finally the articles are rinsed in distilled water and allowed to dry.

3. Reasons technique is not recommended by CAST

3.1 The principal reason that Oil Red O is not recommended by CAST is because it is not as effective as physical developer. Although papers referenced above [4,5,8] indicate the reverse to be true, these experiments have been performed using single, sebum-rich marks, which are not truly representative of what may be encountered on real exhibits. Subsequent experiments using 'natural' fingerprints and depletion series of marks [6] are in general accord that:

- Oil Red O is not effective on marks older than four weeks;
- Oil Red O is not effective on marks exposed to prolonged immersion in water;
- some solvents used in ninhydrin and 1,8-diazafluoren-9-one (DFO) formulations outside the UK (e.g. petroleum ether) may dissolve the constituents targeted by Oil Red O and therefore it cannot be used in sequence after these processes.

3.2 A small-scale study carried out by CAST on marks known to be one year old confirmed that physical developer was a far more effective reagent and that Oil Red O developed very few marks on articles of this age.



a) b)

Palm print approximately one year old a) treated with physical developer b) treated with Oil Red O

3.3 In addition to this, although Oil Red O involves fewer processing steps overall than physical developer, it may actually take up to 90 minutes for marks to develop and therefore the whole process may actually be slower in many cases.

3.4 For these reasons, CAST does not currently (2011) see any operational benefit in recommending Oil Red O as a replacement for, or in sequence with, physical developer.

4. References

1. **Sutcliffe, L.** (2000) *Lipid Reagents*, HOSDB Placement Year Report. London: Home Office.

2. **Castello, A., Alvarez, M., Miquel, M. and Verdu, F.** (2002) 'Long lasting lipsticks and latent prints', *Forens. Sci. Comm*., vol. 4 (2).

3. **Beaudoin, A.** (2004) 'New Technique for Revealing Latent Fingerprints on Wet, Porous Surfaces: Oil Red O', *J. Forens. Ident*., vol. 54 (4), pp 413–421.

4. **Rawji, A. and Beaudoin, A.** (2006) 'Oil Red O Versus Physical Developer on Wet Papers: A Comparative Study', *J. Forens. Ident*., vol. 56 (1), pp 33–52.

5. **Guigui, K. and Beaudoin, A.** (2007) 'The Use of Oil Red O in Sequence with other Methods of Fingerprint Development', *J. Forens. Ident*., vol. 57 (4), pp 550–581.

6. **Salama, J., Aumeer-Donovan, S., Lennard, C. and Roux, C.** (2008) 'Evaluation of the Fingermark Reagent Oil Red O as a Possible Replacement for Physical Developer', *J. Forens. Ident*., vol. 58 (2), pp 203–237.

7. **Horobin, R. W.** (1981) 'Structure-staining relationships in histochemistry and biological staining. Part 3. Some comments on the intentional and artifactual staining of lipids', *Acta histochemica*, Supp.-Band XXIV, S, pp 237–246.

8. **Wood, M. A. and James, T.** (2009) 'Latent fingerprint persistence and development techniques on wet surfaces', *Fingerprint Whorld*, vol. 35 (135), pp90–100

9. **Wood, M. A. and James, T.** (2009) 'ORO: The Physical Development Replacement?' *Sci. & Jus.*, vol. 49 (4), pp 272–276.

5.13 Other lipid specific reagents

5.13.1 Ruthenium tetroxide (RTX)

1. History

1.1 The use of ruthenium (and osmium) tetroxide for fingerprint development has been reported since the 1920s [1]. In its early application the process was extremely dangerous to use, requiring ruthenium crystals to be heated in a water bath at temperatures not exceeding 50ºC. Explosions could occur if heating was too rapid or the temperature exceeded 50ºC, making the technique unsuitable for use in most laboratories [2].

1.2 The risk of explosion while fuming ruthenium tetroxide was overcome by the discovery of a chemical method for producing fumes by Mashiko *et al.*[3]. In this technique a solution consisting of 0.1g of ruthenium chloride (III) hydrate in 100mL of water was added to a second solution containing 11.3g of ammonium cerium (IV) nitrate in 100mL of water. The fumes generated in this reaction were circulated with a development chamber using a fan, and the authors demonstrated that sebaceous fingerprints could be developed on both porous and non-porous surfaces. Some work on sequential treatment was carried out, showing that ruthenium tetroxide must be used before ninhydrin and 1,8-diazafluoren-9-one (DFO), but cannot be used in sequence with physical developer. Some interference with superglue and Gentian Violet (basic violet 3) processing was also observed.

1.3 However, it was found difficult to generate sufficient quantities of fumes by the chemical reaction process and Mashiko and Miyamoto [4] later proposed a solution consisting of 0.25g per 100mL of tetradecafluorohexane (C_6F_{14}), which was applied to articles via spraying directly from a glass bottle through a nozzle. Solution dipping was also proposed for exhibits such as adhesive tapes. Wilkinson *et al.* [5] investigated the use of ruthenium tetroxide solution for the development of fingerprints on skin and although the process was found capable of developing marks, these appeared to be of lower contrast than marks produced using other techniques, and could not be lifted.

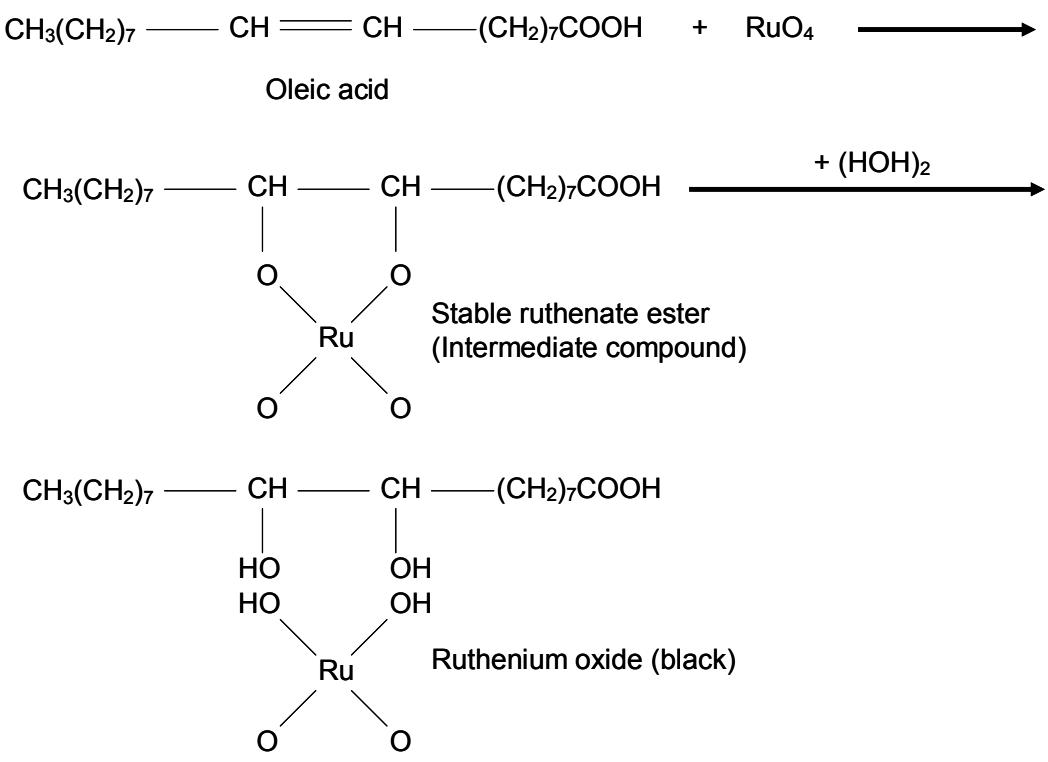
1.4 Mashiko later developed ruthenium tetroxide as a commercial product and has advertised its use in fingerprint journals, [6] although there has been ongoing debate about the safety of the process [7,8].

1.5 In the one comparative study carried out to date, Mashiko's commercial product was not used for cost reasons and the researchers attempted to prepare solutions by dissolving ruthenium tetroxide fumes in carrier solvents of 1-methoxynonafluorobutane (HFE7100) or 2,3-dihydrodecafluoropentane (HFC4310mee). The best results were obtained from HFE7100, which gave a solution of equivalent effectiveness to the commercial formulation. Ruthenium tetroxide

solution was then spray applied and the results obtained compared with those obtained from spray application of iodine solution and powdering. In these trials ruthenium tetroxide was only found to be the best process for very fresh marks on wallpaper and paint. For marks over one day old, performance decreased significantly. Ruthenium tetroxide could not be used in sequence with powders, and inhibited the take-up of fluorescent dye in marks developed using superglue.

