A comprehensive experimental study of industrial, domestic and environmental interferences with the forensic luminol test for blood

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Received 8 October 2002; revised 31 January 2003; accepted 6 February 2003

ABSTRACT: This paper presents the first comprehensive and quantitative study of substances that interfere with the forensic luminol test for blood. Two hundred and fifty substances have been selected on the basis of modern lifestyles and of contiguity with crime scenes. The intensity of the chemiluminescence produced by each substance has been measured relative to that of hae-moglobin and the peak wavelength shift has also been determined. The following is a short list of nine substances that produce chemiluminescence intensities comparable with that of haemoglobin: turnips, parsnips, horseradishes, commercial bleach (NaClO), copper metal, some furniture polishes, some enamel paints, and some interior fabrics in motor vehicles. Care needs to be taken when the luminol test for blood is used in the presence of these substances. Copyright © 2003 John Wiley & Sons, Ltd.

KEYWORDS: forensic science; presumptive blood test; chemiluminescence; luminol; interfering catalysts

INTRODUCTION

We have previously published brief accounts (1, 2) of substances which interfere with the chemiluminescent luminol test for blood (3, 4, 5). This test is widely used throughout the world by forensic specialists when they investigate crime scenes where interpersonal violence may have occurred. The test often involves spraying a whole room or house with a mixture (1) of luminol (Figure 1) in an alkaline solution with either hydrogen peroxide or sodium perborate. Catalysts such as potassium ferricyanide or copper sulphate, which very greatly enhance the emission, are deliberately omitted from the forensic mixture. The mixture then emits bright sparkles of blue light when it is sprayed on droplets of blood containing haemoglobin, which exerts a strong catalytic effect on the emission.



Figure 1. The chemical structure of luminol (5-amino-2,3-dihydro-1,4-phthalazinedione)

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Contract/grant sponsor: PathCentre, Western Australia. Contract/grant sponsor: University of Western Australia.

The mechanism of luminol catalysis

It is well known that the luminol reaction produces significantly brighter chemiluminescence in the presence of a catalyst (6, 7, 8). However, the mechanism behind this catalysis is not well understood. In their comprehensive book, *Chemiluminescence in Organic Chemistry*, Gundermann and McCapra (6) spend little time on the mechanism of catalysis of the luminol reaction, saying simply that catalysis appears to occur due to the presence of metal ions such as Cu²⁺, or metal complexes such as haemoglobin.

In recent times, there is still a general lack of understanding of the mechanism of catalysis. It is believed, on the whole, that the catalyst (usually in the form of a metal ion) forms some sort of intermediate complex or radical, which in turn leads to the excited state of luminol. Ojima and Nonoyama (7) suggested the formation of a ternary complex containing a ligand, a copper (II) ion, and the 3-aminophthalate anion, which then reacts with hydrogen peroxide to form the excited state. Even more recently (2001), Lin *et al.* (8) argued that the Co(II)-ethanolamine immobilized resin catalysed the formation of a superoxide radical, which then acted upon the luminol to produce chemiluminescence.

Interferences with the forensic luminol test for blood

The forensic luminol test has a long history (3, 4, 5) as a presumptive test for blood at the early stages of a crime

Table 1. Methods used for the preparation of interfering substances for testing

Type of interfering substance	Method of preparation* [†]
Liquid (i.e. juices, paints, beverages)	10 drops (using a dropping pipette) of the liquid were applied to a sheet of Whatman No. 541 filter paper
Spray liquids (i.e. spray paints, fragrances)	Each sample was squirted twice onto Whatman No. 541 filter paper
Large solids (i.e. flooring and automobile off-cuts)	These samples were tested as they were, with no preparation
Small solids (i.e. tablets)	Samples were mounted on the adhesive side of Tesa [®] packaging tape
Smearable substances (i.e. vegetable pulps, hair products)	Samples were smeared onto Whatman No. 541 filter paper

*[†]See Notes to tables.

scene investigation. However, some commonly occurring substances catalyse luminol chemiluminescence even when blood is absent, thus producing false-positive results.

