

The 20th Conference of
Korean Society of Forensic Science

새로운 잠재혈흔 검출키트
헤마세인(Hemascein™)의 유효성 검
토

Validation of novel latent bloodstain detection kit Hemascein™

손부남¹, 임시근², 박정기¹, 정성희¹

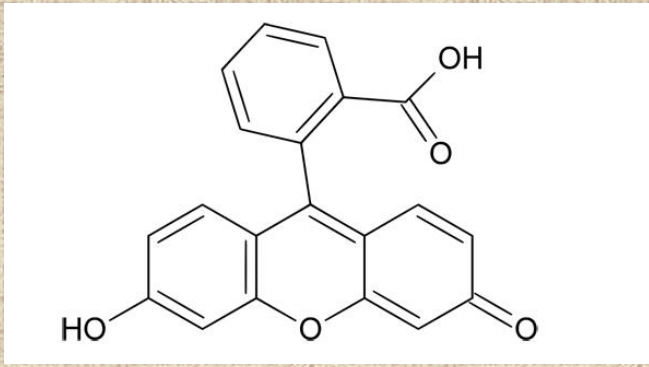
**Son Bu-Nam¹, Lim Si-Keun², Park Jung-Gi¹,
Jung Sung-Hee¹**

¹경상남도경찰청 과학수사계, ²국립과학수사연구소 유전자분석과
¹Gyeonnam Provincial Police Agency, Scientific Investigation Section
²National Institute of Scientific Investigation, DNA Analysis Department

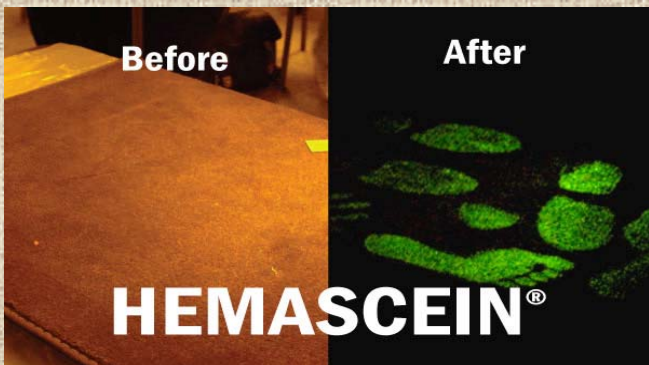
ABSTRACT

Blood stains are one of the effective evidence in murder, robbery and other violence crimes and can be the decisive evidence to solve the criminal matters. However, because murders are becoming more sophisticated and murderers try their best not to leave the blood stains, it is very hard to find blood stains these days. Until now, Luminol was often used to search for hidden blood stains, but its short luminescence time and influence of DNA discernment have led to the development of Luminol derived BlueStar[®] reagent. In this experiment, it is comparing Hemascein[™], the new blood stain search reagent made by Abacus Diagnostics with BlueStar[®] and Luminol in order to figure out for its significance and its effectiveness in spots of crimes. First, in terms of illuminant reaction, it measured its sensitiveness, strength of reaction, duration hours, and transferable reactions. By using various sources of light and filter, it made its best conditions to search reactions in Hemascein[™], and photo shoots. Also, it compared and measured the variations of reactions in porosity materials (absorbable) such as clothing and paper box and non porosity materials (non-absorbable) such as film. Lastly, it looked for cautions in terms of its usage of Hemascein[™], in the actual spots of crimes.

What is Hemascein™



The aqueous formulation of **fluorescein** reveals latent bloodstains based on the peroxidase-reduction of a reduced colorless reagent sprayed over the targeted area. An area suspected of containing latent bloodstains is sprayed with fluorescein using **ABASpray™**. The fluorescein reagent reduced with **hydrogen peroxide** (colorless) is immediately oxidized to **fluorescein** by the presence of **blood-associated proteins** and will fluoresce when excited by a light source **between 415 and 480 nm**.



| | |
|---|--|
| Sensitivity in detecting latent bloodstains | 2 to 5 times when compared to other tools in use |
| DNA recovery of diluted blood | Yes |
| Recovers finer detail | Yes |
| Longevity, time for documentation | Highest when compared to other tools in use |
| Stability | 7 yrs (Room Temperature) |
| Safety | Fluorescein is routinely used in humans |
| Identify if human origin | Yes (Hematrace®) |
| Formulation | Fluorescein containing formulation (aqueous) |
| Requires dark room and eyes to adjust to darkness | No |

