

Hematoxylin

(Gill's Formula)

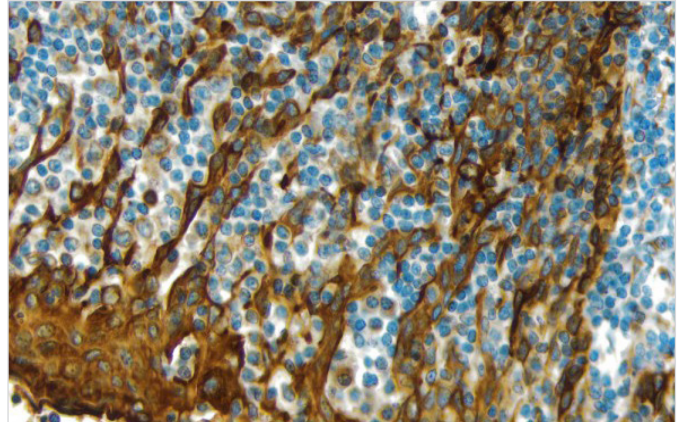
Cat. No. H-3401

Storage 15–25 °C

Description This ready-to-use nuclear counterstain is designed to be used after completion of immunohistochemical staining or for routine histology. Nuclei in stained sections will be blue.

Components

Product Name	Volume
Hematoxylin (Gills Formula)	500 ml



Tonsil: Cytokeratin (m), ImmPRESS® Anti-Mouse IgG Kit, ImmPACT® DAB substrate (brown). Counterstained with Hematoxylin (blue).

Instructions for Use

1. Rinse slides in tap water.
2. Immerse slides in Hematoxylin or apply counterstain directly to slide, completely covering the tissue section.
3. Incubate sections with Hematoxylin for 1–5 minutes.
4. Rinse sections with running tap water until rinse water is colorless.
5. Differentiate by dipping slides 10 times in acid rinse solution (2 ml glacial acetic acid plus 98 ml of deionized or distilled H₂O) followed by 10 dips in tap water.
6. Incubate slides in appropriate bluing solution.*
7. Mount sections in aqueous or permanent mounting media as per standard protocols.

* Bluing solutions:

(a) Add 1.5 ml NH₄OH (30% stock) to 98.5 ml of 70% ethanol (not recommended for substrates sensitive to alcohols, e.g. AEC, ImmPACT AEC, ImmPACT AMEC Red substrates). Incubate for 1 minute and rinse briefly (10 to 20 dips) in tap water.

(b) Add 0.3 ml NH₄OH (30% stock) to 100 ml tap water. Dip 3–5 times and rinse briefly (10 to 20 dips) in tap water.

Notes

- For quick hematoxylin staining without a bluing step, please use Hematoxylin QS (H-3404).
- Counterstain intensity should be optimized for each tissue type, antigen unmasking protocol, and immunocytochemical staining intensity desired.
- Generally, hematoxylin solutions produce a more intense nuclear stain with age. Staining times can be adjusted to achieve the desired nuclear intensity.
- See reverse side for substrate compatibility.
- Use ordinary precautions to avoid contact with skin and eyes.
- This product is for research use only.

Counterstain/Substrate Compatibility Table

This table is designed as a reference to determine the optimal counterstain/substrate combination for your application. Considerations should be given to tissue type, antigen unmasking protocol, and other detection parameters to achieve the desired staining intensity.

Substrate	Catalog Number	Hematoxylin and Hematoxylin QS H-3401 and H-3404	Methyl Green H-3402	Nuclear Fast Red H-3403
ImmPACT® DAB (brown)	SK-4105	Excellent Contrast	Excellent Contrast	Fair Contrast
ImmPACT DAB EqV	SK-4103	Excellent Contrast	Excellent Contrast	Fair Contrast
DAB (brown)	SK-4100	Excellent Contrast	Excellent Contrast	Fair Contrast
DAB-Ni (gray-black)	SK-4100	Excellent Contrast	Fair Contrast *	Good Contrast
ImmPACT AEC (red)	SK-4205	Excellent Contrast	Counterstain Incompatibility **	Color Incompatibility
ImmPACT AMEC Red (red)	SK-4285	Excellent Contrast	Counterstain Incompatibility **	Color Incompatibility
AEC (red)	SK-4200	Excellent Contrast	Counterstain Incompatibility **	Color Incompatibility
TMB (blue)	SK-4400	Color Incompatibility	Counterstain Incompatibility	Excellent Contrast
ImmPACT VIP (purple)	SK-4605	Fair Contrast	Excellent Contrast	Poor Contrast
Vector® VIP (purple)	SK-4600	Fair Contrast	Excellent Contrast	Poor Contrast
ImmPACT SG (blue-gray)	SK-4705	Poor Contrast	Good Contrast	Excellent Contrast
SG (blue-gray)	SK-4700	Poor Contrast	Good Contrast	Excellent Contrast
ImmPACT NovaRED® (red)	SK-4805	Excellent Contrast	Excellent Contrast ***	Color Incompatibility
Vector NovaRED (red)	SK-4800	Excellent Contrast	Excellent Contrast ***	Color Incompatibility
ImmPACT Vector Red (magenta)	SK-5105	Excellent Contrast	Excellent Contrast	Color Incompatibility
Vector Red (magenta)	SK-5100	Excellent Contrast	Excellent Contrast	Color Incompatibility
Vector Black (black)	SK-5200	Excellent Contrast	Excellent Contrast *	Excellent Contrast
Vector Blue (blue)	SK-5300	Color Incompatibility	Good Contrast	Excellent Contrast
BCIP/NBT (indigo)	SK-5400	Color Incompatibility	Excellent Contrast *	Excellent Contrast

* This substrate shows a slight decrease in sensitivity following the methyl green protocol. This decrease can be minimized by reducing the heat incubation and acetone rinse times in the methyl green protocol.

** Substrate dissolves in acetone wash.

*** A slight color change in ImmPACT NovaRED and Vector NovaRED reaction product may be seen using methyl green.