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PAPER

CRIMINALISTICS

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Detection of Latent Bloodstains Beneath Painted Surfaces Using Reflected Infrared Photography

ABSTRACT: Bloodstain evidence is a highly valued form of physical evidence commonly found at scenes involving violent crimes. However, painting over bloodstains will often conceal this type of evidence. There is limited research in the scientific literature that describes methods of detecting painted-over bloodstains. This project employed a modified digital single-lens reflex camera to investigate the effectiveness of infrared (IR) photography in detecting latent bloodstain evidence beneath a layer or multiple layers of paint. A qualitative evaluation was completed by comparing images taken of a series of samples using both IR and standard (visible light) photography. Further quantitative image analysis was used to verify the findings. Results from this project indicate that bloodstain evidence can be detected beneath up to six layers of paint using reflected IR; however, the results vary depending on the characteristics of the paint. This technique provides crime scene specialists with a new field method to assist in locating, visualizing, and documenting painted-over bloodstain evidence.

KEYWORDS: forensic science, infrared photography, bloodstains, forensic photography, invisible radiation, latent evidence

Blood evidence recovered at crime scenes is generally considered as having a high degree of forensic value. Several contemporary techniques may be used to detect the presence of blood at scenes where violent crimes were committed (1–3). However, when blood evidence has been tampered with, hidden, or covered up, it becomes increasingly difficult to detect. A common question facing forensic practitioners when examining blood is whether a red-brown stain is actually blood. This may lead to further questions such as whether or not the blood is of human origin, and if so, whether the bloodstains can be linked to a particular individual using DNA. Furthermore, bloodstain pattern analysis (BPA) may also be used to assist in reconstructing the event. Because bloodstain evidence has such an intrinsic forensic value, perpetrators frequently attempt to conceal or remove this type of evidence from the scene (4). One method of concealing blood spatter is to paint over the wall or surface where the stains are located.

Painting over bloodstains on walls will conceal blood evidence beneath the paint layers. This makes detection of bloodstains more difficult during crime scene examinations. There is limited research described in the forensic science literature that investigates the visualization of bloodstains located beneath painted surfaces. Some research suggests a nondestructive technique using monochromatic light sources such as a Polilight™ (5); however, some work also suggested that the blood reagent luminol could still produce a chemiluminescence reaction even when the blood is located beneath a layer of paint (6,7). Previous studies have also demonstrated that paint does not appear to cause false positive results with luminol (8). While there are a limited number of published studies involving painted-over bloodstains in the context of crime scene investigation, extensive research has been completed in the field of art conservation in regards to visualizing artifacts beneath paint layers. The application of infrared (IR) photography has been used in conservation practices to evaluate the authenticity of paintings by looking beneath its uppermost paint surface (9).

IR radiation is capable of penetrating visually opaque material to reveal object states that the naked eye cannot see. It is also capable of discerning between different material that may appear optically similar under visible light but produce distinct differences when irradiated by IR. These variations occur because objects can absorb, reflect, or transmit IR by varying degrees (10). The two main uses for using IR within art conservation practices are (i) to detect any alterations or restoration work that has been made to the painting and (ii) to visualize any preliminary drawings or markings beneath the painted surface that were used to form the composition of the artwork. These original markings also referred to as "underdrawings" may act as a signature of the painting and are usually not present in forgeries. IR has proven to be a successful technique in this context and can record artifacts that are beneath the paint surface (11).

Previous research into the interaction between IR radiation and paint determined that a layer of white paint could increase the degree of transmission when illuminated by IR radiation. Mairinger (12) suggested the following factors that increase IR penetration through paint:

> i) the greater the wavelength of the incident radiation, ii) the smaller the thickness of the paint layer, iii) the smaller the number of particles in the layer (pigment/volume concentration) and iv) the lesser the difference of refractive indices of pigment and medium (12, p. 42).

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A practical study into the transmission of IR radiation through paint pigments determined that certain combinations of optical filters are able to penetrate some pigments, whereas other paints were completely opaque and that multiple pigmented paints impair IR penetrability (13).

IR photography has already found numerous applications within forensic science practices. Early research identified IR as being capable of visualizing bite marks on human skin (14) as well as the assessment of inks during document examination (15). Recent studies have found IR useful in the visualization of gun shot residue patterns on dark and multicolored clothing (16) as well as detecting and visualizing the presence of a tattoo that had been removed by laser surgery (17). IR is applied within the BPA discipline with the most common use assisting the visualization of latent bloodstains on dark surfaces (18). Other recent studies using IR involve the detection of blood on different types of clothing, fabrics, and patterns (19) and identifying bloodstains on black fabric using various blood dilutions (20). The most noteworthy conclusion to draw from the scientific literature is that blood is capable of absorbing IR radiation. Chun-Yen Lin et al.'s (20) research indicated that blood continued to absorb IR radiation up to 1/8th of its original concentration.

Previous studies have indicated two key points relating to this work: (i) that paint layers, depending on pigment and thickness, are capable of allowing IR transmission and (ii) that blood absorbs IR radiation. Combining these two concepts produce a likely hypothesis suggesting it is possible for IR to visualize a latent bloodstain located beneath a layer of paint. Theoretically, IR radiation will transmit through certain paint pigments, thus increasing the transparency of paint layers, be absorbed by blood, while the background reflects IR. These differences in spectral response (transmission, absorption, and reflection) are recorded by IR and latent bloodstains could be detected.

This project employed a modified digital single-lens reflex (SLR) camera, converted to exclusively record IR wavelengths, to investigate the effectiveness of IR in detecting latent blood beneath layers of paint. A qualitative evaluation was completed by comparing images taken from a series of samples using IR and standard visible light photography. Quantitative image analysis was also conducted to verify the photographic results.