Source: Abacus Diagnostics

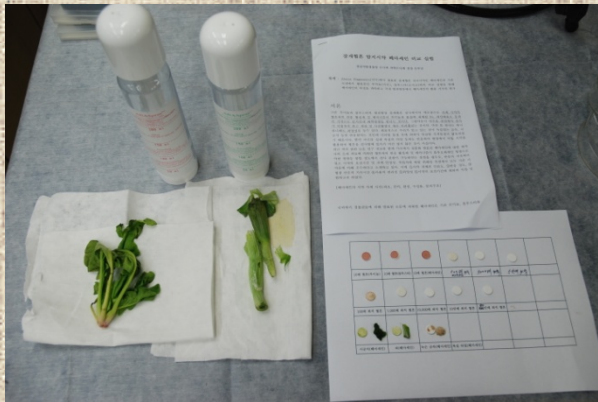
Preparing HemaScein™ working solution



1. Making HemaScein™ Stock Solution; Add 5ml of distilled water into the HemaScein™ powder vial.
2. Making HemaScein™ Working Solution; Dilute 1ml of HemaScein™ Stock solution with 100ml of distilled water in ABASpray™ bottle “A”.
3. Prepare 100ml of 3% hydrogen peroxide solution in ABASpray bottle “B”.



Preparing sample and test method



- Bloodstain Sample: Filter paper stained by diluted (by distilled water) blood were dried for 5 days in indoor condition.
- Light Source and Camera: Spectrum9000(485nm), B+W099IR Filter, Nikon D80, 60mm prime lens
- Photography: w/light and w/o light 5-30 sec. after spray.
- Transferable test sample: Spinach, Green Onion, Metal Rust Stain, Ceramic Tile, Wall Paint.

Different reactions for various illuminants and filters

| | 470nm B+W 061 3× (G) | 470nm B+W 090 5× (R) | 470nm B+W 099 (O) IR | 470nm B+W 022 2× (Y) | 505nm B+W 099 (O) IR |
|-----------------------|-------------------------|-------------------------|-------------------------|-------------------------|-------------------------|
| × 100 | | | | | |
| B l o o d | 485nm B+W 061 3× (G) | 485nm B+W 090 5× (R) | 485nm B+W 099 (O) IR | 530nm B+W 090 5× (R) | 530nm B+W 099 (O) IR |
| | | | | | |

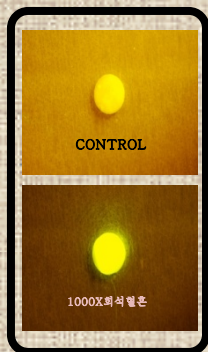
Tested with SL350, Spectrum 9000, Crime light, Blue light (zillight), Angsem (Alt-light), Mega Max, Blue Max, etc. with illuminant tools and variations of wavelength. **The best combination of wavelength and barrier filter was 485nm and B+W 099 IR.**

Sensitivity Determination

HemaScein™

| | ×50 | ×100 | ×500 | ×1,000 | ×5,000 |
|----------------|-----|------|------|--------|--------|
| Room Light ON | | | | | |
| Room Light OFF | | | | | |

| | ×10,000 | ×50,000 | ×100,000 | ×500,000 | ×1,000,000 |
|----------------|---------|---------|----------|----------|------------|
| Room Light ON | | | | | |
| Room Light OFF | | | | | |



Able to measure at least up to 1/100,000 diluted bloodstains. Better ID under room light OFF. There was no light-ON condition for BlueStar® and Luminol as it was not possible to test them with light ON.



