

1.01384.0100  
1.01384.9025

Microscopy

Brilliant cresyl blue solution

for the staining of reticulocytes and trichomonads for microscopy



In Vitro Diagnostic Medical Device



This “Brilliant cresyl blue solution - for the staining of reticulocytes and tricomoads for microscopy” is used for human-medical cell diagnosis and serves the hematological investigation of sample material of human origin. It is a staining solution that makes target structures (by staining) in hematological specimen materials evaluable for diagnostic purposes.

Principle

The regenerative capacity of erythrocytes can be detected by counting of reticulo-cytes.

The measurement of substantia granulofilamentosa (ribonucleoproteins) is pos-sible with fresh, non-fixed, young erythrocytes (supravital staining).

Four stages of substantia granulofilamentosa maturation can be distinguished depending on the stage of reticulocyte development: coiled skein (I), incomplete network (II), complete network (III) and granular form (IV). In peripheral blood the development stages III and IV are found most commonly.

When stained with brilliant cresyl blue the reticulocytes display a black-blue net-work or black-blue dots.

Sample material

Anticoagulant venous blood, in exceptional cases capillary blood

Reagents

Cat. No. 101384  
Brilliant cresyl blue solution  
for the staining of reticulocytes and tricomoads for microscopy

100 ml, 25 l

Also required:

Cat. No. 106404 Sodium chloride for analysis  
EMSURE® ACS,ISO,Reag. Ph Eur

500 g, 1 kg,  
5 kg

Sample prepration

The sampling must be performed by qualified personnel.

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. Fol-low the manufacturer’s instructions for application / use.

Reagent preparation

Physiologic saline solution

For preparation of approx. 1000 ml solution mix and dissolve:

Sodium chloride	8.5 g
Distilled water	991.5 g

Brilliant cresyl blue working solution

Dilute the Brilliant cresyl blue solution, Cat. No. 101384, 1: 80 to 1:200 with physiologic saline solution. For the best dilution make a test before.

The diluted staining solution should be filterd before use.

Procedure

Single tests

Draw 20 µl blood and 20 µl brilliant cresyl blue working solution into a hemoglo-bin pipette and fill into a small sealable container. Mix thoroughly and after about 30 min prepare a thin smear.

Tests in series

Prepare thin smears of brilliant cresyl blue working solution on microscope slides using a glass rod. Air-dried slides prepared in this way can be stored for 2 - 3 weeks.

For reticulocyte counts smear a small drop of blood quickly over the stain layer, and immediately place the still wet preparation in a moist chamber (Petri dish with damp filter paper). Leave for 5 - 10 min and then allow to dry in air.

Counting under the microscope

Count the reticulocytes per 1000 erythrocytes with oil immersion under the micro-scope following a meandering pattern. In order to avoid confusion when counting it is advisable to place a reticulocyte counting grid subdivided into small squares (or a square paper diaphragm) in one of the two eye pieces.

In peripheral blood the development stages III and IV are found most commonly. When stained with brilliant cresyl blue the reticulocytes display a black-blue net-work or black-blue dots.

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

Result

In peripheral blood the development stages III and IV are found most commonly. When stained with brilliant cresyl blue the reticulocytes display a black-blue net-work or black-blue dots.

The reticulocyte count is expressed in relation to 1000 counted erythrocytes (i.e. as ‰). If the erythrocyte count is low, then the absolute reticulocyte count/ µl is used.

Calculation

Reticulocyte count [cells/µl] = 
$$\frac{\text{erythrocyte count}/\mu\text{l} \times \text{reticulocyte count}(\text{‰})}{1000}$$

Normal range

	‰	reticulocyte count/µl
Adults	5 - 15	25 000 - 75 000
Newborn babies	20 - 60	100 000 - 300 000

Technical notes

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

Use a standard hemoglobin pipette and grid for counting reticulocytes.

The diluted staining solution should be filterd 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 meth-ods.

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

Storage

Store the Brilliant cresyl blue solution - for the staining of reticulocytes and trico-monads for microscopy at +15 °C to +25 °C.

Shelf-life

The Brilliant cresyl blue solution - for the staining of reticulocytes and tricomoads for microscopy 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 +15 °C to +25 °C.

The bottles must be kept tightly closed at all times.

Capacity

approx. 1000 applications / 100 ml

Additional instructions

**For professional use only.**

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

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 labora-tory guidelines.

## Instructions for disposal

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

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](http://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

Cat. No.	101368	Brilliant cresyl blue zinc chloride double salt for microscopy Certistain®	25 g
Cat. No.	104699	Immersion oil for microscopy	100-ml dropping bottle, 100 ml, 500 ml
Cat. No.	106009	Methanol for analysis EMSURE® ACS, ISO, Reag. Ph Eur	1 l, 2.5 l, 5 l
Cat. No.	106404	Sodium chloride for analysis EMSURE® ACS, ISO, Reag. Ph Eur	500 g, 1 kg, 5 kg

## Hazard classification

Cat. No. 1.01384.0100

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. 1.01384.0100

C.I. 51010	1 g/l
C.I. 52040	3 g/l
NaCl	9 g/l

1 l = 1.01 kg

## Other IVD products

Cat. No.	101383	Wright's eosin methylene blue solution for microscopy	100 ml, 500 ml, 2.5 l
Cat. No.	109204	Giemsa's azur eosin methylene blue solution for microscopy	100 ml, 500 ml, 1 l, 2.5 l
Cat. No.	111674	Hemacolor® Rapid staining of blood smear staining set for microscopy	1 set
Cat. No.	116300	LEUCOGNOST® ALPA Detection of the alkaline leukocyte phosphatase activity in leukocytes	12 units

## Literature

1. Löffler, H., Rastetter, J., Haferlach, T, Atlas der klinischen Hämatologie, 2004, Springer-Verlag Berlin Heidelberg
2. Conn's Biological Stains: A Handbook of Dyes, Stains and Fluorochromes for Use in Biology and Medicine, 10th Edition, (ed. Horobin, R.W. and Kiernan, J.A). Bios, 2002



Consult instructions for use



Manufacturer



Catalog number



Batch code



Caution, consult accompanying documents



Use by YYYY-MM-DD



Temperature limitation

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