

TB AURAMINE O REAGENT

IVD *In vitro* diagnostic medical device



For use in TB-Stain Auramine O kit INSTRUCTIONS FOR USE

REF Catalogue number: TBAO-OT-100 (100 mL) TBAO-OT-250 (250 mL) TBAO-OT-500 (500 mL) TBAO-OT-1L (1000 mL) TBAO-OT-2.5L (2500 mL)

Introduction

Many bacterial cells are easily stained by using simple dyes or Gram stain. However, a few strains of bacteria, such as *Mycobacteria* and *Nocardia* cannot be stained using simple dyes (the results may vary significantly if successfully stained). Cell walls of the *Mycobacteria* strain contain fatty acids which make them hard to stain. In order to stain the cell wall, a higher concentration of dye or a longer period of heating is required. However, once stained, the dye is ever more difficult to remove from the cells. Those bacteria are called acid-fast because they retain their primary color even after being treated with acid alcohol (3% HCl alcohol solution). Fluorescence has been used to detect acid-fast bacteria for many years. This method is more sensitive than the Kinyoun method. It takes less time to interpret the results. Auramine O, Acid alcohol as a differentiation medium and potassium permanganate as a counterstain are used in this method.

Product description

- **TB AURAMINE O REAGENT** - For staining acid-fast bacteria using fluorescence method

Other slides and reagents that may be used in staining:

- High-quality glass slides for use in histopathology and cytology, such as VitroGnost SUPER GRADE or one of more than 30 models of VitroGnost glass slides
- BioGnost's immersion media, such as Immersion oil, Immersion oil, types A, C, FF, 37, or Immersion oil Tropical Grade

Preparing the section for staining

Note: preparation of the section depends on the type of the section.

Preparation is not necessary for all types of samples. The samples must be fixated using heat using Bunsen burner or oven.

SPUTUM

- The sputum sample must be treated with a preparation containing hypochlorite in order to isolate the mycobacteria from the surrounding mucus.

LUMBAR PUNCTION SAMPLES, SEDIMENTS

- After finishing the process of enrichment, smear the sample on the glass slide and let it dry.

HISTOLOGICAL SECTIONS

- Clear the sample with intermedium; in xylene (BioClear) or in a xylene substitute (BioClear New).
- Rehydrate the section through series of descending alcohol solutions (Histanol 100, Histanol 95).

Sample staining procedure

	Solution	Preparation time (min)
1.	Apply TB Auramine O reagent	15
2.	Rinse with tap water	10
3.	Apply TB Decolorizer Fluorescent	1
4.	Rinse with tap water	5
5.	Apply TB Permanganate reagent	5
6.	Rinse with tap water and dry (except during histology sections staining)	5
	Note: do not dry the samples for histology sections; dehydrate it through series of ascending alcoholic solutions (Histanol 95, Histanol 100) and clear the section with intermedium; xylene (BioClear) or xylene substitute (BioClear New). Apply an appropriate BioMount medium for cover glass covering/mounting on the section immediately after clearing .	

Results

Acid-fast bacteria - yellow-green (viewed through microscope with a fluorescent filter)

Background - black

Note

Time periods of staining procedures are not standardized. Intensity of staining depends on the period of immersion in the dye. Real staining protocol depends on personal requests and priorities.

Preparing the sample and diagnostics

Use only appropriate instruments for collecting and preparing the samples. Process the samples with modern technology and mark them clearly. Follow the manufacturer's instructions for handling. In order to avoid mistakes, the staining procedure and diagnostics should only be conducted by authorized and qualified personnel. Use only microscope according to standards of the medical diagnostic laboratory. In order to avoid an erroneous result, a positive and negative check is advised before application.

Safety at work and environmental protection

Handle the product in accordance with safety at work and environmental protection guidelines. Used solutions and out of date solutions should be disposed of as special waste in accordance with national guidelines. Chemicals used in this procedure could pose danger to human health. Tested tissue specimens are potentially infectious. Necessary safety measures for protecting human health should be taken in accordance with good laboratory practice. Act in accordance with signs and warnings notices printed on the product's label, as well as in BioGnost's material safety data sheet.














Storing, stability and expiry date

Keep the TB Auramine O reagent in a tightly sealed original packaging at temperature of +15°C to +25°C. Do not keep in cold places, do not freeze and avoid exposing to direct sunlight. Date of manufacture and expiry date are printed on the product's label.

References

1. Madison B (2001). "Application of stains in clinical microbiology". *Biotech Histochem* **76** (3): 119-25.
2. Ryan KJ, Ray CG (editors) (2004). *Sherris Medical Microbiology* (4th ed.). McGraw Hill.
3. Margaret A. Bartelt, 2000: Diagnostic Bacteriology: A Study Guide, F.A. Davis Company.

TBAO-OT-X, V2-EN2, 03 July 2019, AK/IŠP

	Refer to the supplied documentation		Storage temperature range		Number of tests in package		Product code		European Conformity
	Refer to supplied instructions		Keep away from heat and sunlight		Valid until		Lot number		Manufacturer
	For <i>in vitro</i> diagnostic use only		Keep in dry place		Caution - fragile				



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