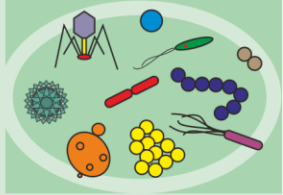
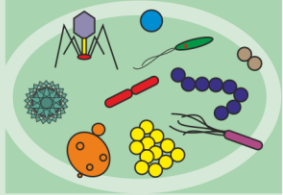


1. practice

Gram staining, negative staining, native preparation

Fidrich (2018)

Gram staining

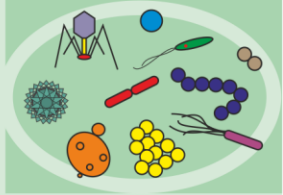


- differentiate **G+** a **G-** bacterial cells

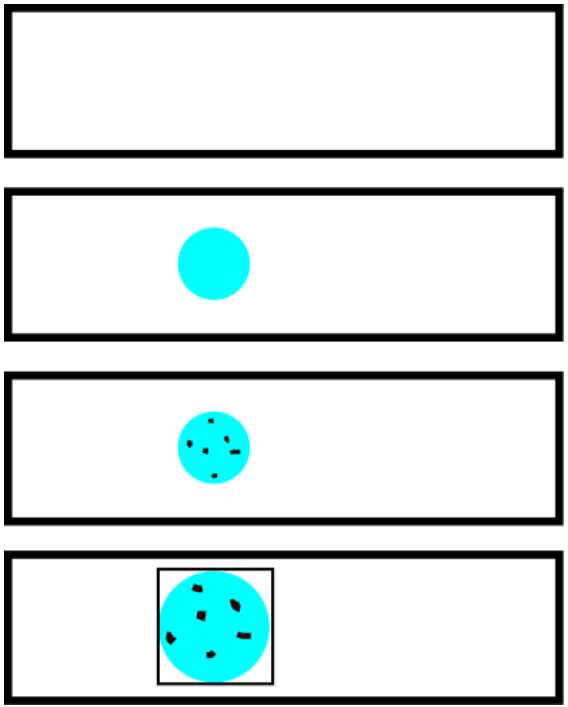
- Fix dry microscopic slide by the flame after air dry

- Crystal violet (1 min)
  - Rinse with H_2O
- Lugol solution (30 s)
  - Rinse with H_2O
  - Wash the preparation by ethanol (10-15 s)
- Safranin (1 min)
  - Rinse with H_2O

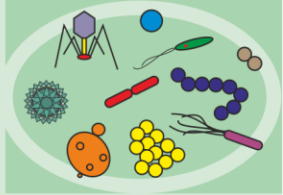
Native preparation



1. Drop of water
2. Transfer small ammount of cells in drop of water
   - Do not smear the drop
3. Cover drop with cover slide
4. Observe
   - Bright field
   - Phase contrast

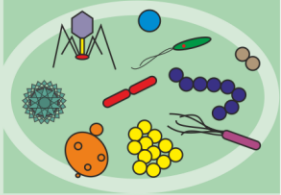


Negative staining



- Nigrosin
  1. Drop of nigrosin + loop of water + culture
  2. spread over the slide with another slide
  3. Allowed to air dry
- Kongo red
  1. Drop of Kongo red + culture
  2. spread over the slide with another slide
  3. Allowed to air dry
  - 1% HCl on dry microscopic slide

What to observe ?



- Native preparation (2)
  - 2x *Bacilli*
- Gram staining (2)
  - 1x pure culture
  - 1x mix of cultures (2 or more)
- Negative staining (2)
  - 1x with Nigrosin
  - 1x with Kongo red

***Bacilli*:**

Bacillus sphaericus
Bacillus cereus
Bacillus megaterium
Paenibacillus polymyxa

Cocci:

„Azotobacter vinelandii“
Leuconostoc mesenteroides
Sporosarcina ureae
Staphylococcus aureus
Micrococcus luteus

Rods:

Serratia marcescens
Escherichia coli

***Archaea*:**

Haloarcula hispanica

Eukaryota:

Saccharomyces cerevisiae
Yarowia lipolytica