

Technical Note No. 653

Computer Accession No. 65851

MASTER COPY

The Enhancement of Marks Made in Blood With
3,3',4,4'-Tetraaminobiphenyl

Javaid I Hussain and C Anthony Pounds

Central Research and Support Establishment,
Home Office Forensic Science Service,
Aldermaston, Reading, Berkshire, RG7 4PN

March 1989

British Crown Copyright 1989

PTG

Technical Notes are LIMITED
circulation items.

They are intended for use by
authorised Forensic Science
Laboratories in the United Kingdom
only. In general, they are not
available for release to other
organisations.

DISTRIBUTION:-

HQ Forensic Science Service
HOFSL Aldermaston
HOFSL Birmingham
HOFSL Chepstow
HOFSL Chorley
HOFSL Huntingdon
HOFSL Wetherby
FSL Metropolitan Police
FSL Belfast
FSL Aberdeen
FSL Edinburgh
FSL Glasgow
CRSE

SUMMARY

The use of 3,3',4,4'-tetraaminobiphenyl (TAB) as a reagent for the enhancement of faint bloodmarks has been investigated. TAB was found to be capable of enhancing blood marks on a range of different surfaces.

INTRODUCTION

Faint bloodmarks may be enhanced by a number of reagents which react with different blood components. Amido black [1,2] and other dyes [3] enhance bloodmarks by staining blood proteins. Ninydrin [1] can enhance bloodmarks by reacting with amino acids and to a lesser extent, with proteins present in blood. Bloodmarks may also be made more readily visible by appropriate illumination so they appear fluorescent against a non-fluorescent background or vice versa [1].

One component of blood, namely the haeme component of the protein haemoglobin, can be readily detected by a number of reagents including leucomalachite green, benzidine, and Kassel Mayer's reagent. These reagents are very sensitive and have been or still are in use as presumptive tests for blood [4]. The mechanism of action of these three reagents is similar and involves the formation of a coloured product from the interaction of the reagent with haemoglobin and hydrogen peroxide.

In addition to its use as a presumptive test for blood, leucomalachite green has also been employed as a means of enhancing faint bloodmarks including fingerprints and shoemarks left in blood [5]. Unfortunately, when used for this purpose leucomalachite green although very sensitive, has two major disadvantages. Firstly, the reagent must be dissolved in a volatile solvent system containing a high concentration of acetic acid and applied to the bloodmark as a fine spray. As a consequence this treatment should be undertaken in a fume cupboard or the operator should wear adequate safety equipment. Secondly, the coloured product which is formed can diffuse away from its site of production so that fine detail is blurred and lost.

3,3',4,4'-Tetraaminobiphenyl (TAB) is a derivative of benzidine commonly referred to as 3,3'-diaminobenzidine, which has been employed as a histological indicator of peroxidase activity [6] and is known to give a reaction with haemoglobin. TAB reacts with haemoglobin in the presence of hydrogen peroxide to form a brown insoluble product which does not diffuse from its site of formation. The reagent is employed as an aqueous solution and no organic solvents are required in its application.

These factors make TAB a suitable reagent for the enhancement of faint bloodmarks and we have investigated its use for this purpose.

EXPERIMENTAL

3,3',4,4'-Tetraaminobiphenyl tetrahydrochloride was obtained from the Sigma Chemical Company Limited, Poole, Dorset, England.

Bloodmarks were laid on a number of different surfaces including paper, glass, polythene, wood, metal and paint and were fixed with either 5-sulphosalicylic acid [6], methanol [1], or heat [1]. The bloodmarks were then enhanced by immersion in solutions containing different concentrations (0.01-0.5% w/v) of tetraaminobiphenyl tetrahydrochloride in 0.2M Tris-HCl buffer pH7.0, and different concentrations (0.015-1.0% v/v) of hydrogen peroxide. After incubation with TAB (maximum enhancement was usually achieved after 2.5 minutes) the bloodmarks were washed thoroughly with water, allowed to dry and then examined.

In a further set of experiments bloodmarks laid on a number of different surfaces were enhanced using TAB and compared with the enhancement resulting from treatment with amido black staining [2].

RESULTS AND DISCUSSION

Maximum enhancement was obtained when marks were immersed in a 0.1% w/v solution of TAB in buffer with a hydrogen peroxide concentration of 0.15% v/v. Maximum enhancement was usually achieved after 2-5 minutes immersion in the TAB solution. The TAB solution was unstable and solutions which were more than 30 minutes old were discarded.

Under these conditions it was found that TAB was an effective and sensitive reagent for the enhancement of faint bloodmarks. Use of the reagent increased the contrast between the blood and the background and resulted in a dark brown bloodmark. The product formed by the interaction of TAB with blood is insoluble and cannot diffuse away from its site of formation. Fine detail is thus preserved with no observable blurring or fuzziness at edges.

TAB is light sensitive and it was noted that some non-specific background darkening was evident when porous surfaces such as paper and wood were left exposed to light after treatment. Similarly the reagent solution will darken on standing and should thus be light-protected as much as possible before actual use.

Blood-stains which have been fixed with methanol [1] or with 5-sulphosalicylic acid [6] could be successfully treated with TAB. In general however blood-stains fixed by heat were less suitable for TAB treatment. This is due to the fact that heat treated marks were usually not as stable as methanol or acid fixed marks and some loss of blood material occurred while the marks were immersed in the TAB solution.

Comparison of bloodmarks treated with amido black (the present recommended method of enhancing fingermarks in blood) and with TAB are shown in Figure 1. In general the two methods develop the bloodmark to the same extent. However as TAB produces little non specific background staining no destaining step is required. With amido black considerable destaining is required to produce the visible mark. Thus TAB is considerably more convenient to use than amido black. This is particularly evident with porous surfaces such as paper and untreated wood.

