Microscopy

Pyronine G (C.I. 45005)

for microscopy Certistain®



In Vitro Diagnostic Medical Device



25 g

for DNA and RNA staining in methyl green-pyronine G staining in histological sections

This staining dye "Pyronine G (C.I. 45005) - for microscopy Certistain®" is used for human-medical cell diagnosis and serves the purpose of the histological investigation of sample material of human origin. It is a dry staining dye that is used to prepare a staining solution, that when used together with other in vitro diagnostic products from our portfolio makes target structures in histological specimen materials (by fixing, where necessary embedding, staining with the above pyronine G solution, counterstaining, mounting) evaluable for diagnostic purposes.

Principle

Pyronine G belongs to the group of xanthene dyes.

Pyronine G is used together with methyl green in a one-step method for staining of DNA (green) and RNA (red).

Also in flow cytometry, pyronine G is described as a dye for staining RNA and DNA.

Sample material

Sections of Carnoy-fixed, paraffin-embedded tissue (3 - 5 μm thick paraffin sections) are used as starting material.

Reagents

Cat. No. 1.07518.0025

Pyronine G (C.I. 45005)

for microscopy Certistain®

Color Index No.: 45005

Color Index Name: -

Also required:

Cat. No.	100063	Acetic acid (glacial) 100% anhydrous for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 l, 2.5 l
Cat. No.	100983	Ethanol absolute for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 I, 2.5 I, 5 I
Cat. No.	101990	1-Butanol for analysis EMSURE® ACS,ISO,Reag. Ph Eur	l, 2.5 l, 4 l, 10 l
Cat. No.	102445	Chloroform 1 for analysis EMSURE® ACS,ISO,Reag. Ph Eur	l, 2.5 l, 4 l, 10 l
Cat. No.	115944	Methyl green zinc chloride double salt (C.I. 4259 for microscopy Certistain®	90) 25 g

Sample prepration

The sampling must be performed by qualified personnel.

Fixation

Fix histological slides in the conventional manner with Carnoy's fixing solution. For preparation of Carnoy's fixing solution mix:

Ethanol absolute	6 parts
Chloroform	3 parts
Acetic acid 100 %	1 part
mix	

All samples must be treated using state-of-the-art technology.

All samples must be clearly labeled.

Suitable instruments must be used for taking samples and their preparation. Follow the manufacturer's instructions for application / use.

Reagent preparation

Methyl green solution

For preparation of approx. 100 ml solution mix:

Methyl green zinc chloride double salt (C.I. 42590) Certistain®	2 g	
Distilled water (warm)	100 ml	
dissolve		
extract with chloroform and discard the organic phase		
filter the aqueous staining solution		

Pyronine G solution

For preparation of approx. 100 ml solution mix:

Pyronine G (C.I. 45005) Certistain®	2 g	
Distilled water	100 ml	
dissolve		
extract with chloroform and discard the organic phase		
filter the aqueous staining solution		

Methyl green-pyronine G working solution

For preparation of approx. 50 ml solution mix:

Methyl green solution	7.5 ml	
Pyronine G solution	12.5 ml	
Distilled water	30 ml	
mix		

The freshly prepared staining solutions should be filtered before use.

Procedure

DNA/RNA staining

Deparaffinize histological slides in the conventional manner and rehydrate in a descending alcohol series.

The slides should be allowed to drip off well after the individual staining steps, as a measure to avoid any unnecessary cross-contamination of solutions.

The stated times should be adhered to to guarantee an optimal staining result.

Slide with paraffin section		
Distilled water	1 min	
press in filter paper		
Methyl green-pyronine G working solution	6 min	
Distilled water	rinse	
press in filter paper		
1-Butanol	5 min	
1-Butanol	5 min	
Xylene	5 min	
Mount the xylene-wet slides with e.g. Entellan® new and cover glass.		

After dehydration (ascending alcohol series) and clearing with xylene, histological samples can be mounted with water-free mounting agents (e.g. DPX new or Entellan® new) and a cover glass and and can then be stored.

The use of immersion oil is recommended for the analysis of stained slides with a microscopic magnification >40x.

