

BIOSCHIFF REAGENT

IVD In vitro diagnostic medical device



Schiff's reagent for detection of aldehydes and mucous substances

For use in microscopy and electrophoresis

INSTRUCTIONS FOR USE

REF Product code: BS-OT-30 (30 mL) BS-OT-100 (100 ml) BS-OT-500 (500 ml) BS-OT-1L (1000 mL) BS-OT-2.5L (2500 mL)

Introduction

BioSchiff reagent is a colorless solution that changes to violet (magenta) in the presence of aldehydes. The intensity of the color obtained depends on the amount of reactive glycol structures in the tissue. It is prepared by the reduction of pararosaniline by using sulfuric acid. Schiff's reagent is used with various chemical methods, and one of the most common and most widely used ones is the PAS staining (Periodic Acid Schiff). The PAS staining is based on oxidation reaction with the presence of periodic acid and Schiff's reagent. Periodic acid makes the molecules containing glycol groups create aldehydes affected by Schiff's reagent that stains them violet (magenta). This method is most commonly used in liver and muscle cells testing. Schiff's reagent can be used for DNA detecting according to Feulgen.

Product description

- **BIOSCHIFF REAGENT** - Pararosaniline, hydrochloric acid and sodium metabisulfite solution with added stabilizer.

Other slides and reagents that may be used in staining:

- Fixative such as BioGnost's neutral buffered formalin: Formaldehyde NB 4%, Formaldehyde NB 10%
- Dehydrating/rehydrating agent, such as BioGnost's alcohol solutions: Histanol 70, Histanol 80, Histanol 95 and Histanol 100
- Histopathology staining reagent, such as BioGnost's Hematoxylin G3, Hematoxylin ML
- Oxidation reagent, such as BioGnost's Periodic acid, 0.8% solution
- Reagents for preparing sulfite solution, such as BioGnost's Sodium metabisulphite, solution and HCL reagent, P.A.S.
- Differentiation reagent, such as BioGnost's Clearing agents, such as BioClear xylene or a substitute, for instance BioNene on the limonene basis or 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 or one of more than 30 models of BioGnost's glass slides
- Covering agents for microscopic sections and mounting cover glass, such as BioGnost's BioMount, BioMount High, BioMount M, BioMount New, BioMount DPX, BioMount DPX High, BioMount DPX Low, BioMount C, BioMount Aqua, Canada Balsam or MountQuick Tube medium
- VitroGnost cover glass, dimensions range from 18x18 mm to 24x60 mm
- Immersion oils, such as BioGnost's Immersion oil, Cedarwood oil, Immersion oils types A, B, NVH, FF and 37

Preparation of additional solutions used in staining

- Sulfite solution
Mix 10 mL of sodium metabisulphite solution with 10 mL of HCL reagent, P.A.S., add 200 mL of tap water, then mix.

Preparing 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 (BioNene, BioClear New)
- Infiltrate and fit the sample in paraffin (BioWax Plus, BioWax 56/58, BioWax Blue, BioWax Micro)
- Cut the paraffin block to 3-5 μ m slices and place them on a VitroGnost glass slide

The PAS histologic sections staining procedure

1.	Deparaffinize the section in xylene (BioClear) or in a xylene substitute (BioClear New)	3 exchanges, 10 min each
2.	Rehydration 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.	Rinse in tap water	5 min
6.	Treat with Periodic acid, 0.8% solution	5-10 minutes
7.	Rinse in distilled (demi) water	
8.	Treat with BioSchiff reagent	10-15 minutes
9.	Treat with sulfite solution	3 exchanges, 2 min each
11.	Rinse under tap water	3 min
12.	Staining with Hematoxylin G3 or Hematoxylin ML	10 seconds to 2 min
13.	Rinse under tap water	3 min
14.	Dehydration using 70% alcohol (Histanol 70)	2 exchanges, 30 seconds each
15.	Dehydration using 95% alcohol (Histanol 95)	2 exchanges, 30 seconds each
16.	Dehydration using 100% alcohol (Histanol 100)	30 seconds
17.	Dehydration using 100% alcohol (Histanol 100)	2 min
18.	Clear the section in xylene (BioClear) or in a xylene substitute (BioClear New)	2 exchanges, 5 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.

Note

Time periods of staining processes are not entirely standardized and they approximately correspond to clinical and laboratory practical experience. Intensity of staining depends on the period of immersion in the dye. Real staining protocol depends on personal requests and priorities.

Result

Blue - nuclei

Violet - polysaccharides, glycogen, neutral mucopolysaccharides, mucoproteins, glycoproteins, glycolipids, phospholipids, basement membrane, collagen

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 BioSchiff reagent in a tightly closed original package at a room temperature. Do not freeze and avoid exposing to direct sunlight. Date of manufacture and expiry date are printed on the product's label.

References

1. Bancroft, J. D. et Gamble, M. (2002): Theory and Practice of Histological Techniques, 5th ed., Churchill Livingstone, London.
2. Kiernan, J.A. (1999): Histological and histochemical methods: Theory and practice, 3rd ed., Butterworth Heinemann, Oxford, UK.
3. Kodousek, R. (1969): A new, rapid method of preparing Schiff's reagent, *Histochemical Journal*, 1, p 277-278.

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	Refer to the supplied documentation		Storage temperature range		Number of tests in package		Product code		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 number		Manufacturer		
	For <i>in vitro</i> diagnostic use only		Keep in dry place		Caution - fragile						