It is unfortunate that, although some interferences with the luminol test have been discussed qualitatively (4, 5, 9, 10), it is rare to find any quantitative study that compares the intensity of the chemiluminescence catalysed by a particular substance with that catalysed by blood. This is particularly unfortunate because a number of forensic personnel who use the luminol test professionally tend to reject certain chemiluminescence glows on a very informal and non-quantitative basis—often on the grounds that the glow intensity is qualitatively much weaker than that of blood (11).

One indirect exception to the above comments is provided by the early work of Kraul and Meyer in 1941 (12). They published a small list of peak wavelength differences caused when various interfering substances catalyse the luminol test. Curiously, although they did publish wavelength shifts, they did not publish any intensity differences.

We have previously published some preliminary work (1, 2), which provided quantitative information about intensity differences and wavelength differences associated with a small set of interfering substances. One of the ironies of the field, pointed out in one of these publications (1), is that a common cleaning agent (sodium hypochlorite solution), which may often be used in an attempt to remove blood from the crime scene, itself catalyses the luminol chemiluminescence.

We now believe that the time is ripe for a major investigation of a wide range of substances that may produce interferences with the luminol test, which are associated with modern lifestyles, and which may thus often be found at crime scenes. This is the aim of the present study.

METHODS

The standard luminol solution was prepared with the following chemicals; 0.01 g luminol (Aldrich), 0.5 g Na_2CO_3 (Merck), 0.13 g $NaBO_3$ ·4H₂O (Aldrich) and

10 mL distilled water. The 150 g/L haemoglobin solution used consisted of 1.5 g haemoglobin (Sigma) dissolved in 10 mL aqueous solution containing 0.2 mol/L NaOH solution.

As the samples tested for catalytic effect on the luminol solution were either liquids or solids, a variety of methods were used for preparing the samples for testing. In most cases, the interfering substances were mounted on filter paper, as were the haemoglobin stains. However, in the case of large solid objects, the actual surface was used instead, after it had been machined to the standard sample area viewed by the photomultiplier. The methods of preparation used are shown in Table 1.

The testing procedure was the same as described by Quickenden and Cooper (1), and Quickenden and Creamer (2). All samples were mounted the same distance away from the photomultiplier tube, and the photomultiplier had the same area of view at all times. Each sample was sprayed with the standard luminol solution, and any resultant chemiluminescence was detected by a photomultiplier tube (EMI 9635 QAM) after being passed through a calibrated circular graded interference filter (Barr and Stroud CGS2), which monochromated the emitted light. The resultant output was passed through an inverting operational amplifier and was displayed as a scan on a computer monitor.

RESULTS AND DISCUSSION

Table 2 (and the accompanying footnotes) presents all of the results obtained in the present study and, for completeness, a small set of results transferred from a previous publication (2). The selection of the various categories in Table 2 was carried out with some thought for the lifestyles in modern society, and the interaction of these lifestyles with typical crime scenes. Advice was taken from law enforcement officers involved in the forensic area in the selection of these categories and the selection of the individual items in Table 2.

As we are searching for substances that mimic the effect of blood (haemoglobin) in the forensic luminol test, the intensity of luminol emission in the presence of each substance is given as both the recorded

