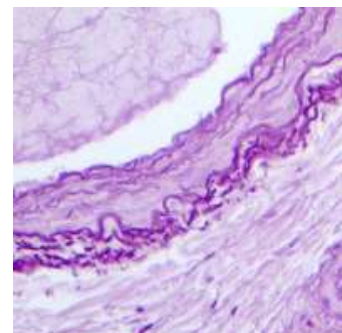


Orcein Solution

Description

Orcein Solution may be used in histology procedures for the visualization of Hepatitis B surface Antigen (HBsAg), elastic fibers, and copper associated proteins. HBsAg appears as irregular shaped aggregates in the cytoplasmic region of the cells. This reagent may be used on formalin-fixed, paraffin-embedded or frozen sections.



HBsAg	Dark Brown/Purple
Elastic Fibers	Dark Brown/Purple
Copper Assoc. Proteins	Dark
Purple Background	Light Reddish/Purple

Uses/Limitations

Not to be taken internally.
 Histological applications.
 Do not use if reagents become cloudy.
 Do not use past expiration date.
 Use caution when handling reagents.
 Non-Sterile.

Control Tissue

Known hepatitis positive liver.
 Lung for elastic fiber.
 Any well fixed tissue cut 3-5 microns.

Precautions

Avoid contact with skin and eyes.
 Harmful if swallowed.
 Follow all Federal, State, and local regulations regarding disposal.

Procedure

1. Prepare Oxidizing Solutions immediately prior to beginning procedure.

Combine: 50 ml Distilled Water
 5 ml Potassium Permanganate
 3 ml Sulfuric Acid Solution (3%)

Mix.

2. Deparaffinize sections if necessary and hydrate to distilled water.
3. Incubate slide in freshly prepared Oxidizing Solution for 10 minutes.
4. Rinse slide briefly in running tap water followed by 1 dip in distilled water.
5. Incubate slide in Oxalic Acid Solution (2%) for 10 minutes.

6. Note: Section should be colorless following this step.
7. Rinse slide for 1 minute in running tap water followed by 2 dips in distilled water.
8. Apply adequate Orcein Solution to cover tissue and incubate slide in Orcein Solution for 2 hours.
9. Note: Check periodically and apply additional stain as needed to avoid drying.
10. Rinse slide in Alcohol, Reagent (70%).
11. Apply Differentiating Solution for 3-5 seconds.
12. Dip slide in Alcohol, Reagent (70%) and check slide microscopically for proper differentiation.
13. Note: Repeat step 8 if necessary.
14. Dehydrate quickly in 3 changes of absolute alcohol.
15. Clear, and mount in synthetic resin.

References

1. Deodhar K.P., Tapp E., Scheuer P.J. Orcein staining of Hepatitis B Antigen in paraffin sections of Liver Biopsies. *Journal of Clinical Pathology*; vol. 28: pages 66-70, 1975.
2. Salaspuro, M., Sipponen, P. Demonstration of an intracellular copper-binding protein by Orcein staining in long-standing cholestatic liver diseases. *Gut*, 1976, volume 17: pages 787-790.