

# TB CARBOL FUCHSIN REAGENT

IVD *In vitro* diagnostic medical device



For use in TB-Stain Cold and TB-Stain Hot kit

## INSTRUCTIONS FOR USE

REF Catalog number: TBC-OT-100 (100 ml) TBC-OT-250 (250 ml) TBC-OT-500 (500 ml) TBC-OT-1L (1000 mL) TBC-OT-2.5L (2500 mL)

### Introduction

TB Carbol Fuchsin Kinyoun reagent is used in staining acid-fast bacteria such as *Mycobacteria* and *Nocardia* strains that cannot be stained using simple dyes (or provide different results if successfully stained). Cellular wall of the *Mycobacteria* strain contains waxy substance - mycolic acid. Those are beta-hydroxy carboxylic acids with chains containing up to 90 carbon atoms. Its resistance to acidity is associated with mycolic acid chain length. TB Carbol Fuchsin contains phenol that allows the dye to penetrate the mycobacterium cell wall by heating, and the dye is almost impossible to remove from the cell during decolorization. The reagent is used for special staining according to Ziehl-Neelsen which is the most renowned and most widely used method of identification of tuberculosis bacteria.

### Product description

- **TB CARBOL FUCHSIN REAGENT** - Primary stain reagent for identification of acid-fast bacteria

### Other slides and reagents that may be used in staining:

- Glass slides used in microbiology, such as VitroGnost ECONOMY GRADE or glass slides used in cytology, such as VitroGnost STANDARD GRADE or high quality glass slides used in histopathology, such as VitroGnost SUPER GRADE or one of more than 30 models of VitroGnost glass slides
- Decolorizer solution for use in staining methods according to Ziehl-Neelsen, such as BioGnost's TB Decolorizer
- Counterstain solution for use in staining methods according to Ziehl-Neelsen, such as BioGnost's Methylene Blue Loeffler reagent
- Counterstain solution for use in staining methods according to Kinyoun, such as BioGnost's TB Malachite Green reagent
- BioGnost's immersion media, such as Immersion oil, Immersion oil, types A, C, FF, 37, or Immersion oil Tropical Grade

### Preparing the sample for staining

- Transfer the sample on a clean glass slide using a sterilized smear loop  
Note: Acceptable samples include sputum, lumbar puncture sample, sediment or a histological section
- Spread the sample evenly across the glass slide using 1-2 drops of saline solution
- Fix the sample using the Bunsen burner after drying by wriggling the glass slide through the cone of flame for 2-3 times  
Note: Samples can be fixated in an oven at temperature 100°C-110°C for 20 min
- Cool the glass slide and begin the process of staining  
Note: If the sample is a histological section, it should be applied using standard histological techniques

### Sample staining procedure

#### According to Ziehl-Neelsen

1.	Cover the samples completely with the TB Carbol Fuchsin reagent. Carefully heat the glass slide containing the sample and dye on the bottom side of the slide using the Bunsen burner until evaporation occurs. Keep the slide hot for 5 min. Do not let the dye boil.	5 min
2.	Rinse with tap water until the dye destains.	
3.	Cover the sample using using TB Decolorizer and let it set for 15-30 seconds (depending on the sample thickness).	15-30 seconds
4.	Rinse with tap water.	
5.	Stain the sample using BioGnost's Methylene Blue Loeffler reagent	30 seconds
6.	Rinse with tap water thoroughly.	
7.	Dry the section	

#### According to Kinyoun

1.	Cover the samples completely with the TB Carbol Fuchsin reagent.	5 min
2.	Rinse with tap water until the dye destains.	
3.	Cover the sample using using TB Decolorizer and let it set for 15-30 seconds (depending on the sample thickness).	15-30 seconds
4.	Rinse with tap water.	
5.	Stain with TB Malachite Green reagent	60 seconds
6.	Rinse with tap water thoroughly.	
7.	Dry the section	

### Result

Acid fast bacteria - red

Background - blue

### Note

Microbiology staining procedures are not standardized and they depend on standard operating procedures of individual laboratories and the experience of the personnel conducting the staining procedure. Intensity of staining depends on the period of immersion in the dye. Depending on personal requests and standard laboratory operating procedures, sample processing and staining can be carried out according to other protocols.

### 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 TB Carbol Fuchsin reagent in a tightly sealed original packaging at temperature of 15 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* (4<sup>th</sup> ed.). McGraw Hill.
3. Margaret A. Bartelt, 2000: Diagnostic Bacteriology: A Study Guide, F.A. Davis Company.

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	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				



BIOGNOST Ltd.  
Medjugorska 59  
10040 Zagreb  
CROATIA  
[www.biognost.com](http://www.biognost.com)

