BIOGNOST®

# BRILLIANT CRESYL BLUE SOLUTION

IVD *In vitro* diagnostic medical device CE

## Solution for staining reticulocytes in blood

## INSTRUCTIONS FOR USE

REF Product code: BCB–OT–100 (100 mL) BCB-OT-500 (500 mL) BCB–OT–1L (1000 mL)

### Introduction

Reticulocyte count in peripheral blood determines the erythropoietic activity, reticulocyte counting is the basic and one of the most commonly used methods in diagnostic hematology. Reticulocytes are juvenile erythrocytes containing the remains of basophilic ribonucleoproteins. The remains present material which Brilliant cresyl blue solution binds to and in turn creating granulated or fibrous blue-black precipitate. Juvenile reticulocytes have the greatest concentration of precipitated material; lower amount of granules or short fibers may be microscopically detected while observing older reticulocytes. This method uses fresh, non-fixed juvenile erythrocytes.

### Product description

- **BRILLIANT CRESYL BLUE SOLUTION** – Solution for hematological and cytological testing of blood samples

### Other products that may be used in staining:

- Glass slides used in hematology, such as VitroGnost STANDARD GRADE or high quality glass slides used in histopathology and cytology, such as VitroGnost SUPER GRADE or one of more than 30 models of VitroGnost glass slides

### Testing sample

- Uncoagulated venous blood (with added EDTA) or capillary blood

### Staining procedure

#### Staining in tube

- Mix equal volumes of blood and staining solution in a glass tube (for instance, 20 $\mu$L of blood + 20 $\mu$L of Brilliant Cresyl Blue solution)
- Let it set for 30 min. Mix again
- Prepare a blood smear on a glass slide
- After drying on air, view the smear microscopically without previous fixation or counterstaining

#### Glass slide staining

- Prepare thin smear of Brilliant Cresyl Blue solution on a glass slide by using a glass stick. Slides dried on air may be kept for up to 2 weeks.
- Mix a small drop of blood over the smear.
- In order to avoid the preparation getting dry, place it immediately in an incubation chamber for 5-10 min
- Dry the preparation. View the smear microscopically without previous fixation or counterstaining

### Result

Stained reticulocytes display a dark blue network or dark blue dots.

Reticulocyte counting

The number of reticulocytes is expressed as the ratio to 100 counted erythrocytes, i.e. as a percentage

Normal reticulocyte values (%):

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|---|---|
| Grownups: | 0.5-1.5 |
| Newborns: | 2-6 |

### Note

Time periods of staining processes are not entirely standardized in clinical and laboratory practical experience. Time periods specified in the instruction approximately correspond to a longtime work practice with optimal results. Intensity of staining depends on the period of immersion in the dye. Real staining protocol depends on personal requests and priorities. Microscopical smear must be properly smeared and stained. Most of the mature reticulocytes contain only a few granules or fibers, making counting more difficult. Cells containing Pappenheimer bodies (found in peripheral blood after surgical procedure of removing a part of entire spleen) may make differentiation between mature and juvenile reticulocytes. However, Pappenheimer granules are often present in form of an individual dot and are stained in a darker hue (compared to reticulate precipitate).

### Preparing the sample and diagnostics

Use only appropriate instruments for collecting and preparing the samples. All the samples must be processed with the most modern technology and be visibly marked. Follow the manufacturer's instructions for handling. In order to avoid mistakes, staining must be conducted by a trained professional. Only trained medical personnel may make a diagnosis. Use only microscope according to standards of the medical diagnostic laboratory. In order to avoid an erroneous result, a positive and negative check is advised before application.

### 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 which is available on demand.

**Storing, stability and expiry date**

Keep Brilliant Cresyl Blue solution in a tightly closed original package at a temperature of +15 to +25 °C. Do not keep in cold places, do not freeze and avoid exposing to direct sunlight. Date of manufacture and expiry date are printed on the product's label.

**References**


1. Carson, F. L. (2007), Histotechnology, 2nd ed. Singapore
2. Cook, D. J. (2006): Cellular pathology, 2nd ed. Banbury: Scion Publishing Ltd.
3. Kiernan, J. A. (2008) Histological and histochemical methods, 4th ed. Bloxham: Scion Publishing Ltd.



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|  | Refer to the supplied documentation |  | Storage temperature range |  | Number of tests in package | REF | Product code | CE | European Conformity |  | BIOGNOST Ltd.<br>Medjugorska 59<br>10040 Zagreb<br>CROATIA<br>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 | | | | | | |