# KPL TMB Membrane Peroxidase Substrate System

| Catalog No. | Size |
|---|---|
| 5420-0025 (50-77-00) | 440 mL |

## DESCRIPTION

TMB Membrane Peroxidase Substrate system is a 3 component system which develops a dark blue precipitate on membrane sites bearing horseradish peroxidase. It is not recommended for microwell or immunohistochemical staining assays.

## FORM

5420-0025 (50-77-00) is a 3-component system consisting of the following:
2 x 100 mL KPL TMB Peroxidase Substrate
2 x 100 mL KPL Peroxidase Substrate Solution B
1 x 40 mL KPL TMB Membrane Enhancer

## STORAGE /STABILITY

Store components at 2-8°C. Stable for a minimum of 1 year from date of receipt when stored at 2 - 8°C. KPL TMB Peroxidase Substrate may develop a yellow tinge over time which does not affect product performance.

## CONTENT

KPL TMB Peroxidase Substrate contains 3,3',5,5'-tetramethylbenzidine at a concentration of 0.4 g/L in an organic base. KPL Peroxidase Substrate Solution B contains $H_2O_2$ at a concentration of 0.02% in a Citric Acid buffer. KPL TMB Membrane Enhancer is a proprietary formulation.

## APPLICATIONS

KPL TMB Membrane Peroxidase Substrate System is a precipitating substrate ideally suited for use in blotting procedures. The substrate may be adapted for use as a soluble substrate for ELISA by omitting the KPL TMB Membrane Enhancer.

## USE

Preparation: Mix one part of the KPL TMB Membrane Enhancer with five parts each of the KPL TMB Peroxidase Substrate and the KPL Peroxidase Substrate Solution B in a glass container prior to use (i.e. 5 mL Enhancer + 25 mL TMB Peroxidase Substrate + 25 mL Peroxidase Substrate Solution B, to yield a total volume of 55 mL).
The substrate solution should be clear. Warm to room temperature before use.

Recommended Substrate Volume: Immerse membrane in substrate using approximately 1 mL substrate per 10 $cm^2$ membrane.

Substrate Development: Following incubation with peroxidase labeled conjugate, wash membrane thoroughly. Incubate membrane in substrate for 5 - 15 minutes or until desired color is achieved. Incubation times will vary depending on your assay.

To Stop Substrate Reaction: Once desired color is achieved stop reaction by immersing membrane in reagent quality water for 20 - 30 seconds. The reaction should be stopped before background color becomes too intense resulting in insufficient contrast between positive staining and background.

Membrane Storage: Dry membrane thoroughly, seal with clear plastic and store in the dark to minimize fading.

To Reduce Substrate Sensitivity: High background or fading of the color in a few hours are signs of overreaction with KPL TMB Membrane Substrate. To reduce the intensity of the substrate reaction, it is recommended that the conjugate and/or antibodies in the immunoassay be further diluted. Dilution of the substrate is not recommended.

## PRODUCT SAFETY AND HANDLING

See SDS (Safety Data Sheet) for this product.

| RELATED PRODUCTS | CAT. NO. |
|---|---|
| KPL 1 Component TMB Membrane Peroxidase Substrate | 5420-0027 (50-77-03) |
| KPL Milk Diluent/Blocking Solution | 5140-0011 (50-82-01) |
| KPL Wash Solution | 5150-0008 (50-63-00) |

The product listed herein is for research use only and is not intended for use in human or clinical diagnosis.