J Forensic Sci, September 2007, Vol. 52, No. 5
doi: 10.1111/j.1556-4029.2007.00539.x
Available online at: www.blackwell-synergy.com

TECHNICAL NOTE

Rebecca Hoile,[1] *B.Sc.; Simon J. Walsh,*[2] *B.Sc. (Hons); and Claude Roux,*[3] *Ph.D.*

Bioterrorism: Processing Contaminated Evidence, the Effects of Formaldehyde Gas on the Recovery of Latent Fingermarks*

ABSTRACT: In the present age of heightened emphasis on counter terrorism, law enforcement and forensic science are constantly evolving and adapting to the motivations and capabilities of terrorist groups and individuals. The use of biological agents on a population, such as anthrax spores, presents unique challenges to the forensic investigator, and the processing of contaminated evidence. In this research, a number of porous and nonporous items were contaminated with viable anthrax spores and marked with latent fingermarks. The test samples were then subjected to a standard formulation of formaldehyde gas. Latent fingermarks were then recovered postdecontamination using a range of methods. Standard fumigation, while effective at destroying viable spores, contributed to the degradation of amino acids leading to loss of ridge detail. A new protocol for formaldehyde gas decontamination was developed which allows for the destruction of viable spores and the successful recovery of latent marks, all within a rapid response time of less than 1 h.

KEYWORDS: forensic science, microbial forensics, anthrax spores, decontamination, formaldehyde, fingermark

While intelligence reports rate the likelihood of an attack other than an explosive device as medium to low risk (Australian Security and Intelligence Organization, personal communication) it was the bioterrorism-related attacks in October 2001, in which letters laced with *Bacillus anthracis* (Anthrax) spores were distributed through the United States Postal Service, which drew attention to the fact that we could no longer remain complacent about biological agents and their potential for use in large-scale terrorist acts. The experience of the American anthrax attacks highlighted a limited preparedness, not only from the perspective of Public Health but also in terms of subsequent forensic investigation and the specific challenges that biological agents place on forensic specialists required to process contaminated scenes, and collect and analyze the evidence they contain.

Biological threat agents are defined as; "pathogenic (disease causing) organisms and toxins from a biological source" (1). Bacteria, viruses and biological toxins constitute the three main classes of biological agents. Each has specific characteristics which enable them to invade, infect or otherwise harm its target causing disease and/or death. In many cases, biological toxins can also replicate and spread from host to host. Unlike chemical agents, such as Sarin nerve gas used by the Aum Shinrikyuo cult to attack public places and subways in Japan (2–5), biological agents will show no immediate effect, have no distinguishable color, smell or taste and no single identifying signature to indicate their presence.

Biological agents are by comparison, easier to obtain, cheaper to produce per kilogram, more effective in their distribution, and include some of the most infectious and lethal substances known to man. Of the numerous pathogenic organisms which could be classified as potential bioterrorism agents, *B. anthracis* (anthrax) still remains the biological weapon of choice. This is due to its ability to produce an endospore, enabling it to form protection from desiccation, UV light and most household chemicals.

Unlike chemicals which may dissipate, biodegrade, or change structure to a less harmful form, biological spores remain viable, retaining an ability to cause disease upon exposure. It is therefore difficult to process a scene contaminated with a biological agents such as, *B. anthracis* due to the level of protective equipment which must be worn whilst attending the scene.

The reality is that not all evidence recovery will take place *in situ* and therefore it will be necessary to remove items of interest, decontaminate them and then proceed to analysis.

The recovery of traditional forensic evidence such as latent fingermarks forms an integral part of the investigation process. Considering the possibility that potential evidence has been contaminated with a highly stable and resistant entity, this may disrupt the traditional sequence and analysis requiring the addition of a decontamination step. It is not yet clear what effect the decontamination process will have on the recovery and/or analysis of such evidence?

One common decontamination procedure involves the use of formaldehyde. Formaldehyde (CAS No: 50-00-0; including formalin and paraformaldehyde) is a common chemical used in industries such as hospitals, mortuaries, film processing and textiles (for fabric treatments and leather tanning). Formaldehyde gas is primarily used in the decontamination of spaces or biological containment equipment like biological safety cabinets. It is classified as a category two carcinogen risk (phase R49), and is moderately toxic following

[1]Forensic Counter Terrorism and Disaster Victim Identification Unit, New South Wales Police, I Charles St Parramatta, NSW, Australia.
[2]Forensic & Technical Services, Australian Federal Police, GPO Box 401, Canberra, ACT, 2601, Australia Centre for Forensic Science, NSW, Australia.
[3]University of Technology, Sydney, NSW, Australia.

