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Simultaneous detection and image capture of biological evidence using a combined 360° camera system with single wavelength laser illumination

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Abstract

Forensic investigators frequently utilise light sources to detect and presumptively identify biological evidence. The instrumentation typically deploys single or multiple wavelength exposures at various intensities, which interact with constituents of biological material, initiating fluorescence or improving contrast between the material and substrate. Documentation using sketches and/or photographic approaches follows detection, which are essential for scene reconstruction. Recent research has demonstrated the simultaneous detection and capture of biological evidence using a 360° camera system combined with an alternate light source exhibiting broad wavelength ranges of light. Single wavelength light sources reportedly offer enhanced sensitivity, due to the increased light intensity and narrower bandwidth of light, although their combined use with a 360° camera system has not yet been explored.

Samples of human blood, semen, saliva, and latent fingerprints were deposited on to a variety of substrates. A 360° camera system combined with a laser light source was used to detect and capture the samples. Ten participants were asked to detect the samples on images of the substrates without ground truth knowledge. It was possible to detect and capture biological evidence, although success varied according to substrate colour and light intensity. Advantageously, presumptive screening for biological fluids and the simultaneous location and visualisation of such evidence as part of a 360° panorama of the scene for contextual purposes was permitted. There was no fluorescent response from the fingerprints, although the oblique lighting effects appeared sufficient to aid mark detection in some circumstances. The use of single wavelength illumination clearly facilitates identification of a range of forensically important material. When coupled with a 360-degree camera, this allows for simultaneous identification and recording of such evidence in the context of the whole environment.

Keywords: *Laser light source; biological evidence; detection; 360-degree photography; high dynamic range; fluorescence*

Highlights:

- 360° camera adapted using Laser Light Source to visualise biological fluids
- At higher intensity outputs the opportunity to locate semen and saliva was improved
- Laser light source was generally ineffective at detecting latent fingerprints
- Preliminary results demonstrate successful proof of concept for combined method
- Method allows more dynamic recording of spatial placements of biological fluids

1. Introduction

Biological evidence, including blood, semen, saliva, vaginal secretions, urine and fingermarks are commonly encountered evidence types which can be found at crime scenes. As part of crime scene investigations it is necessary to determine and document the location and distribution of such evidence within the crime scene, as this can provide valuable intelligence information as to what may have occurred at the scene through reconstructing a sequence of events. The detection of biological evidence can also facilitate the identification of persons present at the scene, through DNA or the examination of friction ridge detail. It can be difficult to detect biological evidence on account of the state of the evidence 'in situ'. This may include, the quantity and/or concentration of the biological fluid, the nature of the substrate, or the properties associated with the evidence type. For example, saliva may be less obvious than blood on lighter coloured substrates.

In order to detect biological evidence of this nature, Forensic Investigators (FIs) can utilise filtered light analysis using alternate light sources (ALS). They offer simple and non-invasive methods for the enhancement and presumptive testing of such evidence [1-4]. An ALS typically allows the selection of different wavelengths of light (~300 nm – 900 nm) to help visualise evidence otherwise invisible to the naked eye, based on the response received from the object of interest. An ALS can consist of a light emitting diode (LED) or a laser light source [5]. LED sources often exhibit a range of wavelengths simultaneously, whereas laser light sources often exhibit a single wavelength of light [6, 7]. Tuneable light sources allow a FI to select the wavelength of light required to induce fluorescence from the target evidence and change barrier filters to enhance the evidence against its background [8]. Different wavelengths and barrier filters can be combined to create the optimum combination that will allow the biological evidence to be enhanced and visualised [8]. It is also possible to detect multiple sources of biological evidence using the same lighting conditions.

The intensity of the light output of ALS's varies between models and manufacturers. The intensity of the light is important because it can affect the operational use of the instrument in terms of health and safety, the effectiveness of the instrument's ability to detect evidence, and the cost of instrument purchase. The intensity output of the instrument may also affect the distance with which the operator is required to stand during its operation. For example, a limited power output may require the operator to be positioned closer to the item being examined. Should evidence be detected, the FI would be required to document the evidence using photographic methods. In this instance, the camera would also need to be positioned appropriately to effectively capture the evidence. Research into the feasibility of lasers as alternate methods for detecting biological fluids has provided laser systems which are lower cost and more portable than their earlier heavy and cumbersome counterparts [6]. In recent years their reduced cost and size coupled with their ability to examine larger areas within crime scenes has seen a resurgence in interest by the forensic community.

Previous research has demonstrated that laser light sources are reportedly more effective in the detection of biological fluids than LED based light sources, due to narrower bandwidths exhibited by the laser, which can effectively target the evidence under examination, and the increased levels of intensity offered [9]. Intensity is used to define the quantity of light that is emitted from the source. Research conducted by Auvdel [6] evaluated the detection capabilities of laser and UV methods for detecting biological fluids. The research demonstrated that the laser was shown to be a more effective method for detecting biological stains than the UV method. However, Auvdel [6] stated that a higher success rate for the detection of biological stains was achieved through a combination of both laser and UV light sources. In contrast, James *et al.* [7] found that alternate light sources such as high intensity lights and laser lights were comparable in their ability to detect biological fluids, and that the choice of which to adopt is based upon costs, portability and ease of use, for example.