2. Theory

2.1 Ruthenium tetroxide (and the closely related process osmium tetroxide) develops fingerprints by reacting across the carbon double bonds present in unsaturated fatty acids in fingerprint residues. The reaction product is a black hydrous oxide that allows the fingerprint to be visualised.



Reduction of ruthenium tetroxide by reaction with unsaturated fatty acids, (adapted from equivalent reaction for osmium tetroxide [2]).

2.2 The same reaction will occur whether ruthenium tetroxide is applied by fuming or in solution.

3. Reasons technique is not recommended by CAST

3.1 The Home Office Centre for Applied Science and Technology (CAST) does not recommend the use of ruthenium tetroxide because it is not as effective as other available processes and there are health and safety concerns about its use.

3.2 HOSDB has not carried out any comparative studies on ruthenium tetroxide because of health and safety concerns raised by other researchers. In the only published comparative study involving ruthenium tetroxide published to date [9], the reagent was found to be less effective than both powders and iodine-benzoflavone spray.

3.3 With regard to the health and safety aspects, there has been published debate about whether ruthenium tetroxide is toxic or not. There is also confusion as to whether the material safety data sheet (MSDS) data referred to in information supplied with the commercial product are for ruthenium dioxide or ruthenium tetroxide. CAST has reviewed the chemical literature available on the toxicity of ruthenium tetroxide and at best the substance has not been fully evaluated. Until this has been satisfactorily resolved CAST does not intend to carry out comparative trials or to recommend the process for operational use.

4. References

1. **Morris, J. R.** (1974) *An Examination of the Chemical Literature on Fingerprint Technology for the Period 1890 to August 1974*, AWRE SSCD Memorandum 359, October. Aldermaston: Atomic Weapons Research Establishment.

2. **Olsen, R. D.** (1975) 'The Oils of Latent Fingerprints', *Fingerprint and Ident.*, January, pp 3–12.

3. **Mashiko, K., German, E. R., Motojima, K. and Colman, C. D.** (1991) 'RTX: A New Ruthenium Tetroxide Fuming Procedure', *J. Forens. Ident*., vol. 41 (6), pp 429–436.

4. **Mashiko, K. and Miyamoto, T.** (1998) 'Latent Fingerprint Processing by the Ruthenium Tetroxide Method', *J. Forens. Ident*., vol. 48 (3), pp 279–290.

5. **Wilkinson, D. A., Watkin, J. E. and Misner, A. H.** (1996) 'A Comparison of Techniques for the Visualisation of Fingerprints on Human Skin including the Application of Iodine and α-Napthoflavone', *J. Forens. Ident*., vol. 46 (4), pp 432–451.

6. **Mashiko, K.** (2005) 'Safe New Formulation for Ruthenium Tetroxide (RTX)', *Fingerprint Whorld*, vol. 31 (121), pp 144–146.

7. **Blackledge, R. D.** (1998) 'Re: 'Latent Print Processing by the Ruthenium Tetroxide Method', *J. Forens. Ident*., vol. 48 (5), pp 557–559.

8. **Mashiko, K.** (1999) 'Letter – Re: "Latent Print Processing by the Ruthenium Tetroxide Method", *J. Forens. Ident*., vol. 49 (2), pp 111–112.

9. **Flynn, K., Maynard, P., Du Pasquier, E., Lennard, C., Stoilovic, M. and Roux, C.** (2004) 'Evaluation of Iodine-Benzoflavone and Ruthenium Tetroxide Spray Reagents for the Detection of Latent Fingermarks at the Crime Scene', *J. Forens. Sci*, vol. 49 (4), pp 707–715.

5.13.2 Osmium tetroxide

1. History

1.1 Osmium tetroxide was already being proposed as a reagent for developing fingerprints on paper in the early 1900s. By 1920, Mitchell [1] was able to describe two application techniques, namely 'osmic acid', a 1% aqueous solution of osmium tetroxide brushed onto a document, and osmium tetroxide fuming, where the paper exhibit was held over a dish of the boiling 1% aqueous solution. The 'osmic acid' solution treatment was stated to produce black marks if the surface was kept moist whilst exposed to sunlight, whereas the prints produced in the fuming process were grey. A further fuming process was later proposed, involving placing osmium tetroxide crystals in a small, shallow glass dish within a fuming cabinet and adding ethyl ether or carbon tetrachloride [2]. It was essential not to apply heat in this process because of the risk of an explosion.

1.2 Later researchers used pre-prepared ampoules of osmium tetroxide within a fuming cabinet, and used a sensitising chemical called 5-norbornene-2-carbonyl chloride in vapour form as a pre-treatment to produce additional linkages for the osmium tetroxide to react with [3].

1.3 Bones [4] carried out a detailed assessment of the osmium tetroxide fuming process, looking at different environments for the fuming process (air, argon), different development conditions (light, dark, vacuum) and the effects of ageing and humidity on the quality of prints developed. It was concluded that the process was equivalent to ninhydrin in sensitivity, and that the optimum processing conditions were in an air environment and in darkness. It was also shown that osmium tetroxide could develop handprints on fabrics, although there was negligible ridge detail visible.

1.4 Smith Jr [5] later proposed the osmium tetroxide fuming technique for the development of fingerprints on adhesive tapes, including medical

tapes and strapping tapes. The exhibits were processed in air and stored in the dark; progressive darkening of the substrate was observed if exhibits were exposed to the light, and this could obscure marks.

1.5 In the early 1980s the Home Office Scientific Research and Development Branch (HO SRDB) included osmium tetroxide in a comparative study of techniques for development of fingerprints on fabrics [6], which included vacuum metal deposition and radioactive sulphur dioxide. Of these techniques osmium tetroxide, both as a fuming process and in solution, proved significantly less effective than radioactive sulphur dioxide and vacuum metal deposition, and no further work was carried out on this reagent.

2. Theory

2.1 The theory associated with osmium tetroxide is identical to that described for ruthenium tetroxide above. Osmium tetroxide reacts across the carbon double bonds in the unsaturated fatty acids within fingerprint deposits to form intermediate osmate ester compounds that finally produce the black osmium dioxide compound [7].

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend the osmium tetroxide process because of the highly toxic nature of the substance. In comparative studies that have been carried out it has not proved to be any more effective than any other process currently (2011) in use.

4. References

1. **Mitchell, C. A.** (1920) ‘The Detection of Fingerprints on Documents’, *Anal*., vol. 45, pp 122–129.

2. **Olsen Sr, R. D.** (1978) *Scott’s Fingerprint Mechanics*, ISBN 0-398-06308-7. Springfield, Illinois, USA: Charles C. Thomas.

3. **Kerr, F. M.** (1978) ‘Using Osmium Tetroxide to Develop Latent Fingerprints’, *RMCP Gazette*, vol. 40 (3), pp 28–29.

4. **Bones, C.** (1974) *Report on Industrial Training during the Period April–September 1974 at AWRE Aldermaston*, SCS Report No. 431. Aldermaston: Atomic Weapons Research Establishment.

5. **Smith Jr, D. W.** (1977) ‘A Practical Method for the Recovery of Latent Impressions on Adhesive Surfaces’, *Ident. News*, vol. 27 (10), October.

6. **Albinson, R. A.** (1984) *The Development of Latent Fingerprints on Fabric*, Draft HO SRDB Report No.72/84 (unpublished). London: Home Office.

7. **Olsen, R. D.** (1975) 'The Oils of Latent Fingerprints', *Fingerprints and Ident.*, January, pp 3–12.

5.13.3 Europium chelate

1. History

1.1 The use of lanthanide series elements in fingerprint detection has been considered in a range of techniques. The attraction of these elements is that they can form fluorescent complexes with large Stokes shifts, meaning that they can be illuminated in the ultraviolet region of the spectrum and emit in the red/infra-red region. The decay time during fluorescence is also longer than many other fluorescent species, making them useful in time-resolved imaging applications and for visualising fingerprints on fluorescing backgrounds.