*This research has been presented as an oral presentation at the 18th Australian and New Zealand Forensic Science Symposium, 2006; and also as a poster at the 4th European Academy of Forensic Science Conference in Helsinki, 2006.

Received 30 Sept. 2006; and in revised form 25 Mar. 2007; accepted 8 April 2007; published 7 Sept. 2007.

acute exposure via inhalation, dermal or oral routes (6). Formaldehyde is toxic to a range of microorganisms and in high concentrations is known to kill bacteria, viruses, fungi, and parasites (7). Consequently it has long been used as a disinfectant in many industries and is still the most common and cost effective method of biological decontamination within the Public Health arena.

Formaldehyde vapor is an extremely effective biocidal agent. It acts as an alkylating agent, inactivating microorganisms by reacting with carboxyl, amino, hydroxyl, and sulfhydryl groups of proteins as well as amino groups of nucleic acid bases. A number of factors influence the efficiency of fumigation. Two such factors are temperature and relative humidity. Fumigation is most effective above a temperature of 20°C and relative humidity of 65%. At temperatures below 18°C formaldehyde fumigation is less effective (8). Formaldehyde gas has long been recognized as an effective sporicidal agent and the recommended concentrations and exposure times are well documented for the decontaminated of biological safety cabinets. The standard method employed for decontamination of biological safety cabinets includes the reaction between 30 mL or formalin and 10 g of potassium permanganate to create vaporous formaldehyde, with an exposure time of between 6 and 12 h (7).

This research seeks to evaluate the effects of using formaldehyde gas as a biological decontaminate, on the subsequent recovery of a principle forensic evidence type, latent fingermarks.

Generally, latent fingermarks are rendered visible by either physical adherence of an environmental contaminate (i.e., powdering), or via chemical or physico-chemical enhancement, creating a contrast by color or fluorescence. In this work, porous and nonporous items (paper and glass) were contaminated with viable spores, decontaminated with formaldehyde gas and then the success of a number of commonly used fingermark enhancement techniques was evaluated. A number of available fingermark recovery techniques were selected for each substrate, including those that target both water and nonwater soluble constituents of the fingermarks.

Materials and Methods

The project was divided into three phases. Phase 1 set out to determine the time needed to expose contaminated items to formaldehyde gas to decontaminate them. Test items were contaminated with a known quantity of *Bacillus* spores and exposed to formaldehyde gas for different periods of time.

Phase 2 involved determining the effect of formaldehyde gas on amino acids present in latent fingermarks. A 0.1 M solution of the amino acids was prepared in 10-fold serial dilutions. The limit of detection based on a reaction of amino acids with fingerprint recovery techniques before and after decontamination was assessed.

The objectives of phase 3 were to determine the effects of the decontamination process on the recovery of latent fingermarks by a selection of common methods, namely:

Porous item—paper

- DFO
- Ninhydrin
- 1,2-Indanedione
- Physical developer

Nonporous item—glass

- Cyanoacrylate (CA) Basic yellow 40
- Grey powder
- Magna black powder

A more detailed description of the procedures for each phase of experimentation is provided below.

Phase 1: Effects of Formaldehyde Gas on Spore Survival (Sample and Formaldehyde Preparation)

Test samples consisted of porous and nonporous items (A4 photocopy paper cut into small strips and glass microscope slides respectively), labeled in triplicate for each of the set exposure times (10, 15, 20, 25, 30, 40, 45 min, 1, 2, 3, 4, 5, 6, 12 h)

Three formaldehyde treatment methods are described and were prepared as follows;

1. Reaction of 30 mL formalin (37% w/v) with 10 g $KMNO_4$ crystals at a relative humidity of 65%. This is a standard preparation used in industry for the decontamination of biological safety cabinets (9). Because of the detrimental effects of this standard concentration (unpublished observation); a number of varying concentrations were prepared based on previously determined reactions.
2. Reaction with 15 mL formalin (37% w/v) with 5 g $KMNO_4$ crystals at a relative humidity of 65%.
3. Reaction with 15 mL formalin (37% w/v) and 7.5 g $KMNO_4$ crystals at a relative humidity of 65%. These reactions were prepared within a small tin placed inside the class III biological safety cabinet.

