

ALCIAN BLUE SOLUTION pH 2.5



IVD In vitro diagnostic medical device

Classified acc. to Regulation (EU) 2017/746 - Class A device

Solution for staining acidic mucopolysaccharides

INSTRUCTIONS FOR USE

Basic UDI-DI	385889212HPC30708STARVF		
EMDN code	W01030708		
REF	Catalog number	Volume	UDI-DI
AB2-OT-100		100 mL	03858888820343
AB2-OT-500		500 mL	03858888820794
AB2-OT-1L		1000 mL	03858888820800



Intended use and test principle

Alcian Blue 8GX is a positively charged dye molecule that electrostatically binds to negatively charged acidic groups in tissues, such as carboxyl groups ($-\text{COO}^-$) and sulfate groups ($-\text{SO}_4^{2-}$), whereby acidic mucins and glycosaminoglycans are stained blue to turquoise blue. For this reason, this dye is used for the demonstration of glycosaminoglycans in mucins, for staining amyloid, cysteine, and for polychromatic staining of mast cells using the Alcian blue-safranin method. It is also used for the determination of bacterial species and detection of bacterial capsules. Alcian Blue solution pH 2.5 is a component of BioGnost's Alcian Blue – P.A.S. kit and Alcian Blue pH 2.5 kit. The Alcian Blue pH 2.5 kit enables good staining and visualization of acidic mucins without staining sulfated mucins. The Alcian Blue – P.A.S. kit enables differentiation of neutral and acidic mucins, glycogen, and glycoproteins.

Product description

- ALCIAN BLUE SOLUTION pH 2.5** – a solution of Alcian Blue 8GX powder dye with pH adjusted to 2.5

Example of staining with Alcian Blue solution pH 2.5 as a component of the Alcian Blue–PAS kit:

Additional reagents and materials that can be used in the method

- Fixatives, such as BioGnost's neutral buffered formaldehyde solutions: Formaldehyde NB 4%, Formaldehyde NB 10%
- Dehydration/rehydration agents such as BioGnost's alcohol solutions: Histanol 70, Histanol 80, Histanol 95, and Histanol 100
- Clearing agents, such as BioClear xylene or BioClear New, an aliphatic hydrocarbon-based xylene substitute
- Infiltration and embedding agents such as BioGnost's granulated paraffins BioWax 52/54, BioWax 56/58, BioWax Plus 56/58, BioWax Blue
- Microscopic slide covering agents and cover glass mountants such as BioGnost's BioMount, BioMount High, BioMount M, BioMount New, BioMount New Low, BioMount DPX, BioMount DPX High, BioMount DPX Low, BioMount DPX New, BioMount C, BioMount Aqua
- VitroGnost slides and coverslips for use in histopathology and cytology
- Immersion oils such as BioGnost's Immersion Oil, Immersion Oils types A, C, FF, 37, or Immersion Oil Tropical Grade
- Other components of the Alcian Blue–P.A.S. kit: Sodium tetraborate solution, Periodic acid 0.8% solution, BioSchiff reagent, Hematoxylin ML, Sodium metabisulfite, solution, HCl reagent, PAS

Preparation of histological sections for staining

- Fix (Formaldehyde NB 4%, Formaldehyde NB 10%) and process the tissue sample
- Embed the tissue in a paraffin block (BioWax 52/54, BioWax 56/58, BioWax Plus 56/58, BioWax Blue)
- Cut the paraffin block into 4-6 micron thin sections and mount on a VitroGnost microscope slide

Preparation of the additional solution needed in the staining process

- Sulfite solution

Note: make the sulfite solution directly **before** use! Preparation of sulfite solution in a volume of approximately 80 mL (for a Hellendahl jar): Mix 3.75 mL of Sodium metabisulfite, solution with 3.75 mL of HCl reagent, PAS. Then add 75 mL of tap water to the solution and mix.

Sample staining procedure

Pour the reagents into glass staining jars (type Coplin, Hellendahl, or Schifferdecker) and return them to the original bottles after staining. Close well. Filter the reagents as needed.

