

## Alcian Blue- PAS (ABPAS) Stain

### PRODUCT INFORMATION:

SSP008 100ml Ready to use  
 SSP008 250ml Ready to use  
 SSP008 500ml Ready to use

### PERFORMANCE CHARACTERISTICS:

**Staining Interpretation:**  
**Acid Epithelial Mucins** : Blue  
**Neutral Epithelial Mucins** : Magenta  
**Mixed (Acid & Neutral) Mucins** : Purple

**Laboratory Use Only** 

### 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 delivery and stability, replace the dispenser cap after every use and immediately place the vials at room temperature away from sunlight in an upright position.

### SPECIMEN PREPARATION

**Recommended positive controls:** Small intestine, Appendix, Colon, Large Intestine, Stomach

**Sample preparation and fixation:** Formalin-fixed, Paraffin-embedded tissue sections of 3- 5 µm thickness

### PRECAUTIONS

1. Normal precautions exercised in handling laboratory reagents should be followed.
2. This product should be used by qualified and trained professional users only.
3. The product contains Alcohol and is classified as highly-flammable, must be kept away from ignition sources
4. It can cause serious eye and skin irritation. Refer to Material Safety Datasheet for any updated risk, hazard or safety information.
5. Dispose of waste observing all local, state, provincial or national regulations.
6. Do not use reagents after expiration date
7. Use protective clothing and gloves, while handling reagents
8. Avoid microbial contamination of reagents as it may lead to incorrect results

### MATERIALS REQUIRED, BUT NOT PROVIDED

- Xylenes
- Graded alcohols (50%, 70%, 95%, Absolute)
- Bluing solution
- DPX Mountant
- Microscopic slides (Positively charged)
- Slide holder
- Jars
- Cover slips
- Coplin jars
- Drying Oven (70° ± 5° C)

### STAINING PROCEDURE

1. Bake the sections in Drying oven at 70° C for 20 minutes.
2. Deparaffinize and hydrate to distilled water.
3. Incubate the tissue with Alcian Blue (Reagent A) pH 2.5 for 30 minutes.
4. Wash in running tap water for 5 minutes; rinse in distilled water.
5. Incubate the slides with Periodic acid (Reagent B) for 5 minutes.
6. Rinse in distilled water.
7. Add Schiff's reagent (Reagent C) to the slides in dark chamber and keep it for 30 minutes.
8. Wash in running tap water for 5 minutes; rinse in distilled water.
9. Add Modified Mayer's Hematoxylin (Reagent D) for 1-2 minutes.
10. Wash in running tap water for 5 minutes.
11. Dehydrate, clear, and cover slip with the DPX Mountant.

### QUALITY CONTROL

The recommended positive tissue controls for Alcian Blue-PAS (ABPAS) stain are tissue sections of Liver, Kidney and Colon.

### PERFORMANCE CHARACTERISTICS

Alcian Blue-PAS (ABPAS) for Acid Epithelial Mucins and stromal mucins stains Blue color, Neutral Epithelial Mucins and Glycogen stains Magenta color and Acid and Neutral Mixture stains Purple color.

### SUMMARY AND EXPLANATION

#### For laboratory use only

Alcian Blue-PAS (ABPAS) stain is intended for laboratory use to identify, by light microscopy, mucins in tissue samples. This is a combined method utilizing the properties of both PAS and Alcian blue methods to demonstrate the full complement of tissue proteoglycans. It is used to differentiate between acidic epithelial mucins (sialomucin, sulfomucin) are stained with the Alcian Blue technique and neutral epithelial mucin and glycogen are stained by the PAS reaction.

The rationale of the technique is that by first staining all the acidic mucins with Alcian blue, those remaining acidic mucins which are also PAS positive will be chemically blocked and will not react further during the technique. Those neutral mucins which are solely PAS positive will subsequently be demonstrated in a contrasting manner. Where mixtures occur, the resultant colour will depend upon the dominant moiety.

### PRINCIPLE OF THE PROCEDURE

Alcian blue is any 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 color is due to the presence of copper in the molecule. The 3% acetic acid solution (pH2.5) with alcian blue is believed to form salt linkages with the acid groups of acid mucopolysaccharides.

The periodic acid acts as oxidizing agent which oxidizes compounds having free hydroxyl groups or amino/alkyl amine groups. The tissue sections are first oxidized using periodic acid which oxidizes the vicinal bonds in these sugars, breaking the carbon-carbon bonds resulting in the pair of aldehydes. The oxidation step has to be regulated as to not further oxidize the aldehyde groups.

The aldehyde groups are detected by Schiff's reagent when exposed to it. The Schiff's reagent reacts with the aldehyde groups forming colorless, unstable dialdehyde compound which transforms to insoluble magenta colored complex by restoration of quinoid chromophoric grouping.

### REAGENTS PROVIDED

| Kit Contents                             | Product Code | Storage Conditions | Pack Sizes |       |       |
|--|--------------|--------------------|------------|-------|-------|
|  |              |                    | 100ml      | 250ml | 500ml |
| Alcian Blue Solution (Reagent A)         | SS005        | RT                 | 100ml      | 250ml | 500ml |
| Periodic Acid Solution (Reagent B)       | IPS018       | 2-8°C              | 100ml      | 250ml | 500ml |
| Schiff's Reagent (Reagent C)             | SS003        | 2-8°C              | 100ml      | 250ml | 500ml |
| Modified Mayer's Hematoxylin (Reagent D) | PS020        | RT                 | 100ml      | 250ml | 500ml |

#### TROUBLESHOOTING

1. Follow the specific protocol recommendations according to data sheet provided
2. 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
3. Do not allow the section to dry out during the entire staining process
4. Excessive or incomplete counterstaining may compromise the interpretation of the results
5. 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

Authorized and skilled personnel only may use the product. The clinical interpretation of any test results should be evaluated within the context of the patient's medical history and other diagnostic test results. A qualified pathologist must perform the evaluation of the test results. There are no warranties, expressed or implied, which extend beyond the description. PathnSitu is not liable for property damage, personal injury, time or effort on economic loss caused by this product.

#### BIBLIOGRAPHY

1. Bancroft, John D., and Marilyn Gamble. *Theory and Practice of Histological Techniques*. 6th ed. Oxford: Churchill Livingstone Elsevier, 2008. 173-174.
2. Carson, Freida L., and Christa Hladik Cappellano. *Histotechnology: A Self-instructional Text*. 4th ed. Chicago: ASCP Press, 2015. 150-151.
3. Romeis - Mikroskopische Technik, Editors: Mulisch, Maria, Welsch, Ulrich, 2015, Springer-Verlag Berlin Heidelberg.
4. Conn's Biological Stains: A Handbook of Dyes, Stains and Fluorochromes for Use in Biology and Medicine, 10th Edition, (ed. Horobin, R.W. and Kiernan, J.A). Bios, 2002.

#### EXPLANATION OF SYMBOLS

LOT- Lot number / Batch number



Expiry



Storage limitation

Laboratory Use Only

RT- Room Temperature