Spore Preparation. A powdered preparation of *Bacillus thuringiensis* var. *kurstaki* (BT) spores was diluted to the value of 1.4×10^7 spores/g or 140 cfu/μL. A 1-μL aliquot was inoculated onto each test sample and allowed to air dry. A 10-μL aliquot was also inoculated onto the paper and allowed to air dry. Positive controls were prepared by swabbing a random selection of inoculated test areas, prior to decontamination, to establish the corresponding viable spore count. Swabs were then collected following formaldehyde exposure, from both the 1 μL and 10 μL areas of inoculation, using moistened Dacron swabs lawned onto blood agar (BA) plates. Colony counts were conducted after 24 h O_2 incubation at 36°C.

Exposure to Formaldehyde Gas. Fumigation was conducted within a Class III biological safety cabinet using appropriate protective wear for the chemical classification. All test items were placed on racks; glass slides were placed on a horizontal rack within the cabinet and the paper was secured with tape to racks standing upright within the cabinet. This was done to ensure thorough exposure to all surfaces. The formaldehyde preparations were placed into the centre of the cabinet and exposed to the spontaneous reaction. Internal relative humidity of 65% and temperature between 18 and 24°C were monitored using a weather gauge, (Kestel® Nielsen and Kellerman). Test items were removed from the cabinet at each set exposure time and placed within a fume hood. Test samples were then exposed to 5 mL and 2.5 mL (respective methods) of ammonia (NH_4) to complete the neutralization process.

Phase 2: Effects of Formaldehyde Gas on Amino Acids

Preparation of Amino Acids. Four amino acids (glycine, serine, alanine and ornithine) were prepared into a 0.1 M working stock. A 1:10, 1:100, and 1:1000 serial dilution of each working stock solution was prepared using distilled water. An A4 sheet paper was divided equally into quarters along the long axis to provide rows for the neat, 1:10, 1:100, and 1:1000 dilutions. The paper was divided equally into quarters along the short axis to provide columns that acted as repeats. Each amino acid had dedicated sheets which were labeled with the amino acid name, date, and type of

fingermark development technique samples would be exposed to (either ninhydrin or 1,2 indanedione, because of their affiliation with amino acids). Different sheets were created for different exposure times (control, 15 min, 30 min or 45 min). Both control and test sheets were inoculated with 10 μL of the 0.1 M working stock solution and each of the serial dilutions and allowed to air dry for 20 min. Each of the test sheets were placed onto racks within the Class III biological safety cabinet for the designated exposure times.

Formaldehyde Gas Preparation and Exposure. The standard concentration method (30 mL formalin 37% w/v with 10 g $KMNO_4$ crystals at a relative humidity of 65%) for formaldehyde gas production was tested. All test sheets were exposed to the gas within the Class III biological safety cabinet for the set times (15, 30 and 45 min), removed and placed into a fume hood where the sheets were exposed to 5 mL of NH_4 solution to complete the neutralization process.

Amino Acid Development. A control sheet for each of the amino acids and their dilutions was treated using ninhydrin and 1,2 indanedione. For ninhydrin each sheet was dipped into the solution and allowed to air dry within a fume cupboard. Development of the amino acid spots appeared over time, varying in intensity from dark purple to pink depending on the concentration. Sheets labeled for 1,2-indanedione development were dipped into the working solution prepared as above and allowed to air dry. The dried sheets were then subjected to indirect heat for 10 sec. Amino acids appear as pink spots at the conclusion of this treatment.

Phase 3: Effects of Formaldehyde Gas on Fingermark Recovery

Sample Preparation and Fingermark Deposition—Porous Samples—Sheets of A4 photocopy paper were prepared with a standard template indicating a row for each consecutive day, from day 7 to day 1 with boxes repeated across the page four times, representing replicates for each day. Using three volunteers, a latent fingermark was placed into each square beginning with samples deposited 7 days prior to the enhancement experiment. Each test sheet was labeled with the fingermark technique used for development, either ninhydrin, DFO, 1,2- indanedione or physical developer.

Sample Preparation and Fingermark Deposition—Nonporous Samples—Glass microscope slides were selected for the nonporous substrate. Each slide was labeled with the age of latent fingermark (day 7 through to day 1) and the technique of development (CA or powder). Seven days prior to the experiment a latent fingermark was placed onto each slide and repeated in triplicate for each time period.

