Haematoxylin (Mayers)

S058

Haematoxylin (Mayer's) is recommended for Immunohistochemical and cytochemical Staining (as Nuclear Counter Stain) (PAS Staining Procedure). It may also be used for routine Haematoxylin and Eosin Staining.

Composition**

Propritory

Directions

A.Nuclear counter stain for Immunohistochemical staining:

1. Complete individual staining procedure (as desired). Rinse the slide with deionized water.

2. Stain the tissue section or the cell preparation with hematoxylin for 30-60 seconds.

3. Rinse with water to remove excess reagent.

4. Place in bluing reagent (alkaline solution such as a weak ammonia solution, 0.08% in water) until stain is blue (approximately 30 seconds).

5. Rinse in deionized water.

6. Section can be mounted in aqueous mounting media.

B.Hematoxylin and Eosin staining:

1. Prepare 95% alcohol solution.

2. Deparaffinize the tissue section and hydrate to water or fix and hydrate frozen sections.

3. Stain tissue section or cell preparation for 30-60 seconds with hematoxylin.

4. Rinse with water to remove excess reagent.

5. Place in bluing reagent until the stain is blue.

6. Rinse in deionized water.

7. If alcoholic eosin is used, place slide in 95% alcohol for 30 seconds.

8. Place eosin counter stain for 30-60 seconds.

9. Dehydrate in two changes each of reagent 95% alcohol, absolute alcohol and xylene for 2 minutes each.

10. Mount with synthetic mounting medium and examine the slide under microscope.

Principle And Interpretation

Hematoxylin is extracted from the heartwood of the logwood tree, Hemtoxylin campechianum. Hematoxylin (Mayer's) solution contains the dye, hematin and the Aluminum potassium sulfate as a mordant which provides the stain colour (blue). Sodium iodate acts as an oxidizing agent and glacial acetic acid controls the pH of the Solution.

Hematoxylin (Mayer's) reagent is alcohol free and is suitable for the use with all chromogens commonly used in immunohistochemical application. It can be used as a progressive or as a regressive stain. For histochemical purposes, the progressive staining is commonly used in which dye selectively stains the nuclear chromatin without staining cytoplasmic structures. Slides are left in the hematoxylin solution only long enough to stain the nuclei. The excess dye should be removed by 'bluing' of the tissue. Initially the tissue sections are coloured either purple or reddish purple, on exposure to alkaline solution, the tissue section takes on the characteristic blue colour.

Hematoxylin-Eosin is the commonly used stain, which is specific for certain substances of diagnostic importance. Here, acid reacting components of the cell combine with alkaline dyes and the alkaline area react with acid dyes. The stain is applicable staining of amyloid, lipids, inorganic substances such as iron and calcium, pigments like melanin and hemosiderin, carbohydrates and mucopolysaccharides.

Quality Control

Appearance
Red coloured solution.

Clarity
Clear without any particles.

Microscopic Examination
Staining is carried out staining characteristics is observed under microscope by using oil immersion lens.

Results
Nuclei : Blue

Storage and Shelf Life

Store below 30°C in tightly closed container and away from bright light. Use before expiry date on label.

Reference

1. Clark G (Ed)1981, Staining Procedures 4th edition, Published for the Biological Stain Commission by Williams & Wilkins, London.
2)Bauer J.D., Ackermann P.G. and Toro G. (Eds.), 1974, Clinical Laboratory Methods, 8th ed., The C.V. Mosby Co., St. Louis.

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