Interfering substance	Mean peak wavelength shift from haemoglobin (nm)	Replicate peak intensities (arbitrary units)	Mean intensity (% of haemoglobin value)
Fruit and vegetables			
Turnip (pulp)	3 ± 4	134, 166, 170, 230	74 ± 35
Parsnip (pulp)	8 ± 5	106, 112, 143, 170	56 ± 23
Horseradish (pulp)	3 ± 4	33, 40, 60, 61	20 ± 12
Turnip (smear)	13 ± 6	28, 36, 37, 42	15 ± 5
Carrot (pulp)	5 ± 4	20, 22, 25, 25	10 ± 2
Onion (pulp)	1 ± 4	8, 10, 17, 21	6 ± 4
Social interferences			
Cigarette ash	14 ± 6	12, 22, 22, 24	8 ± 4
Red pen ink (Biro [®])	13 ± 5	15, 19, 23, 24	8 ± 3
Cigarette smoke (from automobile)	11 ± 9	12, 14, 14, 15	6 ± 1
Cigarette smoke (from smoking room)	11 ± 14	12, 12, 14, 15	5.5 ± 2
Surfaces, coatings and cleaners			
Copper metal	2 ± 10	239, 254, 255, 255	106 ± 10
Enamel paint (Dulux [®])	9 ± 4	227, 235, 238, 245	100 ± 10
125 g/L NaClO _(aq)	9 ± 4	174, 179, 210, 230	84 ± 22
Dark green spray paint (Taubman [®])	22 ± 3	149, 180, 183, 255	81 ± 34
Wooden-furniture polish (Goddard's [®])	11 ± 23	44, 47, 47, 53	20 ± 4
Blu Tak (Bostik [®])	12 ± 17	11, 23, 34, 34	11 ± 8
Laminated chipboard	1 ± 27	22, 26, 26, 30	11 ± 3
Aluminium metal	3 ± 15	17, 24, 29, 29	10 ± 5
Chipboard	8 ± 10	13, 14, 18, 25	7.5 ± 4
Computer cover	9 ± 22	15, 16, 18, 20	7 ± 2
Terracotta tile	9 ± 2	9, 13, 15, 17	5.5 ± 2.5
Automobiles			
Roof lining	12 + 7	45 45 45 70	22 + 11
1992 Ford Laser [®]	13 ± 7	45, 45, 45, 70	22 ± 11
1982 Mitsubishi Sigma [®]	6 ± 17	7, 11, 11, 23	5.5 ± 4.5
1986 Mitsubishi Magna®	10 ± 16	9, 12, 12, 14	5 ± 2
Door lining 1986 Mitsubishi Magna [®]	16 + 10	12 12 12 12	5 ± 1
Seat fabric	16 ± 19	12, 12, 13, 13	3 ± 1
1986 Mitsubishi Magna [®]	19 ± 15	8, 12, 12, 16	5 ± 3
1980 Mitsubishi Magna [®] (sedan)	19 ± 13 13 ± 12		5 ± 3 5 ± 2.5
e ()	13 ± 12	7.5, 11, 11, 13	$J \pm 2.3$
Toiletries	12 + 0	0 17 10 10	6514
Hair wax (Wella [®]) Lipstick	13 ± 9	8, 17, 18, 19	6.5 ± 4
Black Opal [®] Caramel Crème	13 ± 17	13, 17, 22, 22	7.5 ± 3.5
Black Opal [®] Ebony White	10 ± 17 10 ± 22	7, 12, 13, 14	5 ± 2.5

Table 2. Spectral measurements showing all substance	s analysed for interference	e with the luminol test for b	lood detection ^{‡§} .
Errors shown are 95% confidence intervals in the mean			

^{‡§||}See Notes to tables.

photometric measurement and the normalized value relative to that of haemoglobin.

A second feature of Table 2 is the presentation of the deviation of the wavelength maximum of the chemiluminescence of the test substance from that produced by haemoglobin. The wavelength differences were measured with the hope that some systematic difference might have enabled the rejection of chemiluminescence catalysed by substances other than blood. Unfortunately, the data in Table 2 showed that this hope was not realised, as most of the peak wavelengths in Table 2 do not deviate very significantly from the peak maximum of the chemiluminescence catalysed by haemoglobin. The intensity data relative to haemoglobin are of much greater importance.

The mean chemiluminescence intensities for each of the interfering substances listed in Table 2 were obtained from the replicate determinations listed in the table. In the case of the haemoglobin, a greater number of replicates were used (eight) as the mean intensity of 236 ± 12 arbitrary units provided a comparison point for the intensities of all the interfering substances. All the wavelength shifts listed in Table 2 are the mean of eight replicates, and are measured relative to the mean value of 438 ± 2 nm obtained for haemoglobin. The errors quoted are the 95% confidence interval in the mean values.

Interfering substance	Mean peak wavelength shift from haemoglobin (nm)	Replicate peak intensities (arbitrary units)	Mean intensity (% of haemoglobin value)
Copper metal	2 ± 10	239, 254, 255, 255	106 ± 10
Enamel paint (Dulux [®])	9 ± 4	227, 235, 238, 245	100 ± 10
125 g/L NaClO _(aq)	9 ± 4	174, 179, 210, 230	84 ± 22
Dark green spray paint (Taubman [®])	22 ± 3	149, 180, 183, 255	81 ± 34
Turnip (pulp)	3 ± 4	134, 166, 170, 230	74 ± 35
Parsnip (pulp)	8 ± 5	106, 112, 143, 170	56 ± 23
Roof lining (1992 Ford Laser [®])	13 ± 7	45, 45, 45, 70	22 ± 11
Horseradish (pulp)	3 ± 4	33, 40, 60, 61	20 ± 12
Wooden-furniture polish (Goddard's [®])	11 ± 23	44, 47, 47, 53	20 ± 4

Table 3. Spectral measurements showing the major interferences with the luminol test for blood detection[‡]. Errors shown are 95% confidence intervals in the mean values

[‡]See Notes to tables.