Control samples were prepared and marked with latent fingermarks, for both the nonporous and porous substrates for each of the development techniques to be tested. Control sets were setup to compare the recovery of latent marks both pre- and postdecontamination with formaldehyde gas. Spore survival was also tested pre- and postdecontamination via bacterial culture.

All fingermark test samples were placed in the Class III biological safety cabinet (Fig. 1) and exposed to the formaldehyde gas at set intervals; 15, 20, 30, 45, 60 min, and every hour after up to 12 h. The test sheets were removed from the cabinet and transferred to the fume hood where they were exposed to the NH_4 vapor prior to fingermark development.

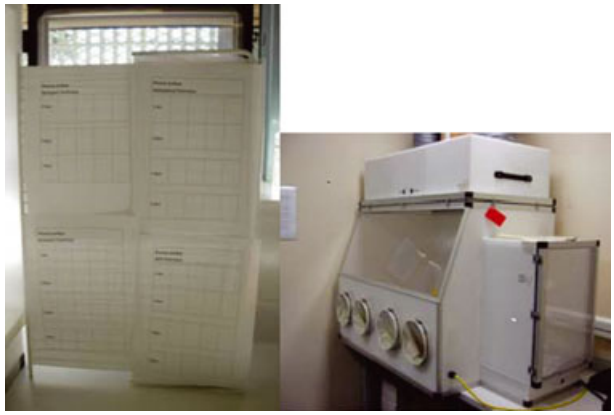

FIG. 1—*Photographs of the Class III biological safety cabinet and rack with test sheets.*

Fingermark Development Techniques—Fingermark development techniques were undertaken at the NSW Police Forensic Services Group (FSG) Fingerprint laboratory following standard operating procedures. The methods are outlined below.

Grey and Magna Black Powder—The color of the powder was selected based on the contrast required. Either of the powders was dusted over the surface of the glass slides with a squirrel hair fingerprint brush to produce visible fingermark ridges.

Cyanoacrylate Fuming—CA vapor polymerizes on greasy, moist latent fingermarks to form a hard polymer. The glass slides were placed onto a rack inside the fuming cabinet. Fifteen drops of superglue (CA) was added to a tray placed on the heating mat at the rear of the cabinet. Water was placed into a tray at the base of the cabinet. The cabinet was set for a relative humidity of 80% for a time of 30 min (based on standard operating procedures).

Ninhydrin—A working solution of ninhydrin (final concentrations; 3% ninhydrin stock, 5% v/v isopropyl alcohol in 92% v/v HFE 7100) was prepared by dissolving ninhydrin stock solution in Klenasol/HFE 7100 (nonpolar solvent carrier). Test and control samples were immersed in the working solution, allowed to air-dry and subjected to indirect heat via an iron press for 10 sec. A purple color change indicates a positive result.

1,8-Diazafluoren-9-one—The paper test and control samples were immersed in a working solution of DFO (final concentrations; 0.035% 1,8-diazafluorenone, 2.5% dichloromethane, 6% methanol, 0.5% glacial acetic acid and 90% HFE 7100), air-dried and immersed again. Once dry they were exposed to indirect heat from an iron for 10 sec. Marks were observed with the Polilight lamp (505 nm) and photographed with a barrier filter at 530 nm using a digital Nikon F3 camera.

1,2-Indanedione—A working solution of indanedione was prepared by mixing 2.5 g 1,2-indanedione with 25 mL acetic acid, 225 mL ethyl acetate and 2250 mL HFE 7100. The test and control samples were immersed in the working solution, allowed to air-dry and then immersed again. Once dry the samples were then subjected to indirect heat via the iron press. Fingermarks were observed under the Polilight lamp (530 nm). Fingermarks were photographed using a 590 nm barrier filter with an excitation wavelength of 530 nm.

Physical Developer—The physical developer process is comprised of three major constituents; 100 mL redox solution A (3% w/v ferric nitrate, 8% w/v ferrous ammonium sulphate, 2% w/v citric acid, 4% w/v detergent-surfactant stock solution in water), 5 mL silver nitrate solution B (20% w/v silver nitrate in distilled

water) and 500 mL maleic acid prewash solution. All three solutions are stored at 4°C, made fresh on the day of analysis and brought to room temperature prior to processing. Five clean glass trays are required for development; 1) distilled H_2O, 2) maleic acid prewash solution, 3) dH_2O, 4) physical developer mix (solution B is slowly added to solution A gently stirring) and 5) dH_2O. The test and control samples for this technique were individually immersed in tray 1) for 10 min then transferred to tray 2) for 5 min, rinsed in tray 3) before being added to the physical developer (tray 4) individually until a silver-grey contrast was visible. Samples were then washed in tray 5) and allowed to air dry. Developed marks were photographed with a digital Nikon F3 camera.

