

## **TMB Peroxidase EIA Substrate Kit**

**Catalog Number 172-1067**

**Contents**                      Solution A:            900 ml of 3,3',5,5'-tetramethylbenzidine in aqueous DMF.  
Refer to enclosed MSDS.

   Solution B:            100 ml hydrogen peroxide solution.

**Storage**                      The contents of this kit should be stored at 4 °C.

- Use**
1. Allow reagents to come to room temperature before using.
  2. Immediately before use, prepare substrate solution by combining 9 ml of solution A with 1 ml of solution B. Mix well.
  3. After the last rinse following incubation with peroxidase conjugate, add 100 µl/well of substrate solution. Gently shake plate during color development to mix contents of wells.
  4. Develop until deep blue color of reaction product appears. Read plate at 655 nm.
  5. Reaction may be stopped by adding 100 µl/well 1 N H<sub>2</sub>SO<sub>4</sub>. The color reaction will turn yellow. Read plate at 450 nm.

- References**
- Bos, E. S., *et al.*, *J. Immunoassay*, **2**, 187-204 (1981).
- Goka, A. K. J. and Farthing, M. J. G., *J Immunoassay*, **8**, 29-41 (1987).

This product is intended for research use only. It is not intended for clinical diagnostic purposes.