

Protocol for Hematoxylin and Eosin staining

> Materials

- Hematoxylin solution
- Eosin solution
- 70% Ethanol
- 95% Ethanol (95%)
- Absolute Ethanol (99.5%)
- Tissue clear solution (e.g. Histolab-clear)
- Slides
- Slides support
- Coating/protection solution (e.g. Pertex, Mowiol)
- Coverslips
- Forceps

> Protocol

1. Incubate slides in Hematoxylin solution for 3 min
2. Transfer slides to bath with tap water
3. Rinse with tap water (cold) for more than 5 min (continuous flow)
4. Incubate in Eosin solution for less than a minute
5. Transfer to bath with distilled water (dunking)
6. Rinse in 70% Ethanol (dunking)
7. Rinse in 96% Ethanol (dunking)
8. Incubate/rinse in absolute Ethanol (1 min)
9. Incubate/rinse again in absolute Ethanol (1 min)
10. Optional: Incubate in tissue clear solution (e.g. Histolab-clear) 2 x for 1 min each
11. Mount slides with coating/protection (e.g. Pertex, Mowiol)
12. Air dry for a minimum of 2hrs (better overnight)
13. Store at room temperature