Results and Discussion

Formaldehyde Gas as a Biological Decontaminate

Bacillus thuringiensis spores were exposed to the standard formaldehyde decontamination for set intervals, up to and including 12 h. More than 1 h exposure demonstrated excellent sporicidal activity with no colony growth recorded at intervals between 1 and 12 h (results not shown). Figure 2 compares the standard fumigation method (30 mL 37% w/v formalin, 10 g $KMNO_4$) with one of the trial methods (15 mL 37%w/v formalin, 5 g $KMNO_4$) and the new fumigation method (15 mL 37%w/v formalin, 7.5 g $KMNO_4$). The standard method was successful at rendering the spores nonviable after 25 min yet destructive to amino acids. The spore viability remained high with the trial method because of the incomplete reaction of the two key components which reduced the amount of available toxic vapor (unpublished observation). Experimentation with different ratios lead to the development of a new fumigation method which demonstrated a 10-fold reduction in viable spores after 30 min exposure, with no growth recorded after 35 min. This result varied greatly from the recommended time between 6 and 12 h exposure for the fumigation of biological safety cabinets.

While the exact concentration of any bacterial contaminant may be unknown at the time of testing, it is assumed that the item is contaminated with sufficient particles to cause harm and therefore requires decontamination. Figure 3 compares viable spore recovery from both 1 μL and 10 μL inoculums, where 1 μL represents moderate spore contamination (1.4×10^4 cfu/mL) and 10 μL represents gross spore contamination (1×10^{10} cfu/mL). At an exposure time of 35 min a 95% reduction in spore viability was recorded from the 10 μL swabs and a 99% reduction in viable spores from the 1 μL inoculum with no growth recorded for either concentration after 40 min exposure. Therefore while the concentration of initial spore contaminant may be high, the effective decontamination time

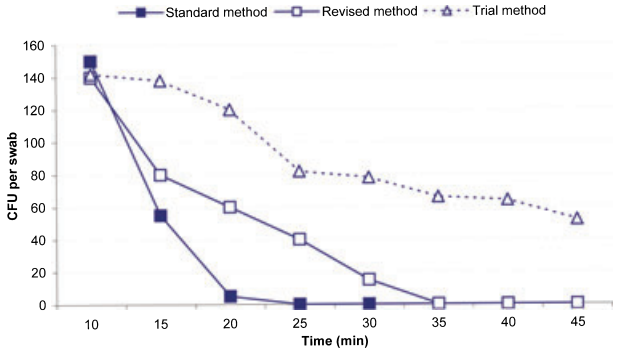

FIG. 2—*The effects formaldehyde concentrations on spore survival.*

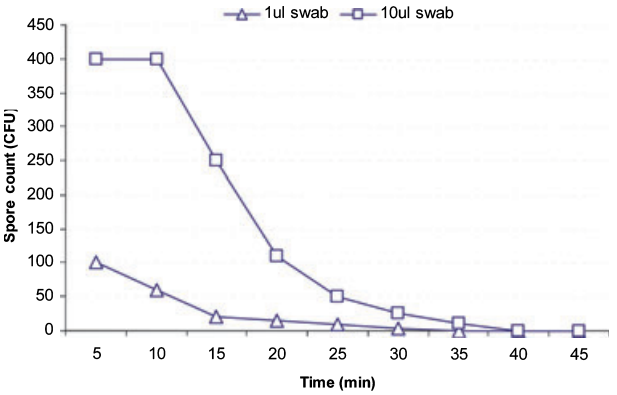

FIG. 3—*The effects of spore load on effective exposure time.*

did not alter relative to the degree of contamination. These results indicate that an effective minimum sporicidal exposure time is 40 min and that extended exposure may not be required for adequate kill rates.

Effect of Formaldehyde on Amino Acids

The use of the standard formaldehyde concentration and exposure time recorded detrimental effects on the recovery of latent fingermarks. One of the main constituents of latent fingermarks is amino acids (10) and therefore the effect of formaldehyde gas on these products was established. Serial dilutions for each amino acid; glycine, serine, alanine, and ornithine, were exposed to the standard decontamination concentration for 15, 30 and 45 min respectively. Each test sheet was treated using 1,2-indanedione and ninhydrin as described earlier. The effects of fumigation on these enhancement techniques were evaluated against the four amino acids (Table 1). The sensitivity or limit of detection was evaluated by comparing reactions with each dilution series (neat, 1:10, 1:100, and 1:1000).

