H111 In Situ Detection of Latent DNA Using Nucleic Acid Binding Dyes and an Alternative Light Source

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After attending this presentation, attendees will gain an appreciation of a method to detect latent DNA using nucleic acid binding dyes and its application in forensic science.

This presentation will impact the forensic science community by improving the collection of latent evidence by developing a presumptive test that detects DNA present at crime scenes, thus allowing a more targeted approach to swabbing for Short Tandem Repeat (STR) analysis.

DNA is deposited onto a surface by touch, yet few means have been developed for its *in situ* detection. Collecting touch DNA-type samples can be difficult as the DNA is not directly targeted, leading to many samples containing no DNA that leads to a waste of expensive reagents and kits. A range of dyes are available that bind to DNA at high specificity and the use of these dyes to detect latent DNA on various substrates and as a biological stain for forensic evidence is reported here. Six common nucleic acid-binding dyes were selected due to their increase in fluorescence in the presence of double-stranded DNA and their effectiveness in detecting latent DNA on surfaces was determined.

It has been suggested that epithelial cells are sloughed off the skin surface and can be transferred onto various substrates by touch, known as "touch DNA."[1] It has been postulated that these cells are keratinized and lack nuclei; the DNA present on the surface is either present as a free molecule (cell-free DNA) or within a cell membrane transferred from DNA-rich sources, for example, touching the eyes, nose, and mouth.[2-5] DNA contributed by any biological source of DNA, such as saliva, blood, and from touch, that cannot be seen by the naked eye refers to latent DNA. Currently, there are presumptive tests for saliva (phadebas which detects alpha-amylase), for semen (acid phosphatase, an enzyme reaction), and for blood (luminol which reacts with iron found in hemoglobin); however, none of these presumptive tests detect the latent DNA present within these biological samples.[6,7] Currently, there are no presumptive tests for touch DNA, either.

For the detection of latent fingermarks, there are many techniques available such as powder dusting, cyanoacrylate fuming, silver nitrate, ninhydrin, indandione, and many others; however the more sensitive the technique the less applicable it is to the crime scene; items need to be taken to the laboratory for testing and often have a long reaction time.[7] The methods applicable for crime scene testing, such as powder dusting, are not highly sensitive.[8] Current methods for fingermark enhancement generally work by interacting with amino acids; there are currently no methods that enhance fingermarks by detecting the DNA present.

In this experiment, common biological samples were stained with six selected dyes (GelGreen™, GelRed™, RedSafe™, SYBR® Green I, Diamond™ Dye, and EvaGreen™) to look at what was fluorescing in the samples such as hair, saliva, skin, and blood. The dyes were also applied to surfaces where latent DNA was present in fingermarks and as cell-free extracted DNA to determine the sensitivity of the dyes. The dye/DNA complex was detected using an alternative light source, the Polilight® (PL500), with an excitation wavelength of 490nm and emission through a 530nm or a 555nm interference filter.

Diamond™ dye, commonly used for gel staining, was found to be one of the more sensitive dyes and also had a longer lasting fluorescent signal that could still be detected after a month of applying the dye. This is advantageous because, if forensic evidence needs to be re-examined, then no additional dye is required to view the DNA present.

In conclusion, this study provides evidence that latent DNA can be detected with the use of nucleic acid binding dyes on substrate surfaces such as glass as a quick and sensitive method for detection. Furthermore, this research may be used to aid in development of a presumptive test for detecting latent DNA at crime scenes.

References:

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Latent DNA, Binding Dyes, Fingermarks