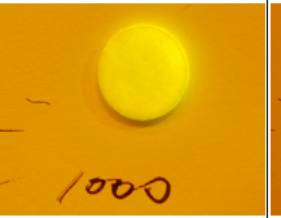
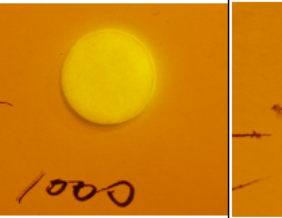
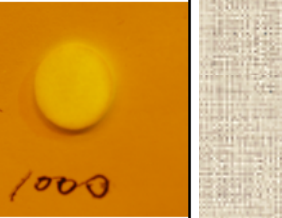
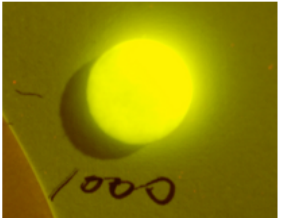
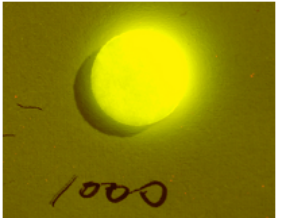
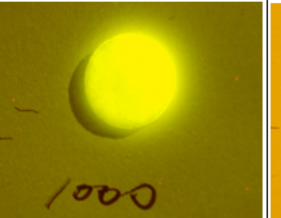
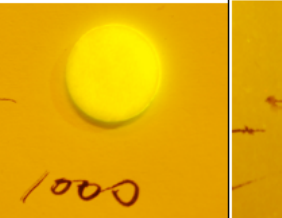
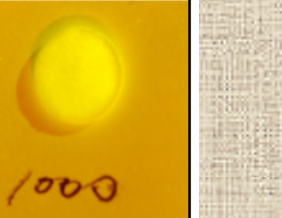
Luminol

| Control | ×50 | ×100 | ×500 | ×1,000 | ×5,000 |
|---------|---------|----------|----------|------------|--------|
| | | | | | |
| ×10,000 | ×50,000 | ×100,000 | ×500,000 | ×1,000,000 | |
| | | | | | |

| Control | ×50 | ×100 | ×500 | ×1,000 | ×5,000 |
|---------|---------|----------|----------|------------|--------|
| | | | | | |
| ×10,000 | ×50,000 | ×100,000 | ×500,000 | ×1,000,000 | |
| | | | | | |


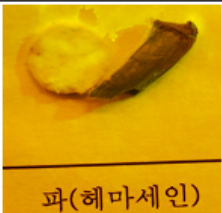

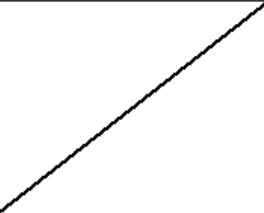
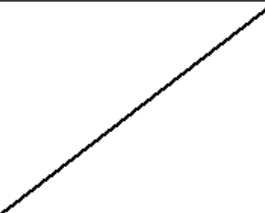
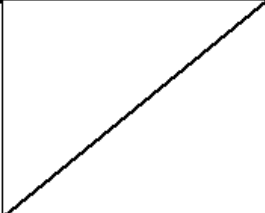
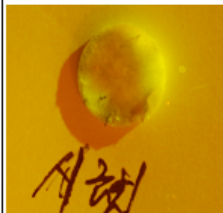

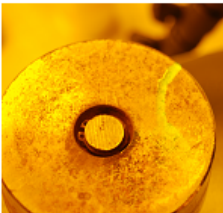
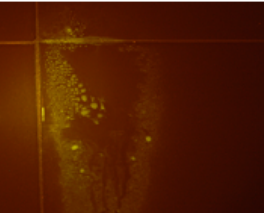
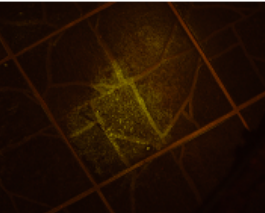

BlueStar®

Duration of HemaScein™ Reaction with 1/1,000 diluted blood

| | 10 minutes pass 485nm | 20 minutes pass 485nm | 30 minutes pass 485nm | 30 minutes pass 455nm | 60 minutes pass 485nm |
|----------------------|--|--|---|--|--|
| Room Light ON |  |  |  |  |  |
| Room Light OFF |  |  |  |  |  |

Able to measure its fluorescent reactions even after
an hour (For Bluestar, it lasts no longer than 60
seconds)

Evaluation of Transferable Reactions

| | Spinach | Green Onion | Metal Rust | Tile | | Painted Concrete Wall |
|----------------|--|--|--|---|---|---|
| | | | | Wall Tile | Floor Tile | |
| Room Light ON |  시금치 |  파(헤마세인) |  녹슨 금속(헤마세인) |  |  |  |
| Room Light OFF |  시금치 |  파(헤마세인) |  |  |  |  |

According to makers' related transferable resources, BlueStar® has false positive reactions in 5% of bleach, tomato, potato, tomato sauce mixed with meat, red onion, and peas but Luminol and HemaScein™ hadn't those false positive reactions. This report took some samples those were frequently reported in Korean crime scene as the source of false positive reaction with Luminol and BlueStar®.

