



Hematoxylin and Eosin Staining System

Pureview® Staining System,

Greater slide throughput, 2 year shelf life

The **Pureview™ Staining System** is designed to produce precise, consistent H&E staining. Staining times in hematoxylin and eosin will depend on the use and the personal preference of the diagnosing physician and the intensity of nuclear staining will increase as the time in stain increases. The hematoxylin and eosin stains can have an expected throughput of 2,000-2,500 slides per 500mL bottles while the bluing and clarifier solutions should be changed more often. Actual results may vary depending on control of carry over into each solution, length of time stains are left exposed and the staining equipment being used. As a general rule, we recommend changing the hematoxylin and eosin stains once per week if throughput is not achieved. The following staining guidelines below will help establish a custom protocol for your specific needs.

Hematoxylin 660+™ (16600 & 16600-4) **Hematoxylin 660+™(XL) (16600-XL & 16600-XL4)**

Hematoxylin 660+™ is a proprietary, progressive hematoxylin formulated to produce crisp, well-delineated nuclear chromatin while eliminating background staining. Recommended staining times in **Hematoxylin 660+™** range from 2 to 6 minutes. **Hematoxylin**

660+™ is an excellent nuclear stain for routine Hematoxylin and Eosin staining, frozen sections and IHC. It does not contain mercury and does not require filtering before use.

Hematoxylin 660+™(XL) is formulated as a more intense version of **Hematoxylin 660+™** for darker, quicker staining making it an excellent choice for either routine or frozen section staining. It does not contain mercury or alcohol.

Focus™ Aqueous RTU (16703 & 16703-4) **Focus™ Aqueous Concentrate (add to water) (16703C & 16703-C4)** **Focus™ Alcoholic RTU (16703-AL & 16703-AL4)** **Focus™ Alcoholic Concentrate (add to alcohol) (16703-AL-C & 16703-AL-C4)**

Focus™ Clarifiers are an aqueous (or alcoholic) clarifying agent formulated to be used with **Hematoxylin 660+™** and **Hematoxylin 660+™(XL)** to increase cell transparency and to help eliminate any background staining. One of the **Focus Hematoxylin 660+™(XL)** clarifying solutions should be used to selectively remove non-specific staining, including background caused by water bath adhesives or charged slides, without affecting nuclear staining.

After hematoxylin staining, excess hematoxylin is rinsed from the tissue section and slides are exposed to **Focus™** for 30 seconds. If more transparency is desired or if non-specific staining is present, the exposure





time can be increased. **Focus™** is supplied in a ready to use (RTU) formulation and is also available as a concentrate. (Aqueous and Alcoholic versions)

PH Blue™ RTU (16702 & 16702-4)
PH Blue™ Concentrate (16702-C & 16702-C4)

PH Blue™ is a buffered bluing reagent with a pH of 8.0 to assure bluing of hematoxylin stained tissue sections. This buffering system allows the solution to be more resistant to the effects of carryover and remain in the ideal bluing pH range of 7.0-8.0 longer for consistent results. (higher pH may result in tissue detaching from slides “floaters” and lower pH ranges can result in unclear nuclear detail. Suggested bluing time is 1 minute; after bluing, rinse tissue sections in running water for 1 minute or in 2 changes of deionized water. Excess bluing reagent must be completely removed prior to eosin staining. **PH Blue™** is available as a concentrate and in a working solution that requires no dilution.

Eosin 615™ (16601 & 16601-4)
Eosin 615™ LITE (16601-LT & 16601-LT4)
Eosin 615™ TriChrome (16601-TC & 16601-TC4)
Eosin 615™ Phloxine (16601-PH & 16601-PH4)

Eosin 615™ series are available in four different versions to accommodate varying preferences in cytoplasmic staining. The standard Eosin 615 series Eosin is an alcoholic eosin that allows for exceptional differentiation and contrast between cytoplasmic components and nuclei. **Eosin 615™** stains cytoplasm various shades of pink and red. Prior to staining in **Eosin 615™**, tissue sections should be rinsed in alcohol. The concentration of the alcohol rinse can range from 70% to 95%.

Staining time in **Eosin 615™** depends on the use and personal preference of the diagnosing physician. The following are guidelines that will help establish a custom protocol for your specific needs. When staining routine hematoxylin and eosin sections, good results can be obtained with **Eosin 615™** with a staining time of 30 to 90 seconds. A staining time of 30 seconds is recommended as an initial staining time, with an increase of 30 second intervals until desired color and density of the cytoplasm is achieved.

Eosin 615™LITE is recommended when less differentiation/lighter staining of the cytoplasm with a more pastel appearance is preferred. **Eosin 615™TriChrome** produces three distinct varying cytoplasmic shades and **Eosin 615™Phloxine** is formulated to produce “a more reddish” coloration in the cytoplasm, especially in connective tissue components.

Recommended automated and manual histology staining procedure for **Pureview® Hematoxylin 660+™ (Series) & Eosin 615™ (Series) Stains**

Initially deparaffinize tissue sections in 3 changes of Xylene* of 3 minutes each.

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| 1. 100% Ethanol | 1 minute |
| 2. 100% Ethanol | 1 minute |
| 3. 100% Ethanol | 1 minute |
| 4. 80% Ethanol | 1 minute |
| 5. Running H2O Wash | 1 minute |
| 6. Hematoxylin 660+™ | 3-6 minutes |
| 7. Running H2O Wash | 1 minute |
| 8. Focus® | 30 seconds |



9. Running H₂O Wash 1 minute
10. **PH Blue™** 1 minute
11. Running H₂O Wash 1 minute
12. 80% or 95% Ethanol 30 seconds
13. **Eosin 615™** 30-90 seconds
14. 100% Ethanol 1 minute
15. 100% Ethanol 1 minute
16. 100% Ethanol 1 minute
17. Xylene* 1 minute
18. Xylene* 1 minute
19. Xylene* 1 minute
20. Mount and coverslip with **CoverSeal®**

***Zero Xylene™** or **ClearAway Citrus™** may be used in place of Xylene. Times in Xylene substitutes may need to be extended.

