

## Alcian Blue Stain Kit

### PRODUCT INFORMATION:

#### REF

SSP007 100ml Ready to use  
SSP007 250ml Ready to use  
SSP007 500ml Ready to use

### PERFORMANCE CHARACTERISTICS:

**Staining Interpretation:**  
**Nuclei** : Pink to Red  
**Acid Mucins:** Blue  
**Cytoplasm** : Pale Pink

### SUMMARY AND EXPLANATION

#### For laboratory use only

Alcian Blue Stain Kit is intended for use in the histological visualization of sulfated and carboxylated acid mucopolysaccharides and sulfated and carboxylated sialomucins (glycoproteins) which can be secreted by various connective and epithelial tissue tumors. The tissue parts that specifically stain with this dye become blue to bluish green after staining and are called "Alcianophilic." These can be combined with H&E staining, PAS staining, and van Gieson staining methods. The results should be interpreted along with positive and negative controls by trained or qualified personnel.

### PRINCIPLE OF THE PROCEDURE

Alcian blue is a member of a family of polyvalent basic dyes that are water-soluble. Alcian blue is a large planar phthalocyanine molecule with a copper atom in the center. The molecule also contains four basic isothiuronium groups, which carry a positive charge. The positive charge imparted by these groups results in the attraction of the alcian blue dye molecules to the anionic sites in mucin molecules. The blue colour is due to the presence of copper in the molecule. The 3% acetic acid solution (pH 2.5) with Alcian Blue is believed to form salt linkages with the acid groups of acid mucopolysaccharides.

Nuclear fast red solution is an acid dye that is used as a nuclear counterstain. It must be combined with a moderate, such as aluminium sulfate. Then the overall ionization is positive, and it stains nuclei.

### REAGENTS PROVIDED

| Kit Contents                          | Product Code | Storage Conditions | Pack Sizes |       |       |
|---------------------------------------|--------------|--------------------|------------|-------|-------|
|                                       |              |                    | 100ml      | 250ml | 500ml |
| Alcian Blue Solution (Reagent A)      | SS005        | RT                 | 100ml      | 250ml | 500ml |
| Nuclear Fast Red Solution (Reagent B) | SS006        | RT                 | 100ml      | 250ml | 500ml |

### STORAGE AND HANDLING

**Storage Recommendations:** Store at Room Temperature. When stored at the appropriate conditions, the reagents are stable until expiry. **Do not use the reagents after expiration date provided on the vial.**

To ensure proper reagent performance delivery and stability, replace the dispenser cap after every use and immediately place the vials at recommended storage conditions away from sunlight in an upright position.

During transport, short-term exposure to 2-8°C does not affect product performance.

### SPECIMEN PREPARATION

#### Recommended positive controls:

Formalin-fixed paraffin-embedded as well as frozen tissue sections of Colon, Appendix, Small Intestine

#### Sample preparation:

- Formalin-fixed, paraffin-embedded tissue sections of 3-5 µm thickness

### PRECAUTIONS

- Normal precautions carried out in handling laboratory reagents should be followed
- This product should be used by qualified and trained professional users only
- It can cause serious eye and skin irritation. Refer to Material Safety Datasheet for any updated risk, hazard or safety information
- Dispose of waste observing all local, state, provincial or national regulations
- Do not use reagents after expiration date
- Use protective clothing and gloves, while handling reagents
- Avoid contamination of reagents as it may lead to incorrect results

### MATERIALS REQUIRED, BUT NOT PROVIDED

- Xylenes
- Graded alcohols (50%, 70%, 95%, Absolute)
- DPX Mountant
- Microscopic slides (Positively charged)
- Slide holder
- Cover slips
- Coplin jars
- Distilled water

### STAINING PROCEDURE

- Bake the tissue sections in drying oven at 70°C for 20 minutes.
- Deparaffinize and rehydrate the tissue sections.
- Stain with Alcian Blue Solution (Reagent A) for 30 minutes.
- Wash in running tap water for 2 minutes.
- Rinse in distilled water.
- Counter stain with Nuclear Fast Red Solution (Reagent B) for 2-3 minutes.
- Wash in running tap water for 1 minute.
- Dehydrate using graded alcohols (70%, 95%, 100%) for 2 minutes each.
- Clear in xylenes and mount with DPX mountant.