Following detection and visualisation of biological evidence it must be extensively documented to demonstrate its location and distribution within the environment in context to its surroundings. Digital photography is commonly utilised to document biological evidence 'in situ'. In order to successfully

capture a fluorescent response from the target biological evidence, fluorescence filters can be fitted over the digital cameras existing lens to block the excitation wavelengths of light.

As an alternative to standard digital photography, 360° camera systems may be used as part of crime scene documentation. Such systems reportedly aid the documentation process by reducing the risk of failing to document evidence present at the scene, reducing contamination via persons present at the scene, and allowing post photographic documentation, such as measuring and further examination. Previous research by Sheppard *et al.* [10] has successfully combined the use of a 360° camera system with an LED based light source for simultaneous detection and capture of biological evidence. This type of examination may offer rapid screening opportunities to the investigator, with real time information available for use, and the opportunity for data relating to spatial relationships between evidence (in this case biological fluids), to be effectively captured. Given the reported advantages associated with the use of laser based light sources, the research team were interested in establishing whether a similar approach could be effectively used to detect and capture biological evidence. Similarly, given the high intensity output of the laser light source, its capability for the detection of latent fingerprints was also of interest to the team, which has not been previously considered.

2. Method

2.1 Biological fluid preparation and deposition

In line with ethical requirements of the host institution and in accordance with health and safety procedures, human semen was obtained from one male donor, aged 26. Human saliva was obtained from a female donor aged 24 and human blood was obtained from a male donor, aged 52. Samples of semen and saliva were collected into separate 100 ml Thermo Scientific™ Sterilin™ Polystyrene Containers and labelled accordingly. All biological fluid samples were collected on the morning of the study and were immediately stored in a fridge at 3°C for 1 hour. White cotton, dark blue cotton and coloured cardboard (160g/m²; red, orange, yellow, green, blue and violet in colour) were utilised as the substrates for fluid deposition and were cut into approximate 5 cm x 5cm square swatches.

Using Biohit Proline® automated pipettes, 5 and 250 µL of semen and saliva were deposited onto each substrate type. The pipette was held directly above the substrate and the biological fluid deposited at a 90° angle to the substrate. A single drop of each biological fluid was deposited onto each swatch. An Accucheck lancet was used to prick the donor's finger and a droplet of blood was applied to each of the substrates. Samples were left to dry under ambient conditions (approximately 18°C) for 24 hours prior analysis.

2.2 Fingerprint deposition

Four participants (2 male, 2 female) of various ages deposited five consecutive latent fingerprints onto white ceramic tile, white photocopier paper, window glass, and white textured plastic. Participants were asked to refrain from washing their hands in the hour preceding deposition, and to rub their fingers together immediately prior to deposition, in order to distribute the friction ridge residue. Participants were asked to contact the distal phalanx of each finger onto the substrate, without formal control of force, surface area, angle or contact time. Different fingers were used for each substrate.

2.3 Examination of biological fluid and fingerprint samples using filtered light analysis

A specialist 'trauma room' at the host institution was utilised for all investigations as it provided an environment, which limited contamination from other biological fluids, and allowed for examination in complete darkness. Walls in this room were covered with lining paper to remove the reflectivity and to ensure that the walls were more representative of common household environments. All swatches were adhered to the wall lining paper using double-sided sticky tape, in the approximate centre of one wall. Fingerprint samples were examined horizontally on a table due to the mass of the samples (the substrates were too heavy).

The environment was illuminated using a TBL Mini laser (Figure 1, Tech-Long Industry Ltd.) at each of the following monochromatic wavelengths of light: 405nm, 445 nm and 532 nm. During exposure the biological samples were photographed using a SceneCam 360° camera (Figure 2, Spheron VR AG). The TBL Mini laser was positioned adjacent to the SceneCam, as shown in Figure 3. This was to allow for a complete 360° rotation of the camera, and to avoid the light from the laser being obstructed by the tripod. The camera was initially positioned 150 cm away from the swatches and the fingerprint samples. For the 405 nm (+/- 1 nm) and 445 nm (+/- 1 nm) wavelengths, a 495 nm (GG495) longpass camera filter (62 mm) was adhered over the existing fisheye lens on the 360° camera using Duct tape™ to allow induced fluorescence to be observed. For the 532 nm (+/- 1 nm) laser wavelength, a 580 nm (NoIR Laser Shields) longpass camera filter (72 mm) replaced the 495 nm filter in accordance with best practice guidelines [11]. In addition, three different intensities were examined; referred to further as low, medium and high. The camera was then calibrated according to the manufacturer's instructions and a 360° panorama was taken at the maximum resolution of 50 megapixels (Spheron SceneCam User Manual, 2007). The fingerprints were photographed using a DSLR camera of 24 megapixels (MP) with appropriate filters attached, as described for the 360° camera method.