Throughout this study, several objectives were investigated and included (i) standardizing or calibrating exposure values (EVs) between the specially modified IR camera and a standard camera, to optimize image quality and provide an accurate comparative image analysis; (ii) developing a method to quantify the effectiveness of IR in detecting and visualizing blood beneath layers of paint; and (iii) investigate the effects of variables such as different types of paints, different colors, and interferences.

Materials and Methods

Spectral Sensitivity of the Digital Camera

This project utilized a specially modified Canon EOS 10D™ digital SLR camera (Canon U.S.A., Inc., Lake Success, NY) that was converted to record exclusively within the IR spectrum. The camera modification was conducted by a commercial photographic engineering company. Digital cameras are naturally sensitive to the IR region because of the spectral sensitivity of the semiconductors used by digital cameras. However, IR blocking filters are installed over these sensors to improve optical performance and assist chromatic aberration correction by attenuating the IR wavelengths recorded by the sensor. Modification involved replacing the IR blocking filter with an IR transmission filter directly onto the sensor. The replacement filter is a long pass optical filter equivalent to a Wratten 87C (IR transmission > 820 nm). For the remainder of this article, the modified Canon EOS 10D™ SLR camera that photographs exclusively within the IR spectrum will be referred to as the IR camera and the standard unmodified camera used to photograph control images will be referred to as the VIS camera.

Calibrating Exposure Values and Camera Settings

To enable accurate comparative evaluation between each type of recorded image (IR and visible light), each photograph was taken at a consistent EV. Owing to each recording technique using different components of the electromagnetic spectrum and using different optical filters on each camera sensor, calibration between exposures is necessary. A critical component of this experimental design was to determine "equal" exposures for both standard white light and IR techniques. An experiment was undertaken to ensure that equivalent exposures could provide a meaningful comparison between each recording method.

Testing EVs consisted of taking a series of images using a Kodak™ Q13 (Rochester, NY) test target standard. Cameras were attached to a copy stand and evenly lit using two tungsten halogen lights. Several test images were taken of the Q13 test target with a range of altering exposures. The images were analyzed using Adobe Photoshop™ (Adobe Systems Inc., San Jose, CA) by first removing all color information and then measuring the segment defined as "M" on the Q13 target. This value is considered as having an 18% reflectance and is a standard reflectance value in photographic science. The eyedropper tool was used to measure the pixel brightness, which has a numerical range for an 8-bit image between 0 and 255 (256 different brightness values). By analyzing the "M" segment, EVs for each photographic method were chosen where the "M" was closest to match the 18% gray value, which consisted of a brightness value of 128 (or 256/2). For the IR camera, this exposure was calculated to be *f*/2.8 at 1/250th sec (EV 11) and *f*/3.5 at 1/320th sec (EV 11.9) for the VIS camera at 100ISO. The camera settings used throughout the experiment are detailed in Table 1.

Sample Preparation

Sheets of 10 mm plasterboard were sourced from a local hardware store. A white acrylic primer was applied to the plasterboard to seal the surface and provide an undercoat similar to a wall surface. A single sheet of plasterboard was also used as a control surface and was not primed. The primer was left to dry for a minimum of 12 h before any further sample preparation was completed. After the plasterboard was sealed, it was cut into a series of 200 × 200 mm sized pieces.

TABLE 1—*Camera settings for each camera used in the experiment.*

	IR Camera	VIS Camera
Lens	50 mm macro	50 mm macro
f/stop	2.8	3.5
Shutter speed	250	320
ISO	100	100
"u" distance	0.8 m	0.8 m
EV	11	11.9

Stencil Construction

A perspex stencil was constructed to assist in placing the bloodstains on the sample surface. The stencil positioned the bloodstains in the same arrangement on each sample to aid in the quantitative image analysis component of the research. The perspex stencil included a series of small holes aligned in a 3 × 3 grid. The holes aligned to a grid with total dimensions of 100 × 100 mm, and each hole was *c.* 50 mm horizontally or vertically away from one another. The stencil also had four smaller holes located peripherally to this grid, large enough for a pin to be inserted. Once the template had been drawn onto the perspex surface, 10-mm holes were drilled for each blood hole and 2 mm for the peripheral holes. A small red triangle adhesive was attached to the front of the stencil to ensure it consistently faced the same direction while applying blood to the sample surface. Four large magnetic weights were also attached to the bottom of the stencil to create a gap between the stencil and the plasterboard while pipetting blood to prevent pooling. A schematic of the stencil design is shown in Fig. 1.

Detection of Blood Beneath Paint

Three test conditions were investigated in this study and include types of white paint, colored paints, and interferences. The three types of white paint selected were acrylic, oil-based, and white spray paint; the acrylic colored paints selected were black, red, yellow, blue, green, orange, and purple; and the interferences tested included using undiluted blood, diluted blood to 1/100th and 1/1000th, cleaning up the blood after dropping it on the plasterboard, using water in lieu of blood, having no undercoat present, and then using white acrylic paint on a black undercoat and black acrylic paint on a black undercoat. A blank sample where no blood was applied to the sample surface was also used as a control. The basic procedure for preparing all samples, painting, and photography is outlined below.

The perspex stencil was placed over a prepared piece of plasterboard and secured. Four pins were inserted through the peripheral holes in the stencil to create an indentation in the plasterboard. The stencil was then removed and four black map pins were placed securely into the indentations. A control photograph (C0) of the sample was then taken using both the IR and the VIS cameras. The cameras were attached to a copy stand, and the samples were evenly lit with two tungsten halogen lamps that were set up permanently for the study. All samples at each stage of the project were photographed on a piece of black cardboard with a linear scale, a Q13 target, and the respective sample number. The layout of how samples were photographed is shown in Fig. 2.