Ninhydrin has long been used for the development of latent fingermarks on porous items such as paper; its effectiveness to react with amino acids is evident with strong reactions for all amino acids at 0.1 M and medium reactions visible for 1:10 dilutions, for

TABLE 1—*Reaction of amino acids with a standard preparation of formaldehyde gas.*

	Ninhydrin				1,2-Indanedione			
Amino acid	Control	15 min	30 min	45 min	Control	15 min	30 min	45 min
Glycine neat	xxx	xxx	xxx	xxx	xxx	x	x	x
1:10	xx	xx	xx	xx	xx	Pol x	Pol x	Pol x
1:100	x	x	x	x	Pol x	Pol x	Nil	Nil
1:1000	Nil	Nil	Nil	Nil	Nil	Pol x	Nil	Nil
Serine neat	xxx	xxx	xxx	xxx	xxx	x	x	x
1:10	xx	xx	xx	xx	x	Pol x	Pol x	Pol x
1:100	x	x	x	Nil	Pol x	Nil	Nil	Nil
1:1000	x	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Ornithine N	xxx	xxx	xxx	xxx	xxx	Pol x	Pol x	Pol x
1:10	xx	xx	xx	xx	xx	Pol x	Pol x	Pol x
1:100	x	Nil	x	x	Pol x	Nil	Nil	Nil
1:1000	Nil	Nil	Nil	Nil	Nil	Nil	Nil	Nil
Alanine neat	xxx	xxx	xxx	xxx	xxx	Pol x	x	Nil
1:10	xx	xx	xx	xx	x	Pol x	Pol x	Pol x
1:100	x	x	x	X	Pol x	Nil	Nil	Nil
1:1000	x	Nil	Nil	Nil	Pol x	Nil	Nil	Nil

xxx, strong reaction to amino acid; xx, medium reaction; x, faint reaction; Pol x, visible with Polilight lamp; Nil, no reaction.

each exposure time. These results are consistent with the control samples indicating no detrimental effect from formaldehyde at these concentrations. Yet as the concentration of amino acids decreases differences between the test and control samples appear, with the exception of glycine, which demonstrated no change between the control and test samples. Serine, ornithine, and alanine all demonstrated a loss of amino acid reactivity at either the 1:100 or 1:1000 dilution indicating formaldehyde interaction at these low concentrations.

By comparison 1,2-indanedione, has demonstrated raised sensitivity to changes in the environment in previous experiments (11) and likewise Table 1 indicates similar findings with reduction of visible amino acids evident after only 15 min exposure to formaldehyde gas.

The effect of each of the porous techniques used is outlined in Fig. 4 where both DFO and 1,2-indanedione demonstrate increased sensitivity to formaldehyde treatment compared with ninhydrin.

Effect of Formaldehyde on Fingermark Development Techniques

The fingermark development methods selected represent the range of techniques currently being utilized within major crime laboratories. While it may be possible to develop latent marks at the crime scene, it will not always be possible to collect forensic evidence *in situ* within CBR contaminated incidents, because of environmental factors which could potentially affect both the operator and the integrity of the mark itself (12). In such circumstances it is advisable to remove the item from the scene so that specialized processing and development can occur within the optimal conditions of a fingerprint laboratory. All techniques utilized in this research have been carried out as they would be within a laboratory—subsequent to the hazardous material, namely the bacterial spores, having been destroyed (via decontamination) and thereby no longer posing a risk to the operator.

Test samples for both porous and nonporous substrates were subjected to both the standard and revised methods of formaldehyde decontamination for the predetermined effective exposure time of 40 min. The nonporous enhancement techniques (fingerprint powder and CA fuming) were largely successful (75% and 87.5%, respectively) at recovering latent marks from the glass slides postdecontamination (Fig. 4). Varying the decontamination procedure from the standard to the revised method had no effect on either nonporous technique. The porous techniques however demonstrated widely varying results. Physical developer was the only enhancement method that was robust to the decontamination process, showing minimal or no effect on the success of enhancement following both the standard and revised decontamination methods. The ability of the three other porous enhancement techniques (ninhydrin, DFO and 1,2-indanedione) to detect latent marks was more profoundly compromised by the decontamination process. For all these three techniques, no latent marks were recovered postdecontamination, using the standard formaldehyde concentration. The revised decontamination method had a less severe effect, particularly for ninhydrin development (66% successful recovery rate). Latent marks exposed to the revised decontamination treatment were still able to be recovered following DFO and indanedione treatment however the successful recovery rates were reduced (33% and 8%, respectively).

