MAYER’S HEMATOXYLIN SOLUTION

INTENDED USE

Mayer’s Hematoxylin Solution is commonly used after immunohistochemistry or cytchemistry staining as a nuclear counterstain. It may also be used for standard hematoxylin and eosin (H&E) staining, but it is more commonly used where acid alcohol differentiation, or exposure to alcohol, might destroy the stained cytoplasmic component. Mayer’s Hematoxylin Solution is formulated without alcohol, and as such will not dissolve out AEC (3-amino-9-ethylcarbazole), alkaline phosphatase/Fast Red chromogen or other soluble colored products. Mayer’s Hematoxylin Solutions are for “In Vitro Diagnostic Use.”

PRECAUTIONS:

Filter Mayer’s Hematoxylin Solution before each use. Solution is then ready to use.

PREPARATION:

DETERIORATION:

until expiration date shown on the label. Do not return used solution to stock bottle.

STORAGE AND STABILITY:

Hematoxylin, a common nuclear stain, is isolated from an extract of logwood (Haematoxylon campechianum). The first successful biologic application of hematoxylin was described by Bohmer in 1865. Mayer introduced his formulation in 1903. Since then numerous formulations have appeared. Of these, Harris’, Gill’s, Mayer’s and Weigert’s have retained popularity. Before hematoxylin can be used as a nuclear stain, it must be oxidized to hematin and combined with a metallic ion (mordant). Most successful mordants have been salts of aluminum or iron. Generally, hematoxylin solutions are classified as progressive or regressive based on dye concentration. Progressive stains (e.g. Harris hematoxylin) have a lower concentration of dye and selectively stain nuclear chromatin without staining cytoplasmic structures. The desired intensity is a function of time. If staining times are excessive, a regressive stain might act similarly to a regressive stain solution. Staining with regressive stains generally requires more time than staining with regressive stains. Regressive stains (e.g. Harris hematoxylin) color all stainable tissue components (nuclear and cytoplasmic) intensely. To arrive at the correct staining response, excess dye must be removed from the tissue section. After sufficient differentiation, a properly defined section will demonstrate nuclear staining, but will not stain cytoplasmic structures.

The final step in hematoxylin staining is the “blueing” of the tissue section. Initially tissue sections are colored either purple or a reddish purple. After exposure to alkaline solutions (warm tap water [if slightly alkaline], dilute ammonia water, Scott’s tap water substitute, or lithium carbonate), the tissue section takes on the characteristic blue color of a hematoxylin stained slide.

REAGENT

MAYER’S HEMATOXYLIN SOLUTION, Catalog No. MHS
(MHS1-100ML / MHS16-500ML / MHS32-1L / MHS80-2.5L / MHS128-4L)
Certified hematoxylin (1.0 g/l), sodium iodate (0.2 g/l), aluminum ammonium sulfate·12 H2O (50 g/l), chlorydral hydrate (50 g/l) and citric acid (1 g/l).

STORAGE AND STABILITY:

Store reagent at room temperature (18–26°C) protected from light. Reagent is stable until expiration date shown on the label. Do not return used solution to stock bottle.

DETERIORATION:

Discard if staining times becomes excessive or solution turns brown.

PREPARATION:

Filter Mayer’s Hematoxylin Solution before each use. Solution is then ready to use.

PRECAUTIONS:

Normal precautions exercised in handling laboratory reagents should be followed. Dispose of waste observing all local, state, provincial or national regulations. Refer to Material Safety Data Sheet and product labeling for any updated risk, hazard or safety information.

PROCEDURE

PROCEDURE 1:

HEMATOXYLIN AND EOSIN STAINING

1. Prepare a 95% alcohol solution by adding 5 ml deionized water to 95 ml Reagent Alcohol or Ethanol (100%).
2. Deparaffinize to water or fix and hydrate frozen sections.
3. Stain in Mayer’s Hematoxylin Solution until nuclei are blue. Rinse in warm running tap water. If Alcoholic Eosin is to be used: Place in Reagent Alcohol, 95%. Place in Eosin Y Solution Counterstain.
4. Rinse in distilled water. If Alcoholic Eosin is to be used: Place in Reagent Alcohol, 95%. Rinse in running tap water or dilute alkaline solution until nuclei are blue.
5. Rinse in deionized water.
6. If any portion of the stain is alcohol soluble, mount in aqueous mounting media. If stain is alcohol insoluble, dehydrate in alcohol, clear in xylene or xylene substitute and mount in resins medium.
7. Place in Reagent Alcohol, 95%.
8. Place in Reagent Alcohol, Aqueous or Alcoholic with Phloxine...
9. Deparaffinize to water or fix and hydrate frozen sections.
10. Stain in Mayer’s Hematoxylin Solution 1–5 minutes. Rinse in running tap water or dilute alkaline solution until nuclei are blue.
11. Rinse in deionized water.
12. If any portion of the stain is alcohol soluble, mount in aqueous mounting media. If stain is alcohol insoluble, dehydrate in alcohol, clear in xylene or xylene substitute and mount in resins medium.

EXPECTED RESULTS

Nuclear chromatin should be blue. Nucleoli should be visible. Cytoplasm will display various shades of pink to pink-orange (depending upon the counterstain used) and red blood cells will be red.

7. Adding new stock to depleted working solutions of Mayer’s Hematoxylin or Eosin is not recommended.
8. Avoid excessive water carry-over into Mayer’s Hematoxylin.
9. Positive control slides should be included in each run.
10. The data obtained from this procedure serves only as an aid to diagnosis and should be reviewed in conjunction with other clinical diagnostic tests or information.

REFERENCES


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