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Advancing Semen Detection Techniques: Utilizing STK®Sperm Tracker to Emphasize Traces and Potentially Define Time Since Deposition (TSD)

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Recently, a new innovative technology has emerged for semen detection: STK®Sperm Tracker (AXO Science). This reagent is a non-toxic presumptive test, specifically designed to highlight male acid phosphatase (PA) present in human seminal fluid. Upon interaction with this enzyme, the reagent triggers a reaction that, after subsequent exposure to UV-365 nm lamp irradiation, enables the observation of persistent blue fluorescence.

It is crucial to test new methodologies and stay updated on the latest developments

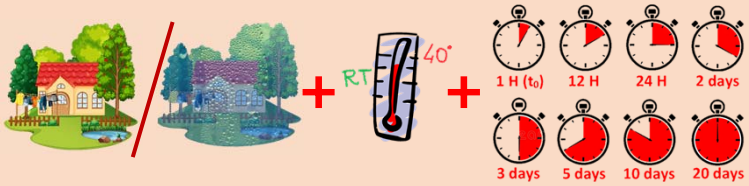
TRY NEW THINGS

Evaluate the efficiency of STK®Skin in emphasizing and detecting semen traces across various scenarios and environments

Mock samples were created using pig ears along with natural and bleached human hairs.

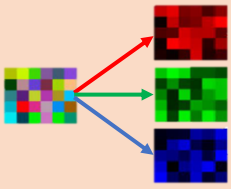
Fresh sperm was collected from at least 3 donors (to minimize interindividual variability) and a 50 µL sample was laid down in triplicate on each surface.

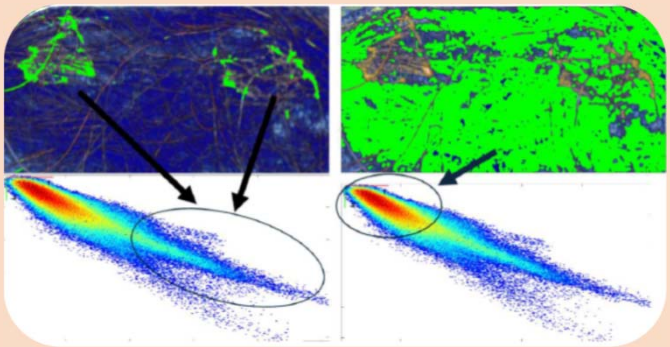
Different deposition times and environmental conditions were simulated.



The emphasized traces were then photographed in a standardized manner using VILBER VL-6.L UV lamp (365nm) *for exposure*.

Subsequently, exploratory analyses, including pixel-level and image-level approaches on RGB raw data, along with Principal Component Analysis (PCA), were conducted by computing the colorgramns on the RGB collected images.





Each graph shows the scores of PC1/PC2 and PC3 and each point representing a pixel from the original image. The color reflects pixel density, where red shows high density and blue shows low density. The PC1/PC2 scores plot effectively differentiates seminal fluid areas from the background. Background scores cluster on the left at low PC1 and PC2 values, while trace pixels form a distinct cluster on the right at high PC1 and PC2 values.

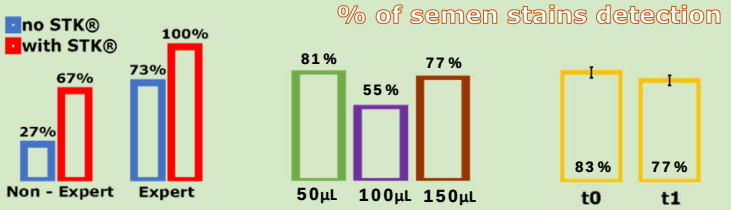
The analysis showed different results between pig ears and hair. STK® Skin was less effective on hair probably because the trace dispersed throughout the hairs, unlike with skin samples.

RGB imaging only considers color properties, which can make identifying traces treated with STK® Skin difficult if their color is too similar to the rest of the sample. More advanced imaging systems like hyperspectral imaging could be used to implement the analysis.

Evaluate the efficiency of STK®Spray in emphasizing and detecting semen traces across various scenarios and environments

Double-blind experiments were carried out: both Non-Expert and Expert operators participated to compare accuracy and sensitivity of STK®Spray in detecting traces of semen on different surfaces.

Fresh sperm samples were collected from at least 3 donors and varying volume of samples were spotted on different surface. The detection was carried out at two different times: immediately after deposition and seven days after deposition.



The STK®Spray has proven to be more efficient in semen detection compared to the examination conducted solely with Alternative Light Sources (ALS). A significant reduction in the risk of false positives associated with ALS has also been observed.

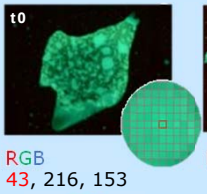
The stains' volume and the detection time since deposition do not significantly influence the detection of traces with STK®Spray. It would be interesting to further explore using minimal volumes of semen traces or mixing them with other biological fluids

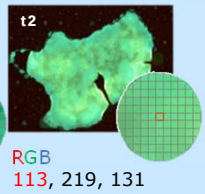
Examine variation in fluorescence intensity over Time Since Deposition

During the chemometric analysis, an exciting colorimetric change in STK® fluorescence has been revealed over TSD.

50 µL of semen were spotted (x 5) onto rigid blue-plastic supports (to ensure consistent material and color reference) and stored at room temperature. The evidences were emphasized with STK®Spray in 4 different times since deposition.

The matrix of signals was analyzed using PCA method and TSD has been revelead as a significant source of variability: in the positive PC1 cluster t0 samples are grouped while sample from t2 and t3 are grouped in the negative PC1. The loadings of PC1 reveal that the intensity of the red channel plays a crucial role: low red values result in positive loadings, clustering samples at t0 in positive PC1.





The trend was also observed in RGB values comparison: chromatic variation is due to an increase in the red index over time. This variation over time could be determined by a decrease in the activity of acid phosphatase, the target of the reagent

This would to develop a multivariate regression model that can estimate the time since deposition of a unknown trace