BIOGNOST®

ACRIDINE ORANGE powder dye, C.I. 46005

IVD *In vitro* diagnostic medical device CE

Basic Orange 14

For staining acid mucins, apoptosis detection, differential staining of RNA and DNA

INSTRUCTIONS FOR USE

REF Product code: AO-P-25 (25 g)

Introduction

Histology, cytology and other related scientific disciplines study the microscopic anatomy of tissues and cells. In order to achieve a good tissue and cellular structure, the samples need to be stained in a correct manner. Acridine Orange powder dye is intended for microscopy staining using different methods. It is used as fluorescent stain for differential DNA and RNA staining. Other uses include acid mucins staining and apoptosis detection. Acridine Orange is also used in cytogenetics for displaying DNA and DNA-rich structures during C-banding of chromosomes.

Product description

- **ACRIDINE ORANGE** – Powder dye for creating dye solution for differential staining of RNA and DNA acc. to Bertalanffy

Other preparations and reagents used in preparing the dye solution:

- Anhydrous potassium dihydrogen phosphate for buffer solution (KH_2PO_4)
- Anhydrous disodium hydrogen phosphate for buffer solution (Na_2PO_4)

Preparing the dye solution

Phosphate buffer solution:

- Basic solution A – 0.1 M of potassium dihydrogen phosphate:
  Dissolve 13.61 g of KH_2PO_4 in 1000 ml of distilled/demineralized water
- Basic solution B – 0.1 M of disodium hydrogen phosphate:
  Dissolve 17.8 g of $Na_2HPO_4 \times 2H_2O$ in 1000 mL of distilled/demineralized water
- Preparation of buffer solution, pH 6:
  Mix basic solutions A and B in ratio of 8.5 : 1.5*
  *850 mL of solution A mix with 150 mL of solution B

Basic Acridine Orange dye solution:

- Dissolve 0.1 g of Acridine Orange powder dye (by mixing) in 100 ml of distilled/demineralized water

Working Acridine Orange dye solution:

- Dissolve basic powder dye solution with phosphate buffer in 1:10 ratio

Result

Structures with abundant DNA - green to yellow-green fluorescence
Structures with abundant RNA - red fluorescence

Note

The mentioned formulation is only one of the ways of preparing the dye solution. Acridine Orange dye is most commonly used according to Bertalanffy. Depending on personal requests and standard laboratory operating procedures, the dye solution can be prepared 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. Only use a 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 a 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 Acridine Orange powder dye in a tightly closed original package at a temperature between 15°C and 25°C. Keep in dry places, do not freeze and avoid exposure to direct sunlight. The expiry date is stated on the product's label.

References

1. Conn, J. (1977): *Biological Stains*, 9th ed. Baltimore: Williams and Wilkins Co.
2. Carson, F. L., Hladik, C. (2009): *Histotechnology: A Self-Instructional Text*, 3rd ed., Chicago: ASCP Press
3. Von Bertalanffy, L., Maisin, M., Maisin, F. (1956): Use of acridine-orange fluorescence technique in exfoliative cytology, *Science* 124:1024-1025.

AO-P-25, V2-EN1, 30 September 2015, VR/IŠP

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						