The stencil was then placed back onto the plasterboard using the map pins to guide it to its original position. Two hundred microliters of defibrinated horse blood (sourced from commercial biological supplier) that had been diluted to 1/10th of its original concentration was dropped directly through each of the nine holes in the stencil using a micropipette. The stencil was removed and the blood was left to dry for a minimum of 48 h. Once the blood had dried, the layer zero photograph (L0) was taken using both IR and VIS cameras. After photographing the L0 layer, the four map pins were removed from the plasterboard and a single layer of selected paint was applied to the sample using small disposable paint rollers. To maintain a level of consistency, the painting technique was standardized. Thirty milliliters of paint was considered a "single paint layer" and covered the entire surface of the plasterboard. The pins were then placed back into their original positions, and the paint was left to dry for a minimum of 24 h. Once the paint had dried, the layer one photograph (L1) was taken in the same manner as the L0 photograph. This process was repeated when applying layers two through to six (L2–L6). After the photography had been completed, visual examination and image analysis were conducted.

A slightly altered method was required when applying the white spray paint. Once both the C0 and L0 photographs were taken, the can of paint was shaken for *c.* 2 min before spraying and for a further 10 sec every few minutes during application. The can was held *c.* 150–200 mm from the surface and the paint was applied by moving back and forth in even strokes. This procedure was then repeated to apply layers two through to six (L2–L6).

A slightly altered method was also needed when introducing interferences to the samples as changes were made to the way in which the paint or blood was applied to the sample surfaces. All interferences used white acrylic paint unless otherwise specified; one sample used undiluted blood instead of 1/10th diluted blood; two samples used 1/100th and 1/1000th diluted blood, respectively; one sample involved allowing the blood to dry before wiping it off with a cleaning product before applying the first layer (L1) of paint; one sample substituted water for blood; one sample used a plain piece of plasterboard without any primer applied; one sample involved the plasterboard having a black undercoat before applying layer one through to six (L1–L6) of white acrylic paint; and one sample involved the plasterboard having a black undercoat before applying layer one through to six (L1–L6) of black acrylic paint.

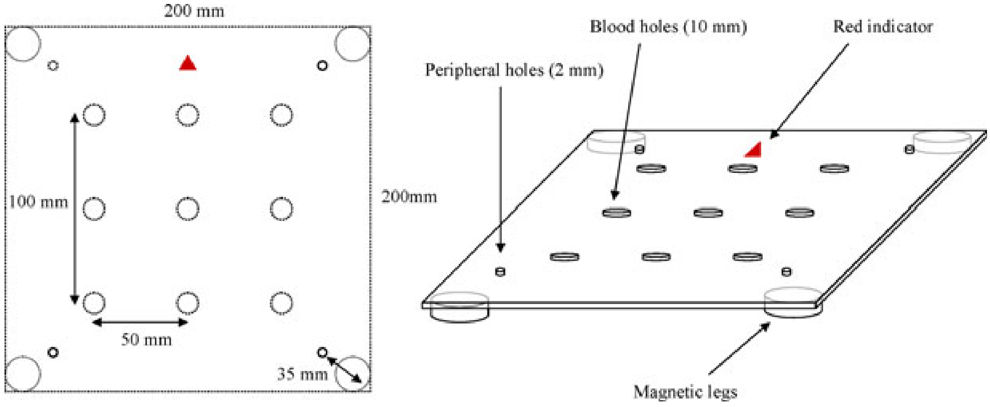

FIG. 1—*Schematic of the stencil design.*

FIG. 2—*Layout of samples photographs; (from top to bottom) a linear scale, the plasterboard sample with four black map pins in place, the Q13 test target and the sample number.*

Visual Examination of Photographs and Image Analysis

The qualitative analysis component of this study involved a visual comparison of the resultant photographs of all samples at each stage of the painting process. By visually examining the images on a calibrated computer monitor, the paint layer at which the bloodstain became latent was recorded, and the success of the technique was rated.

Image analysis used a combination of Adobe Photoshop^{TM} and ImageJ^{TM} (National Institutes of Health, Bethesda, MD) to quantify the effectiveness of this technique. Adobe Photoshop^{TM} was used to prepare the photographs for analysis, and ImageJ was used to record pixel brightness values from the image. Image analysis was completed for the C0, L0, L1, L2, and L3 photographs of each sample. Each photograph to be analyzed was cropped into a 1000 × 1000 pixel image by using the pins on each sample as a guide to form a square. The image was then transformed into an 8-bits/channel image by selecting Image > Mode and by checking both “Grayscale” and “8-bits/channel” so that all color information was discarded and to maintain consistency during analysis. The image was then opened in ImageJ and the linear tool was used to take six individual readings at the 150, 500, and 850 pixel mark, both horizontally and vertically, and ensuring it crossed through three bloodstains each time. By selecting Analyze > Plot Profile on ImageJ, a profile of the line was graphed and the brightness values were saved into Excel spreadsheets. Each pixel is assigned a brightness value (a numerical value from 0 to 255) that represents its brightness on a gray scale. A value of 0 (zero) refers to a pixel being completely black and a value of 255 refers to a pixel being completely white. By generating a plot profile, graphic information can be turned into empirical data, and visual changes across the image, such as brightness, can be recorded.

Results

The results from this study are expressed using two methods (i) a qualitative evaluation using a visual comparison between images taken using IR and standard visible light photography and (ii) a quantitative method using image analysis (mean pixel brightness values).

Comparison of photographs using IR and white light photography demonstrated that IR could detect latent bloodstains beneath up to six layers of paint. Certain paints, such as black and purple acrylic, allowed greater IR transmission than other paints used in this study. Bloodstains located beneath white acrylic could not successfully be photographed using IR after a few layers had been applied to the sample. In cases where the blood remained visible beneath multiple layers when photographed using both standard and IR, such as with red acrylic paint, IR did improve the visualization of the bloodstains. Figure 3 summarizes the information obtained from the qualitative analysis of all test conditions investigated and reports the layer where the bloodstain was last visible. Figure 4 features a series of photographs that were taken during the study to demonstrate the comparison of results between reflected IR and visible light photography.