Cyanoacrylate, physical developer and fingerprint powders, all target the greasy and moist, or oily components of the fingermark. Using these reagents numerous fingermarks were developed regardless of latent mark age or whether they had been treated pre- or postdecontamination. In fact, recovery was more successful with older marks. While powders are the easiest to use and can be more readily applied to recover marks *in situ*, more sensitive techniques are normally applied within the laboratory.

Figure 5 demonstrates the high quality of marks recovered both pre- and postdecontamination using physical developer, with this individual mark being divided in half and having one half exposed to formaldehyde (standard concentration) for 40 min.

The three remaining techniques, ninhydrin, 1,2-indanedione and DFO are popular and sensitive techniques for the development of fingermarks from porous substrates such as paper. Yet the exposure to the standard formaldehyde gas concentration for 40 min led to no latent marks being recovered. There was only evidence of a pale mark where the original mark was placed. Latent mark visualization using these three techniques is dependant on the presence (and hence the concentration) of amino acids. The total amount of amino acids in a mark has been reported to be between 0.3 and 2.59 mg/L, with serine, glycine, ornithine, and alanine being the most abundant (13). Techniques such as ninhydrin, DFO and 1,2-indanedione target the amino acid composition of the mark generating a colored reaction upon application, the fact the no latent marks were recovered using these techniques indicates interference or destruction of the amino acid component of the mark during the decontamination process. This is consistent with the results obtained in Phase 2 of this study and illustrated in Table 1, by varying degree of degradation of four amino acids when exposed to formaldehyde.

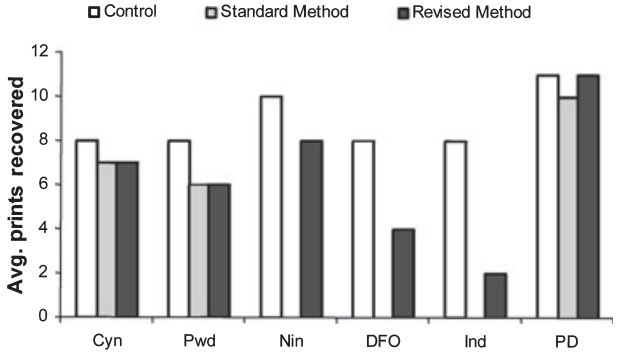

FIG. 4—*Effects of formaldehyde on latent fingermark recovery techniques.*

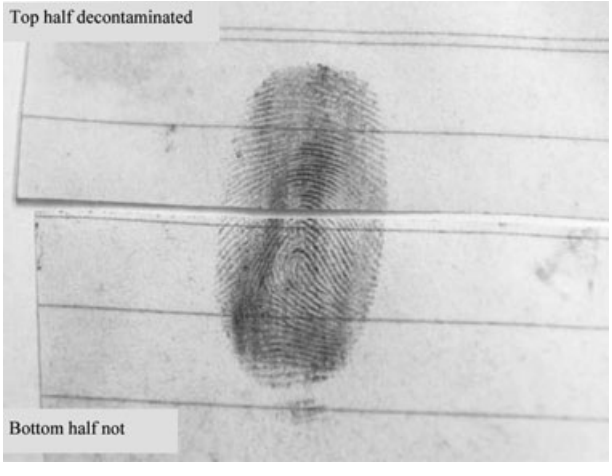

FIG. 5—*Development of print using physical developer after decontamination treatment with formaldehyde (standard concentration).*

By comparison the revised fumigation method, when applied to the test samples, allowed for effective recovery of latent marks on the nonporous test samples for each of the methods tested. We demonstrated earlier that the revised decontamination method remained effective in destruction of bacterial spores. Of the four porous methods selected physical developer, while time consuming, was the most sensitive with a 91% successful recovery rate, followed by ninhydrin (66%), DFO (33%), and 1,2-indanedione (8%). Recovery rates are dependant on development techniques as well as the quality and composition of the latent mark (unpublished observation).

