



PRODUCT SPECIFICATION SHEET

Gentian Violet (SI014)

Use

Gentian Violet is used for Gram's staining.

Principle

The Gram stain is a differential staining technique most widely applied in all microbiology disciplines laboratories. It is one of the most important criteria in any identification scheme for all types of bacterial isolates. Different mechanisms have been proposed to explain the gram reaction. There are many physiological differences between gram-positive and gram-negative cell walls (1). Ever since Christian Gram has discovered Gram staining, this process has been extensively investigated and redefined. In practice, a thin smear of bacterial cells is stained with gentian violet, then treated with an iodine containing mordant to increase the binding of primary stain (2). A decolorizing solution of alcohol or acetone is used to remove the gentian violet from cells which bind it weakly and then the counter stain (like safranin) used to provide a colour contrast to those cells that are decolorized. The gram-positive organisms or cells have more mucopeptide in their cell walls as compared to gram-negative ones. Gram-negative bacteria have more content of polysaccharides and lipo-proteins in their cell walls. The polymers of glycerol or ribitol phosphate called as teichoic acids are also found in the cell walls of gram-positive organisms but are very less or almost not present in gram-negative organisms. In a properly stained smear by gram staining procedure, the gram positive bacteria appear blue to purple and gram negative cells appear pink to red.

Storage And Stability

1. Store the bottle in dry, cool and dark place.
2. The shelf life of reagents is as per the expiry date mentioned on the reagent bottle labels.

Procedure

1. Transfer the specimen to a clean microscope slide. Prepare a smear and air dry.
2. Heat-fix the slide by passing through the flame and place on a staining rack with smear side up.
3. Flood the slide with **Gentian violet** (SI014) and time for 1 minute.
4. Rinse with tap water and flood with **Gram Iodine** (SI016).
5. Wait for 1 minute and rinse with water.
6. Decolorize with **Gram Decolorizer** (SI 002) until no more color is released. Immediately rinse with tap water.
7. Flood the slide with **Gram Safranin** (SI028) for 30 seconds and rinse.
8. Wipe any stain that remains off the back of the slide and blot dry or air dry completely.

PRECAUTIONS

1. For Invitro Diagnostic use only
2. Overheating of organisms during fixing process may disintegrate organisms on the smear.
3. Decolorising process timing depends upon the thickness of smear and adequate allowance should be given depending upon the tube of the specimen.
4. Product to be used only by adequately trained and qualified laboratory personnel.



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QUALITY CONTROL

Appearance: Purple Colored solution

Microscopic Examination

Using *Gentian Violet* as a stain, gram staining is carried out and staining characteristics of microorganisms are observed under microscope by using oil immersion lens.

Run controls using 18-24 hour standard ATCC cultures of known gram-positive and gram-negative microorganisms. The following test strains are recommended:

| Organism | Expected Results | |
|---|---------------------|---------------------|
| <i>Staphylococcus aureus</i> ATCC 25923 | Gram-positive cocci | Dark purple colored |
| <i>Escherichia coli</i> ATCC 25922 | Gram-negative rods | Red or pink colored |

Packaging

Product Name : **Gentian Violet**

Product Code : **SI014**

Available Pack sizes : **500ml/125ml**

Further Information

For further information please contact your local MICROMASTER Representative.



MICROMASTER LABORATORIES PRIVATE LIMITED

SI014PSS QAD/FR/024,Rev.00

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