### QUALITY CONTROL

The recommended positive control for Alcian Blue stain Kit is tissue sections of Colon, Appendix and Small Intestine.

### PERFORMANCE CHARACTERISTICS

Alcian Blue Stain Kit stains **Acidic Mucosubstances** in Blue color, **Nuclei** in **Pink to Red** color and **Cytoplasm** in **Pale Pink** color.

### TROUBLESHOOTING

- Follow the specific protocol recommendations according to data sheet provided
- Tissue staining is dependent on the handling and processing of the tissue prior to staining. Improper fixation, tissue processing, freezing, thawing, washing, drying, heating, sectioning or contamination with other tissues or fluids may produce artifacts, reagent trapping or inaccurate results
- Do not allow the section to dry out during the entire staining process
- Gently mix all the reagents prior to use.
- Excessive or incomplete counterstaining may compromise the interpretation of the results
- If unusual results occur, contact PathnSitu Technical Support at +91-40-2701 5544 or E-mail: [techsupport@pathnsitu.com](mailto:techsupport@pathnsitu.com)

### LIMITATIONS AND WARRANTY







- This product is intended for use only by authorised, trained, and qualified personnel.
- A qualified and trained pathologist/personnel must interpret the results of the test.
- Interpretation of test results must be made in conjunction with relevant background information and additional laboratory findings.
- Always use the recommended volume and concentration of reagents to ensure complete coverage of the tissue section and to minimise the risk of false-positive or false-negative results.

5. Use appropriate buffers, instruments, consumables, and incubation conditions as recommended to achieve optimal staining performance.
6. It is strongly recommended to include known positive and negative controls when performing the test to ensure the validity of results.
7. The product has been validated on formalin-fixed, paraffin-embedded (FFPE) tissues. The end user must establish performance on other tissue types.
8. Unexpected results may occur in untested tissues due to inherent variability in tissue components.
9. False-positive reactions may occur due to insufficient washing, inappropriate protocol conditions, or other contributing factors.
10. In instances where the staining pattern or localisation differs from the specifications outlined in this datasheet, please get in touch with technical support for guidance.
11. Maintain the product under the recommended storage conditions to preserve reagent stability and performance.
12. Do not use reagents that appear cloudy, discoloured, or show signs of contamination. Discard any components showing signs of deterioration.
13. Alcian blue may also bind to acidic structures such as nucleic acids, mast cell granules, or cartilage matrix, reducing specificity.
14. This product is intended for single-use application only. Once applied to a tissue section, reagents should not be recovered or reused, as this may compromise test integrity and specificity.
15. PathnSitu makes no warranties beyond those expressly stated in the product description.
16. PathnSitu shall not be liable for property damage, personal injury, time or effort, or economic loss arising from the use of this product.
17. Please refer to the complete datasheet for all instructions, precautions, and additional product limitations.
18. For detailed information and specifications on individual components, please refer to Product Material Safety Data Sheet (MSDS)

### BIBLIOGRAPHY

1. Schenk, Eric (January 1981). "Notes on Technique: Note from the Biological Stain Commission a Newly Certified Dye—Alcian Blue 8GX". *Stain Technology*. 56 (2): 129–131.
2. "Alcian Blue". *The I.C.I. Journal*: 59–60. March 1948.
3. Kuo YR et al. Recipient Adipose-Derived Stem Cells Enhance Recipient Cell Engraftment and Prolong Allotransplant Survival in a Miniature Swine Hind-Limb Model. *Cell Transplant* 26:1418-1427 (2017).

### EXPLANATION OF SYMBOLS

|   |                           |   |                  |
|---|---------------------------|---|------------------|
|  | Lot number / Batch number |  | Expiry           |
|  | Storage limitation        | RT  | Room Temperature |
|  | Date of manufacture       |  | Catalogue number |
|  | Manufacturer address      |   |                  |