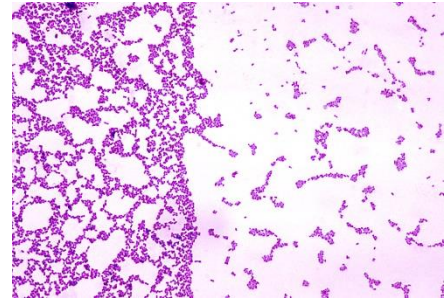


Cell Structure: Bacteria on Teeth

Dental caries or cavities are the result of bacterial action. Yes, sugar definitely plays a role in cavities, but bacteria are the real culprits. Bacteria use their capsule, pili and fimbriae to stick to surfaces like your teeth. Then you feed them every day, probably even several times a day. Bacteria share the food you eat. They digest sugars in your food to produce acids which are released onto the surface of your teeth. Over time the tooth's enamel weakens and cavities form. The bacterium primarily responsible for dental caries is *Streptococcus mutans*. Similarly, plaque which forms along the gum line and between teeth is the hardened residue of bacterial action. About 70% of the dry mass of plaque deposits are bacteria with the remaining 30 % consisting of glycoproteins, polysaccharides and other macromolecules. Plaque begins as a biofilm. A community of bacteria including *S. mutans* and others that live around your teeth create the biofilm. Over time the biofilm becomes hardened into plaque. Plaque is hard and protects the bacteria from routine brushing allowing the bacteria and the acids they produce to erode your teeth.



S. mutans photomicrograph by
CDC/Dr. Richard Facklam

Materials

Methylene blue

Microscope slides

Staining tray

Distilled water

Bibulous paper/ paper towel

Bunsen burner

Striker

Toothpick

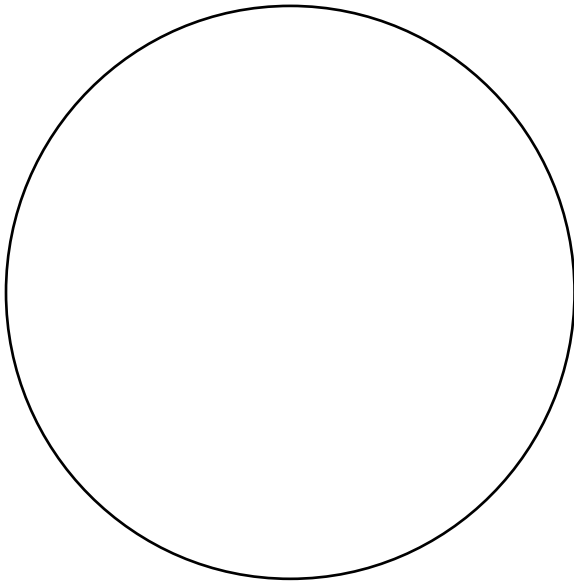
Slide clips or clothespin

Microscope

1. Obtain a slide and toothpick.
2. Light your Bunsen burner. Open the valve on the bottom of the burner about ½ way open. If there are air baffles, open those halfway. Turn on the gas at the benchtop. Place the striker near the top of the burner and ratchet the striker back and forth until the burner lights. Adjust the flame so that there is a tight blue cone in the center of the flame and a medium flame with no red or bright orange color.
3. Gently rub the toothpick along your gum line and between your teeth. DO NOT jab at your gum. You should not have any blood on your toothpick. Do not worry if you don't see anything.
4. Add a small drop of water to the center of the glass slide.
5. Rub the end of toothpick in the center of the glass slide. Spread the drop and sample to create a smear approximately 1.5 cm in diameter.
6. Allow the slide to air dry.
7. Pick up the slide with the slide clip or clothes pin. Pass the slide through the flame of the Bunsen burner 4 or 5 times to heat fix the slide. Heat fixing denatures proteins in the cells so that the cells stick to the glass slide. Do not hold the slide in the flame. This will incinerate your sample and may cause your slide to shatter. Once your slide is heat fixed, turn off the gas valve at the bench top.
8. Place the slide on the staining tray and allow it to cool for 2 minutes.
9. Apply methylene blue to the slide. Stain the slide for 5 minutes.

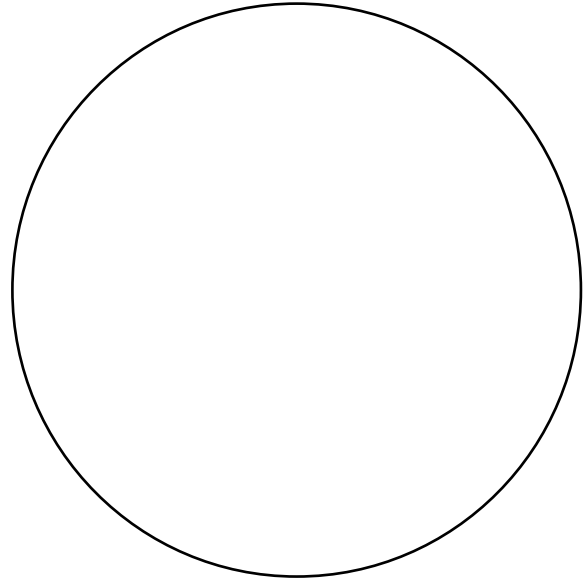
10. After 5 minutes tip the slide to drain the stain away. Gently wash the slide with distilled water for 20 seconds. Do not squirt water directly at the smear. Squirt the water above the smear and allow the water to wash down over the smear.
11. Place the slide in the book of bibulous paper or between several pieces of paper towel. Gently blot dry.
12. Place the slide on the microscope. Make sure your smear is closest to the objective and that you have not placed the slide on the stage upside down. Start observing the slide at 4x. Use the mechanical stage controls to move the slide from side to side and front to back. You are looking for what appears to be light blue dust. Remember, bacteria are very, very small. Once you have found some stain color on the slide, center the color in your field of view and increase the magnification. Fine adjust the slide to bring the image into sharp focus. Center the image again and increase the magnification to 400X. The bacterial shapes and arrangements (single cells, pairs of cells, chains or clusters of cells) should be visible.
13. After viewing your slide, dispose of the slide and the toothpick in the biohazard container.

You should be able to find bacteria and possibly epithelial cells. Draw what you see below. Remember to exaggerate the size of the specimen.



Magnification: _____

Describe the organisms shape & arrangement:



Magnification: _____

Describe the organisms shape & arrangement:

1. Why was it important to heat fix the slide?

2. Why was it important to stain the slide?
3. Go to the internet and look up the meaning of strepto- and coccus. Write the meaning of these word roots below.
4. From the results of your stain and your research on word roots, do you suspect *S. mutans* was present on your slide? Why or why not?