# The Effects of Substrate, Blood Volume, and Washing on the Performance of Luminol, Bluestar® Forensic, and Bluestar® Forensic Magnum

Jane G. Mak, Amy N. Brodeur, and Kevin R. Kosiorek

Boston University School of Medicine
Department of Anatomy & Neurobiology, Biomedical Forensic Sciences
72 East Concord Street, Boston, MA 02118

#### Introduction

The use of chemiluminescent and fluorescent latent blood detection reagents are typically used during the processing of a crime scene. One application that is not widely used is the use of these reagents to locate blood on evidence that has been submitted to a forensic laboratory, such as clothing. Bloodstains can be difficult to locate on dark colored clothing or clothing with patterns that obscure stains. This study evaluated the use of luminol, Bluestar® Forensic, and Bluestar® Forensic Magnum for this purpose.

Three variables were tested: substrate, blood volume, and washing. Human blood from anonymous donors was used.

The substrates tested were four different black fabrics composed of 100% cotton, 100% cotton denim, 100% polyester, and 50% polyester, 50% acrylic. Swatches of each substrate were stained with 16µL of blood and air dried.

The blood volumes tested were 0.5µL, 1.0µL, and 2.0µL. Swatches of black, 100% cotton fabric were stained and air dried.

The detergents used for washing were hypoallergenic liquid laundry detergent and liquid laundry detergent with bleach alternative. Swatches of black, 100% cotton fabric were stained with 16µL of blood and air dried. Machine washing was simulated in a beaker with a magnetic stirrer.

All swatches were created in quadruplicate.

## Methods

The Weber preparation of luminol was used and the working solution was prepared immediately prior to use. (1)

Bluestar® Forensic, and Bluestar® Forensic Magnum were prepared according to manufacturer's instructions immediately prior to use.

Reagents were applied to swatches in complete darkness using pressure pump spray bottles. When chemiluminescence was observed, the spraying was stopped and the timer was immediately started. The timer was stopped when chemiluminescence was no longer observed.

Photographs were taken immediately after the timer was started using a 64 second exposure and a f-stop of 2.7.

# Reference

1. Weber, K. Die Anwendung der Chemiluminescenz des Luminols in der gerichtlichen Medizin und Toxikologie I. Der Nachweis von Blutspuren. Deutsche Zeitschrift für Gerichtliche Medizin1966; 57: 410-423.

# Effects of Substrate on Reagents

Table 1: Average Chemiluminescence Duration (Seconds ±2SD)

				<b>50</b> %
		100% Cotton	100%	Polyester,
	100% Cotton	Denim	Polyester	50% Acrylic
Luminol	$90.7 \pm 72.0$	$85.4 \pm 70.3$	$128.9 \pm 61.9$	281.8±160.5
Bluestar®	71.7±28.5	101.1±85.3	162.6±112.8	416.2±55.5
Bluestar®	110.8±110.1*	176.1±205.1	429.9±185.3	$1218.7 \pm 171.3$
Magnum	110.0 - 110.1	110.1 - 200.1	443.3 - 100.3	1410.1 - 111.0

<sup>\* -</sup> Average of three swatches. One swatch was determined to be an outlier using the Q Test.

Figure 1:
Top to Bottom: Luminol, Bluestar®, Bluestar® Magnum
Left to Right Cotton, Denim, Polyester, Polyester/Acrylic