Figure 1: TBL Mini Laser (Tech-Long Industry Ltd.)



Figure 2: SceneCam 360-degree camera (Spheron VR AG.)

The panoramas were uploaded to the complimentary SceneCenter software as .sph files. The fingerprint photographs were viewed as JPEG files. Each panorama/photograph was visually examined to determine whether the biological samples or fingerprints were present at each of the conditions described.

2.4 An investigation to determine the accuracy of detection of biological fluids and fingerprint samples using a triple mini laser

Ten participants were provided with 8 x 5 grids, which were designed to replicate the position of the swatches on the wall during examination. Participants were asked to examine the panoramas and to reproduce the distribution and appearance of any staining that they were able to detect onto the grids. No information as to the number or location of the existing droplets was provided. For the fingerprint samples, 2 x 2 grids were provided, and participants were asked to indicate which areas of the grid contained any friction ridge detail. All panoramas and JPEG images were examined on the same computer and screen. The computer monitor had a 15.4" colour display with 2880 x 1800 native resolution at 220 ppi.



Figure 3: TBL Mini laser positioned adjacent to the SceneCam

3. Results and Discussion

Table 1 demonstrates the combined percentage scores for all ten participants for blood detection on all substrates. The results show that it was generally possible to simultaneously detect and photograph blood at all wavelengths and intensity outputs for the red, orange and yellow cardboard, and white cotton substrates using a laser light source combined with a 360° panoramic camera system. A few participants did not detect blood in some instances (2 out of 10 participants).

No participant was able to detect blood on the green cardboard or dark blue cotton substrates. On the blue and purple cardboard substrates, successful detection of blood was possible at higher intensities of light. As the wavelength increased, some participants were also able to detect blood at low and medium intensity outputs.

Table 1: Percentage scores for blood detection

Substrate	Wavelength								
	405 nm			445 nm			532 nm		
	Low	Medium	High	Low	Medium	High	Low	Medium	High
Red Cardboard	80 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Orange Cardboard	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Yellow Cardboard	100 %	100 %	100 %	100 %	100 %	90 %	90 %	100 %	90 %
Green Cardboard	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %
Blue Cardboard	0 %	0 %	0 %	0 %	40 %	90 %	0 %	0 %	0 %
Purple Cardboard	0 %	0 %	80 %	0 %	60 %	90 %	20 %	20 %	50 %
White Cotton	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Dark Blue Cotton	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %

The detection of blood on the lighter substrates was unsurprising given that these substrates offered sufficient contrast to permit visualisation. On these substrates it was possible to detect blood with or without the light source, as shown in figure 4. This was also evident for the remaining substrates, with the exception of the dark blue cotton, where the use of a light source was considered to be detrimental to the detection of blood. The authors recognise that the use of a light source is not necessarily essential for the detection of blood, given that in contrast to semen and saliva, blood absorbs radiation rather than reflects radiation, and so does not produce a fluorescent response. Given the fluorescent properties of semen and saliva, filtered light analysis for their detection is considered to be essential. What these results demonstrate is that during routine substrate examination for biological fluids, it is possible to detect and capture blood in addition to alternative fluids using this approach in some circumstances. As it is possible for blood to be enhanced using fluorescent reagents such as fluorescein, it is entirely possible that on the darker substrates blood would have been visible and captured as part of a 360° panorama, had such a reagent been used and exposed to the laser light source. There is evidence to suggest that infrared light can offer the user a more effective means of locating blood on darker substrates due to the increased levels of contrast between the background and the fluid [12, 13]. However, the purpose of this research was to investigate simultaneous detection of biological fluids, and their opportunities for capture using a 360° camera system, and anecdotally crime scene investigators do not use infrared for the detection of alternative biological fluids, such as semen and saliva.

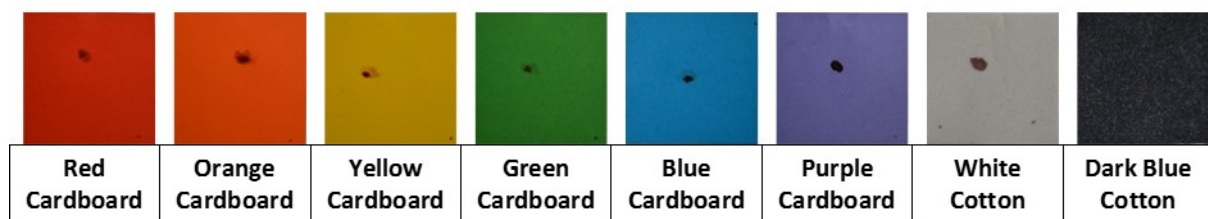


Figure 4: Blood droplets deposited onto each substrate without light source exposure.