[In a limited experiment bloodmarks laid on pigs skin were also successfully enhanced using TAB].

CONCLUSIONS

Bloodmarks fixed with methanol or 5-sulphosalicylic acid can be enhanced with a 0.1% w/v solution of 3,3',4,4'-tetraaminobiphenyl tetrahydrochloride in 0.2M Tris-HCl buffer pH7.0 containing 0.15% v/v hydrogen peroxide. In certain circumstances the use of tetraaminobiphenyl reagent can result in better enhancement than that achieved with amido black.

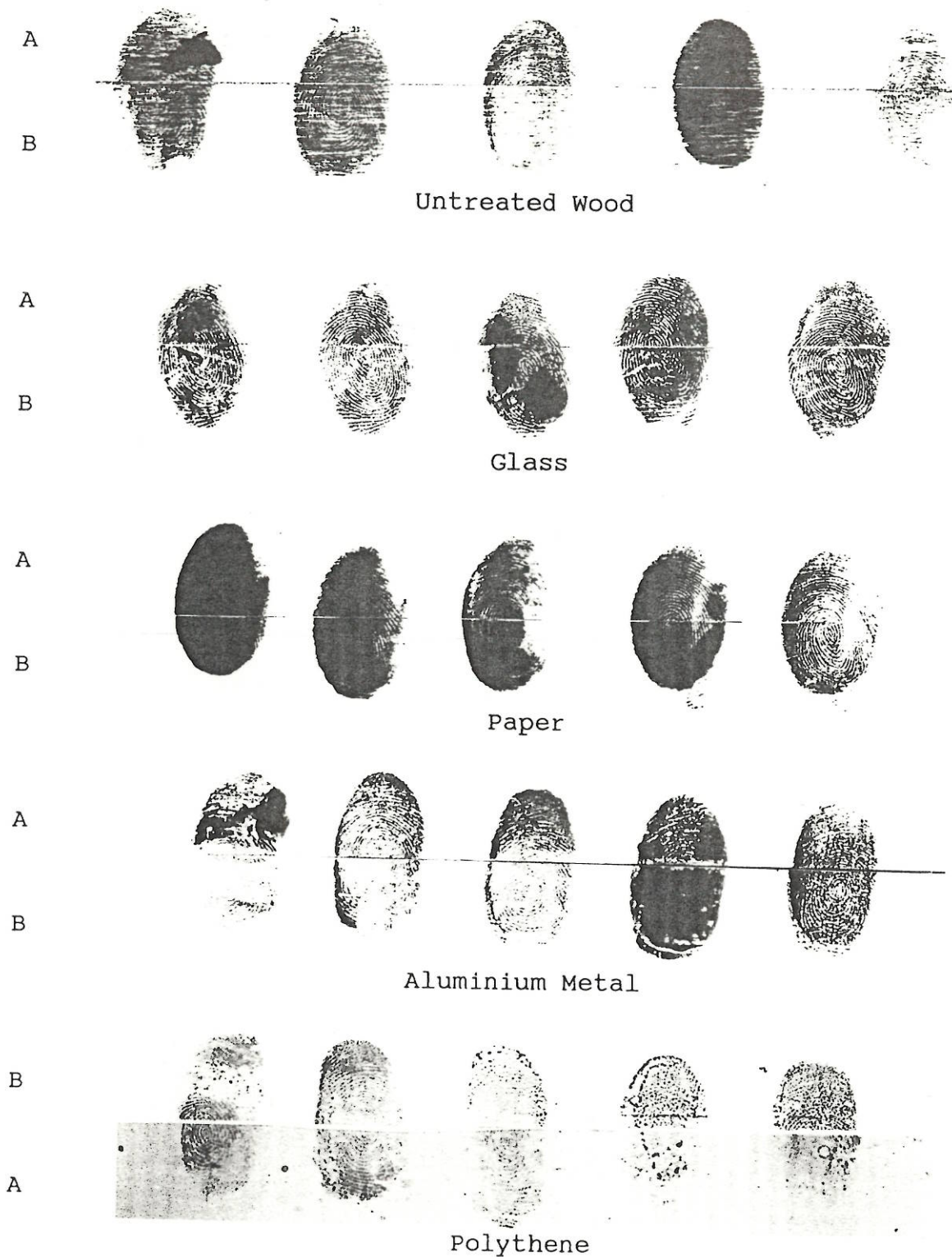


Figure 1: Comparison of bloodmarks treated with (A) Amido Black and (B) 3,3',4,4'-Tetraaminobiphenyl.

REFERENCES

- [1] Manual of Fingerprint Development Techniques, Home Office Scientific Research and Development Branch, 1986.
- [2] Hussain JI and Pounds CA. The Enhancement of Marks in Blood. Part 2: A Modified Amido Black Staining Technique. CRSE Report (in press).
- [3] Barnett KG, Bone RG, Hall PW and Ide RH. The Use of Water Soluble Proteins Dyes for the Enhancement of Footwear Impressions in Blood on Non-Porous Surfaces. Part 1. Technical Note 629, 1988.
- [4] Owen GW. A Comparison of Some Presumptive Tests for Blood. CRSE Report 84, 1973.
- [5] Metropolitan Police Forensic Science Laboratory. Private Communication.
- [6] Hussain JI and Pounds CA. The Enhancement of Marks in Blood. Part 1. 5-Sulphosalicylic Acid: An Convenient and Effective Fixative for Marks in Blood. CRSE Report 649, 1988.

10

10

10

10