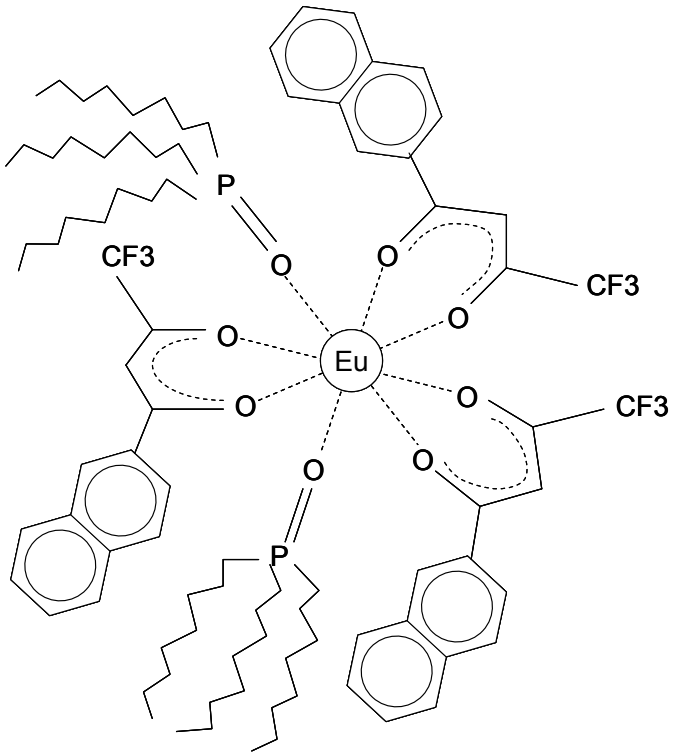
1.2 Initial studies into the potential of these elements for fingerprint detection utilised europium salts as complexing agents for the post-treatment of marks developed using ninhydrin [1]. However, it was recognised that europium complexes also had potential for use as a superglue dye, especially in circumstances where background fluorescence caused problems and a large Stokes shift was desirable [2-4]. The dye was successfully applied to superglue marks developed on multicoloured surfaces and on skin. Dyes were dissolved in methyl ethyl ketone [2,3] or petroleum ether [4].

1.3 Later researchers have considered europium chelates as a fingerprint development reagent in their own right, producing a range of formulations that can either be applied by spraying or as a solution that exhibits can be dipped into [5-9]. Bright, fluorescent marks were successfully developed on both porous and non-porous items in laboratory trials, although these were not replicated when the technique was applied to casework.

2. Theory

2.1 The theory associated with the europium chelate reagent is that the europium complex is in some way attracted by the lipid components of the fingerprint deposit and absorbed into it from solution. Wilkinson [7] suggests that the presence of methanol may aid the transfer process from solution into the fingerprint. Methanol partially dissolves in the lipids of the fingerprint residue, and because the europium complex is water insoluble and prefers the hydrophobic environment of the fingerprint lipids, some of the complex is transferred with the methanol. Once

absorbed by the lipids, the water molecules attached to the europium complex are displaced and replaced by various lipid-based ligands. The resultant structure is a fluorophore and will fluoresce when illuminated with light of an appropriate wavelength.



Structure of biological fluorophore [7].

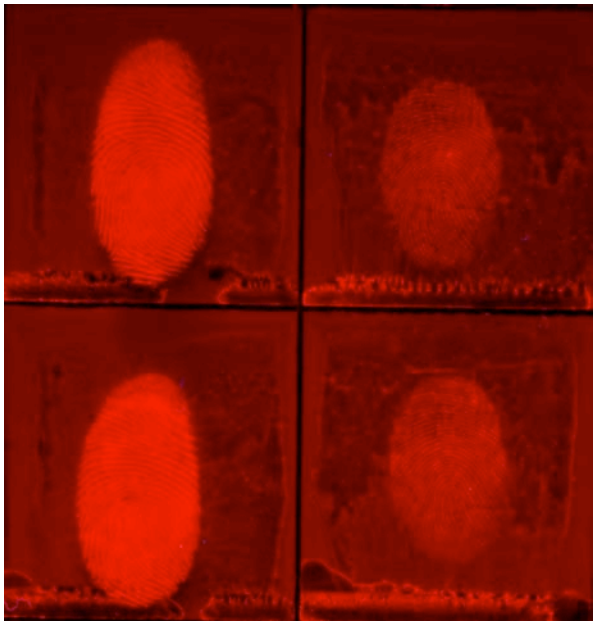
2.2 The bulky fluorophore structure protects the europium from the aqueous environment of the biological medium (in this case the water present in the fingerprint residue). A detergent is added to further isolate the europium ion from the water molecules.

2.3 The formula proposed by Wilkinson [7] is made up as a two- part system and is as follows:

Solution A – 23mg europium chloride hexahydrate;
300mL distilled water;
2mL Tergitol 7.

Solution B – 42mg thenoyltrifluoroacetone;
50mg trioctyl phosphine oxide;
700mL methanol.

2.4 The two solutions are then mixed together for 30 minutes, and articles to be treated are immersed in the resultant solution for 5 seconds then washed in water and allowed to dry.



Sebaceous marks deposited on a ceramic tile and developed using europium chelate.

3. Reasons technique is not recommended by CAST

3.1 CAST does not recommend the process because it has not yet been evaluated. The main reason for this is that reports from other researchers indicate that the performance of the reagent on older marks is poor, therefore it is unlikely to provide advantages over any currently (2011) recommended process. The main solvent used in the existing formulation is methanol, which is not preferred by HOSDB because of its flammability and toxicity. If the process were to be recommended, some reformulation work would be required to see if the methanol content could be reduced or eliminated.

4. References

1. **Menzel, E. R. and Mitchell, K. E.** (1990) 'Intramolecular Energy Transfer in the Europium-Ruhemann's Purple Complex: Application to Latent Fingerprint Detection', *J. Forens. Sci*, vol. 35 (1), pp 35–45.

2. **Misner, A., Wilkinson, D. and Watkin, J.** (1993) 'Thenoyl Europium Chelate: A New Fluorescent Dye with a Narrow Emission Band to Detect Cyanoacrylate Developed Fingerprints on Non-porous Substrates and Cadavers', *J. Forens. Ident*., vol. 43 (2), pp 154–165.

3. **Wilkinson, D. A. and Watkin, J. E.** (1993) 'Europium Aryl-β-Diketone Complexes as Fluorescent Dyes for the Detection of Cyanoacrylate Developed Fingerprints on Human Skin', *Forens. Sci. Int*., vol. 60, pp 67–79.

4. **Lock, E. R. A., Mazella, W. D. and Margot, P.** (1995) 'A New Europium Chelate as a Fluorescent Dye for Cyanoacrylate Pretreated Fingerprints – EuTTAPhen: Europium ThenoylTrifluoro Acetone Ortho-Phenanthroline', *J. Forens. Sci*, vol. 40 (4), pp 654–658.

5. **Allred, C. E., Murdock, R. H. and Menzel, E. R.** (1997) 'New Lipid-specific, Rare Earth-based Chemical Fingerprint Detection Method', *J. Forens. Ident*., vol. 47 (5), pp 542–556.

6. **Allred, C. E. and Menzel, E. R.** (1997) 'A Novel Bio-conjugate Method for Latent Fingerprint Detection', *Forens. Sci. Int*., vol. 85, pp 83–94.

7. **Wilkinson, D.** (1999) 'A One-step Fluorescent Detection Method for Lipid Fingerprints; $Eu(TTA)_3.2TOPO$', *Forens. Sci. Int*., vol. 99, pp 5–23.

8. **Caldwell, J. P., Henderson, W. and Kim, N. D.** (2001) 'Luminescent Visualisation of Latent Fingerprints by Direct Reaction with a Lanthanide Shift Reagent', *J. Forens. Sci*, vol. 46 (6), pp 1332–1341.

9. **Li, C., Li, B., Yu, S., Gao, J. and Yao, P.** (2004) 'Study on the Direct Developing of a Latent Fingerprint Using a New Fluorescent Developer', *J. Forens. Ident*., vol. 54 (6), pp 653–659.

5.14 Radioactive sulphur dioxide

1. History

1.1 The potential application of radioactive sulphur dioxide ($^{35}SO_2$) for the development of latent fingerprints was first reported by Grant *et al.* in 1963 [1] during the course of investigations into the resistance of paper to attack by atmospheric pollution. They observed that when developing autoradiographs of paper treated with SO_2, spots could be seen on the paper that on closer examination were identified as fingerprints. A further publication by Grant *et al.* [2] gave more background detail on the method used. Radioactive SO_2 was measured into an evacuated flask and the pressure raised to atmospheric by the addition of air at a controlled humidity of 66%. The paper sample was exposed to the gas mixture for 12 hours, then placed against x-ray film for 1 week. Other experiments carried out by the researchers demonstrated that ageing of the marks reduced the chances of fingerprint development. It was also found that alkaline fillers in the paper could give rise to heavy SO_2 take-up by the background and that metal impurities also picked up SO_2.

1.2 The results of further research into the technique were reported by Spedding in 1971 [3]. He suggested that SO_2 was reacting with the lipids present in fingerprint deposits and noted that reactions occurred with oleic and linoleic acids. Prints were also developed on paper that had been wetted. Spedding *et al.* also issued a more detailed report [4], providing details of the apparatus used for development of fingerprints. This consisted of a treatment box within which samples could be hung on a rail. Humidity inside the box was raised to 60%, radioactive SO_2 introduced and the samples exposed for 30 minutes before being removed and placed in contact with x-ray film. Trials were also conducted for a range of paper types, comparing the effectiveness of SO_2 for fingerprint development with that of ninhydrin and iodine. SO_2 was found to be the most effective technique across the range of paper types investigated. Spedding *et al.* also considered the potential effects of the SO_2 technique on subsequent development techniques, in particular the 60% humidity and SO_2 concentration used. It was considered that the humidity could be detrimental to the subsequent use of silver nitrate, but that other techniques should be unaffected. The report also suggested that radioactive SO_2 could prove a useful technique for the development of fingerprints on fabrics.

