Nigrosin (10%)

Intended use
Nigrosin is used for negative staining of bacteria.

Summary
Nigrosin is a mixture of synthetic black dyes made by heating a mixture of nitrobenzene, aniline and aniline hydrochloride in the presence of a copper or iron catalyst. Nigrosin is used for negative staining of bacteria, as well as the capsule-containing fungus, *Cryptococcus neoformans*. The shapes and sizes of the organisms are seen as colour-free outlines against the dark background. Nigrosin consists of a suspension of fine particles of carbon. This forms a dark background, against which capsules are clearly seen because of displacement of carbon particles. An advantage of using this method, rather than regular positive stains like methylene blue or carbol fuchsin, is that prior fixation by heat or alcohol is not needed, so the organisms are seen in more lifelike shapes.

Principle
Negative staining is one of the many staining techniques that can be employed for viewing bacterial cell morphology and size. The advantages of the negative stain include the use of only one stain and the absence of heat fixation of the sample. Negative staining employs the use of an acidic stain and, due to repulsion between the negative charges of the stain and the bacterial surface, the dye will not penetrate the cell. In negative staining, the results yield to a clear cell with a dark background. The shapes and sizes of the organisms are seen as colour-free outlines against the dark background. Furthermore, negative staining with nigrosin can reveal some microorganisms that cannot be stained by regular methods.

Reagent /Contents
Nigrosin	10.0 g
Formalin	0.5 mL
Distilled water	100 mL

**Formula adjusted, standardized to suit performance parameters

Appearance: Blackish violet coloured solution.

Storage and stability
Store at 15-30°C away from bright light. Use before expiry date on label.

Materials required but not provided
Clean grease-free glass slide, loops, staining rack, Bunsen burner, blotting paper, immersion oil (Cat. No. 207090110025) and microscope.

Type of Specimen
Isolated colony of spore / capsule forming bacteria and fungi.
Clinical specimen: Blood, urine, CSF, pus, wounds, lesions, body tissues, sputum etc.
From environment: Air, water, soil, sludge, wastewater, food, dairy samples etc.

Procedure
1. Place a small drop of nigrosin stain on one side of the clean grease-free slide.
2. Aseptically transfer one loopful of the test sample or saline suspension of growth from culture plate or loopful of the culture into the drop of nigrosin stain and mix gently.
3. Use a second clean grease-free slide, place it over the first slide at an angle of 45-degree to the stain and spread the stain across the slide to make a smear.
4. Allow the slide to airdry and observe under oil immersion lens.

Note: Some breaking of the dye will occur, especially when the drop of dye is large. Breakage in dye will create bright spots and lines, should not be mistaken for bacteria when observing under microscope.

Interpretation of results
A dark background with clear halo surrounding the bacterial cells is observed.

Warranty
This product is designed to perform as described on the label and pack insert. The manufacturer disclaims any implied warranty for use and sale for any other purpose.

Reference
1. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product	Pack Size
207140210125	Nigrosin (10%)	125 mL
207140210250		250 mL
207140210500		500 mL