Relevance of the chemiluminescence wavelength measurements

Despite the above comments, there are a few examples in Table 2 that show a significant movement of the emission peak away from haemoglobin. In the case of sodium hypochlorite (household bleach), it was found previously (1) that quite substantial deviations can occur, and these have been attributed (1) to the simple filter effect caused when coloured and uncoloured substances are used as the interfering material. Unfortunately, in such cases the quantitative wavelength shift caused by the substance depends on local circumstances, such as the concentration of the substance. Because of this, it seems unlikely that a wavelength shift will be uniquely diagnostic for a particular substance.

Nevertheless, diagnostic wavelength shifts could conceivably occur if the catalytic substance was fluorescent and possessed an absorption spectrum that overlapped with the chemiluminescence spectrum of the luminol. Shifts of this type are well known in photochemistry; they usually result in a shift towards longer wavelengths and are not to be confused with simple filter effects, which preferentially remove certain wavelengths from the chemiluminescence spectra of the luminol. Wavelength shifts of the non-filter effect type have not, as yet, been reported for any known interfering substance in the luminol test.

Sample variability and sample preparation in relation to luminescence measurements

One of the problems associated with this type of quantitative analysis, based on the measurement of chemiluminescence intensity, is the heterogeneous nature of the sample. By comparison, chemiluminescence intensity measurements on homogeneous sample situations (e.g. solutions in spectrofluorimeter cells) give much less variability between replicate samples than do those measurements carried out on chemiluminescent surfaces that have a non-uniform gas/solid interface.

The latter is the situation in the present measurements, and every care has been taken to minimize experimental errors arising from incorrect sample positioning and changes in the angle of irradiation and the angle of view of the chemiluminescent surface, from replicate to replicate. Furthermore, an attempt was made to ensure uniform sample application to whatever substrate was used (usually a Whatman No. 541 filter paper). The particular method of preparation for each substance is listed in Table 1.

To add to the above difficulties, further problems arise when solid samples (e.g. of flooring or fabrics) have to be located in the measuring instrument instead of the coated filter paper. Inevitably, a sample of a flooring material, etc. that is cut from different regions of a motor vehicle may have various amounts of surface contaminant, which may affect the chemiluminescence intensity. The option of cleaning such samples individually with suitable solvents was not adopted, as it was decided that the practical requirement in forensic work is to use the unmodified local forensic sample, so that it will behave in the experiment as it would have done *in situ*.

In view of all the above variability and uncertainties, it is not surprising that replicate luminescence intensities from different samples of the same substance should vary from one another by as much as is indicated by the so-called errors in Tables 2 and 3.

Relevance of the chemiluminescence intensity measurements

Most importantly, Table 2 shows a very wide range of intensities relative to haemoglobin for the wide range of substances examined. For this reason, we have subdivided these interferences into several categories, based on the general class of interfering substance. All substances are ranked in descending order of intensity within each category. A histogram of the intensities was used as an aid to establish a small set of practical intensity level subdivisions. As an approximation, that arbitrary scale has been divided into the following categories:

- Strong interference: > 20% the intensity of the haemoglobin standard.
- Weak interference: < 20%.
- Negligible interference: < 5%.

It should be noted that Table 2 shows only the substances that produced a chemiluminescence intensity > 5% that of the haemoglobin standard, as any chemiluminescence below 5% is generally not visible to the naked eye.

Table 3 contains a useful summary of the nine brightest emitters from all the 250 substances tested. These are the substances that have at least 20% of the intensity of the haemoglobin. Special attention should be paid to the possibility of interference from these substances when forensic detection for blood is carried out using the luminol reaction.