Types of White Paint

IR photography was capable of visualizing bloodstains located beneath up to three layers of the selected types of white paint within this test condition. Bloodstains located beneath white acrylic remained detectable under visible light even after three layers had been applied. IR was not able to detect the bloodstains past this point and it did not improve visualization of L1 or L2 in comparison with the images taken using standard photography. Figures 5 and 6 are profiles of the mean pixel brightness values for each photographic method. In both figures, the line representing the blood layer (L0) demonstrates how well blood can be detected from the background before paint layers were applied over the bloodstains. The greater separation between the line representing the control layer (C0) and the lines for each respective paint layer where the blood was located indicates that the bloodstain was detectable after each layer of paint had been applied.

Blood located beneath white oil-based and spray paint was detected using IR after three layers. Under visible light, the bloodstain beneath the oil-based paint was no longer detected after two layers had concealed the sample. Similarly, the blood beneath spray paint disappeared following application of the third layer.

Colored Paints

IR photography visualized bloodstains located beneath six layers of black acrylic paint despite a single coat rendering the bloodstain undetectable when viewed under normal visible light. Because black acrylic paint gave such a positive result, image analysis was extended to include all six layers. Figures 7 and 8 provide a profile of the mean pixel brightness values for each photographic method and demonstrate the difference between the two results. The graphs demonstrate that following the third layer of paint, the application of additional layers caused detection of the bloodstains using IR to reach a limit where it could no longer quantitatively be distinguished from the surrounding background. However, close visual examination of the IR photographs confirmed minor traces of the bloodstain even after the sixth layer had been applied.

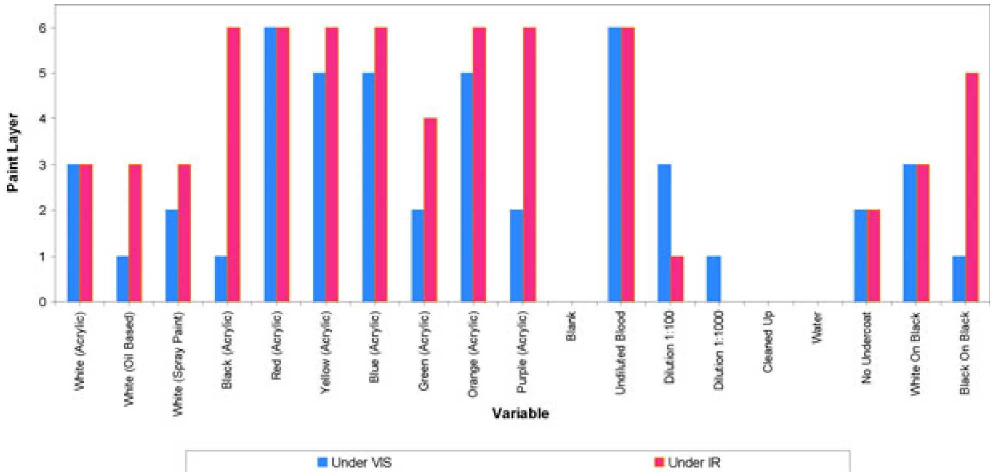

FIG. 3—*Visual examination results when recorded using visible light photography (VIS) and infrared (IR) photography; the paint layer value refers to the layer where the bloodstain was last visible in the photographs.*