Research has examined the effects of light sources for the detection of biological fluids on subsequent identification methods such as DNA analysis. It is critical for any subsequent DNA analysis that any methods used to locate biological fluids are non-destructive. There have been concerns that light source exposure, particularly from UV light, could have adverse effects on the already limited biological material contained within biological evidence which is/could be available for subsequent DNA analysis. Nicholson *et al.* [14] indicated that DNA analysis was most affected by long exposure times to UV light (approximately 3 minutes). Such prolonged exposure resulted in poor recovery of DNA and increased allelic dropout, which is thought to have been attributed to degradation of the biological sample [14]. High doses of UV radiation can cause structural disintegration of DNA [15]. As a result, light sources which exhibit UV wavelengths needs to be used with caution to prevent any damage to DNA within the forensic evidence [2].

The effect of any light source, which utilises wavelengths within the UV range on the biological evidence under examination, must therefore be taken into consideration. Utilising the proposed technique under study could potentially expose biological evidence to such wavelengths for prolonged periods of time. The chosen resolution of the panorama will affect the time taken to scan an environment with higher resolutions increasing the time of the scan from 30 seconds up to 12 minutes depending on the light conditions within the environment. As a result, higher resolutions will undoubtedly expose biological fluids to certain wavelengths of light for times, which could have adverse effects on the DNA within the samples.

Although on the lighter coloured substrates the blood detection rates were high, there were differences between the wavelength conditions. At the 405 nm and 445 nm wavelengths the presence of blood

was more obvious, attributed to the fluorescence of the substrates under these conditions, which increased the levels of contrast, which was not seen at the 532 nm wavelength. This observation did not affect the detection rates exhibited by the participants, although logically this is likely to affect blood detection at lower volumes and/or concentrations of blood.

The combined percentage scores for the detection of semen at volumes of 5 μ l and 250 μ l can be viewed in tables 2 and 3.

Table 2: Percentage scores for semen detection at 5 μ l

Substrate	Wavelength								
	405 nm			445 nm			532 nm		
	Low	Medium	High	Low	Medium	High	Low	Medium	High
Red Cardboard	0 %	0 %	0 %	0 %	20 %	80 %	0 %	0 %	60 %
Orange Cardboard	0 %	0 %	0 %	0 %	10 %	70 %	0 %	20 %	60 %
Yellow Cardboard	0 %	0 %	0 %	0 %	10 %	0 %	0 %	10 %	0 %
Green Cardboard	0 %	0 %	0 %	0 %	0 %	80 %	0 %	0 %	0 %
Blue Cardboard	0 %	10 %	50 %	30 %	30 %	90 %	0 %	0 %	0 %
Purple Cardboard	0 %	0 %	0 %	0 %	30 %	90 %	0 %	10 %	20 %
White Cotton	0 %	0 %	0 %	0 %	0 %	80 %	0 %	50 %	50 %
Dark Blue Cotton	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %

Table 3: Percentage scores for semen detection at 250 μ l

Substrate	Wavelength								
	405 nm			445 nm			532 nm		
	Low	Medium	High	Low	Medium	High	Low	Medium	High
Red Cardboard	90 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Orange Cardboard	90 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Yellow Cardboard	80 %	100 %	100 %	100 %	90 %	90 %	100 %	100 %	100 %
Green Cardboard	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Blue Cardboard	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Purple Cardboard	100 %	100 %	100 %	100 %	100 %	100 %	100 %	90 %	100 %
White Cotton	0 %	0 %	0 %	100 %	0 %	90 %	100 %	100 %	100 %
Dark Blue Cotton	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %

The results clearly demonstrate that the rate of semen detection and capture using the 360° camera system increased in the presence of a higher volume of semen. At higher intensity outputs the opportunity to locate semen was improved. This was to be expected, but would highlight the need for higher intensity light exposure to detect lower volumes of semen. As the distance between the apparatus and substrates was fixed, it is not possible to confirm how the results might have differed had the conditions been changed. However, previous research has demonstrated that when the distance between a light source/camera combination and a biologically contaminated substrate was increased to 3 metres, detection and capture was still possible with lighter coloured substrates at 5 μ l [10]. This study involved the use of an LED based light source, with no capacity to increase intensity

output, and the light was emitted in the desired emission wavelength range. On this basis, it is logical to assume that a single wavelength with the ability to increase intensity output would facilitate a more effective means of detecting biological fluids at an increased distance from the stain. Likewise, if the laser light source was positioned at a closer distance to the stain it would be possible to reduce intensity, to allow effective detection and capture of the stain.

