



PHARMA DEVILS

MICROBIOLOGY DEPARTMENT

STANDARD OPERATING PROCEDURE

Title: Gram Staining Procedure

SOP No.:		Department:	Microbiology
		Effective Date:	
Revision No.:	00	Revision Date:	
Supersede Revision No.:	Nil	Page No.:	1 of 2

1.0 OBJECTIVE

To lay down procedure for Gram's staining.

2.0 SCOPE

This SOP is applicable in microbiology laboratory.

3.0 RESPONSIBILITY

Prepared by - Executive Microbiology

Checked by - Assistant Manager Microbiology / QC

Approved by - Head QA, QC

4.0 PROCEDURE

- 4.1 Prepare a thin smear of the culture to be examined on a glass slide and fix it by passing through a flame.
- 4.2 Allow the slide to cool.
- 4.3 Flood the smear with Gram's crystal violet solution for 1 minute.
- 4.4 Drain off the crystal violet solution and flood with Gram's iodine for 1 to 2 minutes.
- 4.5 Wash the smear with 70% ethanol/Gram's decolourizer till all the extra crystal violet stain is washed off.
- 4.6 Wash with potable water immediately.
- 4.7 Flood the smear with safranin solution for 1-2 minutes.
- 4.8 Wash with potable water.
- 4.9 Dry the slide and place a drop of cedar wood oil and examination under oil immersion lens of the microscope.

5.0 SAFETY & PRECAUTIONS

Not applicable.

6.0 REVISION HISTORY

Revision No.	Reason for Revision	Superseded from & Date
00	First Issue	-----



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7.0 REFERENCES

Not applicable.

8.0 ABBREVIATIONS

SOP : Standard Operating Procedure

9.0 ANNEXURES

Not applicable.