



PENROSE MODIFIED BROWN & HOPPS STAIN KIT PROCEDURE

Item# CUPMBKT

(Revised 03/7/18)

**KIT
COMPONENTS
INCLUDED:**

100 ml 1% CRYSTAL VIOLET

100 ml STOCK BASIC FUCHSIN STAIN

100 ml ADVANCED PICRIC ACID/ACETONE

100 ml GRAM'S IODINE

100 ml GALLEGOS SOLUTION

PRINCIPLE: This kit demonstrates gram positive and gram negative bacteria.

SPECIMEN: Any well fixed paraffin embedded tissue cut at 5 microns.

QUALITY CONTROL: American MasterTech Scientific Recommended Control Slide:
Gram Positive / Gram Negative, CSG0225P.

SOLUTIONS:

WORKING BASIC FUCHSIN SOLUTION:

STOCK BASIC FUCHSIN STAIN 2 ml

Distilled Water 50 ml

Prepare just before use; mix thoroughly!

PROCEDURE:

1. Deparaffinize slide in Xylene or Xylene Substitute and hydrate through alcohols.
2. Rinse slide in running Tap water.
3. Rinse slide thoroughly in Distilled water.
4. Place slide in **1% CRYSTAL VIOLET** for 2 minutes.
5. Rinse slide in running Tap water to remove excess stain.
6. Flood slide with **GRAM'S IODINE** for 5 minutes.
7. Rinse slide in running Tap water to remove excess Iodine.
8. Quickly blot slide to remove excess water. (*Do not allow section to dry!*)
9. Differentiate in Acetone (*Not included in kit*) until blue stops running off of section. (*Approximately 2 dips per second is suggested for best results.*)
10. Rinse slide quickly in running Tap water.
11. Place slide in **Working Basic Fuchsin Solution** for 5 minutes.
12. Rinse slide quickly in running Tap water.
13. Place slide in **GALLEGOS SOLUTION** for 5 minutes.
14. Rinse slide quickly in running Tap water.
15. Dip slide **once only** in **ADVANCED PICRIC ACID/ACETONE**.
16. Dehydrate slide quickly through 2 changes of fresh Absolute Alcohol.
17. Clear slide through 3 changes of Xylene or Xylene Substitute.
18. Coverslip using a permanent mounting media.

RESULTS:

Gram Positive Bacteria: **BLUE**

Gram Negative Bacteria: **RED**

Other Tissue: **YELLOW**

REFERENCE: Sheehan DC Hrapchak BB: Theory and Practice of Histotechnology; 1980, 233 - 235.

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