

ImmPACT[™] VIP Peroxidase Substrate Cat. No. SK-4605

Store at 2-8 °C

Burlingame, CA USA

ImmPACT[™] VIP Substrate

The ImmPACT™ VIP Substrate contains all of the reagents necessary to prepare a working solution for staining tissue sections or cells, or for use on nylon and PVDF membranes. In the presence of peroxidase (HRP) enzyme, the ImmPACT™ VIP Substrate produces a purple reaction product.

REAGENTS:

120 ml ImmPACT™ VIP Diluent 3 ml ImmPACT™ VIP Reagent 1 2.5 ml ImmPACT™ VIP Reagent 2 2.5 ml ImmPACT™ VIP Reagent 3 3 ml ImmPACT™ VIP Reagent 4

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 ImmPACT[™] VIP Diluent:
 - Add 3 drops (approximately 90 µ1*) of **ImmPACT**[™] **VIP** Reagent 1
 - Add 3 drops (approximately 75 µ1*) of ImmPACT™ VIP Reagent 2
 - Add 3 drops (approximately 75 μ 1*) of **ImmPACT**TM **VIP** Reagent 3
 - Add 3 drops (approximately 120 µ1*) of ImmPACT[™] VIP Reagent 4
- · Mix well before use

INSTRUCTIONS FOR USE:

After incubation with a peroxidase (HRP) detection system, rinse well. Incubate with the substrate working solution at room temperature for 2-15 minutes. Optimal development times should be determined by the investigator.

Wash slides for 5 minutes in water.

Counterstain, if desired, with Vector® Hematoxylin (Cat. No. H-3401), Vector® Hematoxylin QS (Cat. No. H-3404) or Vector® Methyl Green (Cat. No. H-3402)†. (See counterstain compatibility chart on reverse side.) Dehydrate and permanently mount in a non-aqueous mounting medium such as *VectaMount™* (Cat. No. H-5000) using standard protocols. Do not use recycled alcohols with this product.

† **ImmPACT**[™] **VIP** is not compatible with all methyl green formulations.

NOTES:

Unused working solution is stable up to 14 days if stored at 2-8 °C. Working solution may change color during storage but this will have no effect on the quality or intensity of the staining. If a precipitate forms filter through a $0.45 \,\mu m$ filter.

It is not necessary or recommended to add detergent to $ImmPACT^{TM}$ VIP working solution to reduce surface tension (e.g. for use in an automated stainer).

Prolonged incubation in alcohol or use of alcohol-based differentiating solutions may decrease sensitivity.

This product is not recommended for use on nitrocellulose membranes.

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.

* Drop volumes differ due to solvent compositions.