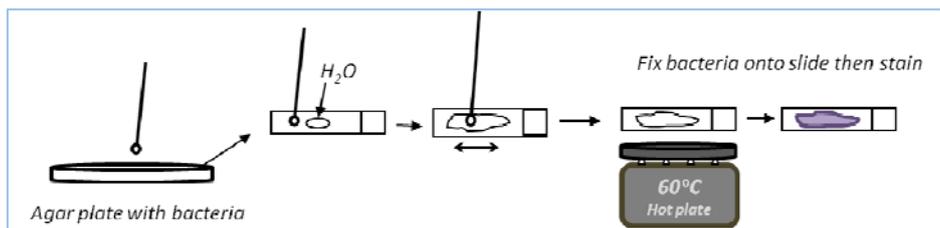


Prepare a bacterial stain for microscopy

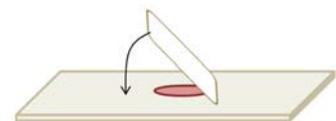
Materials

- Bacterial culture (plate, slope or broth)
- 60°C heat plate
- Glass microscope slides
- Coverslips (22x22mm or 22x40mm)
- Microscope up to 400x magnification (1000x oil immersion if available)
- Water: dropper bottle & squirt bottle
- Toothpicks or bacteriological loops
- Stains: crystal violet, carbol fuchsin
- Paper towel

Procedure

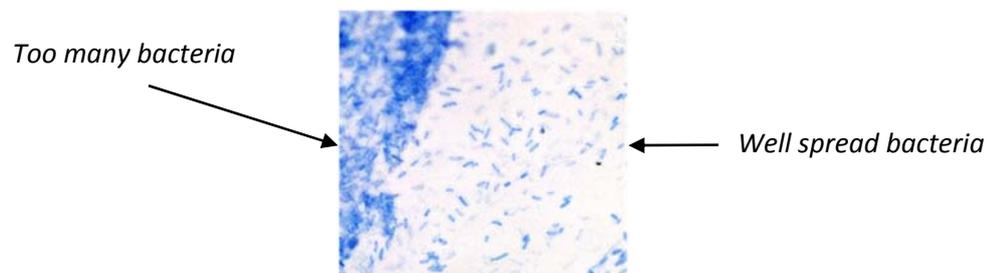


1. Label a slide with the bacterial species
2. Place a small drop of water onto the centre of the slide
3. Using a sterile loop or toothpick, pick a single colony from a plate
If preparing from a broth culture, dip the loop into the broth
4. Spread the bacteria in the water over the central third of the slide
5. Dry the bacteria onto the slide:
 - place the slide onto a 60°C heat block/hot plate or metal tray in an oven **until dry**
6. Cover the bacteria with the staining solution for 1 minutes
7. Rinse excess stain off with water
8. Place the slide between 2 layers of paper towel. Pat down to blot dry
9. Add 1-2 drops mounting solution (e.g. 50% glycerol in water) and add a coverslip (22x40mm); lower the coverslip on an angle to avoid trapping bubbles



Additional points:

Spreading for a good view: if the bacteria are not spread sufficiently, you cannot see individual cells, leading to mis-interpretation of the view. Look to the edges of a heavy smear to see single cells.



Keeping slides: To keep glycerol mounted slides, seal the edges of the coverslip with nail polish.

Permanent mounts can be prepared with a permanent mounting solution like Depex (apply at Step 9 above). This solution should be used in a fume hood, the slide must be dry before application and then left to dry and seal before viewing and storage.