

## TMB EASE™, Ready-to-use

ELISA HRP Substrate  
Cat. No. 5320

### INTRODUCTION

Kem-En-Tec Diagnostics **TMB EASE, Ready-to-use substrate** is a new, highly sensitive and stable substrate with a unique aqueous formulation. This gives maximized safety during use and minimized waste problems after use.

The composition gives a slow assay reaction and is highly suitable for the direct switch from OPD\* based assays.

\* *ortho-phenylenediamine*

TMB\*\* produces a deep blue colour during the enzymatic degradation of H<sub>2</sub>O<sub>2</sub> by horseradish peroxidase and gives a clear yellow colour after addition of stop solution.

\*\* *3, 3', 5, 5'-tetramethylbenzidine*

### REAGENTS

**Substrate buffer** as well as **hydrogen peroxide** are included in this ready-to-use substrate. No other reagents should be added. Recommended stop solution is 0.18 M sulphuric acid.

### THE WORKING PROCEDURE IS VERY SIMPLE

1. The desired amount of substrate is poured into a sealed container and allowed to reach room temperature in the dark.
2. Recommended volumes of 100 µl substrate solution per microtiter well is added.  
Time of development is typically 15-30 minutes. We recommend the plate to be kept in the dark during incubation i.e. wrapped in tinfoil.
3. The blue colour is read directly in kinetic assays at 650 nm. For endpoint assays 100 µl (equal volumes) of 0.18 M sulphuric acid is added and the absorbance is read at 450/620 nm within one hour.
4. If reduced intensity is required it is recommended to shorten the time of development or to dilute the conjugate/antibody in the immunoassay further.

Dilution of the substrate is not recommended.

Contact our Technical Service at [support@kem-en-tec.com](mailto:support@kem-en-tec.com) for recommended instructions, should an even lower reaction rate be the demand.

### RECOMMENDED HANDLING AND STORAGE

The TMB substrates are extremely sensitive to certain handling and storage conditions. Avoid exposure to light, heat and contamination with metal ions or peroxidase. Redispense only into bottles made of High Density Polyethylene (HDPE), amber colour, i.e. Nalgene 2004, and keep the substrate at 2-8 C in the dark. (Dispensing guidelines are available upon request).

### ECO-TEK® BRAND NAME

The **ECO-TEK** brand name stands for environmentally-friendly products.

These products are characterized by offering the highest level of sustainability from replacing toxic components in buffer solutions to wasting less water.