1.3 The wider application of the technique to substrates other than paper was reported in late 1970 [5]. Excellent results were reported for PVC sheet and initial results on fabrics were encouraging. Further studies into the optimum humidity for treatment were presented, with humidities in excess of 60% giving rise to an increase in the uptake of SO_2 in the substrate compared with that in the fingerprint, and therefore being undesirable.

1.4 The initial results obtained for paper exhibits had been encouraging and the technique was used on operational exhibits of types that had previously given poor results with ninhydrin, iodine and silver nitrate. An early operational success was obtained on forged £5 notes [6].

1.5 Research into the technique continued, with the objectives of establishing optimum processing conditions and the range of substrates that radioactive SO_2 could develop marks on. A more detailed study was carried out into fingerprints deposited on paper [7], investigating the effect of storage time (1–6 days) and storage humidity (31–93% relative humidity) on the quality of fingerprints developed using radioactive SO_2, ninhydrin, iodine, silver nitrate and vacuum metal deposition (VMD), then also in a developmental stage. Across the range of conditions studied VMD gave the best results, followed by ninhydrin. In these trials radioactive SO_2 performed relatively poorly. In contrast, studies conducted on dry paper identified SO_2 as being more effective than ninhydrin, silver nitrate and a sequence of ninhydrin followed by silver nitrate [8]. One advantage of the SO_2 process was that it eliminated much of the printed text that could potentially obscure minutiae. An optimum development sequence of SO_2 > ninhydrin > silver nitrate was proposed for paper exhibits.

1.6 The major area of research for the practical application of radioactive SO_2 was the development of fingerprints on fabrics and a comprehensive report into these studies was issued by Wells in 1975 [9]. The equipment used in these studies consisted of a 150 litre Perspex box into which a mixture of radioactive SO_2 and nitrogen gas (N_2) was introduced. The optimum humidity was identified as 65%, but effectiveness fell rapidly in the range 66–75% and 60% was recommended for operational purposes. The addition of ozone into the gas mixture was found to increase SO_2 uptake by the fingerprint and thus reduce autoradiography times. An autoradiography guide was developed for a range of substrates including fabrics, plastic wrappings and banknotes, outlining optimum development times. The use of a dark, sealed enclosure containing desiccant was recommended for storage of exhibits prior to treatment. Fingerprints were successfully developed on a wide range of fabrics, although the quality and number of marks were significantly reduced when ageing conditions involving any degree of high humidity were used. Extended exposure to atmospheric, non-radioactive SO_2 was also thought to desensitise the print. Prints on Melinex film were least affected by these conditions, followed by prints on fabrics, with paper being the most affected. Throughout the studies operational work was performed to see if marks could be developed on real fabric exhibits and parallel studies were also performed on fabrics worn for different periods of time, both next to the skin and as outer garments. It was concluded that for operational work on fabrics, exhibits needed to be dry, of fine weave and not worn next to the skin.

1.7 In the mid-1970s an Atomic Weapons Research Establishment (AWRE) system using compressed SO_2 cylinders was used on adhesive tape

from Irish Republican Army (IRA) improvised explosive devices (IEDs) and numerous fingerprints were found. The rise of Republican terrorism and the planting of IEDs on the mainland led to a need for a method of processing adhesive tape from unexploded devices. The Police Scientific Development Branch (PSDB) worked closely with the anti-terrorist unit and eventually trained members of the unit to use the radioactive SO_2 system. A number of identifications from the terrorists fingerprints on adhesive tape were found. The main reason for the use of SO_2 was that most of the tape was black and a non-destructive method was required in order to carry out other forensic examinations for fibres, hair and mechanical fit. The equipment using a pressurised gaseous source of SO_2 was potentially hazardous and PSDB designed and built a metal-free reaction chamber and control system, and a simple Perspex chamber was developed for treatment of exhibits [10]. The source of SO_2 was changed from pressurised gas cylinders to paper impregnated with radioactive thiourea, which was ignited to release radioactive SO_2 gas. This became the standard system introduced in the UK for operational work although only two other systems were built, one for the Metropolitan Police Forensic Science Laboratory Serious Crimes Unit (MPFSL SCU) and one for the Birmingham Forensic Science Service (FSS). PSDB also developed light-tight sachets based on aluminised Melinex for autoradiography of non-flat items in daylight [11] and further research was carried out to investigate methods for developing marks on curved surfaces [12]. The development of the basic violet 3 (Gentian Violet) transfer technique subsequently reduced the need for SO_2 on tapes.

1.8 Initial studies to investigate the relative effectiveness of SO_2 on adhesive tapes established that marks could be detected on both sides of the tape using this technique. Marks were shown to survive for 64 days on the adhesive side, although survival times were shorter on the non-adhesive side and dependent on whether the tape was stored indoors or outdoors [13]. The technique was found to give excellent operational results on adhesive tapes in several terrorism-related cases in the 1970s, and became regarded as an essential treatment for this type of exhibit in serious and terrorist-related cases [14]. A further comparative trial was carried out on fabrics in the early 1980s, investigating the relative effectiveness of several techniques including SO_2, VMD and osmium tetroxide fuming [15,16]. This study looked at ageing fingerprints on a range of fabrics and types of weave. These studies showed that SO_2 was the most effective of the three techniques, developing appreciably more high quality marks than VMD, the next most effective technique.

1.9 A small operational trial was also carried out with some ridge detail being developed in at least two operational cases on fine synthetic outer garments. A small scale evaluation of SO_2 as an enhancement technique for superglue on synthetic substrates was also carried out and an identifiable, policeman's fingerprints were found on one nylon outer garment. With this operational work showing limited success, thetechnique became mainly limited to use on adhesive tapes. However, the complexity of equipment required to carry out the processing, and the

health and safety issues associated with the use of radioactive isotopes, led to a gradual decline in the operational use of the technique The last operational equipment was decommissioned by the FSS, Lambeth in 2005. CAST holds the equipment in storage in case there is a future requirement to re-investigate the technique, but at present it is unlikely to be restored to operational use at the Sandridge laboratories.

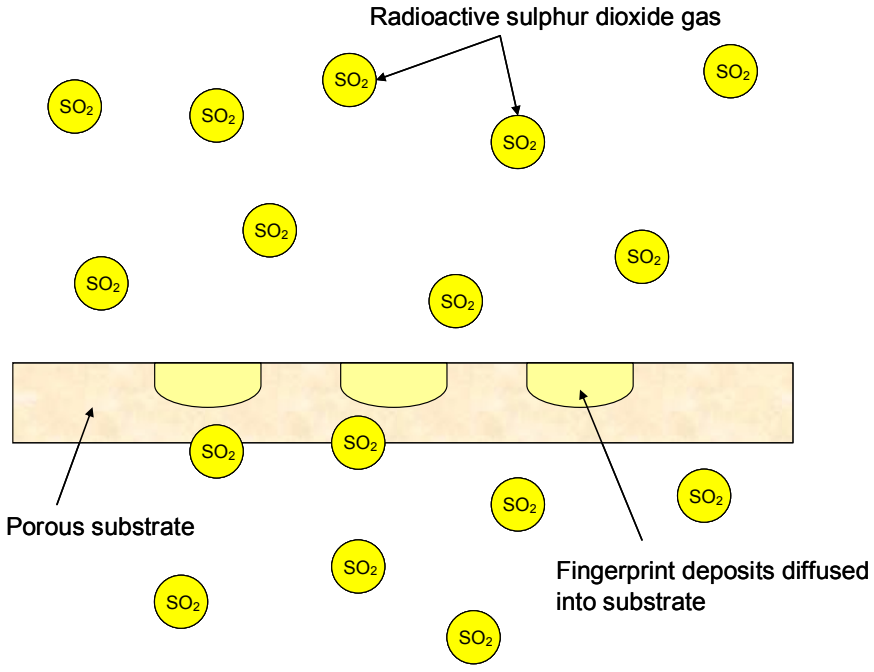
2. Theory

2.1 Wells [9] in his comprehensive report of the radioactive SO_2 process, suggested that several reactions with fingerprint deposits were possible and that a complex combination of these contributed to the fingerprint development process. The mechanisms processed by Wells included the following.

