Gram Staining Solution

CE IVD REF

In Vitro Diagnosis
For Professional Use

【Product Name】

Gram Staining Solution

【Specification】

Manual mear(medicine)，01 Specifications: Crystalline violet staining solution 100 ml, Gram's iodine solution 100 ml,decolourising solution 100 ml, compound red re-staining solution 100 ml; 02 Specifications: Crystalline violet dyeing solution 250ml, Gram iodine solution 250ml, decolorizing solution 250ml,compound red re-staining solution 250ml; 03 Specifications: Crystalline violet dyeing solution 500ml, Gram iodine solution 500ml, decolorizing solution 500ml,compound red re-staining solution 500ml; 04 Specifications:Crystalline violet dyeing solution 1000ml, Gram iodine solution 1000ml, decolorizing solution 1000ml, compound red re-staining solution1000ml. Fw·dyeingmachne, 01 Specification: Crystalline violet dyeing solution 250ml, Gram iodine solution 250ml, decolorizing solution 250ml,compound red re-staining solution 250 ml. 02 Specification: Crystalline violet staining solution 500ml, Gram iodine solution 500ml,decolorizing solution 500ml, compound red dyeing solution 500ml; 03 Specifications: crystalline violet staining solution 1000ml, Gram iodine solution 1000ml, decolorizing solution 1000ml, compound red dyeing solution 1000ml.

【Intended use】

For smear staining of bacteria or fungi.

【Main components】

The product consists of reagent (A): crystal violet staining solution; reagent (B): Gram's iodine solution; reagent (C): decolorizing solution; reagent (D): compound red re-staining solution.

【Contraindication】none

【Inspection Principle】

Due to the difference in the permeability of cell wall to ethanol and its ability to resist decolorization, which is mainly determined by the thickness and structure of peptidoglycan layer, cells stained with crystal violet formed an insoluble complex after treatment with iodine solution, and ethanol could decolorize it. In Gram-negative staining of cells, ethanol or acetone destroyed the outer membrane of the cell wall, damaged the peptidoglycan layer and the plasma membrane of the cells, and the crystal violet and iodine complex leaked out of the cells, and showed red color when it was repeated with other staining solutions. Although the red dye can also enter the purple-stained G+ cells, it is covered by the purple color, so the red color does not appear. In Gram-positive cell staining, ethanol also dehydrates the thick peptidoglycan layer, causing the pores to become smaller. Since the molecules of crystal violet and iodine complex are too large to pass through the cell wall, they are not easy to be removed and thus maintain the purple color.

【Sample Request】

1. Bacteria should be selected from single fresh colonies;
2. Dilution water should be sterile.

【Instructions for use】

Manual colorimetry:

1. Smear: take the bacteria to be examined, apply a thin layer in the center of the slide or add a little sterile water on the slide, take the bacteria and water to mix well and apply a thin layer.
2. Drying: After the smear is dried naturally at room temperature, it can also be slightly warmed on the alcohol lamp to make it dry quickly.
3. Fixation: hold one end of the slide, specimen face up, in the flame of the alcohol lamp on the outside of the rapid back and forth 3 to 5 times, 1s each time, the temperature should not be too high, to prevent the denaturation of bacterial proteins, and then staining after cooling. Can also be fixed with methanol or ethanol.
4. Initial staining: add crystal violet staining solution for 1min, rinse off the staining solution with water.
5. Mordant staining: add drops of Gram's iodine solution and cover the sample, leave it at room temperature for 1min, rinse with water.
6. Decolorization: add drops of decolorizing solution, shake for 15s, until the decolorizing solution does not appear purple, immediately rinse off the decolorizing solution with water to terminate the reaction.
7. Re-staining: add drops of red re-staining solution for 30s, wash with water.
8. Drying. Microscopic examination: put the oil mirror to observe.

For dyeing machines:

(1). Specimen processing

(2). Selection of appropriate staining pattern according to the thickness of the specimen.

(3). Put the staining solution into the staining chamber and adjust the staining parameters of the staining solution. (4). The staining machine stains automatically.

(4). Please refer to the instruction of dyeing machine for detailed operation.

【Interpretation of test result】

Observe the staining results under a microscope:

A Blue to purple colonies observed under the microscope are gram-stain positive bacteria.

B Red colonies observed under the microscope are gram-negative.

【Limitations of the test method】

Some bacteria have chromophilia, which is not always entirely due to staining and should be determined on the basis of the morphology of the organism.

【Precautions, warnings and reminders】

1. Before applying the smear, the back of the sample should be marked with a circle in order to determine the position of the subsequent tests.
2. When removing bacteria, attention should be paid to self-protection. When removing or stoppering the test tube, the mouth of the test tube should be slightly cauterized through the flame, and finally the inoculation ring should be cauterized on the flame to sterilize.
3.When heating the fixed smear, the slide should not be too close to the flame. Generally, the temperature of the slide should not exceed 60 °C, and the back of the slide should not feel too hot when touching the back of the hand.

4. The key of Gram staining is to strictly control the degree of decolorization, and the decolorization time should be judged according to experience. If the decolorization is excessive, positive bacteria can be mistakenly stained as negative bacteria; if the decolorization is insufficient, negative bacteria can be mistakenly stained as positive bacteria.
5. The incubation time of the bacteria to be examined will affect the staining. Positive bacteria will often be negative if the incubation time is too long, or if the bacteria have died or lysed.
6. This product is only used for in vitro diagnosis.
7. Cover the reagent bottle tightly after each use to prevent the reagent from volatilization and contamination.
8. The reagent is corrosive, please pay attention to appropriate protection.
9. For your safety and health, please wear lab coat and disposable gloves.

【Storage conditions and expiration date】

Store at room temperature and away from light.

Valid period: 12 months

【Date of manufacture】

See product box for details.

【Index of Symbols

Symbols	Title	Symbols	Title
CE	CE MARK	REF	Catalogue number
	Manufacturer	EU REP	Authorized representative in the European Community
IVD	In vitro diagnostic medical device		Single use only
	Date of manufacture		Use-by date
LOT	Batch code	Σ	Tests / box
	Consult instructions for use		Upwards
	Keep away from sunlight		Keep dry
UDI	Unique device identifier		

Manufacturer:
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