Result

Chromatinblue-greenCell nucleiredCytoplasmpink to red

Technical notes

The microscope used should meet the requirements of a medical diagnostic laboratory.

When using histoprocessors and automatic staining systems, please follow the instructions for use supplied by the supplier of the system and software.

The freshly prepared staining solutions should be filtered before use.

Remove surplus immersion oil before filing.

Diagnostics

Diagnoses are to be made only by authorized and trained personnel. Valid nomenclatures must be used.

Further tests must be selected and implemented according to recognized methods.

Suitable controls should be conducted with each application in order to avoid an incorrect result.

Store Pyronine G (C.I. 45005) - for microscopy Certistain® at $+5\,^{\circ}$ C to $+30\,^{\circ}$ C.

Pyronine G (C.I. 45005) - for microscopy Certistain® can be used until the stated expiry date.

After first opening of the bottle, the contents can be used up to the stated expiry date when stored at +5 °C to +30 °C.

The bottles must be kept tightly closed at all times.

Additional instructions

For professional use only.

In order to avoid errors, the application must be carried out by qualified personnel

National guidelines for work safety and quality assurance must be followed. Microscopes equipped according to the standard must be used.

Protection against infection

Effective measures must be taken to protect against infection in line with laboratory guidelines.

Instructions for disposal

The package must be disposed of in accordance with the current disposal

Used solutions and solutions that are past their shelf-life must be disposed of as special waste in accordance with local guidelines. Information on disposal can be obtained under the Quick Link "Hints for Disposal of Microscopy Products" at www.microscopy-products.com. Within the EU the currently applicable REGULATION (EC) No 1272/2008 on classification, labelling and packaging of substances and mixtures, amending and repealing. Directives 67/548/EEC and 1999/45/EC, and amending Regulation (EC) No 1907/2006 applies.

Auxiliary reagents

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Cat. No.	100063	Acetic acid (glacial) 100% anhydrous for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 l, 2.5 l
Cat. No.	100974	Ethanol denatured with about 1 % methyl ethyl ketone for analysis EMSURE®	1 I, 2.5 I
Cat. No.	100983	Ethanol absolute for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 l, 2.5 l, 5 l
Cat. No.	101990	1-Butanol for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 l, 2.5 l, 4 l, 10 l
Cat. No.	102445	Chloroform for analysis EMSURE® ACS,ISO,Reag. Ph Eur	1 l, 2.5 l, 4 l, 10 l
Cat. No.	104699	Immersion oil for microscopy	100-ml dropping bottle, 100 ml, 500 ml
Cat. No.	107961	Entellan® new rapid mounting medium for microscopy	100 ml, 500 ml, 1 l
Cat. No.	108297	Xylene (isomeric mixture) for analysis EMSURE® ACS,ISO,Reag. Ph Eur	2.5 l, 4 l
Cat. No.	115944	Methyl green zinc chloride double salt (C.I. 42590) for microscopy Certistain®	25 g

Hazard classification

Cat. No. 1.07518.0025

Please observe the hazard classification printed on the label and the information given in the safety data sheet.

The safety data sheet is available on the website and on request.

Main components of the product

Cat. No. 1.07518.0025

C.I. 45005 $C_{17}H_{19}CIN_2O$

M = 302.67 g/mol

Other IVD products

Cat. No.	100496	Formaldehyde solution 4%, buffered, pH 6.9 (approx. 10% Formalin solution) for histology	350 ml and 700 ml (in wide neck bottle), 5 l, 10 l, 10 l Tritripac®
Cat. No.	101646	PAS staining kit for detection of aldehyde and mucosubstances	2x 500 ml
Cat. No.	105174	Hematoxylin solution modified acc. to Gill III for microscopy	500 ml, 1 l, 2.5 l
Cat. No.	115161	Histosec® pastilles (without DMSO) soldification point 56-58°C embedding agent for histology	10 kg (4x 2.5 kg), 25 kg
Cat. No.	117081	Eosin Y solution 1%, alcoholic for microscopy	11









for use

Catalog number

Batch code



Caution, consult accompanying documents



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