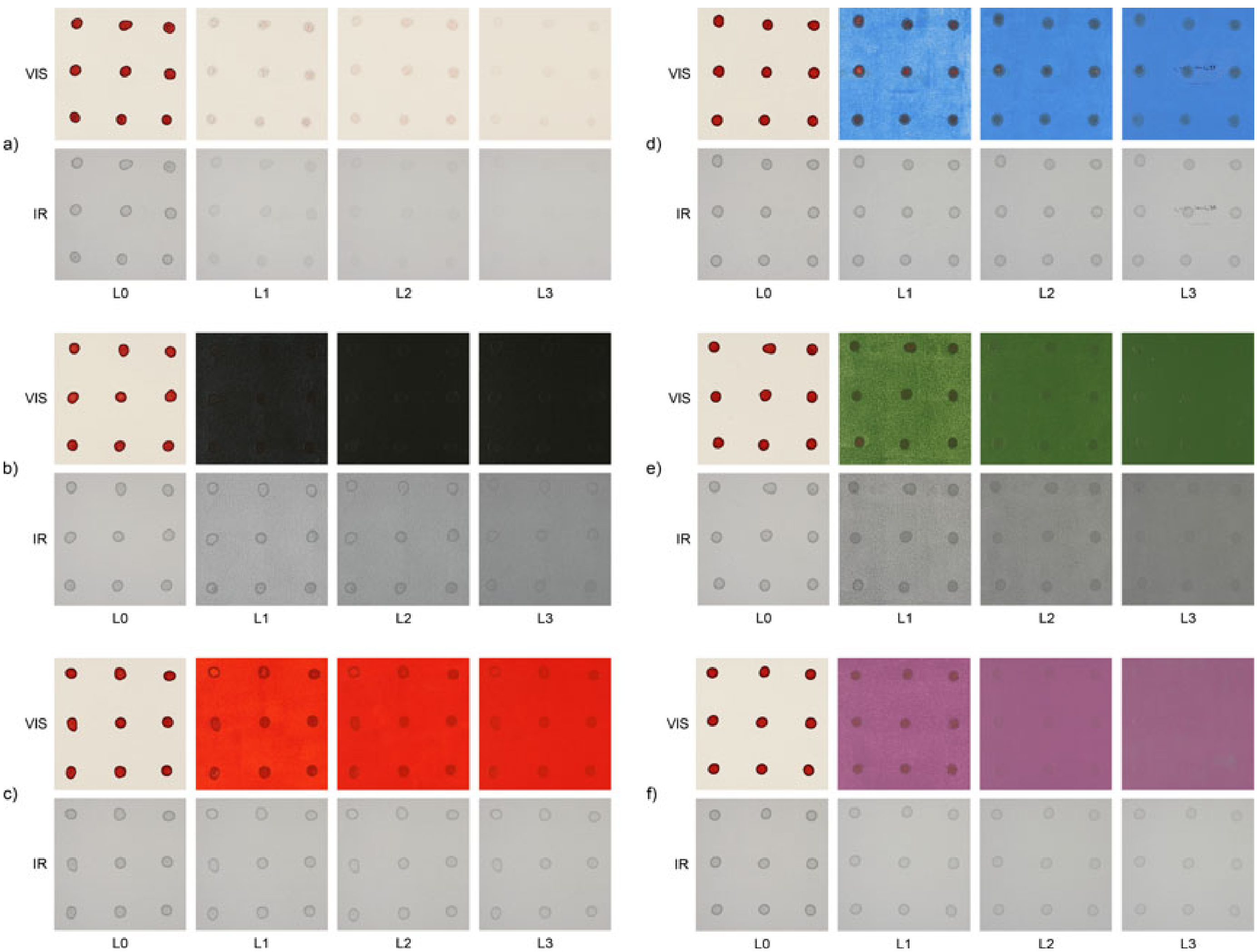

FIG. 4—*Photographs of layers L0, L1, L2 and L3 for (a) white acrylic paint, (b) black acrylic paint, (c) red acrylic paint, (d) blue acrylic paint, (e) green acrylic paint, and (f) purple acrylic paint using infrared (IR) photography and standard visible light photography (VIS).*

Red acrylic paint was incapable of effectively concealing the bloodstains even after six coats had been applied. However, IR improved the contrast between the blood and the background and improved visualization. Yellow, blue, and orange acrylic paint all allowed bloodstains to be detected after six layers had been applied. Green acrylic completely concealed the bloodstains after three layers but it remained visible in the IR photographs until the fourth layer. IR could visualize bloodstains located beneath six layers of purple acrylic compared to three layers to conceal the bloodstains using standard photography.

Interferences

On samples where no bloodstain was present, including the blank and the sample that used water in lieu of blood, nothing was detected at any stage of photography using either method. Furthermore, when a bloodstain had dried and was cleaned off, no residual bloodstain or artifacts were detected visually or quantitatively using image analysis after any paint was applied.

Diluted blood decreased the ability of IR to detect the latent bloodstains. Figure 9 visually compares the IR L0 photograph

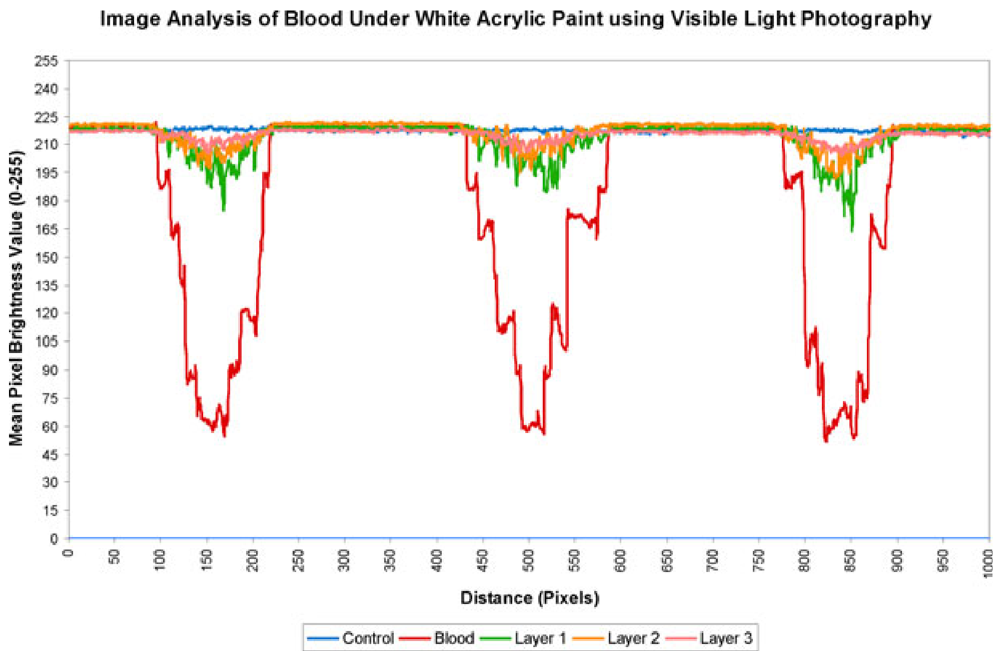

FIG. 5—*Image analysis results (mean pixel brightness value) of paint layers C0, L0, L1, L2 and L3 for white acrylic paint using standard visible light photography (VIS). Bloodstains are located on the 150, 500, and 850 pixel distances and results either side of these markers are the background.*