Brightly emitting interfering substances

The brightest emission from interfering substances is that from copper metal. This emission is as intense as the emission produced from the haemoglobin itself. This means that law enforcement personnel should be particularly cautious when using the luminol test near copper piping and fittings, such as are found in many kitchens, bathrooms and laundries, as well as on copperplated surfaces.

Both the conventional enamel paint and the spray paint also catalyse high chemiluminescence intensities, and as such should also be of concern to forensic investigators. Both paints are common in industrial and household situations, and the spatter pattern of spilt or sprayed paint may well mimic the spatter pattern of blood at a crime scene. It must also be noted that these results, on a limited number of paint samples, suggest that many other paints may indeed give interferences with the luminol blood detection test.

Common household bleach (NaClO_(aq)), is another substance that could interfere with the use of luminol at a crime scene investigation. It has the added complication that the perpetrator of a crime may also use it, as a means of cleaning blood spatter from a crime scene. However, the chemiluminescence emitted by the reaction between bleach and luminol decreases over time, as the bleach evaporates from a surface.

The next obvious interferences come from the pulps of parsnip, turnip and horseradish. These root vegetables are high in peroxidase content, and this is thought (5) to lining of a 1992 Ford Laser[®] produced any reasonable chemiluminescence. The problem of false positives in automobiles is considerable, as vehicles are often the scenes of crimes, and may also be used to transport a body from a crime scene. From a forensic point of view, it is reassuring to see that only one sample from a vehicle produced major chemiluminescence. However, the variability of motor vehicles is enormous, and more extensive investigation needs to be carried out in this area.

be the reason behind the high production of chemi-

The final interfering substance shown in Table 3 is furniture polish. As with the paint samples mentioned earlier, polished wood may often be found in areas susceptible to violent crime, and forensic investigators should note this interference. It may produce the same ambiguities associated with the presence of household bleach, as perpetrators of crimes may use furniture polish in an attempt to clean up a blood spatter.

CONCLUSIONS

A wide-ranging experimental study of interferences with the forensic luminol test for blood has been carried out over a variety of modern lifestyle-related areas. This study of 250 different substances indicates that while the majority (ca. 240) do not produce sufficiently intense chemiluminescence with the luminol reaction to be easily mistaken for blood, nine commonly occurring substances do. These are, turnips, parsnips, horseradishes, commercial bleach (NaClO), copper metal, some furniture polishes, some enamel paints, and some interior fabric in motor vehicles. Care needs to be taken when interpreting results of the luminol test for blood in the presence of such substances. Particular care needs to be exercised when using the test in motor vehicles, where the large diversity of metals, enamel paints and plastic interior linings suggests the need for a further, ongoing study which examines all materials in old and recent model cars produced in the modern global economy.

Acknowledgements

The authors wish to thank the PathCentre of Western Australia, as well as the Faculty of Life and Physical Sciences, Chemistry (School of Biomedical and Chemical Sciences) and the Centre for Forensic Science at the University of Western Australia for their funding of JIC's PhD scholarship.

Notes to tables

*All 'wet' samples were left until dry, before testing.

[†]Both the Whatman No. 541 filter paper and the Tesa[®] packaging tape produce no detectable chemiluminescence when luminol solution is applied.

^{*}For comprehensiveness, all interfering substances published previously by Quickenden and Creamer (2) have been included in the tables of results. *Italics* indicate these.

