# **Diagnopal Dynabioassay Products LTD**



ELISA super-sensitive, one component TMB-Substrate

**Product code: DPSSTMB** 

This product is highly Recommended For getting results from samples with the minimum analyte concentration that produces small amounts of Enzyme-Ab-Target Complex

#### General

TMB (3,3',5,5' tetramethylbenzidine) is a non-carcinogenic benzidine compound in a mildly acidic solution and a chromogen that yields a blue color when oxidized with hydrogen peroxide (catalyzed by HRP) with major absorbances at 370 nm and 652 nm. The color then changes to yellow (measured at 450 nm) upon addition of acid to stop the reaction. TMB is very sensitive and more quickly oxidized than other HRP substrates, resulting in faster color development.

## Form, Stability and storage

**Diagnopal** Super Sensitive ELISA Grade TMB (3,3',5,5') tetramethylbenzidine) is Liquid solution in amber reagent bottle. Store at  $2-8^{\circ}$ C. Do not freeze. Stable for 18 months from date of manufacture when stored at  $2-8^{\circ}$ C.

**APPEARANCE/BACKGROUND**: The expected appearance of the solution is colorless to very light yellow. The background of un-reacted substrate at 650 nm is 0.03 - 0.09.

#### **RECOMMENDED HANDLING**

Diagnopal TMB-Peroxidase Substrate (1- Component) is extremely sensitive to certain handling and storage conditions.

- \*\*Avoid exposure to sunlight, air and extreme temperatures, however exposure to indirect laboratory lighting for 8 hours is not harmful.
- \*\* Never pipette directly from bottle. Pour out needed amount into a plastic reservoir. Do not return excess Diagnopal TMB from reservoir to the primary container.
- \*\* Avoid exposure to silica-based materials including borosilicate glass, metals, bacterial contamination or other oxidizing agents.
- \*\* HDPE and LDPE bottles should be utilized as they are received directly from the manufacturer; washing of bottles is not recommended, but if you need acid washing and several times rinsing with D.W is a must.
- \*\* keep main bottle at 2 8°C and just bring daily use volume to room temperature.

### **Recommended protocol:**

**To start reaction:** Add 100-200  $\mu$ L of Diagnopal TMB-Substrate to each well and incubate 10-20 Min. TMB-Substrate color development following incubation with peroxidase labeled conjugate will occur. Over reaction shall lead to generation of precipitated particles. In these cases, you may need make

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dilution in your sample for quantitative tests or report more than higher than curve standards that you use.

**To Stop Reaction** For optimal performance, stop reaction by adding equal volume of Stop Solution to the microwell plate. This will halt color development and will turn it to yellow.

**To Read Reaction After stopping**, read at a wavelength of 450 nm. Stopped reaction should be read within 10-15 minutes.

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