Drucker Diagnostics
Innovative Solutions for a Healthier World

QBC *F.A.S.T.*™ Auramine O Stain Kit

Instruction Manual

CE

4277-400-053 Rev. I
Revised 2018-11-20

English

QBC *F.A.S.T.*™ Auramine O Stain Kit

Intended Use

For use as a stain for smears made from patient specimens or cultures in the detection or characterization of acid fast bacilli such as *Mycobacterium tuberculosis*.

Summary and Principles

The worldwide incidence of tuberculosis has been on an increasing trend since at least 1990, when the World Health Organization began tracking incidence data[1]. Early and accurate detection of tuberculosis is critical for both effective control and treatment of the disease. The most common method for detection of *Mycobacterium tuberculosis* is the use of sputum smear microscopy[1], which can provide both an initial presumptive diagnosis as well as a quantification of the mycobacterial load.

Acid fast bacilli, such as *Mycobacterium tuberculosis*, can be stained by aniline dyes and are resistant to decolorization by acid and alcohol. When followed by a counterstain, this treatment results in the acid fast bacilli staining with contrast to other organisms and debris that have retained only the counterstain. However, the staining methods classically used for acid fast microscopy result in a smear that can be difficult and time consuming to read. Auramine O and auramine-rhodamine stains have been successfully used for fluorescence based microscopy of mycobacteria. Reports of mechanism of staining are conflicting; these include Auramine O binding to the cell wall of the mycobacteria[2] and the stain binding

to "most if not all" the Auramine O binding to the nucleic acid in the mycobacteria[3]. It has been demonstrated, though, that Auramine O based staining methods are more sensitive than light microscopy methods for the detection of acid fast bacteria[4]. This increase in sensitivity is due, in large part, to the significant contrast the fluorescent stains impart to the acid fast bacilli, which appear green against a dark background. This increase in distinction permits the use of objectives with larger fields-of-view, thereby decreasing the total examination time.

While standard auramine staining methods are a significant improvement over classical methods, the staining process is still time consuming. The QBC *F.A.S.T.* Auramine O Stain Kit simplifies the process further by using a proprietary combination of quenching agent and decolorizer to make a rapid four step process that takes just over two minutes to complete.

Contents

The contents of this kit include the following items:

- QBC *F.A.S.T.* Auramine O Stain (120 mL, 250 mL, or 3.8 L)
- QBC *F.A.S.T.* Decolorizer/Quencher (120 mL, 250 mL, or 3.8 L)
- Package Insert

Warnings and Precautions

For *in vitro* diagnostic use

Human clinical specimens can harbor infectious diseases such as the causative agents of tuberculosis, hepatitis, human immunodeficiency virus (HIV) and others. Universal Precautions local guidelines and regulations should be followed when handling

clinical specimens. All activities that could generate aerosols from clinical specimens should be performed in a biosafety cabinet. Activities that involve culturing of *Mycobacterium tuberculosis* should be performed using Biosafety Level 3 procedures and practices

The chemicals in this kit are hazardous and can be harmful or fatal. Use in a well ventilated area with proper personal protective equipment. Keep product away from open flames. Consult kit MSDS for additional information regarding safety and disposal.

This product is designed to aid in the detection of acid fast bacilli. Sputum smear microscopy and the procedures involved with sample preparation and processing for AFB detection should be performed only by those trained in the techniques involved as well as general laboratory practices and procedures.

Storage Instructions

Store at 2-25ºC. Keep stain protected from light. Do not use beyond expiration date.

Staining Procedure

The *F.A.S.T.* Auramine O Stain Kit can be used with direct and concentrated sputum and culture specimens.

1. Heat fix slide containing specimen smear
2. Cover smear with *F.A.S.T.* Auramine O Stain and let stand for 1 minute
3. Rinse smear gently with deionized or tap water and drain
4. Cover smear with *F.A.S.T.* Decolorizer/Quencher and let stand for 1 minute

5. Rinse smear gently with deionized or tap water and drain Dry slide
6. Examine slide using the QBC ParaLens or equivalent fluorescence microscopy apparatus

Quality Control

QBC Diagnostics Quality Control Slides (Catalog Number 427402) should be used routinely to assess the reagents and staining procedure. Quality control should be run in accordance with applicable governing regulations.

If quality control fails, do not report patient results until cause of failure is rectified. A positive control failure can be indicative of stain reagent degradation. If stain is source of no or low-fluorescing positive control, do not use stain for patient specimens

Expected Results

Mycobacteria and other acid fast bacilli fluoresce green against a dark background when viewed with a fluorescence microscope having a blue excitation and green emission filter configuration (e.g., Excitation filter: 435 - 480 nm; Emission filter: 510 600 nm). All other organisms will not be visible. A fluorescent bacillus is a presumptive identification of *Mycobacterium* spp.

Limitations

Some rapid growing mycobacteria may not fluoresce with this stain. Ziehl-Neelsen, Kinyoun, or other methods should be used on these specimens. Fluorescence of smears will wane over time, so stained specimens should be examined as soon as possible. While a positive result provides evidence of mycobacteria, a negative result does not rule out an infection. Other diagnostic methods such as

culture or PCR should be used for positive identification.

F.A.S.T. Stain reagents can degrade when exposed to excessive heat. Always perform quality control to determine integrity of the stain before reporting patient results. Do not use stain if quality control fails.

Equipment Required But Not Provided

The QBC *F.A.S.T.* Auramine O Stain Kit is designed to work with a fluorescent microscope system capable of exciting specimens from 425-480 nm and transmitting fluorescence of at least 510-600 nm. Additional equipment includes slides, slide warmer or Bunsen burner.

References

1. World Health Organization. (2009) Global tuberculosis control: epidemiology, strategy, financing: WHO Report 2009. WHO Press, Geneva, Switzerland.
2. Baron, E.J. and S.M. Finegold. (1990) Baily & Scott's Diagnostic Microbiology, 8th Edition. The CV Mosby Company, Baltimore, Maryland.
3. Steingart K. R., et al. (2007) Fluorescence versus conventional for sputum smear microscopy for tuberculosis: a systematic review. *Lancet Infectious Disease* 6:570-81.
4. Hanscheid, T. (2008) The future looks bright: low-cost fluorescenct microscopes for detection of *Mycobacterium tuberculosis* and *Coccidiae*. *Transactions of the Royal Society of Tropical Medicine and Hygiene*. Elsevier Ltd., 520-521.

Ordering Information

QBC *F.A.S.T.* Auramine O Stain Kits:

Size	**Catalog No.**
Trial Kit	427422
120 mL Kit	427404
250 mL Kit	427424
3.8 L Kit	427425

Drucker Diagnostics
200 Shady Lane, Suite 170, Philipsburg, PA 16866 U.S.A.
Tel: +1-814-692-7661, www.druckerdiagnostics.com

EC REP
Emergo Europe
Prinsessegracht 20, 2514 AP The Hague, The Netherlands
Tel: (31) (0) 70-345-8570, Fax: (31) (0) 70-346-7299


Manufacturer

EC REP Authorized Representative in the European Community


Use By

REF Catalog Number

IVD In Vitro Diagnostic Medical Device


Temperature Limitation

LOT Batch Code (Lot)


Consult Instructions for Use


Caustic


Flammable