Although not all of the participants were able to detect all droplets of semen on the yellow cardboard substrate, these results were considered to be anomalies, given that the majority of participants did detect the biological fluid at all wavelengths and intensity outputs. With the white cotton substrate the results would suggest that the higher wavelengths of light and intensity's significantly increased participants' ability to detect semen within the panorama. This was surprising given that published literature has suggested that semen fluoresces more effectively at those wavelengths in the region of 400 nm - 450 nm [16, 17]. Many white textiles, such as the white cotton used in this study, contain optical brighteners which naturally fluoresce and as a result can mask any fluorescence from a target biological fluid [6, 17, 18] making it difficult to detect. An examination of the white cotton substrate exposure under all of the lighting conditions would suggest that the detection and capture of semen had been assisted with the reduction in background substrate fluorescence at 532 nm, increasing the contrast. These results are shown in figure 5.





















	Normal Light	Wavelength and Intensity								
		405 nm			445 nm			532 nm		
		Low	Medium	High	Low	Medium	High	Low	Medium	High
5 μ L										
250 μ L										

Figure 5: Semen on white cotton at 5 μ L and 250 μ L exposed to different wavelengths and intensities of light

As with the blood, the quantity of semen present on the substrates affected the ability to visualise semen without a light source. These results can be viewed in figure 6. As expected, with the higher quantity of semen, its visibility was increased on all substrates except for white cotton, where contrast was highly limited. Without a light source, presumptive test information to indicate the source of the staining is absent.








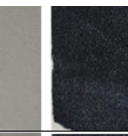



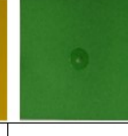
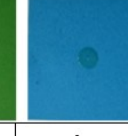
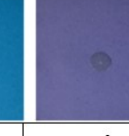

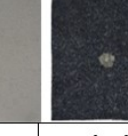
5 μ L								
250 μ L								
	Red Cardboard	Orange Cardboard	Yellow Cardboard	Green Cardboard	Blue Cardboard	Purple Cardboard	White Cotton	Dark Blue Cotton

Figure 6: Semen deposited onto each substrate without light source exposure.

The results for the participant detection of saliva at volumes of 5 μ L and 250 μ L can be viewed in tables 4 and 5.

Table 4: Percentage scores for saliva detection at 5 μ l

Substrate	Wavelength								
	405 nm			445 nm			532 nm		
	Low	Medium	High	Low	Medium	High	Low	Medium	High
Red Cardboard	0 %	0 %	0 %	0 %	0 %	20 %	0 %	0 %	10 %
Orange Cardboard	0 %	0 %	0 %	0 %	0 %	50 %	0 %	0 %	0 %
Yellow Cardboard	0 %	30 %	0 %	0 %	10 %	10 %	0 %	0 %	0 %
Green Cardboard	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %
Blue Cardboard	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %
Purple Cardboard	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %
White Cotton	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %
Dark Blue Cotton	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %	0 %

Table 5: Percentage scores for saliva detection at 250 μ l

Substrate	Wavelength								
	405 nm			445 nm			532 nm		
	Low	Medium	High	Low	Medium	High	Low	Medium	High
Red Cardboard	80 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	90 %
Orange Cardboard	90 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %	100 %
Yellow Cardboard	90 %	100 %	100 %	70 %	90 %	70 %	0 %	0 %	10 %
Green Cardboard	0 %	10 %	0 %	0 %	0 %	20 %	0 %	0 %	50 %
Blue Cardboard	0 %	0 %	0 %	0 %	0 %	70 %	0 %	10 %	40 %
Purple Cardboard	0 %	0 %	10 %	0 %	0 %	80 %	0 %	20 %	40 %
White Cotton	0 %	0 %	0 %	0 %	0 %	10 %	10 %	0 %	0 %
Dark Blue Cotton	10 %	0 %	60 %	0 %	10 %	90 %	0 %	0 %	0 %

According to these results it is difficult to detect and capture saliva stains at 5 μ l volumes. When the volume of saliva was increased to 250 μ l saliva was generally more difficult to detect and capture on the darker substrates and white cotton. Camilleri *et al.* [19] found that the optimal contrast for the visualisation of saliva stains on white cotton surfaces was achieved through an excitation wavelength of 470 nm using interference goggles of 555 nm when using a Polilight. In their study fabric type was found to have no influence on the detection of saliva stains using the Polilight. The type and colour of materials and their designs can influence the detection of biological evidence [20] and can obscure fluorescence of any biological staining present [19]. Many white textiles, such as the white cotton used in this study, contain optical brighteners which naturally fluoresce and as a result can mask any fluorescence from a target biological fluid [6, 17, 18] making it difficult to detect. In all circumstances

a non-detection using the laser light source cannot exclude the presence of biological evidence considering that there was biological evidence deposited in all instances [18, 20, 21].