1.	Deparaffinize in xylene (BioClear) or xylene substitute (BioClear New)	3 exchanges, 2 min each
2.	Rehydrate in 100% alcohol (Histanol 100)	2 exchanges, 5 and 3 min
3.	Rehydrate in 95% alcohol (Histanol 95)	2 min
4.	Rehydrate in distilled/demineralized water	2 min
5.	Immerse in Alcian Blue solution pH 2.5	30 min
6.	Without rinsing, apply Sodium tetraborate, solution	10 min
7.	Rinse under cold running water	5 min
8.	Rinse in distilled/demineralized water	1-2 min
9.	Immerse in Periodic acid, 0.8% solution	5-10 min
10.	Rinse under running tap water	3 min
11.	Rinse the slide with distilled/demineralized water	
12.	Immerse in BioSchiff reagent	10-15 min
	Note: during staining, the jar must be covered to prevent evaporation of SO_2	
13.	Without rinsing, immerse in sulfite solution	3 exchanges, 2 min each
14.	Rinse under running tap water	3 min

15.	Immerse in Hematoxylin ML	1-3 min
16.	Rinse under running tap water	3 min
17.	Dehydrate in 70% alcohol (Histanol 70)	5 dips
18.	Dehydrate in 95% alcohol (Histanol 95)	5 dips
19.	Dehydrate in 100% alcohol (Histanol 100)	2 min
20.	Clear in xylene (BioClear) or xylene substitute (BioClear New)	2 exchanges, 2 min each

Immediately after clearing, apply an appropriate BioMount covering/mounting medium. If BioClear xylene was used, use one of BioGnost's xylene-based mountants (BioMount, BioMount High, BioMount M, BioMount DPX, BioMount C or universal BioMount New). If BioClear New xylene substitute was used, the appropriate mountant is BioMount New. Cover the section with a VitroGnost cover glass.

Result

Mucins - blue turquoise

P.A.S. positive substances - purple (magenta)

Nuclei - blue

Epithelial mucin and cartilage - violet/dark blue

Limitations

This product is intended for professional laboratory use for diagnostic purposes only. Deviation from the staining procedure described in these Instructions for Use may cause differences in staining results.

Sample preparation and diagnostics

Use only appropriate instruments for collecting and preparing the samples. Process the samples using modern technology and mark them clearly. It is necessary to follow the manufacturer's instructions for use. To avoid errors, staining and diagnosis may only be performed by qualified personnel. Use a microscope that complies with medical diagnostic laboratory standards.

If a serious incident occurs during use or as a result of its use, please report it to the manufacturer and/or authorized representative and competent authority.

Safety at work and environmental protection

Handle the product in accordance with occupational health and environmental protection guidelines. Used and expired solutions must be disposed of as special waste following national guidelines. Reagents used in this procedure can pose a danger to human health. The examined tissue samples are potentially infectious, therefore it is necessary to implement human health protection measures in accordance with good laboratory practice guidelines. It is mandatory to read and act according to the information and warning signs printed on the product label and in the Safety Data Sheet, which is available on request.

Storage, stability, and shelf life

Upon receipt, store the product in a dry place and well-closed original packaging at a temperature of +15 °C to +25 °C. Do not freeze or expose to direct sunlight. After first opening, the product can be used until the specified expiry date, if stored properly. The production date and expiration date are printed on the product label.

References

1. Culling, C.F.A. (1974): *Handbook of histopathological and histochemical techniques*, 2nd ed., Butterworth, London, UK.
2. Davey, F.R. and Nelson, D.A. (1977): *Periodic Acid Schiff (PAS) Stain*. IN *Hematology*, 2nd ed., W. J. Williams, E. Buetler, A. J. Erslev, R.W. Rundles, McGraw-Hill, New York, pp. 1630–1632.
3. Hotchkiss, R.D. (1948): *A microchemical reaction resulting in the staining of polysaccharide structures in fixed tissue preparations*, *Arch. Biochem.* 16, p. 131.
4. Sheehan D.C. and Hrapchak, B.B. (1980): *Theory and Practice of Histotechnology*, 2nd ed., CV Mosby, St. Louis, (MO), pp 52, p. 14–167.

Warnings and precautions regarding the materials contained in the product:

Not a dangerous substance or mixture according to Regulation (EC) no. 1272/2008.

AB2-IFU_ENV3, 01.07.2026., IŠP

	Manufacturer		Batch code		Consult Instructions for use		European conformity
	Date of manufacture		Catalogue number		Caution		Unique device Identifier
	Use-by date		Temperature limit		In vitro diagnostic medical device		

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Version	Description / reason for change	Date
3	Revised in accordance with Regulation (EU) 2017/746 - IVDR	01.07.2026.