

Vector® Blue
Alkaline Phosphatase Substrate

Cat. No.: SK-5300

Storage: 2-8 °C

DESCRIPTION

This kit contains all of the reagents necessary to prepare the substrate working solution. Vector Blue Substrate produces a blue reaction product in the presence of alkaline phosphatase (AP) enzyme.

COMPONENTS

<u>Product Name</u>	<u>Volume</u>
Vector Blue Reagent 1	4 ml
Vector Blue Reagent 2	4 ml
Vector Blue Reagent 3	3 ml
Substrate Mixing Bottle	(Empty)

STORAGE:

- Store reagents in original bottles at 2-8 °C
- Avoid storing reagents or working solution in strong direct light

PREPARATION OF SUBSTRATE WORKING SOLUTION:

To 5 ml of 100 mM - 200 mM Tris-HCl, pH 8.2 - 8.5 buffer*

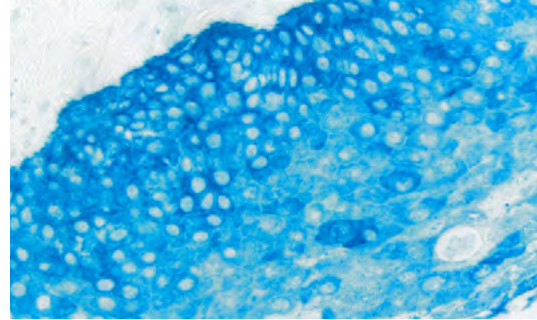
- Add 2 drops (≈80 µl†) of Vector Blue Reagent 1
- Add 2 drops (≈80 µl†) of Vector Blue Reagent 2
- Add 2 drops (≈45 µl†) of Vector Blue Reagent 3

Mix well before use. Use immediately.

* 0.1% Tween 20 in the Vector Blue working solution is recommended, especially for use with antigen retrieved tissues.

† Drop volumes differ due to solvent compositions.

IMPORTANT: Little is known about the toxicity and carcinogenicity of the substrate kit components. Appropriate care should be exercised when using this reagent including gloves, eye protection, lab coats, and good laboratory procedures. Dispose in accordance with local regulations.



Tonsil: Cytokeratin AE1/AE3 (m) detected with ImmPRESS-AP Reagent and Vector Blue AP Substrate (blue). No Counterstain.

INSTRUCTIONS FOR USE:

After incubation with an AP detection system, rinse sections in buffer. Incubate, with the substrate working solution for 20-30 minutes. Optimal development times should be determined by the investigator. Improved staining may be obtained by developing the substrate in the dark.

Wash slides in buffer for 5 minutes. Rinse in water and counterstain, if desired (see chart on reverse).

For permanent, non-aqueous mounting: Vector Blue is partially soluble in xylene; avoid xylene-based clearing agents and mounting media. Dehydrate sections, clear in a xylene-substitute clearing agent such as HistoClear or Clear-Rite 3 and coverslip in a non-xylene-based, non-aqueous mounting media, such as VectaMount® Mounting Medium (H-5000).

For aqueous mounting: Coverslip using an aqueous mounting media such as VectaMount® AQ Mounting Medium (H-5501).

NOTES:

Increasing the incubation time in substrate solution up to four hours may increase staining sensitivity 2-4 times, however background toning may also increase.

Detailed product listings, specifications, protocols and additional information is available on our website: vectorlabs.com