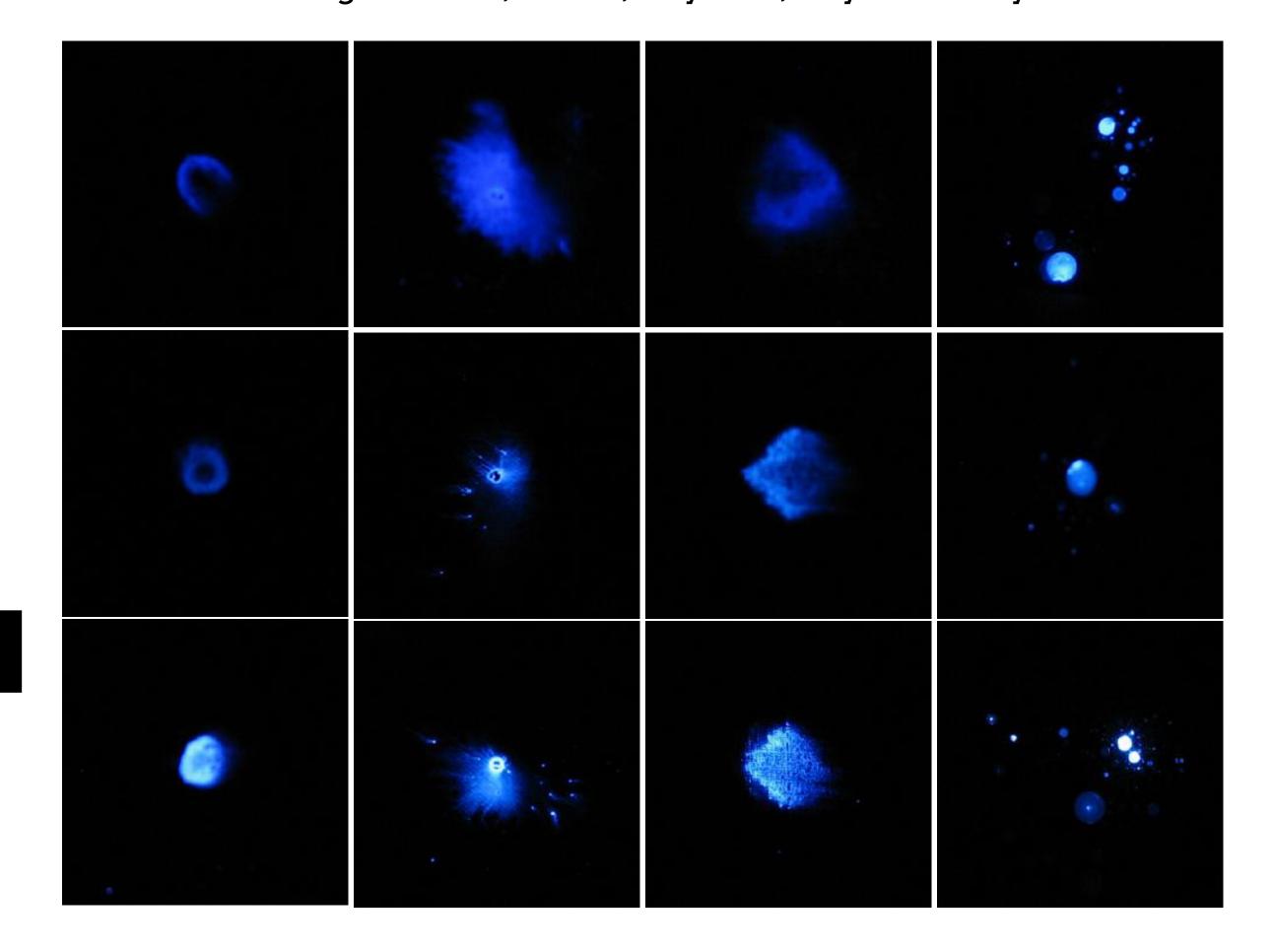


Figure 2:

Top to Bottom: Luminol, Bluestar®, Bluestar® Magnum

 $1.0 \mu \mathbf{L}$ 

 $110.2 \pm 16.3$ 

 $346.4 \pm 118.0$ 

Results

 $0.5 \mu \mathbf{L}$ 

 $95.8 \pm 11.2^{\circ}$ 

Luminol

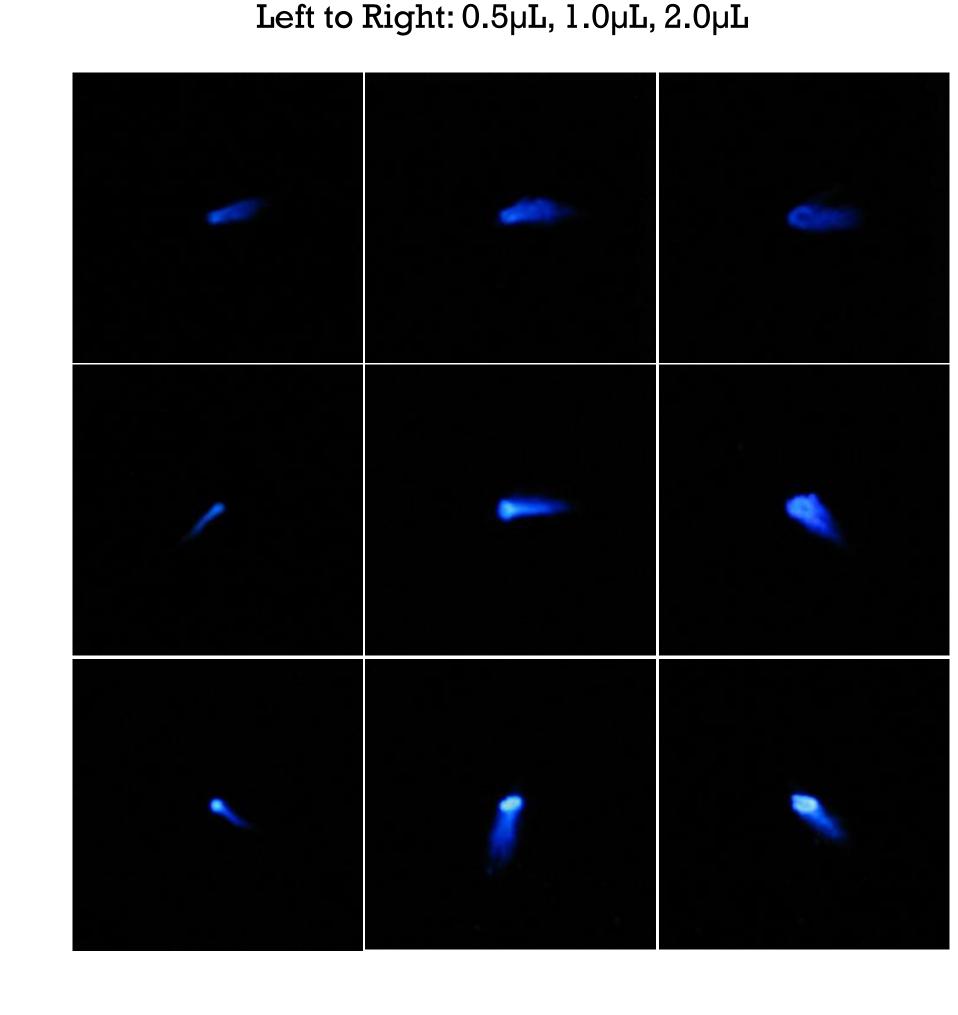
**Bluestar**<sup>®</sup>

using the Q Test.

Effects of Blood Volume on Reagents

**Bluestar® Magnum** | 896.7±80.3 | 1053.2±274.3 | 830.1±215.3

\* - Average of three swatches. One swatch was determined to be an outlier



# Effects of Washing on Reagents

Table 2: Average Chemiluminescence Duration (Seconds  $\pm 2$ SD) Table 3: Average Chemiluminescence Duration (Seconds  $\pm 2$ SD)

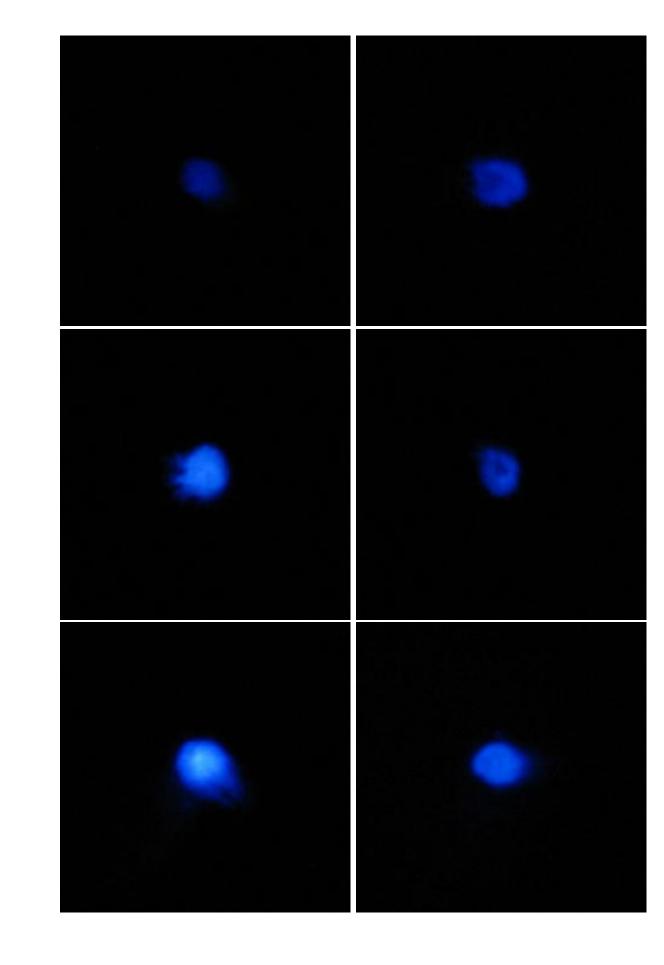
 $2.0 \mu L$ 

 $120.1 \pm 13.0$ 

 $303.5 \pm 98.8$ 

		Bleach
	Hypoallergenic	Alternative
Luminol	173.6±446.5	$125.3 \pm 139.5$
Bluestar®	370.7±281.7	205.7±202.2
Bluestar® Magnum	$578.1 \pm 408.7$	439.5±320.3

Figure 3:
Top to Bottom: Luminol, Bluestar®, Bluestar® Magnum
Left to Right: Hypoallergenic, Bleach Alternative



### Conclusions

Further studies should be done with a larger variety of detergents, stain removers, washing conditions, substrates, and blood volumes.

Even without knowing the full effects of washing, substrate, and blood volume on the reagents, they are useful in a laboratory setting in the same way they are useful at crime scenes. They allow for a more thorough examination of evidence and can be used after other screening methods fail to detect bloodstains.

In this study, Bluestar<sup>®</sup> Forensic Magnum had longer chemiluminescence durations than luminol and Bluestar<sup>®</sup> Forensic. When photographed, the chemiluminescence of Bluestar<sup>®</sup> Forensic Magnum was brighter or had a comparable brightness to luminol and Bluestar<sup>®</sup> Forensic. These results indicate Bluestar<sup>®</sup> Forensic Magnum would be well-suited for detecting bloodstains on clothing.