At higher intensity outputs the opportunity to locate saliva was improved, which concurs with previously published research [19]. Auvdel [6] reported that the use of an argon ion laser (454.5 nm to 514.5 nm with an output power of 18 W) increased the detection rate of saliva by 9 % when compared to detection rates utilising UV light sources (One shortwave UV tube, 254 nm and one longwave UV tube, 366 nm.) [17]. It was believed that the increase in success rate using the laser was attributed in part to the lasers higher intensity of radiation when compared to the light source [6, 17]. This improvement was not aligned to that seen for the semen stains at higher intensity outputs. Saliva is harder to detect than semen due to the lack of solid particulates within the saliva sample, and is known to fluoresce at a lower intensity than semen [2, 19]. Interestingly, it was possible to see the saliva staining at volumes of 250 μ l without the light source on several of the darker substrates, as shown in figure 7, similarly to results found by Wawryk and Odell [22]. In a controlled study, where the origin of the staining is known this result is useful. However, the behaviour of biological staining under specific lighting conditions can be useful as part of presumptive testing.

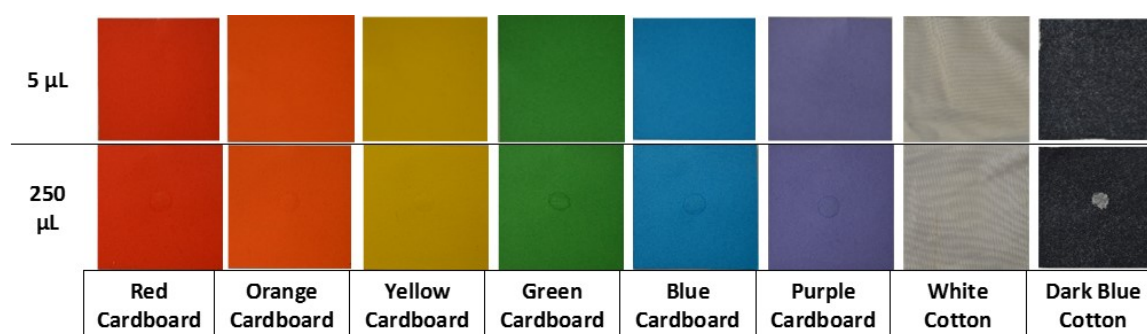


Figure 7: Saliva droplets deposited onto each substrate without light source exposure.

This investigation focused on saliva which had been donated by one participant and further work would seek to utilise more donors as it is known that saliva fluorescence can vary between different donors, as well as within the same individuals, depending on the time of day. This fluctuation is thought to be attributed to different protein contents within the saliva sample [23, 24].

As with a previous research project [10], this research was conducted within a sterile, controlled environment, which does not represent common household environment where crimes are likely to be committed. Alternative agents known to induce false positive results are documented in the literature [8, 17] and therefore it would be expected that the introduction of such agents into the existing methodology would produce similar results. In addition, it is recommended that other biological fluids be investigated, such as vaginal secretions, urine and sweat, to determine the optimum conditions for their successful location and visualisation. In addition, it would be useful to establish the sensitivity of this approach using diluted samples, which could be more reflective of casework samples on a variety of different substrates which incorporate patterned substrates. It would also be useful to monitor the effectiveness of the approach over time and this could form the basis of further work.

This research did not seek to identify the effect of distances of the laser from the biological fluid staining on the ability to successfully detect and visualise the biological staining. To avoid close range searching at crime scenes, research would need to be conducted investigating the effects of the laser light source distance from the target staining and to determine limits of its detection.

The combined results for the participant detection of fingerprints can be viewed in table 6. All samples were viewed under medium intensity. Very few fingerprints were detected on the white

photocopier paper. An examination of the substrate revealed no discernible ridge detail to identify the sample as a friction ridge skin impression, although in some regions vague outlines of marks could be seen. Despite the examination of fresh marks, it is likely that some of the residue had become absorbed into the substrate, reducing their capacity to interact with the light. Also, given the optical brighteners used to white the photocopier paper during its manufacture, the substrate itself fluoresced, masking opportunities for evidence detection. On this basis, this approach was not seen as a reliable means of detecting and capturing latent fingerprint evidence on white photocopier paper.

Table 6: Percentage scores for fingerprint detection

Substrate	Wavelength		
	405 nm	445 nm	532 nm
White Photocopier Paper	10 %	0 %	20 %
White Textured Plastic	40 %	0 %	0 %
White Ceramic Tile	50 %	30 %	100 %
Window Glass	30 %	60 %	100 %

The results suggest that at a wavelength of 405 nm four participants (40 %) detected fingerprints on the white textured plastic, although no ridge detail was present within the marks. At longer wavelengths of light this rate was reduced to 0 %.

