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TMB Substrate

P4: 50 ml
P6: 250 ml

For Research Use Only. Not for Use in Diagnostic Procedures.

Description

TMB Substrate used is ready to use for ELISA detection. Reaction between the substrate and immobilized horseradish peroxidase (HRP) conjugated secondary antibodies in the ELISA wells produces a blue colored solution. Color intensity and development time will vary depending on assay sensitivity and conditions. After reaching the desired color intensity, the reaction is terminated by addition of an acidic STOP solution which changes the solution color from blue to yellow. While the results will remain stable for one hour following termination, the plate should be analyzed promptly on a microwell reader at 450 nm. TMB is light sensitive and is therefore packaged in amber bottles to protect the solution from direct sunlight. Recommended storage temperature is 4°C.

Storage

Store at 4°C.

Background

TMB (3,3', 5,5"-tetramethylbenzidine) is used as a substrate to generate detectable signal in ELISA. The reaction between the TMB substrate and a peroxidase, typically horseradish peroxidase (HRP), produces a measureable color change that correlates with analyte level.

Directions for Use

- (a) After incubation with horseradish peroxidase (HRP) conjugated secondary antibody, wash plates with PBS/0.05% Tween-20.
- (b) Add 100 µl of TMB solution/well. Incubate for specific amount of time or until desired signal is achieved. See product datasheet for recommended incubation times for specific antibody pair.
- (c) Add 100 µl of STOP solution.
- (d) Read Absorbance at 450 nm.

Solutions and Reagents

TMB is a one component system that is ready to use from the bottle.

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