

Staining Human Cheek Cells

Background:

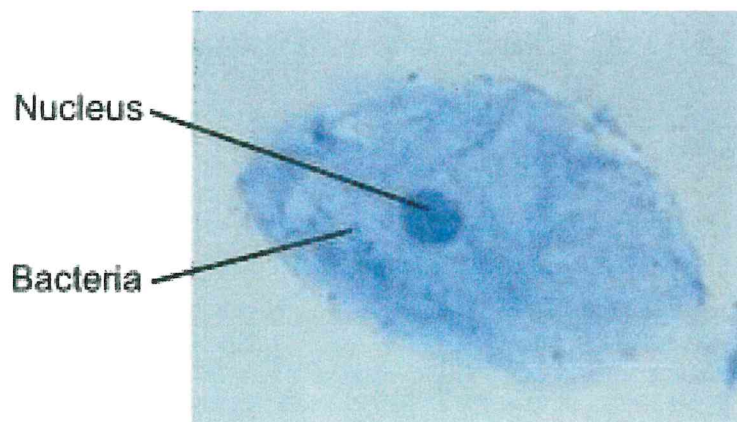
Methylene blue is widely utilized across fields such as chemistry, biology, medicine, and microbiology, serving primarily as a stain to enhance the visibility of tissues in animal, bacterial, and blood samples. This dye enables detailed examination of dead cells, cellular structures, and other tissues in both plants and animals. Biologists often rely on methylene blue to identify microorganisms, which are typically almost transparent. By applying a few drops of this dye to a microscope slide, the otherwise colorless bacterial structures become visible, facilitating their study. In the realm of biology, stains like methylene blue work by attracting and binding to chemicals within biological tissues, thereby highlighting them for observation.

Materials:

1. Glass microscope slides
2. Plastic cover slips
3. Paper towels or tissue
4. Methylene Blue solution (0.5% to 1%) mix approximately 1 part stock solution with 4 parts of water))
5. Plastic pipette or dropper
6. Sterile, individually packed cotton swabs
7. Microscope

Procedure

1. Take a clean cotton swab and gently scrape the inside of your mouth.
2. Smear the cotton swab on the center of the microscope slide for 2 to 3 seconds.
3. Add a drop of methylene blue solution and place a coverslip on top. Concentrated methylene blue is toxic if ingested. Wear gloves and do NOT allow children to handle methylene blue solution or have access to the bottle of solution.
4. Remove any excess solution by allowing a paper towel to touch one side of the coverslip.
5. Place the slide on the microscope, with 4 x or 10 x objective in position and find a cell.



6. Then view at higher magnification.

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Methylene blue stains negatively charged molecules in the cell, including DNA and RNA. This dye is toxic when ingested and it causes irritation when in contact with the skin and eyes.

The cells seen are squamous epithelial cells from the outer epithelial layer of the mouth. The small blue dots are **bacteria** from our teeth and mouth.

Alternative Procedure

Methylene blue is used in gram staining. Gram staining is one of the most important staining methods in microbiology. The predominant color in gram staining is methylene blue. Gram-positive organisms are organisms that preserve the primary color and look purple brown under a microscope. Gram-negative organisms that do not take up primary stains look red under a microscope.

Step 1: The first step in gram staining is to stain the slide with Methylene Blue dye.

Step 2: The following step, often known as fixing the dye, requires employing iodine to produce a Methylene blue-iodine combination to inhibit dye removal.

Step 3: Following that, a decolorizer, usually ethanol and acetone, are used to remove the dye. The capacity of the bacterial cell wall to retain the methylene blue after solvent treatment is the underlying concept of gram staining. Gram-positive bacteria contain more peptidoglycans, whereas gram-negative organisms have more lipids.

Step 4: Initially, all bacteria absorb the methylene blue stain; however, when a solvent is used, the lipid layer of gram-negative organisms is dissolved. Gram-negative bacteria lose the main stain as the lipid layer dissolves. In contrast, solvent dehydrates the gram-positive cell walls, limiting the passage of the methylene blue-iodine combination, and therefore the bacteria stay stained. **The duration of decolorization is an important stage in gram staining because extended contact to a decolorizing chemical can eliminate all stains from both kinds of bacteria.**

Step 5: The last phase in gram stain consists of using basic fuchsin stain to decolorize gram-negative bacteria to provide them with a pink color for better identification. It is sometimes referred to as counterstain. Safranin or basic fuchsin stains are used as a counterstain to help differentiate gram-positive and gram-negative bacteria.