[§]The following substances produced < 5% of the chemiluminescence emitted by the haemoglobin standard. Fruit and vegetables: turnip (juice), pumpkin (pulp, juice), potato, tomato (pulp), parsnip (smear), carrot (juice), watermelon (pulp), banana (smear). Social interferences: tea stain (Tetley®), alcoholic cider (Strongbow®), human mucus, cigarette butts, chicken-covered napkin, beer stain (Emu Export®), coffee stain, rust, human saliva, seasoned wedges container, human faeces, berry sports drink (Powerade®), human perspiration, decongestant tablets (Sudafed®), human sperm, brown wax crayon, milk (Brownes Dairy[®]), aspiring. Surfaces, coatings and cleaners: water-based white paint (Montage®), polyurethane paint (Berger®), sisal natural matting, jute natural matting, rust treatment (Wattyl®), polystyrene, ceramic tile, white spray paint (Dulux[®]), light blue paint (Dulux[®]), purple paint (Wattyl®), stone tile, acrylic coating (Wattyl®), green paint (Wattyl®), deep brown paint (Dulux[®]), hard vinyl flooring, machine oil (Ampol[®]), etching agent (Wattyl®), black plastic, bench top (Marblo®). Automobiles: surface protectant (Armor All®), car cleaner (Polyglaze®), roof lining (1985 Ford Meteor®, 1985 Ford Falcon®, 1987 Mitsubishi Magna® sedan, 1986 Ford Laser®), door lining (1987 Mitsubishi Magna® saloon, 1987 Mitsubishi Magna® sedan, 1985 Ford Meteor®, 1982 Mitsubishi Sigma®), rear luggage compartment lining (1985 Ford Falcon®, 1980s Toyota Land Cruiser®, 1987 Mitsubishi Magna® saloon, 1986 Ford Laser®, 1988 Toyota Hilux®, 1992 Ford Laser®, 1985 Ford Meteor®, 1984 Toyota Corolla®, 1982 Mitsubishi Sigma®, 1987 Mitsubishi Magna[®] sedan), seat fabric (1988 Toyota Hilux[®], 1987 Mitsubishi Magna[®] saloon), floor material (1980s Toyota Land Cruiser[®]). Toiletries: hair mud (Wella[®]), foundation stick (Black Opal[®] Beautiful Bronze), lipsticks (Black Opal® Cinnabar, Black Opal® Red Maple, Black Opal[®] Cashmere), perfumes (Stitch[®] No. 7, Stitch[®] No. 8, Stitch® No. 10, Stitch® No. 14, Rectoverso® Mandarin Musk, Woman Nike®), men's fragrances (Chanel® Antaeus, Chanel® Allure homme, Chanel[®] pour monsieur, Kenzo[®] l'eau par kenzo, Ralph Lauren[®] Polo, Armani[®] Acqua Di Gio, Armani[®] Emporio, Armani[®] eau pour homme, Gucci[®] Rush, Yves Saint Laurent[®] pour homme, Yves Saint Laurent® Jazz, Yves Saint Laurent® Live Jazz, Gerruti® Image, Gerruti® 1881, Hermes® Rocobar, D & G® Masculine, D & G® eau de toilette, Christian Dior® Fahrenheit, Bvigari® Extreme, Oscar dela Renta® Oscar, Givenchy® Insense-Ultramanic, Givenchy® Xeryus, Hugo Boss® Elements Aqua, Hugo Boss® Elements, Hugo Boss® Hugo, Hugo Boss® Boss, Rochas® Man, Davidoff® Cool Water, Noir® Drakkar, Very Valentino[®] for men, Paco Rabanne[®] pour homme, Paco Rabanne® XS, Echt Kolnisch® Wasser No. 4711, Gnerliu® Heritage, Crabtree & Evelyn® Sienna, Lagerfeld® Classic).