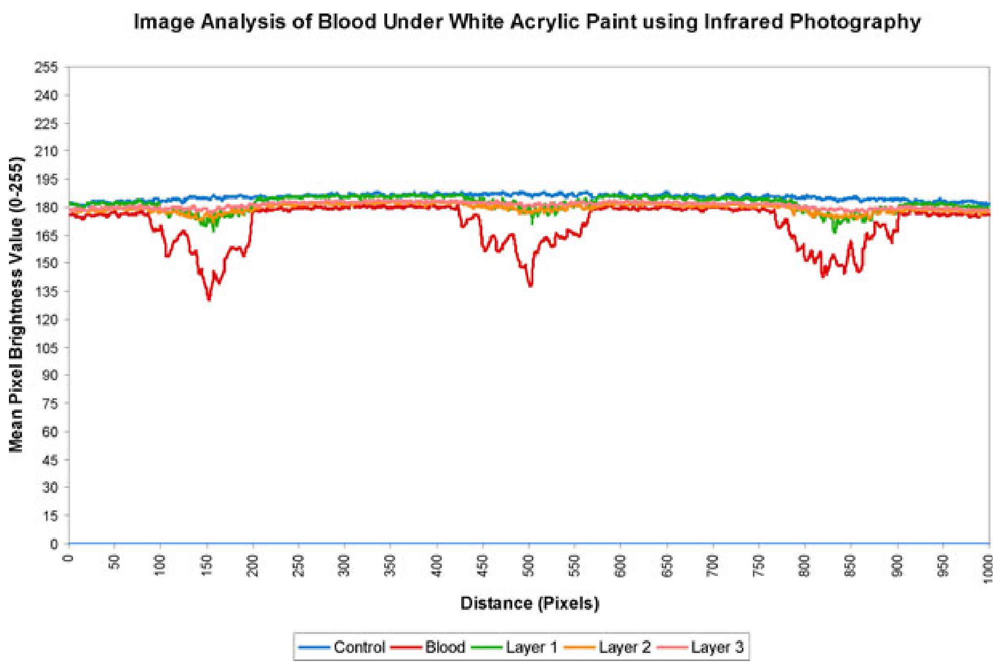

FIG. 6—*Image analysis results (mean pixel brightness value) of paint layers C0, L0, L1, L2, and L3 for white acrylic paint using infrared (IR) photography.*

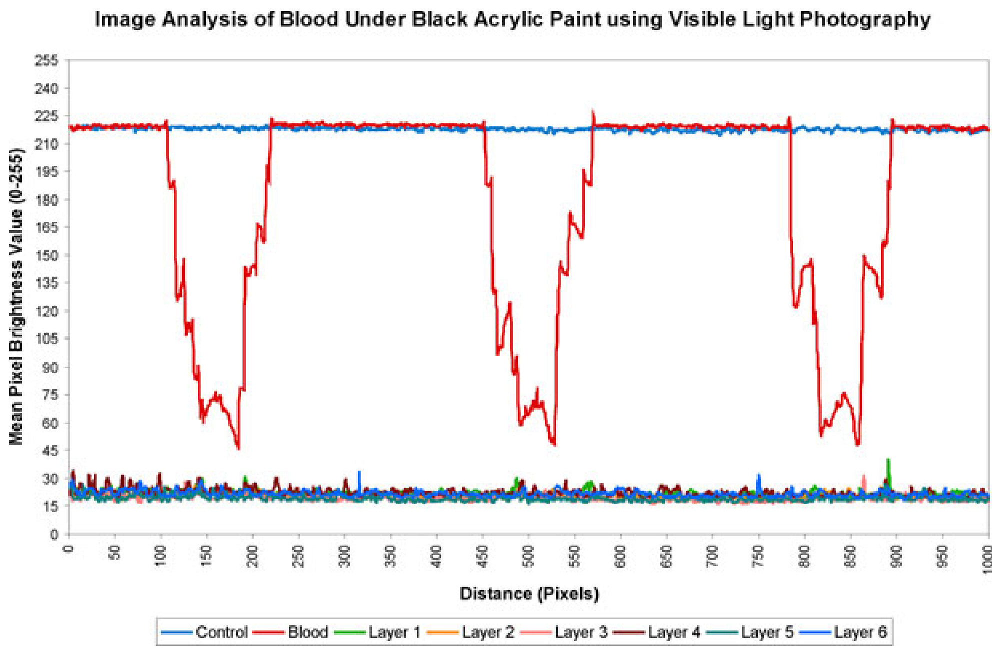

FIG. 7—*Image analysis results (mean pixel brightness value) of paint layers C0, L0, L1, L2, L3, L4, L5, and L6 for black acrylic paint using standard visible light photography (VIS).*

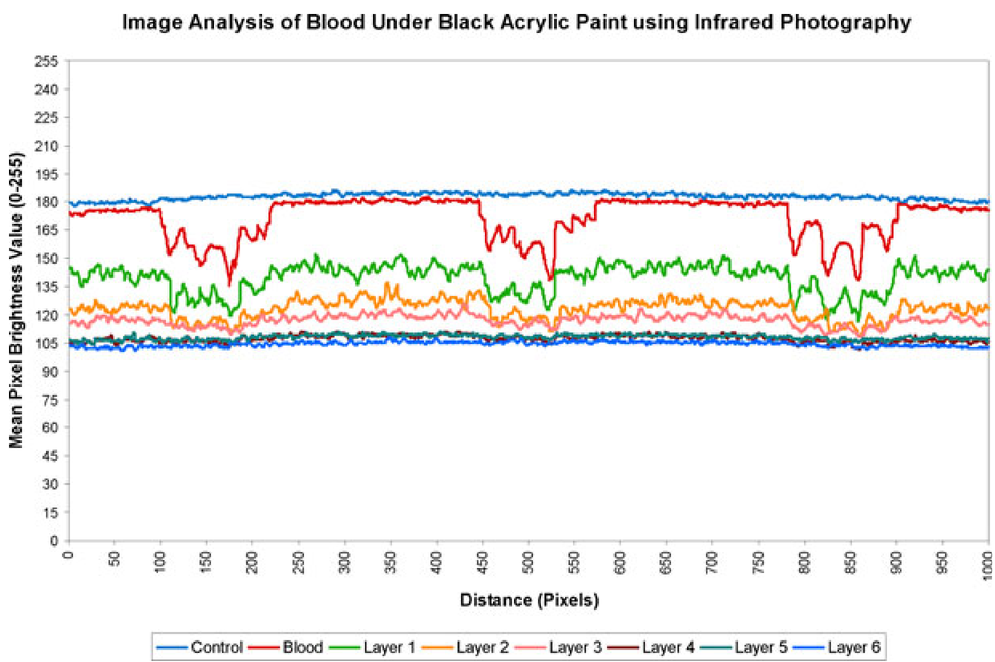

FIG. 8—*Image analysis results (mean pixel brightness value) of paint layers C0, L0, L1, L2, L3, L4, L5, and L6 for black acrylic paint using infrared (IR) photography.*

taken of each of the dilutions. As demonstrated, diluting the blood to 1/100th and 1/1000th greatly impaired visualization when using IR. After a single coat of paint, both bloodstains were concealed. In cases where blood was diluted greater than 1/10th of its original concentration, the bloodstain was more visible when photographed using standard photography in comparison with images taken using reflected IR.

