BIOGNOST®

MOVAT KIT

IVD *In vitro* diagnostic medical device CE

Eleven-reagent kit for visualization of collagen, muscle fibers, reticulin fibers, mucins and fibrins

INSTRUCTIONS FOR USE

REF Product code: MOV-100T (9x30+2x100 mL) MOV-K-100 (11x100 mL)

Introduction

Movat kit is used for visualization of five types of connective tissues in a single staining process. It enables differentiation between collagens, muscle fibers, reticulin fibers, mucins and fibrins, and it also stains nuclei. It is used for diagnosing cardiovascular and pulmonary diseases.

Product description

- **MOVAT KIT**- Eleven-reagent kit for visualization of collagen, muscle fibers, reticulin fibers, mucins and fibrins

The kit contains:	for 100 tests (MOV-100T)	11 x100 mL (MOV-K-100)
Alcian Blue solution pH 2.5	30 mL (AB2-OT-30)	100 mL (AB2-OT-100)
Alkaline alcohol, solution	100 mL (ALA-OT-100)	100 mL (ALA-OT-100)
Hematoxylin, Verhoeff A	30 mL (HEMV-OT-30)	100 mL (HEMV-OT-100)
Ferri reagent, Verhoeff B	30 mL (FRV-OT-30)	100 mL (FRV-OT-100)
Iodine solution, Verhoeff C	30 mL (JODV-OT-30)	100 mL (JODV-OT-100)
Reagent for differentiation, Verhoeff	30 mL (RDV-OT-30)	100 mL (RDV-OT-100)
Acetic acid, 0.5% solution	100 mL (OK05-OT-100)	100 mL (OK05-OT-100)
Sodium thiosulfate, 5 solution	30 mL (NT5-OT-30)	100 mL (NT5-OT-100)
Biebrich Scarlet-Acid Fuchsin reagent	30 ml (BSAF-OT-30)	100 ml (BSAF-OT-100)
Phosphotungstic acid, 5% solution	30 mL (FVK5-OT-30)	100 mL (FVK5-OT-100)
Orange G, 1% solution	30 mL (ORG1-OT-30)	100 mL (ORG1-OT-100)

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 56/58, 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 C, BioMount Aqua
- 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 the 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

Sample staining procedure

NOTE

Apply the reagent so it completely covers the section.

a) using kit for 100 tests (MOV-100T)

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 Alcian Blue solution pH 2.5 (add ≥5 drops)	20 min
6.	Rinse under tap water	5 min
7.	Add Alkaline alcohol, solution (≥5 drops and close in the chamber in order to prevent evaporation)	60 min
8.	Rinse under tap water	10 min
9.	Add 7 drops of Hematoxylin, Verhoeff A, 3 drops of Ferri reagent, Verhoeff B, and 3 drops of Iodine solution, Verhoeff C	15 min
10.	Rinse in distilled (demi) water	5 dips
11.	Add Reagent for differentiation, Verhoeff (≥5 drops) and differentiate the section	3 min
	Note: quickly rinse the section in distilled (demi) water after differentiation and microscopically check for the section for elastin being stained black. Repeat the differentiation if necessary	
12.	Rinse in distilled (demi) water	5 dips
13.	Differentiate in Acetic acid, 0.5% solution	5 dips
14.	Add Sodium thiosulfate, 5% solution (≥5 drops)	1 min
15.	Rinse under tap water	10 min
16.	Rinse in distilled (demi) water	5 dips
17.	Staining using Biebrich Scarlet-Acid Fuchsin reagent (≥5 drops)	3 min
18.	Differentiate in Acetic acid, 0.5% solution	5 dips
19.	Add Phosphotungstic acid, 5% solution	10 min
20.	Differentiate in Acetic acid, 0.5% solution	5 dips
21.	Rinse in distilled (demi) water	5 dips
22.	Stain using Orange G, 1% solution	15 min

23.	Dehydrate using 95% alcohol (Histanol 95)	5 dips
24.	Dehydrate using 100% alcohol (Histanol 100)	2 min
25.	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.

b) using eleven-reagent 100 mL kit (MOV-K-100)

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.	Immerse into Alcian Blue solution pH 2.5	20 min
6.	Rinse under tap water	5 min
7.	Immerse in Alkaline alcohol, solution and close the staining jar in order to prevent evaporation	60 min
8.	Rinse under tap water	10 min
9.	Add 20 mL of Hematoxylin, Verhoeff A, 8 mL of Ferri reagent, Verhoeff B, and 8 mL of Iodine solution, Verhoeff C. Place the section in the incubation tray and cover it in order to prevent evaporation	
10.	Rinse in distilled (demi) water	5 dips
11.	Immerse in the Reagent for differentiation, Verhoeff and differentiate the section	5 min
12.	Note: quickly rinse the section in distilled (demi) water after differentiation and microscopically check for the section for elastin being stained black. Repeat the differentiation if necessary	
	Rinse in distilled (demi) water	5 dips
13.	Differentiate in Acetic acid, 0.5% solution	5 dips
14.	Immerse into Sodium thiosulfate, 5% solution	1 min
15.	Rinse under tap water	10 min
16.	Rinse in distilled (demi) water	5 dips
17.	Stain with Biebrich Scarlet-Acid Fuchsin reagent	3 min
18.	Differentiate in Acetic acid, 0.5% solution	5 dips
19.	Immerse into Phosphotungstic acid, 5% solution	10 min
20.	Rinse in distilled (demi) water	5 dips
21.	Differentiate in Acetic acid, 0.5% solution	5 dips
22.	Stain using Orange G, 1% solution	15 min
23.	Dehydrate using 95% alcohol (Histanol 95)	5 dips
24.	Dehydrate using 100% alcohol (Histanol 100)	2 min
25.	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 and elastic fibers **-** black
Collagen and reticulin fibers - orange
Mucins - blue to green
Fibrins and muscle fibers - red

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 Movat kit 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.

MOV-100T, MOV-K-100, V8-EN4, 11 September 2019, IŠP/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	CE
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						