Conclusion

The results of this research identified the need to develop standard operating procedures for forensic personnel working within the counter terrorism field. It has been observed that standard procedures for bacterial collection, processing and destruction will not always compliment the requirements for analysis of traditional forensic evidence types, such as fingermarks. The results of this study indicate that the standard formaldehyde formulation for bacterial decontamination, while effective at destroying the contaminant, will also affect the recovery of latent fingermarks particularly from porous substrates. A revised method of bacterial decontamination incorporating a reduced concentration of formaldehyde and reduced exposure times has been proposed and evaluated. The revised method enabled successful decontamination of moderately and grossly contaminated evidence within a 40-min exposure period. This research evaluates the effects of formaldehyde gas on current fingermark enhancement techniques and a preferred order of development has been established. The techniques utilized for the development of fingermarks from nonporous items namely CA and powders, demonstrated no reduction in efficacy following exposure to formaldehyde gas. However, recovery of fingermarks from porous items such as paper demonstrated a greater degree of sensitivity to exposure to this biological decontaminant. Based on our observed results we recommend the following processing techniques; ninhydrin or DFO or 1,2-indandedione followed by physical developer, to increase the possibility of successful latent fingermark recovery.

Future research will be conducted in the effects of formaldehyde gas decontamination on the recovery and analysis of DNA and on methods employed for the recovery of latent shoe marks.

Acknowledgments

The NSW Police Major Crime Laboratory and the staff of the specialist Fingerprint Laboratories for their assistance, time and knowledge in the area of latent fingermark recovery techniques. This work would not be possible without the financial support of the New South Wales Police Forensic Services Group.

References

1. Textbook of military medicine medical aspects of chemical and biological warfare. http://www.das.cs.amedd.army.mil/textbook.htm.
2. Morita H, Yanagisawa N, Nakajima T, Shimizu M, Hirabayashi H, Okudera H, et al. Sarin poisoning in Matsumoto, Japan. The Lancet 1995;346(8970):290–3.
3. Okumura T, Suzuki K, Fukuda A, Kohama A, Takasu N, Ishimatsu S, et al. The Tokyo subway sarin attack: disaster management, part 1: community emergency response. AEM 1998;5(6):613–7.
4. Okumura T, Suzuki K, Fukuda A, Kohama A, Takasu N, Ishimatsu S, et al. The Tokyo subway sarin attack: disaster management, part 2: hospital response. AEM 1998;5(6):618–24.
5. Okumura T, Suzuki K, Fukuda A, Kohama A, Takasu N, Ishimatsu S, et al. The Tokyo subway sarin attack: disaster management, part 3: national and international responses. AEM 1998;5(6):625–8.
6. NIOSH pocket guide to chemical hazards. Washington, DC: Department of health and human services CDC, http://www.cdc.gov/niosh/npg/.
7. Lach VH. A study of conventional formaldehyde fumigation methods. J Appl Bacteriol 1990;68:47.
8. Munro K, Lanser J, Flower R. A comparative study of methods to validate formaldehyde decontamination of biological safety cabinets. Appl Environ Micro 1999;65:873–6.
9. Australian/New Zealand Standard 2243.2 safety in laboratories Part 3. Microbiology aspects and containment facilities, 2002, www.nata.asn.au.
10. Lee HC, Gaensslen RE. Advances in fingerprint technology, 2nd ed. Boca Raton, FL: CRC Press, 2001;70–72.
11. Wallace-Kunkel C, Lennard C, Stoilovic M, Roux C. Optimisation and evaluation of 1,2-indanedione for use as a fingermark reagent and its application to real samples. For Sci Int 2006;168(1):14–26.
12. Wilkinson D, Hancock J, Lecavalier P, McDiarmid C. The recovery of fingerprint evidence from crime scenes contaminated with chemical warfare agents. J Forensic Ident 2005;55(3):326–66.
13. Lennard C, Stoilovic M. Application of forensic light sources at the crime scene. In: Horswell J, editor. Practice of crime scene investigation. Boca Raton, FL: CRC Press, 2004:64–73.

Additional information and reprint requests:
Rebecca J. Hoile, B.Sc.
Forensic Microbiologist
Forensic Counter Terrorism and Disaster Victim Identification Unit
NSW Police Headquarters
Level 5 Tower B, 1 Charles Street
Parramatta
New South Wales 2150
Australia
E-mail: hoil1reb@police.nsw.gov.au