

Lactophenol Cotton Blue Staining of Yeast Cells

Introduction: Fungi are widespread in nature, and humans are regularly exposed to the spores from many species. The most commonly encountered fungal diseases are the superficial mycoses which affect the subcutaneous or horny layers of the skin or hair shafts, and cause conditions such as athlete's foot or ringworm. These dermatophytic fungi belong to the Microsporum, Trichophyton and Epidermophyton groups and may appear as yeasts or mycelial forms within the keratin.

When fungi grow in tissue, they may display primitive asexual (imperfect) forms which appear as either spherical yeast or spore forms. These features are important morphologically for identifying different types of fungi. Fortunately, most fungi are relatively large, and their cell walls are rich in polysaccharides.

Principle: The Lactophenol cotton blue solution is made up of a combination of three main reagents, of which Phenol acts as a disinfectant by killing any living organisms, Lactic acid preserves the fungal structures and cotton blue gives colour to the homopolysaccharide chitin present on the fungal cell wall and other fungal structures.

Materials required: Bunsen burner, microscope, glass slides, coverslips, inoculating loop, Lactophenol cotton blue stain and 70% ethanol.

Procedure: On a clean glass slide, a drop of 70% ethanol was taken. Then, the Yeast sample was added to the drop of alcohol with a sterile inoculation loop. The Yeast sample was teased with a needle mounter, to ensure the sample mixed well with the alcohol. One or two drops of Lactophenol Cotton Blue Solution were added with the help of a dropper to the sample before the ethanol dried off. Afterward, the stain was covered carefully with a clean sterile coverslip without making air bubbles to the stain. Then, the slide was focused under the low power objective (10X) of the microscope, followed by high power objective (40X) for a detailed study.

Observation: Fungi are eukaryotic organisms with both macroscopic and microscopic characteristics. Yeast cells with unbroken even edges and round, moist small colonies were observed.

Lactophenol Cotton Blue (LPCB) Staining



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