BIOGNOST®

HCL REAGENT, HEMOGNOST PERLS

IVD In vitro diagnostic medical device

For use with special staining kits in histopathology

INSTRUCTIONS FOR USE

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

HCLH-0T-250 (250 mL)

Introduction

HCL reagent, HemoGnost Perls is used as a part of HemoGnost Perls kit for detection of free ferric ions (Fe³⁺) (not bound to hemoglobin) in cells. It is most often used in bone marrow and spleen cells.

Product description

• HCL REAGENT HEMOGNOST PERLS – Differentiation reagent based on hydrochloric acid for HemoGnost Perls kit.

Example of use of HCL reagent, HemoGnost Perls as a component of HemoGnost Perls 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.
- Covering agents for microscopic sections and mounting cover glass, such as BioGnost's BioMount, BioMount High, BioMount M, BioMount New, BioMount New, BioMount DPX, BioMount DPX High, BioMount DPX Low, BioMount DPX Low, BioMount DPX Low, BioMount C, BioMount Aqua, Canada Balsam
- 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
- BioGnost's immersion media, such as Immersion oil, Immersion oil, types A, C, FF, 37, or Immersion oil Tropical Grade
- Other components of HemoGnost Perls kit: Potassium hexacyanoferrate, solution and Nuclear Fast Red (Kernechtrot) reagent

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 μ m 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.	Mix 30 mL of Potassium hexacyanoferrate, solution and 30 mL of HCL reagent, HemoGnost Perls. Treat the sections with the prepared solution.	20 min
6.	Note: Use fresh solution, discard after use. Carefully rinse in distilled water	
7.	Stain using Nuclear Fast Red (Kernechtrot) reagent	5 min
8.	Rinse in distilled water	
9.	Dehydrate using 70% alcohol (Histanol 70)	2 exchanges, 1 min each
10.	Dehydrate using 95% alcohol (Histanol 95)	2 exchanges, 1 min each
11.	Dehydrate using 100% alcohol (Histanol 100)	2 exchanges, 1 min each
12.	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.

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.

Results

Blue - reactive ferric ions Red - nuclei Pink – cytoplasm

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

Store HCL reagent, HemoGnost Perls in a tightly closed original packaging at temperature stated on the product label. Do not freeze and avoid exposing to direct sunlight. Date of manufacture and expiry date are printed on the product's label.

References

- 1. Culling, C.F.A. (1974): Handbook of histopathological and histochemical techniques, 2nd ed., Butterworth, London, UK.
- 2. Sheehan D.C. et Hrapchak, B.B. (1980): Theory and Practice Histotechnology, 2nd ed., CV Mosby, St. Louis, (MO), pp 52, p 14-167.

HCLH-X, V	1-EN1, 25	February	2019, IŠP/VR

	Refer to the supplied documentation	°C-	Storage temperature range	Σ	Number of tests in package	REF	Product code	C	E	European Conformity	BIOGNOST Ltd. Medjugorska 59 10040 Zagreb	()
[]i]	Refer to supplied instructions	*	Keep away from heat and sunlight		Valid until	LOT	Lot number			Manufacturer	CROATIA www.biognost.com	
IVD	For <i>in vitro</i> diagnostic use only	Ť	Keep in dry place	4	Caution - fragile							