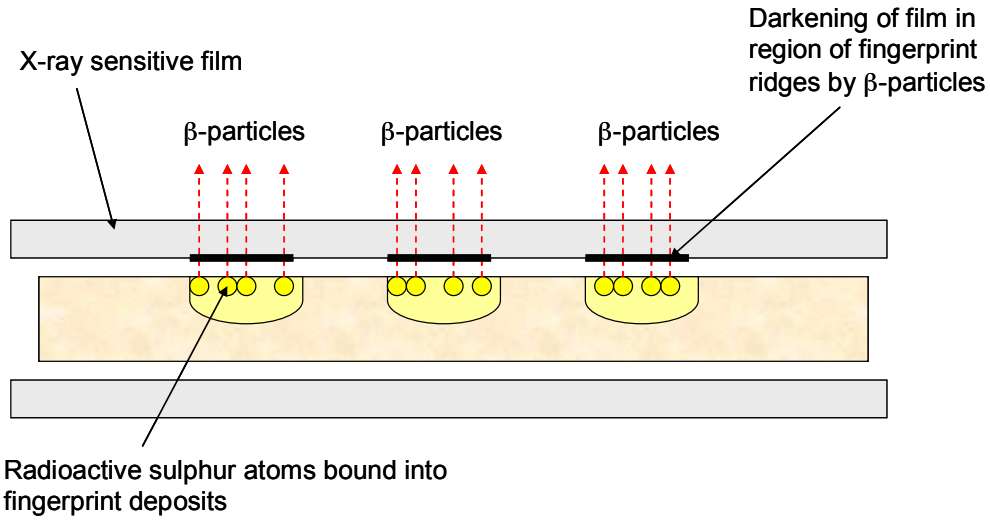
- The fixation of SO_2 as SO_4^{2-} in the water phase associated with sebum and in water adsorbed from the atmosphere due to the hygroscopic nature of the deposit.
- The sensitisation of wettable substrates (e.g. paper, fabric) by adsorbed layers of water molecules directly as a result of contact by fingerprint ridges.
- Reaction(s) with lipids, which may involve the double-bonds of unsaturated free fatty acids, etc.

2.2 The strong dependence of the SO_2 reaction on the relative humidity during treatment tends to support the theory that the main reaction occurring is the water phase fixation mechanism.

2.3 The development of fingerprints by the radioactive SO_2 process and subsequent autoradiography is illustrated schematically below.



a)



b)

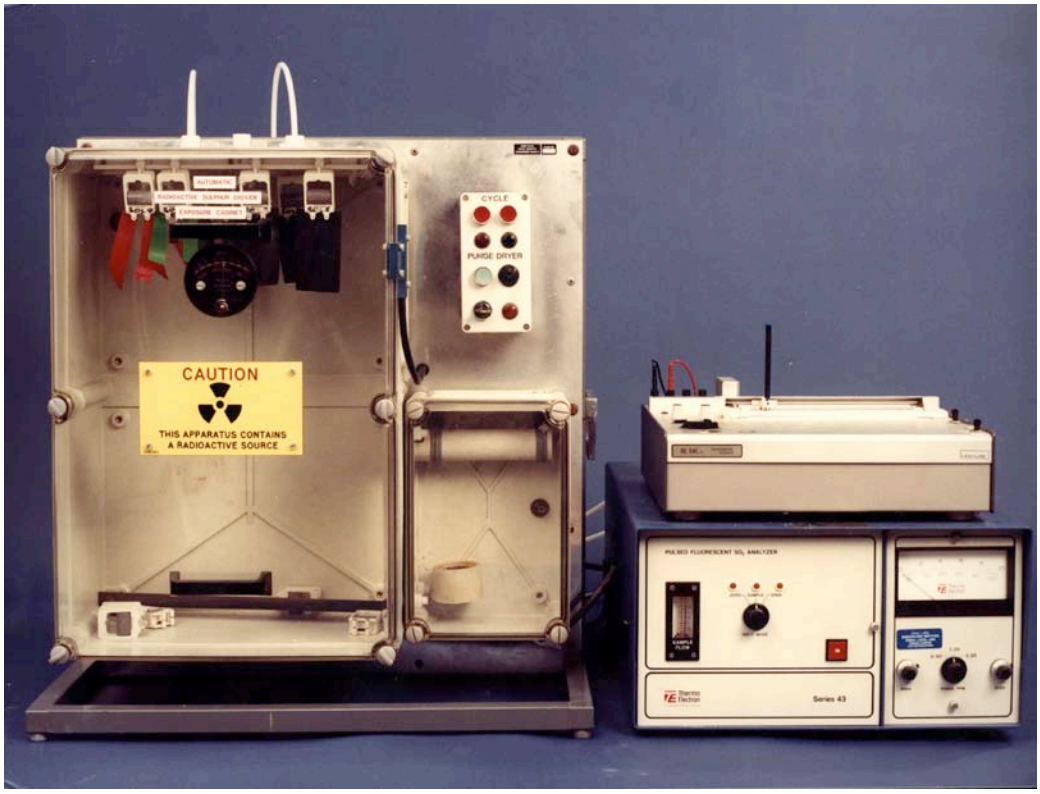
Schematic diagram illustrating the radioactive sulphur dioxide process a) sulphur dioxide gas diffusing through porous substrate and b) autoradiography of sample with radioactive sulphur bound into fingerprint ridges.

3. CAST processes

3.1 The technique ultimately recommended by the Home Office Centre for Applied Science and Technology (CAST) was the combustion of filter paper impregnated with radioactive thiourea in a humidity-controlled cabinet.

3.2 The SO_2 sources were prepared by dissolving radioactive thiourea in water and decanting small aliquots of solution onto discs of filter paper. It is essential to use readily combustible cellulose-based filter papers for this purpose. The concentration of the solution was adjusted to give a concentration of 1mCi (milliCurie) per 50μL, with 5μL being impregnated into each disc to give a disc content of 0.1mCi of thiourea.

3.3 The impregnated disc was then loaded into the crucible chamber of the radioactive SO_2 apparatus. The system used activated charcoal to remove the SO_2. After the normal treatment time of 20 minutes, the gas content of the chamber was passed through the charcoal scrubbing system. A separate chamber containing water was used in the initial humidification phase, which was manually controlled.



Photograph of the radioactive sulphur dioxide apparatus.

3.4 Samples were then suspended in the main chamber, which was sealed and brought to a relative humidity of 55%. The impregnated disc was then ignited and allowed to fill the chamber with the pre-determined concentration of radioactive SO_2 released by combustion. Once the cycle had completed and the SO_2 level had returned to the value before commencing treatment, articles were removed from the chamber, sandwiched between two sheets of x-ray film and then placed in a press. Activity was monitored with a Geiger counter to calculate exposure times, typically seven to ten days.

3.5 The humidity level in the chamber and concentration of radioactive thiourea used in the process were chosen to give the optimum conditions identified in early experimental work. The role of the thiourea in the process was to release SO_2 as a combustion product.

3.6 The process involved constant monitoring of all items of laboratory equipment, clothing and exhibits that came into contact with radioactive material, and the disposal of contaminated articles in an approved fashion.

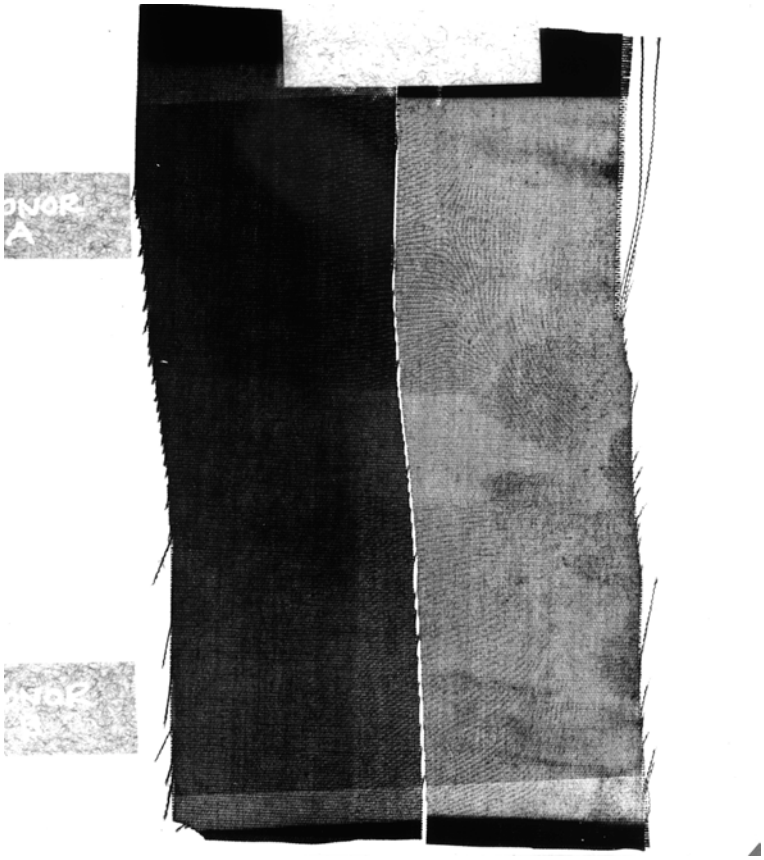
4. Critical issues

4.1 The technique is no longer used operationally and therefore there are no critical issues associated with its use. However, continuous monitoring of radioactivity levels was required when carrying out processing.

