

# FAST GREEN F.C.F. REAGENT

IVD *In vitro* diagnostic medical device



**Reagent for green counterstaining used with Masson-Goldner Trichrome and Paraldehyde Fuchsin staining kits**

## INSTRUCTIONS FOR USE

REF Product code: FGR-OT-100 (100 mL)

FGR-OT-500 (500 mL)

### Introduction

Fast Green F.C.F. reagent is primarily used as counterstain in certain special staining kits, such as BioGnost's Masson-Goldner Trichrome and Paraldehyde Fuchsin kits. Fast Green F.C.F. dye binds with collagen and makes it characteristically green in color.

### Product description

- FAST GREEN F.C.F. REAGENT** - reagent for counterstaining used with Masson-Goldner Trichrome and Paraldehyde Fuchsin kits

**NOTE:** Fast Green F.C.F. reagent is a component of several BioGnost's special staining kits (Masson-Goldner Trichrome and Paraldehyde Fuchsin kits). Staining procedure using Masson-Goldner Trichrome kit is described below. If you are interested in other staining protocols using special stains containing Fast Green F.C.F. reagent, feel free to contact us.

### Example of use of Fast Green F.C.F. reagent as a Masson Golder Trichrome kit component

#### Other materials and reagents 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 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
- Other components of Masson-Goldner Trichrome kit: Bouin's solution (product code BOU-OT-100, BOU-OT-500), Hematoxylin, Weigert A (product code HEMA-OT-100, HEMA-OT-500), Ferri reagent, Weigert B (product code FR-OT-100, FR-OT-500), Biebrich Scarlet-Acid Fuchsin reagent (product code BSAF-OT-100, BSAF-500), P.T.A.-P.M.A. reagent (product code PPR-OT-100, product code PPR-OT-500), Acetic acid, 1% solution (product code OK1-OT-100, OK1-OT-500)

### Preparation of working solution

#### Hematoxylin Weigert working solution

- Mix Hematoxylin, Weigert A and Ferri reagent, Weigert B in 1:1 ratio. Hematoxylin Weigert working solution is stable for approximately 2 weeks. Discard the solution when the nuclei turn brown after staining.

#### 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  $\mu$ m slices and place them on a VitroGnost glass slide.

### Histology sections 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.	Stain using Bouin's solution	60 min at 56° C or over night at room temperature
6.	Cool the section down at room temperature	10 min
7.	Rinse under tap water	10 seconds
8.	Rinse in distilled water	10 seconds
9.	Immerse into Hematoxylin Weigert working solution	5 min
10.	Rinse under tap water	3 min
11.	Stain with Biebrich Scarlet-Acid Fuchsin reagent	2 min
12.	Rinse in distilled water	until the excessive dye is washed off of the section
13.	Treat with PTA-PMA reagent	10 min
14.	Move the section from P.T.A.-P.M.A. reagent into Fast Green F.C.F. reagent without rinsing	
15.	Stain using Fast Green F.C.F. reagent	5 min
16.	Rinse in distilled water	until the excessive dye is washed off of the section
17.	Treat using 1% acetic acid solution	3 min
18.	Dehydrate using 70% alcohol (Histanol 70)	5 dips

19.	Dehydrate using 95% alcohol (Histanol 95)	5 dips
20.	Dehydrate using 100% alcohol (Histanol 100)	2 min
21.	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 VitroGnost cover glass.

### Result

Nuclei - blue-purple  
Muscle fibers, keratin, cytoplasm - bright red  
Collagen, mucus - green  
Erythrocytes - red-orange

### Note

Staining procedures are not standardized and they depend on standard operating procedures of individual laboratories and the experience of the personnel conducting the staining procedure. Intensity of staining depends on the period of immersion in the dye. Depending on personal requests and standard laboratory operating procedures, sample processing and staining can be carried out according to other protocols.

### 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

Keep Fast Green F.C.F. reagent in a tightly sealed original packaging at temperature of 15 to 25°C. Keep in dry 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. Melis, M., Carpino, F., Di Tondo, U. (1989), *Tecniche in anatomia patologica*, Edi Ermes, Milano.
2. Prophet, E.B., Mills, B., Arrington, J., Sobin, L. (1968), *Laboratory methods in histotechnology*, McGraw Hill, Washington D.C.
3. Bancroft, J.D., Gamble, M. (2002), *Theory and practice of Histological Techniques*, Churchill Livingstone, New York.

FGR-X, V1-EN1, 14 January 2019, AK/IŠP

	Refer to the supplied documentation		Storage temperature range		Number of tests in package		Product code		European Conformity	 BIOGNOST Ltd. Medjugorska 59 10040 Zagreb CROATIA <a href="http://www.biognost.com">www.biognost.com</a>	
	Refer to supplied instructions		Keep away from heat and sunlight		Valid until		Lot number		Manufacturer		
	For <i>in vitro</i> diagnostic use only		Keep in dry place		Caution - fragile						