On the white ceramic tile and glass substrates at all of the wavelengths of light some fingerprints were detected, suggesting that detection was related to substrate type. It is important to note however, that this was not due to the marks fluorescing. Detection was attributed to the effects of oblique lighting on the reflective substrates, providing an effective contrast to the marks. This is referred to by Lennard [25], who explains that constituents of latent fingerprint residue with fluorescent properties are typically found at insufficient levels to initiate a suitable fluorescent response. The detection of fingerprints on smooth and porous substrates is only found with substrates that do not themselves fluoresce. At a 532 nm excitation wavelength, all participants were able to detect marks, suggesting that under these conditions detection was most likely, even though, as mentioned, this was not due to a fluorescent response from the fingerprints.

The effectiveness of fingerprint detection was related to differences between the donors. During the creation of the samples measures were taken to ensure that a variety of donors were used according to age and gender, and the use of a depletion series was employed to provide marks of varying mass. It was clear to see that some participants simply deposited marks that were easier to detect, whereas others appeared to deposit no identifiable trace. Given that there was a lack of fluorescent response, and a lack of detection on less reflective and porous substrates, the use of the laser light source was generally ineffective at detecting latent fingerprints in these circumstances. The authors recognise that there are limitations associated with the sample size of participants and substrates under examination, but believe that combined with the biological fluid detection and capture results of this study provide a useful starting point for the use of this technology for documentation. Had the fingerprints been pre-treated with a fluorescent powder or reagent (for example, DFO on the photocopier paper), it seems likely that the laser would have been more effective. The purpose of this research however, was to study the substrates and samples in their raw states, to represent samples likely to be encountered at scenes of crime.

As this technique failed to induce a response from latent fingerprints known to be deposited, the approach has the potential to suggest false negative results to the operator, and should therefore not be relied upon as a means of fingerprint detection, particularly on less reflective or porous substrates. When the samples were examined, often the outline of their fingerprints could be seen, identifying the location of the marks but there was no ridge detail present, suggesting that this approach is unlikely to

be effective at detecting and then capturing ridge detail, and therefore further enhancement and photography to facilitate the use of the marks for human identification would certainly be required.

All substrates utilised within this investigation were plain and as a result the effects of such patterns on the detection of the biological evidence under study could not be examined. Patterned materials may obscure the fluorescence of any biological evidence [19] making them more difficult to detect and capture using a combined laser and 360° photographic system.

4. Conclusion

The aim of this research was to investigate the possibility of combining a 360° camera system with a laser light source for the detection of biological fluids and latent fingerprints on a variety of substrates used to replicate those encountered at scenes of crime. There is currently no technology available to facilitate the detection and capture of biological evidence in this manner, and therefore this innovative research has provided a useful starting point into the development of such technology. This has the potential to allow a more dynamic recording of the spatial placement of biological fluids, and allowed fluids located to be placed in context; this is considered to be a significant improvement over 'still' digital photography. This technique has presented the opportunity to presumptively screen a crime scene for human biological fluids. The study facilitates simultaneous location and visualisation of biological evidence, in addition to capturing a complete 360° view of the entire crime scene. This provides contextual information such as locations of other evidence types (e.g. footwear marks, finger marks).

For all biological fluids generally the higher the intensity of the light source the higher the detection rates were. For substrates containing whitening agents, such as white cotton, care should be taken to not 'bleach' the substrate. LED light sources do not generally allow the intensity of the light to be changed, and therefore detection rates are likely to be higher with this approach. On lighter substrates detection was generally possible across more wavelengths of light. It was also true that detection of some biological fluids was as effectively documented without the use of a laser light source, where the fluid itself offered sufficient contrast to permit its detection and capture. On darker substrates however the opposite was true, and detection relied upon the response of the fluid and/or substrate in order for effective detection. It was also considered important that the presumptive testing element of the response would be lost without the laser. The volume of biological fluid present affected the ability to detect it, particularly with semen, although with saliva the substrate itself was as influential, with more effective detection and capture evident on lighter substrates.

In terms of fingerprint detection, there was no evidence of any fluorescent response from the fingerprint samples, although for heavier fingerprints on reflective substrates the oblique lighting effects of the light source appeared to be sufficient to aid mark detection. The method of capture was insufficient to permit identification from the mark. Care should be taken with this approach given the potential for false negative results, and alternative means of enhancement are recommended.

Informed consent was obtained from all individual participants included in the study.

References

1. Mahajan, M., Zachery, K., Zhang, W., Chen, S., Mansell, J., Brackley, A., Andrews, E. (2015) Brass Board Forensic Crime Scene Survey Camera. U.S Department of Justice.
2. Virkler, K., Lednev, I.K. (2009) Analysis of body fluids for forensic purposes: from laboratory testing of non-destructive rapid confirmatory identification at a crime scene. *Forensic Science International*. 188: 1-17.
3. Lennard, C., Stoilovic, M. (2004) Application of forensic light sources at the crime scene. In: Horswell, J., *The Practice of Crime Scene Investigation*. CRC Press. Boca Raton, Florida, USA. 97-12
4. Powers, L.S., Lloyd, C.R. (2004) Method and Apparatus for detecting and imaging the presence of biological materials. U.S. Patent documents.