5. Application

5.1 Suitable surfaces: Radioactive SO_2 was suitable for use on both sides of adhesive tape and on fabrics. In practice it could be used on both porous and non-porous surfaces, but was restricted to articles small enough to fit inside the reaction chamber.

5.2 The two applications for which radioactive SO_2 is suggested in the *Manual of Fingerprint Development Techniques* [17] are as part of a sequential treatment process for adhesive tapes, and as the principal treatment for fabrics. In theory it was a versatile technique and could be applied to both porous and non-porous surfaces, a potential advantage being that patterned backgrounds that could obscure the developed mark were not visible in the autoradiograph.



Autoradiograph of fabric sample exposed to different environments and treated with radioactive sulphur dioxide.

5.3 When applied to adhesive tapes, the technique was capable of developing marks on both sides of the tape simultaneously, and was also effective on vinyl tapes where techniques such as VMD performed poorly. During the 1970s this type of tape was often found on explosive devices and radioactive SO_2 gave good results, resulting in its continued use on terrorist-related cases until the mid-2000s.

5.4 The technique was shown to be the most effective process for development of marks on fabrics, although in practice no marks with sufficient detail for a positive identification were obtained from operational work.

6. Alternative formulations and processes

6.1 Other vapour phase materials labelled with radioactive isotopes have been considered for the development of fingerprints using autoradiographic methods. Goode *et al*. [10, 18] considered the use of radioactive bromine in two forms, ^{80}Br and ^{82}Br. Bromine was considered for its potential reaction with unsaturated fats in the fingerprint deposit and for the fact that this reaction is rapid. Both isotopes also have a shorter half life than radioactive SO_2, which is advantageous. Fingerprints were successfully developed on a range of paper substrates using radioactive Br_2 and the process shown to be quicker than SO_2 [18]. The quality of the developed prints was shown to be similar to those produced by SO_2, although the contrast of the marks was significantly degraded by exposure to ultraviolet radiation. The technique as originally applied utilised vacuum equipment and this was thought to be clumsy compared with the apparatus used for the more established SO_2 technique. As a consequence, radioactive Br_2 was not pursued further.

6.2 Higgins also reported the use of radioactive iodine (^{128}I) in iodine vapour and in radioactive iodine monochloride (ICl) [19] for the development of fingerprints on paper and again a reduced processing time was achieved compared with SO_2. Although initial trials were successful, the technique was not progressed further.

7. Post-treatments

7.1 No post-treatments are used with the radioactive SO_2 technique other than autoradiography for developing the marks on photographic paper.

8.Validation and operational experience

8.1 <u>Laboratory trials</u>

8.1.1 The largest recorded laboratory trial for radioactive SO_2 was a comparison with VMD on a range of different fabrics representative of over- and undergarments [16]. This trial used six donors, each placing one mark that was split and aged for one day prior to processing. The results are outlined below in terms of individual fabric type, and further summarised in the second table.

Material	**Process**	**Grade of mark**			
		1	**2**	**3**	**4**
Brown, 100% Nylon Warp knit, 1.5 stitches x 3 rows	SO_2	0	2	3	1
	VMD	1	5	0	0
Cream, 100% Silk Standard weave, 3 weft x 3 warp	SO_2	0	3	1	2
	VMD	0	5	1	0
White, 100% Acetate Standard weave, 3 weft x 3.5 warp	SO_2	0	1	4	1
	VMD	4	2	0	0
Grey, 100% Polyester Standard weave, 3.5 weft x 4 warp	SO_2	6	0	0	0
	VMD	5	1	0	0
Cream, 65/35% Polyester/Cotton Standard weave, 3 weft x 4 warp (well worn)	SO_2	6	0	0	0
	VMD	6	0	0	0
White, 65/35% Polyester/Cotton Standard weave, 3 weft x 4 warp	SO_2	3	3	0	0
	VMD	0	4	2	0
White/blue stripe, 65/35% Polyester/ Cotton, Standard weave, 3 weft x 4 warp (well worn)	SO_2	3	3	0	0
	VMD	2	4	0	0
Yellow, 65/35% Polyester/Cotton Standard weave, 3 weft x 4.5 warp (well worn)	SO_2	0	6	0	0
	VMD	2	4	0	0
Red, 80/20% Polyester/Cotton Standard weave, 3 weft x 3.5 warp	SO_2	2	3	1	0
	VMD	3	3	0	0
White, 100% Nylon 'antistat' Warp knit, 1.25 stitches x 2 rows (well worn)	SO_2	0	5	1	0
	VMD	0	6	0	0
White, 100% Nylon Kayser 'antistat', warp knit, 1.5 stitches x 2 rows	SO_2	0	5	1	0
	VMD	0	6	0	0
White, 100% Nylon	SO_2	0	5	1	0

Fine Fare, Warp knit, 2 stitches x 2 rows	VMD	0	6	0	0
White, 100% Nylon 'counterstat', Warp knit, 2 stitches x 2.5 rows	SO_2	1	4	1	0
	VMD	0	4	2	0
White, 100% Nylon Kayser, Warp knit, 2 stitches x 2.5 rows	SO_2	0	6	0	0
	VMD	0	5	1	0
White, 100% Polyester, 4 Float satin weave, 4 weft x 4 warp	SO_2	0	1	2	3
	VMD	0	3	3	0

Material	**Process**	**Grade of mark**			
		1	**2**	**3**	**4**
All fabrics	SO_2	21	47	15	7
	VMD	23	58	9	0

Summary of comparative study carried out between radioactive sulphur dioxide and vacuum metal deposition on fabrics.

8.1.2 The results indicate that that radioactive SO_2 produced about 50% more marks with ridge detail worth initiating a search against (grades 3 and 4) than VMD, and hence radioactive SO_2 was the principal technique recommended for treatment of fabrics. It should be noted that when this trial was conducted in 1984, an optimised superglue technique was not available.

8.1.3 A similar comparison was carried out between radioactive ICl and SO_2, again using six donors, each placing one mark that was split and aged for one day prior to processing. ICl did not develop any marks in this study, but radioactive SO_2 was found to give similar results to the initial trial against VMD.

Material	**Process**	**Grade of mark**			
		1	**2**	**3**	**4**
All fabrics (one-day-old marks)	SO_2	33	34	14	3
	ICl	84	0	0	0
All fabrics (one-week-old marks)	SO_2	45	27	8	4
	ICl	84	0	0	0

Results of further comparative studies between radioactive sulphur dioxide and iodine monochloride on fabrics.

8.1.4 Further comparative trials against osmium tetroxide as a fuming process and as a spray also showed SO_2 to be the most effective process on fabrics.

8.2 Pseudo-operational trials and operational experience

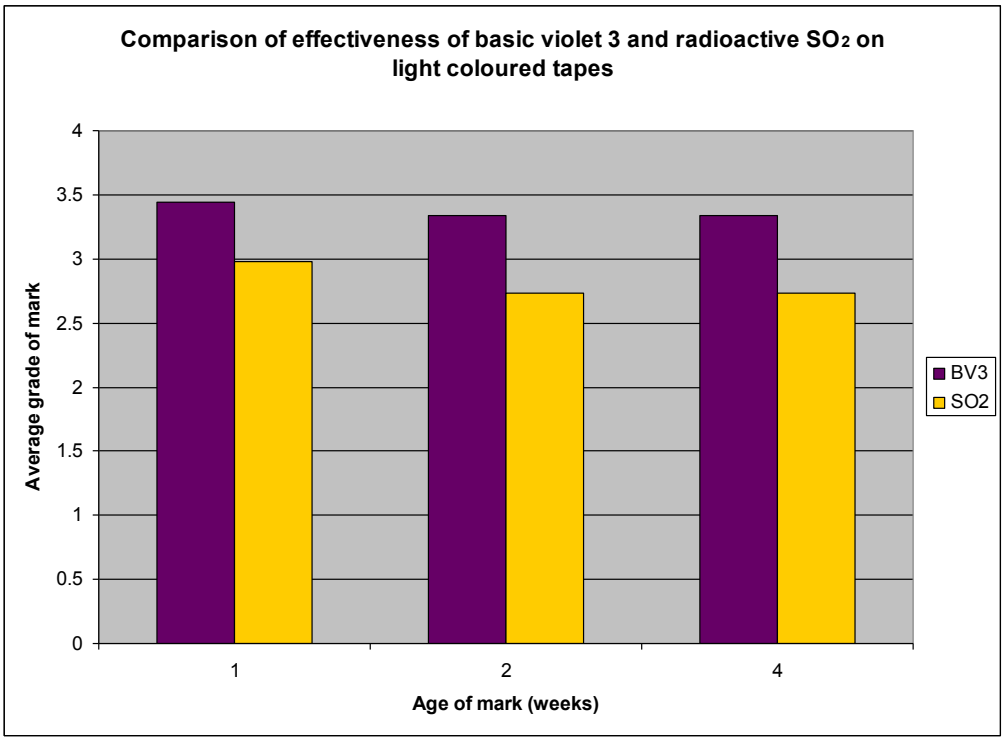
8.2.1 Operational figures for the first years that radioactive SO_2 was used are summarised below.

