



STAINING

Introduction

- A stain is a chemical which can bind with the cellular components and give them color. Different dyes which can be used for staining are basic/cationic dyes (methylene blue, crystal violet, malachite green, safranin) and anionic/acidic dyes (eosin and picric acid).
- Cationic dyes bind with the negatively charged cellular components such as those are present in cell membrane, whereas, anionic dye binds with positively charged cellular components. [1]

Simple staining

Simple staining is the use of single dye which helps in revealing the basic cell shapes and cell arrangements. Stains which can be used in the simple staining are crystal violet, carbolfuchsin, safranin and methylene blue.

Differential staining

It is the use of two or more dyes to differentiate between two different kinds of organisms or two different parts of organisms. Two common methods for differential staining are:

- The gram staining
- The acid fast staining

The gram staining

Gram staining is based on the amount of peptidoglycan present in the cell wall of bacteria.

- Those bacteria which have thick layer of peptidoglycan in their cell wall can retain the primary stain and are known as gram-positive bacteria.
- Those bacteria which have thin layer of peptidoglycan in their cell wall cannot retain the primary stain and are called gram-negative bacteria.

Steps for gram staining:

- i. Bacterial smear is prepared on a slide.
- ii. Crystal violet stain is added on the smear.
- iii. After 1 min. stain is drained and rinsed with water and iodine mordant is added on the smear.
- iv. After 1 min. iodine is drained and slide is rinsed with water.
- v. Decolorizing with the help of alcohol and rinsing immediately
- vi. Safranin is added, remains there for 1 min. and then drained and rinsed.

vii. Slide is looked under compound microscope.

Result interpretations:

Gram positive bacteria appear purple in color, whereas, gram negative bacteria retains the color red(pink) color of safranin dye.