The absence of an undercoat decreased the success of the technique. Two coats of paint completely concealed the bloodstain and no difference in visualization was detected between images using IR and visible light photography. A black undercoat did not affect results when white paint was used as the overcoat. However, the black undercoat with a black overcoat decreased the number of layers that IR could visualize the bloodstains.

Discussion

The aim of this study was to investigate whether bloodstains can be detected or visualized beneath layers of paint when photographed using reflected IR. As stated, within a forensic

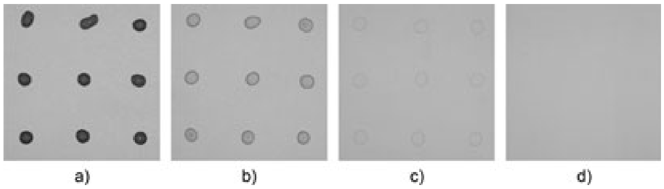

FIG. 9—*Visual comparison of (a) undiluted blood, (b) diluted blood (1/10th), (c) diluted blood (1/100th), and (d) diluted blood (1/1000th). Images taken using infrared (IR) photography.*

context, IR has been used for other types of bloodstain analysis in cases where the blood was difficult to detect because of the lack of contrast between the bloodstain and the surface where it was deposited (18–20). The study illustrated in this paper is somewhat different to previous research. The bloodstains are not sitting on the surface of the substrate but beneath layers of paint. This means that the bloodstain is concealed when viewed under visible light or when photographed using standard photography. The various optical enhancement parameters that apply to IR used in this study are

- *The spectral transmission of the paint*: Because the bloodstains are located beneath layers of paint, the paint layers must be capable of transmitting IR radiation and result in the paint becoming transparent to IR wavelengths. Variations in the chemical composition of the paint, paint color, and thickness of the layers will also alter the spectral transmission qualities of the overlaying paint.
- *The spectral absorption of blood and reflectance of the background*: To record a distinction between the bloodstains and the background, the blood must absorb IR while the background must do the opposite and reflect IR. This difference between the spectral response of the bloodstains and the background is critical. The concentration of a bloodstain also alters the IR absorption properties of blood.
- *The spectral sensitivity of the camera*: The camera must be sensitive to IR radiation. It was necessary to modify a standard digital SLR camera to achieve spectral sensitivity in the IR spectrum. Most digital SLR cameras are not sensitive to this region because an IR blocking filter is installed over the sensor.
- *The spectral transmission of the camera filter*: An IR transmission filter is required to eliminate all visible light and exclusively record wavelengths featuring components of the IR spectrum. An IR transmission filter was installed directly onto the camera's sensor during its modification to allow the exclusive recording of IR radiation.
- *The spectral distribution of the light source*: The light source used for IR must contain components of IR. Tungsten halogen lamps were used throughout this project to illuminate the specimens.

The paints selected for this study represent a small sample of conditions that painted-over bloodstains could be created or concealed beneath. These included using three different types of white paint, colored paints, and introducing variables such as diluting the blood to determine how this would affect the efficiency of IR. The paints selected were chosen because they were among some of the most commonly sold paint and available at local hardware stores. It was not considered viable during this study to test all available paints and colors due to the immense number available.

Of the three types of white paints selected, it was found that white oil-based paint was the most effective in concealing bloodstains when using standard photography, however, it was found that each of the white paints appeared to be very effective in concealing blood, even when photographed using IR. The variation of results achieved from standard visible light photography may be attributed to the chemical make-up of the individual types of paint. The chemical differences between the three paint types may inhibit visible light and IR from penetrating the uppermost layers, thus concealing bloodstains more effectively.

Colored paints were more successful in allowing the detection of bloodstains using IR. Black acrylic paint exhibited the greatest difference when comparing both photographic methods. A single layer of black paint was enough to conceal bloodstains under standard photography, however, even after six layers IR was still able to detect blood.

Purple acrylic paint also produced a strong result, whereas green acrylic inhibited IR penetration. Vandenberg and Oorschot (5) also found green paint to effectively conceal blood, even when viewed using monochromatic light with a central bandwidth of 415 nm. The variation in results among the colored paints indicate that pigment may also play an important role in the effectiveness of whether or not IR can detect a painted-over bloodstain. This further supports the conclusions proposed by Wise (13) who stated that certain pigments allowed greater IR penetration than others did when using IR.

Diluted blood decreased the efficiency of IR to detect a painted-over bloodstain. Undiluted blood was visible after six layers of paint had been applied using both photographic methods but this was attributable to the topographical nature of the sample. By diluting the blood to 1/10th of its original concentration as well as 1/100th and 1/1000th respectively, the level of IR absorption was noticeably weaker than undiluted blood. Diluted blood caused the blood drops to appear lighter. This was most likely due to lower concentrations of hemoglobin molecules being present in the diluted blood and therefore less IR absorption occurred. The results from the diluted samples may represent scenarios where attempts to remove bloodstains have been made and leaving only residual or diluted traces of blood. Water drops were not detectable using standard or IR before or after any stage of the painting process.

Results indicate that the technique worked more effectively on samples where an undercoat was present beneath the bloodstain. The sample without an undercoat caused the blood to appear with less clarity in comparison with samples where an undercoat was applied. An undercoat may provide a more reflective surface and increasing the effectiveness of IR and enhancing the contrast between the bloodstain (IR absorption) and the background (IR reflectance).

