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Chemiluminescence video assisted by chemometrics modeling for forensic identification of blood in crime scene

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The development of advanced chemiluminescent compounds and hematology methodologies has significant implications for forensic science particularly in the detection of evidential residues at crime scenes. This study introduces a novel chemiluminescent (CL) method that utilizes a smartphone to produce digital videos from a chemiluminescent reaction with luminol (5-Amino-2,3-dihydrophthalazine-1,4-dione) reagent and blood. This innovative approach significantly reduces 6 times the reagent consumption, requiring less than 1 mL/0.01 g of sample/chemicals that agree with green chemistry principles. Blood samples used in this study were sourced from bovine liver and human subjects, being the human sample collected by forensic official forensic policve from the criminal police at crime scenes; all samples were subsequently discarded by the criminal police. The frames from a 3 min video were processed using ImageJ software and Color Grab app to generate RGB, HSV, and CMYK pattern recognition, combined with chemometrics modeling. This enabled the differentiation of samples based on positive and negative patterns, effectively preventing false results. The pattern recognition models developed were able to distinguish bovine from human blood, even after dilution, which simulated attempts to hide traces at crime scenes through washing. The method demonstrated an accuracy of 90.30% with only four prediction errors, and presented 100% sensitivity and specificity for the cotton + ceramics class, and 77.78% sensitivity and 93.10% specificity for both the wood and glass classes. Additionally, it was possible to estimate the age of the samples with a precision of 3.6 days. These results were achieved using a new data fusion strategy that facilitated the modeling of digital videos as a combination of frames to enhance model sensitivity and selectivity without increasing model complexity. These results indicate that the developed method is accurate, sensitive, and quick. Supported by these results the method represents a significant advancement in forensic science, offering a practical and efficient solution for crime scene investigations.

information available should be included here]. See DOI: 10.1039/x0xx00000x

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Supplementary material



Figure. S1 - Different substrates. (A) Glass with ketchup, (B) Wood with ketchup (C), Glass with red ink, (D) Glass containing white bubbles due to reaction of blood with luminol and (E) Wood with red ink.



Figure S2 - DIB method and analysis of the wood substrate.

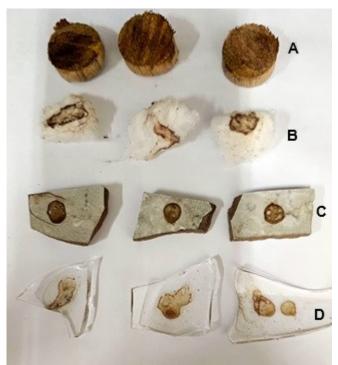


Figure S3 - Different substrates containing blood stains. (A) wood, (B) cotton, (C) ceramics and (D) glass.

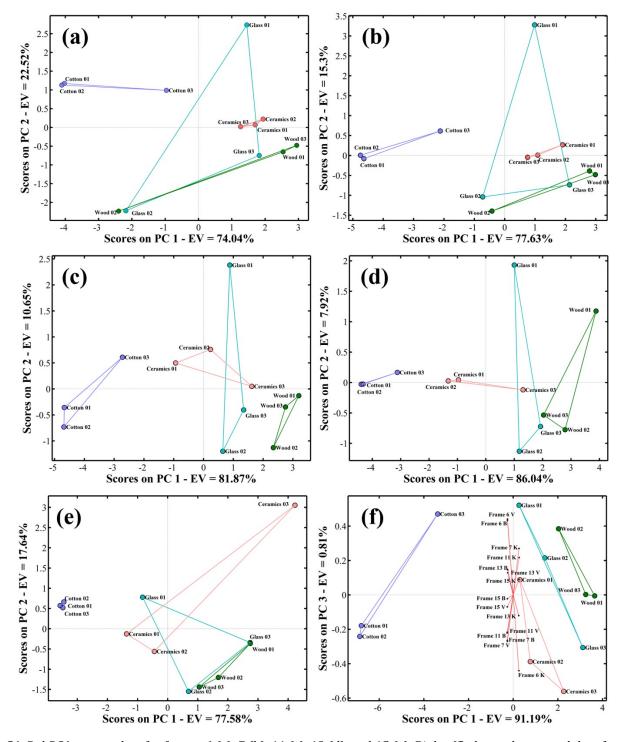


Figure S4. RobPCA scores plots for frames 6 (a), 7 (b), 11 (c), 13 (d), and 15 (e). Biplot (f) shows the merged data from all frames, calculated using the RGB, HSV, and CMYK color models, which provided the greatest differentiation among substrates. Blue, red, green, and cyan circles represent cotton, ceramics, wood, and glass, respectively. B, V, and K represent the color channels used in the analysis: B is the blue channel from the RGB color model, V is the value channel from HSV, and K is the key (black) channel from CMYK. The frames were extracted from 3 min video recordings, and only those with optimal signal quality were selected. Replicates were numbered from 01 to 03 for each substrate.

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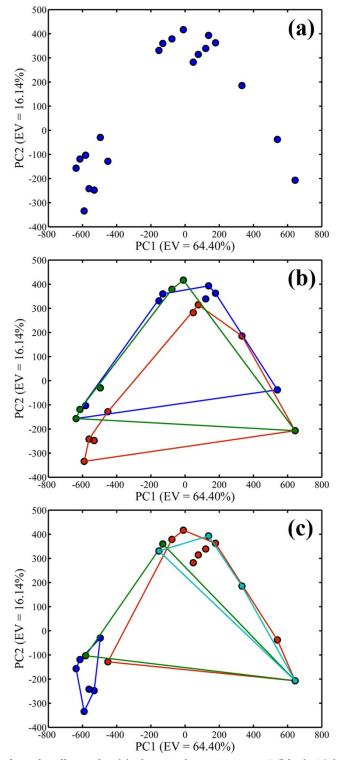


Figure S5. Principal component analysis for all samples (a), the samples separate on 7 (blue), 15 (red) and 21 (green) days with a convex hull (b) and samples for cotton (blue), ceramics (red), wood (green) and glass (cyan) with a convex hull (c).

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