

## H&E Stain Protocol

<i>Station</i>	<i>Solution</i>	<i>Time</i>
1	Xylene	3 minutes
2	Xylene	3 minutes
3	Xylene	3 minutes
4	100% Dehydrant	1 minute
5	100% Dehydrant	1 minute
6	100% Dehydrant	1 minute
7	95% Dehydrant	1 minute
8	Rinse in running tap water	Briefly
9	Deionized or distilled water	Rinse
10	Hematoxylin	2-4 minutes
11	Running tap water	Rinse off excess stain
12	Acid Alcohol	30 seconds to 1 minute
13	Rinse in running tap water	30 seconds (agitate)
14	Bluing Reagent	1 minute
15	Rinse in running tap water	1 minute
16	95% Dehydrant	Rinse
17	Eosin-Y	0.5-3 minutes
18	100% Dehydrant	1 minute
19	100% Dehydrant	1 minute
20	100% Dehydrant	1 minute
21	Xylene	1 minute
22	Xylene	1 minute
23	Xylene	1 minute

### Frozen Section Staining Procedure:

Rinse slides in water rinse for 4-5 dips

1. Hematoxylin 3-5 dips
2. Water Rinse 5-7 dips
3. Bluing Rinse 3-4 dips
4. Water Rinse 3-4 dips
5. 95% Reagent Grade Alcohol 3-4 dips
6. Eosin-Y 1-2 dips
7. Water Rinse 1-2 dips
8. 100% Reagent Grade Alcohol 5-8 dips
9. 100% Reagent Grade Alcohol 5-8 dips
10. Clearing Reagent 10 dips
11. Clearing Reagent 10 dips Coverslip