In photographs taken using IR where the paint still effectively concealed the bloodstain, it was possible that (i) the IR radiation emitted from the tungsten halogen lamps was not strong enough to penetrate particular types of paints, (ii) the layer or layers were too thick to allow IR radiation to successfully transmit through the paint, (iii) a particular chemical constituent in the paint such as the pigment or binder was inhibiting IR penetration, or (iv) a combination of all three reasons. This supports the proposed conditions provided by Mairinger (12) under which the transparency of a paint layer depends on the interaction between IR radiation and the physical dimensions of the layers of paint.

By utilizing both visual comparative examination and image analysis results could be verified. By completing a quantitative analysis, controls were used to ensure that all images were photographed under the same conditions. Controls also ensured samples were exposed to the same amount of light consistently at every stage of the photographic process. However, image analysis was only completed for the control layer, blood layer, and first three layers of paint (C0, L0, L1, L2, and L3). The photographs taken for L4 through to L6 were not processed through ImageJ and were deemed unnecessary. Image analysis plots that contained the first three layers of paint were considered sufficient to demonstrate the success of this technique; however, all photographs were still visually examined and were included in the overall results presented in Fig. 2. The only exception was for black acrylic paint where photographs for L4, L5, and L6 were also analyzed.

Conclusion

Results from this study indicate that bloodstains are capable of being detected beneath layers of paint using reflected IR. By employing qualitative analysis that involved visually examining photographs using both IR and visible light photography, the result indicates that IR could detect bloodstains beneath up to six layers of paint. These results were further supported by a quantitative method that used image analysis to determine the mean pixel brightness of the images and demonstrate the effects multiple layers of paint had on the clarity of bloodstains. Colored paints gave the most successful results in allowing IR to detect painted-over bloodstains. Black paint exhibited the greatest difference between the two recording techniques, and bloodstains were successfully detected after six layers of paint had been applied.

In instances where the blood remained visible following the application of multiple layers of paint, IR increased the contrast between the background and the blood with improved visualization. The application of multiple layers of paint gradually decreased the clarity at which bloodstains could be detected using IR. Furthermore, diluting the blood was found to greatly decrease the level at which IR could detect bloodstains.

This study demonstrates that IR was successful in detecting bloodstains located beneath a number of layers of paint depending on various optical and physical parameters. These parameters include (i) the spectral transmission of the paint, (ii) the physical dimensions of the paint and the particles contained within the paint layers, (iii) variations in the spectral absorption of blood and the spectral reflectance of the background, and (iv) the IR spectral properties of the photographic recording system.

References

1. Barni F, Lewis S, Berti A, Miskelly G, Lago G. Forensic application of the luminol reaction as a presumptive test for latent blood detection. Talanta 2007;72(3):896–913.
2. Miskelly GM, Wagner JH. Using spectral information in forensic imaging. Forensic Sci Int 2005;115(2–3):112–8.
3. Thorogate R, Moreira J, Jickells S, Miele M, Daniel B. A novel fluorescence based method in forensic science for the detection of blood in situ. Forensic Sci Int Genet 2008;2(4):363–71.
4. Bevel T, Gardner RM. Bloodstain pattern analysis with an introduction to crime scene reconstruction, 3rd edn. Boca Raton, FL: CRC Press, 2008.
5. Vandenberg N, Oorschot RA. The use of Polilight® in the detection of seminal fluid, saliva and bloodstains and comparison with conventional chemical-based screening tests. J Forensic Sci 2006;51(2):361–70.
6. Robinson E. Crime scene photography. Burlington, MA: Academic Press, 2007.
7. Bily C, Maldonado H. The application of luminol to bloodstains concealed by multiple layers of paint. J Forensic Identi 2006;56(6):896–905.
8. Quickenden TI, Creamer JI. A study of common interferences with the forensic luminol test for blood. J Biolumin Chemilumin 2001;16 (4):295–8.
9. Seracini M. Diagnostic investigations on the adoration of the magi by Leonardo Da Vinci. In: Gauluzzi P, editor. Mind of Leonardo—the universal genius at work. Florence, Italy: Giunti, 2006;94–101.
10. Porter G. Photography: marks impressions and documents. In: Jamieson A, Moenssens A, editors. Wiley encyclopedia of forensic science. New York, NY: Wiley & Sons, 2009;2036–57.
11. Verhoeven G. Imaging the invisible using modified digital still cameras for straightforward and low-cost archaeological near-infrared photography. J Archaeol Sci 2008;35(12):3087–100.
12. Mairinger F. The infrared examination of paintings. In: Creagh DC, Bradley DA, editors. Radiation in art and archeometry. New York, NY: Elsevier Science, 2000;40–55.
13. Wise D. Through a glass, darkly: practical applications of infrared photography. Melb J Tech Stud Art 2005;2(6):85–94.
14. Wright FD. Photography in bite mark and patterned injury document. J Forensic Sci 1998;43(4):877–80.
15. Ellen DM, Creer KE. Infrared luminescence in the examination of documents. J Forensic Sci 1970;10(3):159–64.
16. Bailey J. Digital infrared photography to develop GSR patterns. Aust J Forensic Sci 2007;39(1):33–40.
17. McKechnie ML, Porter G, Langlois N. The detection of latent residue tattoo ink pigments in skin using invisible radiation photography. Aust J Forensic Sci 2008;40(1):65–72.
18. Raymond MA, Hall RL. An interesting application of infra-red reflection photography to blood splash pattern interpretation. Forensic Sci Int 1986;31(3):189–94.
19. Perkins M. The application of infrared photography in bloodstain pattern documentation of clothing. J Forensic Identi 2005;55(1):1–9.
20. Chun-Yen Lin A, Hsieh H, Tsai L, Linacre A, Lee JC. Forensic applications of infrared imaging for the detection and recording of latent evidence. J Forensic Sci 2007;52(5):1148–50.

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