

Hackam Lab Alcian Blue, pH 2.5

FIXATION

4% Paraformaldehyde, in bed through graded alcohol series to paraffin.

SECTIONS

Paraffin, 5 micrometers

SOLUTIONS

3% Acetic Acid Solution

3ml of Acetic Acid Glacial (Fisher Cat# A38SI-212) add to 97ml water

Alcian Blue Solution

Alcian blue, 8GX

Acetic acid, 3% solution

1.0gm

100.0ml

} Mix overnight and filter through Q8
filter paper

Nuclear Fast Red Solution (Sigma cat no. N3020)

PROCEDURE

- **Warm slides at 60°C for 30' in vacuum incubator (15mm Hg Vacuum)**
 - **Transfer directly to Xylene while they are hot from incubator**
 - Xylene 2x5'
 - 100% ETOH 2x5'
 - 95% 1x5'
 - 70% 1x5'
 - Water 2x5'
1. Place in 3% acetic acid solution for 3 minutes.
 2. Stain in Alcian blue solution for 30 minutes.
 3. Wash in running water for 10 minutes.
 4. Rinse in distilled water.
 5. Counterstain in nuclear fast red solution for 30-60 minutes (check for red nuclear staining).
 6. Wash in running water for 1 minute.
 7. 95% alcohol 2x5' (dips in and out for best results)
 8. 100% alcohol 2x5' (dips in and out for best results)
 9. Xylene 2x5'(dips in and out for best results)
 10. Mount with mixture containing 70% Permount and 30% xylene