Period	Number of cases	Number of articles	Marks developed (cases)	Comments
01/01/1975 – 01/05/1975	4	118	4	Mainly PVC tapes
02/05/1975 – 02/05/1976	28	677	12	Mainly tapes and plastic bags
03/05/1976 – 31/03/1977	49	754	9 + 7 with fragmentary marks	Mainly tapes

Results of casework using radioactive sulphur dioxide in the mid-1970s.

8.2.2 The main successes of the technique were on dark and coloured PVC tape, where none of the techniques then available (basic violet 3, VMD) were capable of yielding marks. Many of these successes were in high-profile cases involving explosive devices.

8.2.3 A laboratory comparison was carried out by PSDB in the late 1970s, comparing basic violet 3 and radioactive SO_2 on a range of light coloured adhesive tapes. The results from approximately 300 graded marks are illustrated below.



Comparison of the relative effectiveness of radioactive sulphur dioxide and basic violet 3 on a range of light coloured adhesive tapes

8.2.4 These results indicate that basic violet 3 is the more effective process, but the processes do not target the same constituents and may be used in sequence. In 1983 HO SRDB treated a series of tape exhibits using radioactive SO_2 and developed 11 marks, only one of which was subsequently detected by basic violet 3.

8.2.5 A further operational case in 1984 gave a further opportunity to assess the sequential processing of tapes from 18 separate exhibits, using basic violet 3 after radioactive SO_2. Results of this exercise are summarised below.

Side of tape	Both processes negative	Both processes developed same ridge detail	SO_2 developed more ridge detail	Basic violet 3 developed more ridge detail
Adhesive	28	0	2	11
Non-adhesive	18	12	10	1

Results of casework using radioactive sulphur dioxide and basic violet 3 on adhesive tape in 1984.

8.2.6 Again, basic violet 3 appeared the more effective process on the adhesive side but it was apparent that the two processes could be used sequentially.

8.2.7 The laboratory trials conducted by HO SRDB and reported above indicated that radioactive SO_2 was the most effective technique for development of marks on fabrics, and selected operational exhibits were treated between 1980 and 1984.

Period	Number of cases	Number of articles	Marks developed (cases)	Comments
01/09/1980 – 01/10/1984	12	13	4	No marks with sufficient ridge detail for identification

Results of casework using radioactive sulphur dioxide on fabrics from 1980 to 1984.

8.2.8 The factors affecting the recovery of identifiable marks were the time lapse before receipt of the exhibit (in many cases greater than one week) and the pattern of the fabric warp/weft obscuring ridge detail. At the present time (2011), the number of points of detail required for identification of a fingerprint are less than they were in the 1980s (when a minimum 16-point standard was in place), and there are digital filtering techniques that can remove the patterned background from the image (such as fast Fourier transforms). Both these factors may have made the marks developed more operationally significant if developed in the current environment.

9. References

1. **Grant, R. L., Hudson, F. L. and Hockey, J. A.** 'Detecting Fingerprints on Paper' (1963) *Nature* 200, p 1348.

2. **Grant, R. L., Hudson, F.L. and Hockey, J. A.** (1963) 'A New Method of Detecting Fingerprints on Paper', *J. Forens. Sci. Soc.*, vol. 4 (2), pp 85–86.

3. **Spedding, D. J.** (1971) 'Detection of Latent Fingerprints with $^{35}SO_2$', *Nature,* 229, pp 123–124.

4. **Spedding, D. J., Rowlands, R. P. and Heard, M. J.** (1970) *A Method for the Detection of Fingerprints on Paper using Sulphur-35 Sulphur Dioxide*, AERE Harwell Memorandum M 2293. Harwell: Atomic Energy Research Establishment

5. **PSDB** (1971) *Report of an Informal Symposium on Fingerprints held at AWRE Aldermaston on 2 December 1970*, Home Office PSDB Report 2/71, March. London: Home Office.

6. **Godsell, J. W.** (1972) *Successful application of the radio-active sulphur dioxide method to development of latent fingermarks on genuine and forged bank notes in operational cases*, Home Office PSDB Technical Note 1/72. London: Home Office.

7. **Godsell, J. W., Vincent, P. G. and Lloyd, D. W.** (1972) *Influence of storage of latent fingerprints in an atmosphere of a given humidity on the choice of the optimum developing agent*, Home Office PSDB Technical Memorandum 3/72. London: Home Office.

8. **Godsell, J. W. and Vincent, P. G.** (1973) *Comparative Study of Radio-Active Sulphur Dioxide, Ninhydrin and Silver Nitrate from the point of view of their Efficiency for Developing Latent Fingerprints on Paper*, Home Office PSDB Technical Memorandum 1/73. London: Home Office.

9. **Wells, A. C.** (1975) *An autoradiographic technique for revealing fingerprints on intractable surfaces using radioactive sulphur dioxide*, AERE Harwell Report R7635, September. Harwell: Atomic Energy Research Establishment.

10. **Goode, G. C., Morris, J. R. and Wells, J. M.** (1977) *Chemical Aspects of Fingerprint Technology: Report for Period April 1976–March 1977*, AWRE SSCD Memorandum No. 510, July. Aldermaston: Atomic Weapons Research Establishment.

11. **Clark, L. S.** (1978) *The design and construction of a beta transmissive sachet for daylight autoradiography*, Home Office PSDB Technical Memorandum 3/78. London: Home Office.

12. **Horn, J. A.** (1981) *The Autoradiography of Water-Soluble Radioactively Labelled Specimens on Curved Surfaces*, MPhil Thesis, June. Polytechnic of Central London.

13. **Jones, R. J. and Clark, L. S.** (1975) *A study of the ageing of latent fingerprints on PVC tape using a radioisotope detection system*, Home Office PSDB Research Note 34/75. London: Home Office.

14. **Smith, B. E.** (1981) 'The role of the Fingerprint Branch at New Scotland Yard working in support of the Police Officers investigating the Provisional IRA bombing campaign in England 1973–76'. *Proceedings of EC Working Group II, Sub-Group on Forensic Science and Related Matters, Fingerprint Symposium*, 4–5 March, London, pp 69–80.

15. **Albinson, R. A.** (1984) 'The development of fingerprints on fabrics – Laboratory simulations using a range of development techniques', *Proceedings of EEC International Fingerprint Conference*, November 27–30, London.

16. **Albinson, R. A.** (1984) *The Development of Latent Fingerprints on Fabric*, Draft HO SRDB Report No.72/84 (unpublished). London: Home Office.

17. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

18. **Goode, G. C., Morris, J. R., Wells, J. M.** (1979) 'The application of radioactive bromine isotopes for the visualisation of latent fingerprints', *J. Radio. Anal. Chem.,* 48, pp 17–28.

19. **Higgins, D. C.** (1980) *The visualisation of latent fingerprints by the use of radioactive iodine monochloride*, AWRE SCS Report No. 548, September. Aldermaston: Atomic Weapons Research Establishment.

5.15 Silver nitrate

1. History

1.1 The use of silver nitrate for the development of latent fingerprints on porous surfaces was first reported at the end of the 19th century, and together with iodine offered the only effective techniques for this type of surface until the use of ninhydrin was proposed in 1954. In the process silver nitrate reacts with the chlorides in the fingerprint to give silver chloride, which is converted to silver metal on exposure to light.

1.2 Various formulations had been reported, utilising both water and alcohol as solvents. The concentration of silver nitrate in these formulations typically varied from 3–10%, often with small additions of nitric acid to the aqueous solutions. In 1969 Cuthbertson carried out an extensive investigation of fingerprint chemistry and utilised the silver nitrate reaction to determine chloride contents in fingerprint deposits [1] and as a consequence of these studies proposed that the optimum silver nitrate concentration was 1%. Below this level there was insufficient reagent to react with the chloride available in the fingerprint and above 10% the background coloration began to become excessive [2]. It was also noted by Cuthbertson that under conditions of high humidity the chlorides in the fingerprint migrated and ultimately the mark became diffuse and undetectable. The operational implications of this study were published by Godsell [3] who recommended that UK police forces adopt the 1% silver nitrate formulation for operational use and ensure that exhibits for treatment were stored in low humidity environments.

