

Staining with Alcian blue at pH 2.5

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Immunogenic glycoprotein components in glycoconjugates (such as mucins) can be discerned by immunohistological methods. Moreover, lectin binding studies with immunohistological methods enable specific detection of carbohydrate moieties in tissue sections. This is because lectins can be conjugated with marker molecules (f.e. enzymes) in a similar way to antibodies in immunohistology. These immunohistological and lectin binding studies, however, may be accompanied by classical histochemical mucin stains for additional informations.

Two major types of mucins can be readily distinguished by histochemical methods, i.e. acid and neutral mucins (glycoconjugates). A significant advance in the histological detection of mucins was achieved by the introduction of stainings such as the periodic acid-Schiff (PAS) and Alcian blue at pH 2.5, Alcian blue at pH 1.0 and high iron-diamine/Alcian blue as staining agents. Moreover, several special methods which induce defined chemical or enzymatic alterations in tissue sections have been developed for the staining of reactive groups in epithelial mucins.

Alcian blue 8GX is primarily used for the detection of acid mucins. Immunostained tissue sections may be processed in parallel to routine histological sections.*

Alcian blue pH 2.5 (Mowry)

Chemicals	Chemical solution
Alcian blue 8 GX (C.I. 74240) Glacial acetic acid Ethanol Distilled water	<ul style="list-style-type: none">• 3% acetic acid solution: 3.0 mL glacial acetic acid dissolved in 97.0 mL distilled water• Alcian blue dye solution: 1.0 g alcian blue dissolved in 100.0 mL 3% acetic acid <p>glacial acetic acid is added to distilled water to give pH 2.5 which corresponds to approx. 3% acetic acid</p> <p>dye solution is filtered prior to use</p>
Staining procedure Sections are passed through distilled water and stained:	

* Dyes and other chemicals in histological staining can be toxic. They must be handled with care

– 3% acetic acid solution	2 min
– Alcian blue dye solution	5-20 min
– distilled water	2 rinses
– 70% ethanol	2 x 1 min
– 95% ethanol	2 x 1 min
– absolute ethanol	2 x 2 min
Slides are cleared in xylene or xylene substitute and mounted in resinous medium under coverglass	

References for further readings

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