"The following substances produced no detectable chemiluminescence on the application of the luminol solution. *Fruit and Vegetables: horseradish (smear, juice), carrot (smear), onion (smear, juice), pumpkin (smear), tomato (smear, juice), watermelon (smear, juice), banana (pulp), apple, beetroot, canned beetroot, lemon, orange. Social interferences: fast food container (McDonald's Big Breakfast[®]), paper packaging (Fish Feast[®]), wet beer (Emu Export[®]), fast food wrapper (Chicken Treat[®]), moist towelette (Chicken Treat[®]), chicken bones with seasoned coating (KFC[®]), energy drink (Red Eye[®]), energy drink (Black Stallion[®]), orange soft drink (Fanta[®]), cola soft drink (Coca Cola[®]), orange and mango juice (100% Just Juice[®]), newspaper print, magazine print, red wine (Evans & Tate[®] 2000 Cabernet Merlot), white wine (Westfield Bronzewing Estate[®] 1997 Verdelho), tomato sauce (Farmland[®]), red felt-tip pen (MonAmi[®]), red chalk (Alpha[®]), human urine, vinegar, paracetamol tablet (Panadol[®]). Surfaces,* coatings and cleaners: foam sheet (2 mm), zinc rich primer (Top Dek®), Venetian blind fabric, red paint (Taubman®), Hessian material, felt material, velvet finish laminate (Formex®), pearl finish laminate (Formex[®]), linoleum flooring (Nathan's[®]), polymethylate sheet, shag-pile carpet, path moss, dralon/viscose furniture fabric (Jacka-Wartley®), fibreboard panelling medium density fibre, polycarbonate sheeting, red house brick, cleaning paste (Gumption[®]), antiseptic solution (Farmland®), fly repellent (Mortein®), latex gloves (Supergloves®), lemon-scented dishwashing liquid, washing detergent (Omo[®]), washing powder (Calgon[®]), gloss acrylic paint (Taubman[®]), matte finish paint (Dulux[®]), flat oil-based paint (Top Dek[®]), vinyl floor polish (Ormonoid[®]), jarrah flooring, soft-backed vinyl, coir natural matting, industrial strength cleaner (CLR[®]), lubricating spray (WD40[®]), bearing grease (Rocol[®] Sapphire), common insecticide (Confidor[®]), linseed oil, gear oil (Castrol[®]), weed killer (Zero[®]), motor oil (Castrol[®]), engine coolant (Valvoline®), mineral turpentine, silicone (Selleys®), adhesive (Araldite[®]). Automobiles: vinyl headrest (1981 Ford Meteor[®]), polypropylene dashboard (1981 Ford Meteor®), rear luggage compartment lining (1986 Mitsubishi Magna®), roof lining (1980s Toyota Land Cruiser®), seat fabric (1985 Ford Falcon®, 1986 Ford Laser®). Toiletries: liquid foundation (Black Opal®), hair-conditioner with Aloe Vera (Alberto[®]), shampoo with Aloe Vera (Alberto[®]), pump soap with Aloe Vera/Camomile (Palmolive®), nail-polish remover (Classics®), sorbolene lotion (Farmland®), perfume (Crabtree & Evelyn® Freesia, Crabtree & Evelyn® Vanilla), men's fragrances (Chanel® Platinum-Egoiste, Christian Dior® Dune, Gnerliu® Vetirer, Kouros® Body, Ralph Lauren® Romance, Yves Saint Laurent® Opium).

REFERENCES

- 1. Quickenden TI, Cooper PD. Increasing the specificity of the forensic luminol test for blood. *Luminescence* 2001; 16: 251–253.
- Quickenden TI, Creamer JI. A study of common interferences with the forensic luminol test for blood. *Luminescence* 2001; 16: 295–298.
- Specht W. Die Chemiluminesenz des Hämins, ein Hilfsmittel zur Auffindung und erkennung forensisch wichturger Blutspuren. Angew. Chem. 1937; 50: 155–157.
- Grodsky M, Wright K, Kirk PL. Simplified preliminary blood testing: an improved technique and a comparative study of methods. *J. Crimin. Law Criminol. Police Sci.* 1951; 42: 95–104.
- Yeshion TE. The forensic application of luminol as a presumptive blood test. In *Proceedings of the 6th International Symposium on Bioluminescence and Chemiluminescence, 1990*, Stanley PE, Kricka LJ (eds). Wiley: Chichester, 1991; 379–384.
- Gundermann KD, McCapra F. Luminol and related compounds. In *Chemiluminescence in Organic Chemistry*. Springer-Verlag: Berlin, 1987.
- Ojima H, Nonoyama K. Catalytic chemiluminescence of luminol by cobalt(III) amine complexes and copper(II) amine complexes. *Aichi Kyoiku Daigaku Hokoku, Shizen Kagaku* 1991; 40: 21–31.
- Lin JM, Shan X, Hanaoka S, Yamada M. Luminol chemiluminescence in unbuffered solutions with a cobalt(II)-ethanolamine complex immobilized on resin as catalyst and its application to analysis. *Anal. Chem.* 2001; **73**: 5043–5051.
- 9. Sutton TP. Presumptive testing for blood. In *Scientific and Legal Applications of Blood Pattern Interpretation*, James SH (ed.) CRC Press: Boca Raton, FL: 1999.
- 10. http//police2.ucr.edu/blood1.htm
- 11. Personal communications to the authors from various professional users of the test.
- Kraul R, Meyer HH. Ist der Nachweis von Blutspuren durch 3-Amino-phthalsäurehydrazid ein Kennzeichnendes verfahren? Angew. Chem. 1941; 54: 213–215.