BIOGNOST®

NUCLEAR FAST RED (KERNECHTROT) REAGENT

IVD *In vitro* diagnostic medical device CE

Reagent for red counterstaining cellular nuclei

INSTRUCTIONS FOR USE

REF Catalogue number: KR-OT-100 (100 mL) KR-OT-250 (250 mL) KR-OT-500 (500 mL)

Introduction

Nuclear Fast Red (Kernechtrot) reagent is primarily used as counterstain solution in certain special kits for staining nuclei using red dye. It most commonly binds with aluminum ions and it is used for staining sperm and spermatids in ejaculate, as well as for identifying PCR tubes in tissue sample embedded in paraffin.

Product description

- **NUCLEAR FAST RED (KERNECHTROT) REAGENT** – Nuclear Fast Red (Kernechtrot) stain solution with aluminum sulfate

NOTE: Nuclear Fast Red (Kernechtrot) reagent is a component of several BioGnost's special kits (HemoGnost Perls kit, Alcian Blue pH 2.5 kit, Reticulin kit, Von Kossa kit). Staining procedure using HemoGnost Perls kit is described below. If you are interested in other staining protocols using special stains using Nuclear Fast Red (Kernechtrot) reagent, feel free to contact us.

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 52/54, 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 Low, BioMount DPX, BioMount DPX High, BioMount DPX Low, BioMount DPX Low Eco, 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
- VitroGnost cover glass, dimensions range from 18x18mm to 24x60mm
- BioGnost's immersion media, such as Immersion oil, Immersion oil, types A, C, FF, 37, or Immersion oil Tropical Grade

Preparing histological sections for staining

- Fix the tissue sample tightly (4% NB Formaldehyde, 10% NB Formaldehyde), 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 52/54, BioWax Plus 56/58, BioWax 56/58, BioWax Blue, BioWax Micro).
- Cut the paraffin block to 4-6 μm slices and place them on a VitroGnost glass slide.

Histological sections staining procedure

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. Note: Use fresh solution, discard after use.	20 min
6.	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 Nuclear Fast Red (Kernechtrot) reagent in a tightly closed original packaging at temperature between 15°C and 25°C. 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.

KR-X, V2-EN2, 15 February 2017, AK/VR

	Refer to the supplied documentation		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						