Schaeffer & Fulton's Spore Stain-Kit

Intended Use:
Schaeffer & Fulton's Spore Stain-Kit is used for staining bacterial spores.

Composition**
Schaeffer & Fulton's Spore Stain A (S028)
Ingredients
- Malachite green 5.000 gm
- Distilled water 100.000 ml

Schaeffer & Fulton's Spore Stain B (S029)
Ingredients
- Safranine O 0.500 gm
- Distilled water 100.000 ml

**Formula adjusted, standardized to suit performance parameters

Directions
1) Prepare a smear of the culture. Air dry it and fix with gentle heat.
2) Flood the entire slide, with aqueous malachite green/Schaeffer and Fulton's Spore Stain A (S028).
3) Steam for 3-6 minutes, and rinse under running tap water.
4) Counterstain with 0.5% aqueous safranin/Schaeffer and Fulton's Spore Stain B (S029) for 30 seconds.
5) Wash with water, dry and observe under oil immersion lens.

Principle And Interpretation
A spore is a dormant form of the bacterium that allows it to survive in drastic environmental conditions. Spores have a tough outer covering made of the protein keratin and are resistant to heat and chemicals. The keratin also resists staining, so extreme measures must be taken to stain the spore. In the Schaeffer-Fulton's method, a primary stain- malachite green is forced into the spore by steaming the bacterial emulsion. Malachite green is water soluble and has a low affinity for cellular material, so vegetative cells may be decolorized with water. Vegetative cells are then counterstained with safranin. Spores may be located in the middle of the cell, at the end of the cell, or between the end and middle of the cell. Spore shape may also be of diagnostic use. Spores may be spherical or elliptical. Members of the genus Corynebacterium may exhibit club-shaped swellings that might be confused with spores. Spore staining distinguishes between true spores and these structures.

Type of specimen
Any isolated colony on primary or subculture plates can be isolated from following specimens. Clinical specimen: Blood, urine, CSF, pus, wounds, lesions, body tissues, sputum etc. From environment: Air, water, soil, sludge, waste water, food, dairy samples etc.

Warning and Precautions
In Vitro diagnostic use only. Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations
1. If over heat fixed, the safranin will replace the malachite green in the spores as well as in vegetative cells.
2. Safranin is easily washed from vegetative cells therefore washing should be minimal.

Performance and Evaluation
Performance of the stain is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Please refer disclaimer Overleaf.
**Quality Control**

**Microscopic examination**
Spore staining is carried out and staining characteristic of the organism is observed under microscope by using oil immersion lens.

**Results**
- Spores: green
- Vegetative cells: red

**Storage and Shelf Life**
Store between 10 - 30°C in tightly closed container and away from bright light. Use before expiry date on label. On opening, product should be properly stored in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use.

**Disposal**
User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (6,7).

**Reference**