

Hematoxylin and Eosin Stain

FOR PARAFFIN-EMBEDDED TISSUE SECTIONS

Stock Solutions:

1% stock Eosin

Eosin Y 1gm
80% EtOH 100ml

Eosin Working Solution

Stock Eosin 45ml
80% EtOH 135ml
Glacial Acetic Acid 900ul

0.1% Acid Alcohol Solution

ddH₂O 500ml
100% EtOH 500ml
HCl 1ml

Harris Hematoxylin

Nonacidified
Shandon
Product # 6765001

1. Deparaffinize slides and hydrate to ddH₂O: (Xylenes 2x5min,
100% EtOH 2x1min, 95% EtOH, 80% EtOH, 70% EtOH, ddH₂O 1min each)
2. Filter undiluted Harris Hematoxylin and Eosin (Whatman #4 filter paper)
3. Immerse sections in filter Harris Hematoxylin for 5min
4. Rinse in 2 changes of ddH₂O
5. Differentiate in 0.1% acid alcohol solution: (EtOH fixed
tissue for about 2sec) (NBF fixed tissue for
about 4sec)
6. Rinse in tap H₂O for 10-15min or until sections turn bright blue
7. Immerse in Eosin for 1min
8. Briefly rinse in tap H₂O
9. Dehydrate in 95% EtOH 2x30sec, 100% EtOH 2x1min, Xylene 2x5min
10. Coverslip with Cytoseal

Results:

Nuclei stained blue.
Cytoplasm stained various shades of pink, identifying different morphologic tissue components.