FUCHSIN ACID REAGENT

IVD *In vitro* diagnostic medical device CE

For use in Mallory Trichrome kit

INSTRUCTIONS FOR USE

REF Catalogue number: FA-OT-100 (100 mL)

Introduction

Fuchsin Acid reagent is used a component of Mallory Trichrome kit. Mallory trichrome staining kit is used for treating the tested microscopic sample using three different stainings with differential counterstaining of two basic parts of the tissue (muscle and collagen fibers) in focus. By staining the sample with Fuchsin Acid acidic dye nuclei and muscles are stained red to pink. Phosphotungstic acid molecule then shuts out Fuchsin Acid dye molecules from collagen and thus enables Aniline Blue to bind, resulting in collagen being stained contrast blue in relation to previously used red dye. Orange G (molecule of the lowest molar mass) stains erythrocytes.

Product description

- **Fuchsin Acid reagent** – aqueous solution of optimally concentrated Fuchsin Acid dye.

Use of Fuchsin Acid reagent as a component of Mallory Trichrome kit:

Other sections and reagents that may be used in staining:

- Fixatives such as BioGnost's neutral buffered formaldehyde solutions: Formaldehyde NB 4%, Formaldehyde NB 10%
- Dehydrating/rehydrating agent, such as BioGnost's alcohol solutions: Histanol 70, Histanol 80, Histanol 95 and Histanol 100
- Clearing agents, such as BioClear xylene or a substitute, such as BioClear New agent on the aliphatic hydrocarbons basis
- Infiltration and fitting agent, such as BioGnost's granulated paraffin BioWax Plus, BioWax 56/68, BioWax Blue, BioWax Micro.
- High-quality glass slides for use in histopathology and cytology, such as VitroGnost SUPER GRADE, VitroGnost COLOR or one of more than 30 models of BioGnost's VitroGnost glass slides

Preparing the histological sections for staining

- Fixate the sample (Formaldehyde NB 4%, Formaldehyde NB 10%), rinse with water and dehydrate through series of ascending alcohol solutions (Histanol 70, Histanol 80, Histanol 95 and Histanol 100).
- Clear the sample with intermedium; in xylene (BioClear) or in a xylene substitute (BioClear New).
- Infiltrate and fit the sample in paraffin (BioWax Plus, BioWax 56/58, BioWax Blue, BioWax Micro).
- Cut the paraffin block to 4-6 μm slices and place them on a VitroGnost glass slide.

Sample staining procedure

Pour the reagents into glass staining jars (Coplin, Hellendahl or Schifferdecker), return to original bottles after staining. Close tightly. Filter the reagents if necessary.

1.	Deparaffinize the section in xylene (BioClear) or in a xylene substitute (BioClear New)	3 exchanges, 2 min each
2.	Rehydrate using 100% alcohol (Histanol 100)	2 exchanges, 5 and 3 min
3.	Rehydrate using 95% alcohol (Histanol 95)	2 min
4.	Rehydrate in distilled (demi) water	2 min
5.	Immerse into Fuchsin Acid reagent	30 seconds
6.	Rinse in distilled (demi) water	until the excessive dye is washed off of the section
7.	Immerse into Phosphomolybdic acid, 1% solution	3 min
8.	Remove excessive reagent from the section using filter paper (without rinsing in distilled water)	
9.	Immerse in Orange G/Aniline Blue reagent	4-6 minutes
10.	Dehydrate using 70% alcohol (Histanol 70)	5 dips
11.	Dehydrate using 95% alcohol (Histanol 95)	5 dips
12.	Dehydrate using 100% alcohol (Histanol 100)	2 min
13.	Clear the section in xylene (BioClear) or in a xylene substitute (BioClear New)	2 exchanges, 2 min each

Immediately after clearing apply an appropriate BioMount medium for covering/mounting on the section. If BioClear xylene was used, use one of BioGnost's mounting xylene-based media (BioMount, BioMount High, BioMount M, BioMount DPX, BioMount C, or universal BioMount New). If BioClear New xylene substitute was used, the appropriate covering agent is BioMount New. Cover the section with a VitroGnost cover glass.

Result

Muscle fibers, cytoplasms, nuclei - red to pink
Collagen - blue
Erythrocytes - orange to red

Note

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.

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 Fuchsin Acid reagent in a tightly sealed original packaging at temperature of 15 to 25°C. Keep in dry 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. Melis, M., Carpino, F., Di Tondo, U. (1989), Tecniche in anatomia patologica, Edi Ermes, Milano.
2. Prophet, E.B., Mills, B., Arrington, J., Sobin, L. (1968), Laboratory methods in histotechnology, McGraw Hill, Washington D.C.
3. Bancroft, J.D., Gamble, M. (2002), Theory and practice of Histological Techniques, Churchill Livingstone, New York.

FA-X, V2-EN2, 13 February 2017, AK/VR

	Refer to the supplied documentation	°C	Storage temperature range		Number of tests in package	REF	Product code	CE	European Conformity		BIOGNOST Ltd. Medjugorska 59 10040 Zagreb CROATIA www.biognost.com
	Refer to supplied instructions		Keep away from heat and sunlight		Valid until	LOT	Lot number		Manufacturer		
IVD	For *in vitro* diagnostic use only		Keep in dry place		Caution - fragile						