5. McClintock, J.J. (2014) Forensic Analysis of Biological Evidence: A Laboratory guide for Serological and DNA Typing. CRC Press. Boca Raton.
6. Auvdel, M.J. (1987) Comparison of Laser and Ultraviolet Techniques Used in the Detection of Body Secretions. *Journal of Forensic Sciences*. Jfsca. 32 (2). 326-345.
7. James, S.H., Nordby, J.J., Bell, S. (2005) Forensic Science: An Introduction to Scientific and Investigative Techniques. Second Edition. CRC Press. Taylor and Francis Group. Boca Raton, Florida, USA.
8. Marin, N., Buszka, J. (2013) Alternative Light Source Imaging. Forensic Photography Techniques. Forensic Studies for Criminal Justice. An Anderson Book. Elsevier.
9. Lee, W-C., Khoo, B-E. (2010) Forensic Light Sources for Detection of Biological Evidences in Crime Scene Investigation: A Review. *Malaysian Journal of Forensic Sciences*. 1. 17-27.
10. Sheppard, K., Cassella, J., Fieldhouse, S., King, R. (2017) The Adaptation of a 360° Camera Utilising an Alternate Light Source (ALS) for the Detection of Biological Fluids at Crime Scenes. *Science and Justice*. 57. (40) 239-249.
11. Forensic Technology Center of Excellence (2018). *Landscape Study of Alternative Light Sources*. U.S. Department of Justice, National Institute of Justice, Office of Investigative and Forensic Sciences
12. Finnis, J., Lewis, J., Davidson, A. (2013). Comparison of methods for visualising blood on dark surfaces. *Science and Justice*. 53. (2). 178-186.
13. De Forest, P.R., Bucht, R., Kammerman, F., Weinger, B., Gunderson, L. (2009) Blood on Black-Enhanced Visualisation of Bloodstains on Dark Surfaces. Research Report Submitted to the U.S Department of Justice. National Criminal Justice Reference Service (NCJRS).
14. Nicholson, J.L., Bille, T.W., Malia, M.J., Bever, R.A. (2005) The Effects of Ultraviolet Light Exposure on DNA Analysis of Fingerprints. Promega Corporation. The Bode Technology Group, Springfield, Virginia. Available at: https://www.promega.com/-/media/files/resources/conference-proceedings/ishi_16/posterabstracts/59nicholson.pdf?la=en. Date accessed: 04/05/2018.
15. Chen, H., Li, R., Li, S., Andreasson, J., Choi, J.H. (2017) Conformational Effects of UV Light on DNA Origami. *Journal of the American Chemical Society*. 139. (4). 1380-1383.
16. Stoilovic, M. (1991) Detection of semen and bloodstains using the Polilight as a light source. *Forensic Science International*. 51: 289-296.
17. Vandenberg, N., van Oorschot, R.A.H. (2006) The use of Polilight in the detection of seminal fluid, saliva, and bloodstains and comparison with conventional chemical based screening tests. *Journal of Forensic Sciences*. 51. (2) 361-370.
18. Kobus, H.J., Sileniek, E., Scharnberg, J. (2002) Improving the effectiveness of fluorescence for the detection of semen stains on fabrics. *Journal of Forensic Sciences*. 47. (4) 819-823.
19. Camilleri, E., Sileniek, E., Henry, J. (2006) Locating Saliva Stains using the Polilight® and SALIgAE® Spray. R & D Report R76. Evidence Recovery and Biology Analytical Groups. Government of South Australia. Forensic Science SA.
20. Sterzik, V., Panzer, S., Apfelbacher, M., Bohnert, M. (2016). Searching for biological traces on different materials using a forensic light source and infrared photography. *International Journal of Legal Medicine*. 130: 599-605
21. Seidl, S., Hausmann, R., Betz, P. (2008) Comparison of laser and mercury-arc lamp for the detection of body fluids on different substrates. *International Journal of Legal Medicine*. 122: 241-244.
22. Wawryk, J., Odell, M. (2005) Fluorescent identification of biological and other stains on skin by the use of alternative light sources. *Journal of Clinical Forensic Medicine*. 12. 296-301.

23. Soukos, N.S., Crowley, K., Bamberg, M.P., Gillies, R., Doukos, A.G., Evans, R., Kollias, N. (2000). A rapid method to detect dried saliva stains swabbed from human skin using fluorescence spectroscopy. *Forensic Science International*. 114. 133-138.
24. Nanda, K.D.S., Ranganathan, K., Umadevi, K.M., Joshua, E. (2011). A rapid and noninvasive method to detect dried saliva stains from human skin using fluorescent spectroscopy. *Journal of Oral and Maxillofacial Pathology*. 15. (1). 22-25.
25. Lennard, C (2001). The Detection and Enhancement of Latent Fingerprints, in: 13th Interpol Forensic Science Symposium, Lyon, France.