GCC Gram Stain Kit SP400

Technical Leaflet

The Gram stain procedure differentiates bacterial cells into 2 groups.
Those cells that retain the primary dye are termed Gram Positive and those which loose the primary dye after decolourisation and take up the counterstain are termed Gram Negative.
This kit contains the reagents required to carry out the Gram technique.

Method
Make a hole in the top of the dispenser nozzle on the reagent bottles or cut off the top 3-5mm.

1. Prepare heat fixed smear by passing the slide, smear side up, 5-6 times through a Bunsen flame.
2. Flood the slide with crystal violet oxalate solution for 1 minute.
3. Wash off excess dye by swirling in a beaker of tapwater for 4-5 seconds.
4. Flood slide with iodine solution, drain off the excess, again flood the slide with iodine solution for 1 minute.
5. Wash the slide in a beaker of tapwater, over-washing is difficult at this stage.
6. Decolourise by running the differentiator solution over the surface of the slide for 5-20 seconds for thin smears and 20-60 seconds for thick smears, until crystal violet no longer bleeds from the smear.
7. Wash in tap water to stop the action of the differentiator.
8. Flood slide with counterstain for 1 minute.
9. Wash very lightly in a beaker of water to remove excess counterstain. Excessive washing at this stage will remove the counterstain from Gram negative organisms.
10. Blot dry and examine.

The method given in this leaflet is designed as a general guide only.

Results: Gram positive organisms - Purple/Black
Gram negative organisms - Shades of red

Storage.
Keep all kit components in the kit box provided, store at room temperature in a darkened cupboard. Replace nozzle caps immediately after use to prevent evaporation and deterioration. Kit will remain usable during its shelf life in these conditions.

Safety Data.
Crystal violet - H302+312 Harmful if swallowed, by skin contact. P260 Do Not breathe vapour. Keep container tightly closed. P262 Do not get in eyes, skin, clothing. IF SWALLOWED - rinse mouth with water, seek urgent medical attention. IF INHALED - move to clear air zone. IF ON SKIN or in EYES - rinse with soap and water and apply eye bath. P314 Get medical attention if you feel unwell.

Gram differentiator - Propanol acetone mixture. H224 Extremely flammable liquid & vapour H302+312+332 Harmful if swallowed, by skin contact and inhalation. H320 Causes eye irritation/damage Keep container tightly closed. P262 Do not get in eyes, skin, clothing. Use in well ventilated place. P280 Wear eye, skin protection. P210 Keep away from heat, sources of ignition. IF SWALLOWED - rinse mouth with water, seek urgent medical attention. IF INHALED - move to clear air zone. IF ON SKIN or in EYES - rinse with soap and water and apply eye bath. P314 Get medical attention if you feel unwell.

Grams iodine - H302+312+332 Harmful if swallowed, by skin contact and inhalation. P260 Do Not breathe fumes. Keep container tightly closed. P262 Do not get in eyes, skin, clothing. IF SWALLOWED - rinse mouth with water, seek urgent medical attention. IF INHALED - move to clear air zone. IF ON SKIN or in EYES - rinse with soap and water and apply eye bath. P314 Get medical attention if you feel unwell.

Gram safranine / fuchsin / neutral red - H305+313 Harmful if swallowed, enters airways by skin contact .Keep container tightly closed. P262 Do not get in eyes, skin, clothing. IF SWALLOWED - rinse mouth with water, seek urgent medical attention. IF INHALED - move to clear air zone. IF ON SKIN or in EYES - rinse with soap and water and apply eye bath. P314 Get medical attention if you feel unwell.

All reagents except Gram differentiator will stain skin and other tissue. If in contact wash with soap & water. For use as an In-Vitro diagnostic laboratory reagent only.

Accidental spillage & Waste Disposal.
Gram differentiator reagent is Highly Flammable. It is volatile in air so spillage vapours may present an explosion hazard in small spaces if mixed with air. Mop up spillage immediately using suitable absorbent granules or paper and remove to outside area for proper disposal (can be evaporated to atmosphere). For differentiator residues and all other reagents in this kit clean the spillage area with detergent & water and run the waste to sewer drains.
Unused reagents can be disposed of in the following manner:
Gram differentiator - evaporate to atmosphere. All other kit reagents - dilute to public sewer with water.

Unsatisfactory performance.
As part of our duty to monitor product performance and part of our policy of continual improvement. Please report to us any unsatisfactory performance you may experience with this product. If any reagent degrades before expiry of shelf life we will replace that reagent free of charge.
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