1.3 The principal issue with the use of silver nitrate as a fingerprint development reagent was the progressive darkening of the background after treatment and research was carried out in the late 1960s and early 1970s in an effort to overcome this. Green [4] investigated the use of alternative silver salts with greater stability to light, and also explored the use of a sodium thiosulphate-acetic acid solution as a fixing process. Morris and Goode [5,6] developed a modified silver nitrate process to overcome both the background darkening and the lack of control over the photochemical development step. The preferred method ultimately proposed by Morris and Goode was to convert the silver chloride to silver sulphide using thiourea, giving a more stable final product. A complexing agent, disodiumethylenediaminetracetic acid (Na_2EDTA), was used in the silver nitrate solution to form complexes with unreacted silver so that it could be washed from the surface more easily. This was found to significantly reduce background darkening [2].

1.4 During the assessment of experimental techniques in the UK in the early 1970s, silver nitrate was used in comparative trials with other processes, including iodine, ninhydrin, radioactive sulphur dioxide and vacuum metal deposition. These trials showed that silver nitrate was the process most adversely affected by storage conditions of high humidity or exposure to moisture [7]. However, if dry storage conditions were used silver nitrate

developed a higher proportion of marks than ninhydrin, although not as many as radioactive sulphur dioxide. However, it must be noted that these experiments were conducted before the heat and humidification protocols were introduced for ninhydrin. Using silver nitrate after ninhydrin was found to produce more marks than either process alone [8]. These results were also confirmed by Caton in 1974, who reported the results of an assessment on over 6,000 paper and cardboard items; 1,617 marks were developed by ninhydrin, with a further 170 developed by subsequent silver nitrate treatment [9].

1.5 Although development of marks was typically carried out using light (ultraviolet or photoflood lighting being recommended), chemical developers could also be used [10]. Products typically used for photographic development were suggested, although the use of an additional immersion stage was not considered desirable because of the potential damage to some types of paper.

1.6 Silver nitrate was also considered as a technique for the intensification of faint ninhydrin marks, using a modified formulation using ethanol instead of water as the solvent [11]. This prevented the diffusion of the amino acids that occurred when the water-based formulation was used and meant that any marks developed using silver nitrate enhanced the existing ninhydrin marks and did not degrade any ridge detail already present. Other researchers have also considered non-aqueous alternatives to silver nitrate, one published formulation consisting of 3% silver perchlorate in toluene [12].

1.7 Other approaches to make the silver nitrate technique more practical were considered, including the use of stopping solutions based on methanol, acetic acid, glycerol and water [13]. This slowed the background darkening effect and negated some of the need for immediate photography and storage of exhibits in the dark. However, the technique was rarely used on paper after the mid-1970s, and although recommended as a reagent for raw wood its use in the UK declined after it was withdrawn from the second edition of the *Manual of Fingerprint Development Techniques* [14]. No further developments have been reported since 1998.

2. Theory

2.1 The theory of the silver nitrate process is that the silver nitrate in solution reacts with the chloride constituents of fingerprint deposits to produce insoluble silver chloride.

$$AgNO_3\ (aq) + NaCl\ (aq) \rightarrow AgCl\ (s) + NaNO_3\ (aq)$$

2.2 Silver chloride is light sensitive and when exposed to ultraviolet light darkens rapidly as metallic silver is formed.

$$AgCl\ (s) + hv \rightarrow Ag\ (s) + \tfrac{1}{2}Cl_2\ (g)$$

2.3 The treated exhibit is therefore exposed to ultraviolet (or white) light to promote development although the optimum exposure time will vary from surface to surface and is not always easy to establish because both the print and the background progressively darken with time. In the case of the background this occurs due to gradual breakdown of unreacted silver nitrate in the porous substrate, and treated exhibits should be stored in the dark to reduce the speed at which this occurs.

2.4 The formulation formerly published by the Home Office Scientific Research and Development Branch (HO SRDB) for operational use on raw wood [15] was as follows.

2.5 Mix 10g of silver nitrate with 500mL of methanol. Immerse article in solution for a maximum of 5 seconds and allow to dry in the dark. Illuminate article and continue exposure until the background starts to darken.

SILVER NITRATE developed fingerprint. Inset: illuminating to develop fingerprints

Development of fingerprints using silver nitrate.

3. Reasons technique is not recommended by CAST

3.1 CAST did recommend and issue the silver nitrate process in the first edition of the *Manual of Fingerprint Development Techniques* [15],

primarily as a process for the development of fingerprints on light coloured, raw wood. It was withdrawn from the manual in subsequent editions [14] because it was considered that physical developer was equally as effective in this application and had no issues associated with progressive darkening of the background on exposure to light.

3.2 On paper items, silver nitrate can develop additional marks if used sequentially after ninhydrin because it is targeting different constituents in the fingerprint deposits. However, chlorides are more affected by moisture and high humidity conditions than many other fingerprint constituents and silver nitrate cannot be used on items that have been wetted. For this reason, physical developer is the preferred method for sequential treatment after ninhydrin because it targets different constituents and can be used on wetted items.

4. References

1. **Cuthbertson, F.** (1969) *The Chemistry of Fingerprints*, AWRE Report No. O 13/69, October. Aldermaston: Atomic Weapons Research Establishment.

2. **Goode, G. C. and Morris, J. R.** (1983) *Latent Fingerprints: A Review of Their Origin, Composition and Methods for Detection*, AWRE Report No. O 22/83, October. Aldermaston: Atomic Weapons Research Establishment.

3. **Godsell, J. W.** (1969) *Chemistry of Fingerprints – A Note on the Investigation of the Chloride Content of Fingerprints and the Implications for Police and Fingerprint Technicians*, HO PRDB Report No. 14/69, September. London: Home Office.

4. **Green, W. D.** (1970) 'Modified Chemical and Physical Methods for the Detection of Latent Fingerprints', *Criminol.*, vol. 5 (16/17), pp 54–62.

5. **Morris, J. R. and Goode, G. C.** (1974) *The Detection of Fingerprints by Chemical Techniques. Report for Period April 1972–March 1974*, AWRE SSCD Memorandum 356. Aldermaston: Atomic Weapons Research Establishment.

6. **Morris, J. R. and Goode, G. C.** (1976) *Chemical Aspects of Fingerprint Technology – Report for Period April 1974–March 1976*, AWRE SSCD Memorandum No. 396, June. Aldermaston: Atomic Weapons Research Establishment.

7. **Godsell, J. W., Vincent, P. G. and Lloyd, D. W.** (1972) *Influence of Storage of Latent Fingerprints in an Atmosphere of a Given Humidity on the Choice of the Optimum Developing Agent*, HO PSDB Technical Memorandum 3/72.London: Home Office.

8. **Godsell, J. W. and Vincent, P. G.** (1973) *Comparative Study of Radio-Active Sulphur Dioxide, Ninhydrin and Silver Nitrate from the point of view of their Efficiency for Developing Latent Fingerprints on Paper*, HO PSDB Technical Memorandum 1/73. London: Home Office.

9. **Caton, H. E.** (1974) 'Physical and Chemical Aspects of Latent Print Development', *Proceedings of the Conference on the Science of Fingerprints*, 24–25 September, 1974, London, UK

10. **Olsen Sr, R. D.** (1978) *Scott's Fingerprint Mechanics*, ISBN 0-398-06308-7. Springfield, Illinois, USA: Charles C. Thomas.

11. **Kirby, F. J. and Olsen Sr, R. D.** (1973) 'Modified Silver Nitrate Formula for Intensifying Faint Ninhydrin Prints', *Forens. Photo.*, vol. 2 (5), pp 15–19.

12. **Kerr, F. M.** (1979) 'A Useful Alternative to Aqueous Silver Nitrate for Fingerprinting', *RCMP Gazette*, vol. 41 (6), pp 8–10.

13. **Price, D. and Stow, K.** (1998) 'A Method for Stopping Over-development of Silver Nitrate Treated Finger and Footwear Marks', *Fingerprint Whorld*, vol. 24 (93), pp 107–110.

14. **Bowman, V. (ed)** (1998 (revised 2002, 2004, 2009)) *Manual of Fingerprint Development Techniques*, 2nd edition. ISBN 1 85893 972 0. London: Home Office.

15. **Kent, T.** (1986) *Manual of Fingerprint Development Techniques*, 1st edition. London: Home Office.