ISSN: 2157-7471
Journal of Plant Pathology & Microbiology

OPEN ACCESS Freely available online

Commentary

Staining Methods: An Overview of Microorganisms and their Applications

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DESCRIPTION

Staining can be defined in microbiology as a technique used to highlight and contrast biological samples at the microscopic level. Staining and dyes are used to enhance specimens at the microscopic level for examination at higher magnification for histopathologic studies and diagnostic purposes. However, staining is not limited to biological samples. It can also be used to study the structure of crystalline polymers. Structural details in organisms cannot be seen with a light microscope due to the lack of contrast. Therefore, dyes are used to stain cells.

Pigments bind to cellular components to create color contrast and enhance visibility. Positively charged (cationic) dyes such as methylene blue, crystal violet, and safranin bind to negatively charged cellular components such as nucleic acids, acidic polysaccharides, and the cell surface of bacteria. Microscopy is a very important tool in microbiology, but it has limitations when it comes to observing cells in general and bacterial cells in particular.

The two most important concerns are resolution and contrast. Resolution is beyond our control, as most bacterial cells are already approaching the resolution limits of most light microscopes. However, contrast can be improved by using different optics. Phase contrast or differential interference contrast microscopy, or by staining the cells or background with a chromogenic dye that not only adds contrast but also gives them color. Microbiology has a wide variety of staining and staining processes. Some involve a single stain and only a few steps, while others use multiple stains and a more complex process. Before starting the staining process, the cells are mounted streaked on glass slides and fixed. A bacterial swab is a small amount of culture spread over a very thin membrane on the surface of the slide. To prevent bacteria from being washed away during the staining process, the smear can be chemically or physically "fixed" to the surface of the slide. Heat-fixing is a simple and efficient method and is accomplished by briefly passing the slides through the flame of a Bunsen burner. This allows the biological material to be almost permanently fixed to the glass surface.

Heat-fixed smears are ready for staining. Simple staining involves adding charge-attracting dyes cationic dyes such as methylene blue or crystal violet or charge-repelling dyes anionic dyes such as eosin or India ink to the smear. Cationic dyes bind to bacterial cells and are clearly visible against a bright background.

Anionic dyes are repelled by cells, causing them to lighten against a stained background. Perhaps the most important feature revealed when staining bacterial cells is cell morphology not to be confused with colony morphology, which is the appearance of bacterial colonies on agar plates. Most heterotrophic and culturable bacteria come in several basic forms. Spherical cells, rod-shaped cells, or rod-shaped cells with bends or twists. There are more diverse forms of archaea and other bacteria found in ecosystems outside the human body.

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Received: 03-Feb-2023, Manuscript No. JPPM-23-20078; **Editor assigned:** 06-Feb-2023, Pre QC No. JPPM-23-20078 (PQ); **Reviewed:** 21-Feb-2023, QC No. JPPM-23-20078; **Revised:** 28-Feb-2023, Manuscript No. JPPM-23-20078 (R); **Published:** 07-Mar-2023, DOI: 10.35248/2157-7471.23.14.660

Citation: Sprague C (2023) Staining Methods: An Overview of Microorganisms and their Applications. J Plant Pathol Microbiol. 14:660.