Perspective
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Microbiological Techniques for Micro-organisms: Staining Methods

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Perspective

Staining is a technique commonly used to improve sample contrast at the microscopic level. Dirt and dyes are widely used in the medical fields of histopathology (microscopy of living tissues) and histopathology, hematology, and cytopathology, which focus on the study and diagnosis of diseases at the microscopic level. Dyes can be used to define biological tissues (eg, to emphasize muscle fibers or connective tissue), cell populations (to classify different blood cells), or organelles within individual cells. Biochemistry requires the addition of class-specific dyes (DNA, proteins, lipids, carbohydrates) to the substrate in order to identify or quantify the presence of a particular compound. Staining and fluorescent labeling serve similar purposes. Biological staining is also used to label cells with flow cytometry and to label proteins and nucleic acids with gel electrophoresis. Optical microscopes are typically used to display stained samples at high magnification using brightfield or epi-fluorescent illumination. Staining is not limited to biological materials and can also be used to study the structure of other materials, such as the lamellar structure of partially crystalline polymers or the domain structure of block copolymers (*In vivo vs. in vitro*). *In vivo* staining (also called vital or *in vivo* staining) is the process of staining living tissue. The contrasting colors of certain cells or structures make it easy to identify and inspect their shape (morphology) or location within cells or tissues. Its usual purpose is to reveal cytological details that may not be visible otherwise. However, staining can also indicate where a particular chemical or reaction takes place within a cell or tissue.

In vitro staining includes staining of cells or structures that have been removed from the biological context. Certain stains are often combined to reveal more detail and features than a single stain alone. Scientists and clinicians can use these standard techniques as a consistent and reproducible diagnostic tool in combination with specific protocols for fixation and sample preparation. Counterstain is a stain that makes cells or structures more visible when the cells or structures are not completely visible in the main stain.

Crystal Violet stains both Gram-positive and Gram-negative bacteria. Treatment with alcohol removes crystal purple color only from Gram-negative bacteria. Safranin as a counterstain is used to stain Gram-negative bacteria that have been decolorized by alcohol. While living in *Ex-vivo*, many cells continue to live and metabolize until they are "fixed." Some dyeing methods are based on this property. Staining that is excluded from living cells but is taken up by dead cells is called vital staining (such as trypan blue or propidium iodide for eukaryotic cells). Those that invade living cells and stain them are known as supravital stains (eg, new methylene blue and brilliant cresyl blue for reticulocyte staining). However, these spots are ultimately more toxic to the organism and more toxic than the other spots. Partially due to toxic interactions within living cells, when supravital staining penetrates living cells, it can produce characteristic staining patterns that differ from staining of already fixed cells (eg,). Appearance of "reticulocytes" and diffuse "multicolored")). To achieve the desired effect, the stain is used in a very dilute solution of 1: 5000 to 1: 500,000. Note that many pigments can be used on both live and fixed cells.

Specific techniques

Gram staning: Primary stain involves crystal violet applied to film then treated with iodine (mordant), alcohol (decolourizer) and counter stained with safranin, characterizes bacteria in one of two groups, Gram positive or Gram negative. If Gram positive appears purple in color or Grams negative appears pink in color.

Endospore (Dornor's method): Primary stain involves malachite green heat fixed to penetrate spores; vegetative cells are counterstained with Safranin. This method detects the presence of endospores in six genera of bacteria. Identification of results if Endospores cells it is observed in green and vegetative cells are in red colour.

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