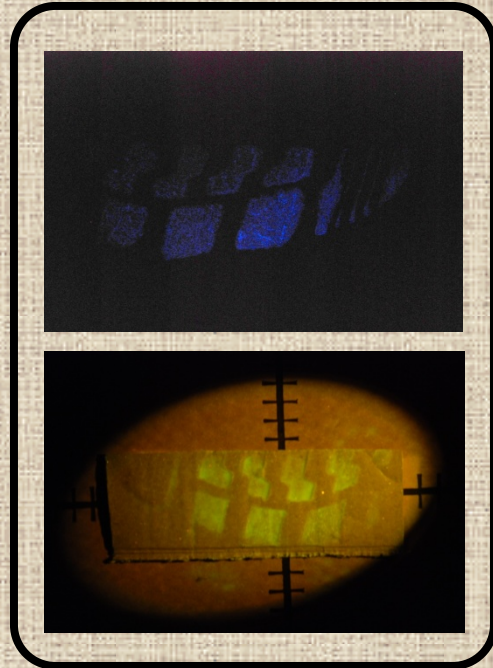
***Special Note: Unlike Luminol, HemaScein™ didn't show transferable reaction with the metal rust.**

Spinach, green onion, tile, concrete wall with paint: similar transferable reactions with BlueStar® and Luminol.

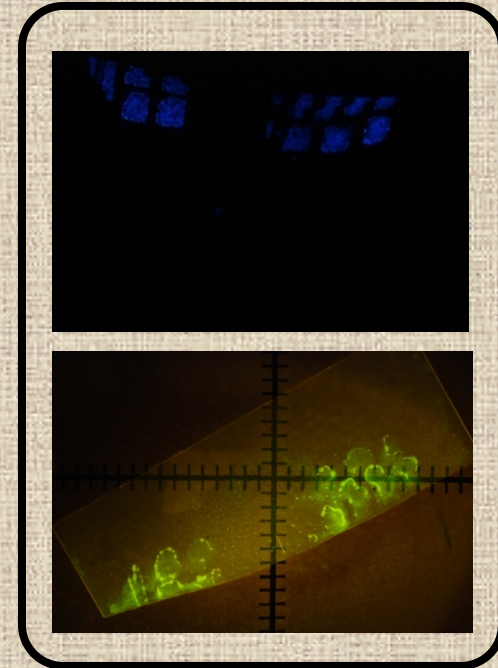
***Caution for bathroom tiles: reagent drops down from the wall and may create strong fluorescence.**

Porous vs. Non-Porous Surfaces

Porous (Cardboard Box)

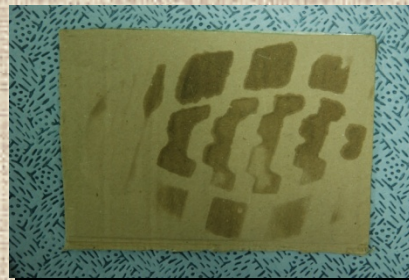


Non-Porous (Plastic Film)



BlueStar®

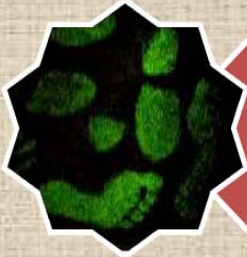
HemaScein™



* 1/100 Diluted Blood

To compensate the weak chemi-luminescence from BlueStar®, camera exposure was increased to 75% to get brighter image.

요약



- Hemascein™ does not need darkness condition to detect latent blood stains. Very useful where unable to cut out environmental lights.
- Hemascein™ shows equivalent or better sensitivity than with Luminol or Bluestar®.



- Better results for blood stains on porous surface such as cardboard box and textile than Bluestar®.
- No particular characteristic against transferable reactions.
- Longer than 1 hour duration of reaction period helps nice photography work.



- Hemascein™ get similar effect on DNA purification and genetic analysis to Luminol and Bluestar
- Non Hazardous
- High Stability and long shelf life