Malachite Green, 1% w/v

S020

Intended use

Malachite Green 1% w/v is used as staining solution in spore staining and simple staining.

Composition**

Ingredients

Malachite green	1.0 gm
Distilled water	100.0 ml

**Formula adjusted, standardized to suit performance parameters

Directions

1. Prepare a smear on a clear, dry glass slide.
2. Allow it to air dry and fix it with gentle heat.
3. Flood the slide with 1% w/v Malachite Green (S020).
4. Allow the stain to be in contact with the smear for 2-3 minutes and heat the preparation for 3-6 minutes and then allow to cool.
5. Wash in slow-running tap water.
6. Counterstain with 0.5% aqueous safranin(S027)/ Schaeffer and Fulton's Spore stain B(S029) for 30 seconds.
7. Wash with water, blot dry and examine under oil immersion objective

Principle And Interpretation

Malachite Green is used for bacterial spore staining by Schaeffer and Fulton's method. It can also use as a simple stain for bacterial cells and in place of methyl green. 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 decolourized 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.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines. For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines . For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards. After use, contaminated materials must be sterilized by autoclaving before discarding.

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 which will prevent differentiation of vegetative cells and spores.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature

Quality Control

- **Appearance :** Dark green coloured solution.
- **Clarity :** Clear without any particles.
- **Microscopic Examination :** Spore staining by Schaeffer and Fulton's method is carried out using Malachite green 1% w/v as one of the stains and staining characteristic is observed under microscope by using oil immersion lens.
- **Results :** Spores are seen as green spherules in red stained rods or red stained debris

Storage and Shelf Life

Store between 10 - 300C 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.

Reference

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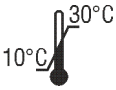

Storage temperature

Do not use if package is damaged

In vitro diagnostic medical device

CE Marking

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