

## PRODUCT INFORMATION SUMMARY

TMB Substrate Solutions:

Solutions Part A and Part B 100 mL (2)

cat no: ES200 100 mL (2) cat no: ES200C 50 mL (2)

## DESCRIPTION:

TMB (3,3', 5,5' -Tetramethyl-Benzidine) substrate solution is used for ELISA applications in immunologic and serologic procedures where HRP (Horseradish Peroxidase) is applied. It is non-carcinogenic.

## HANDLING AND STORAGE:

PRODUCT USE: <u>CAUTION</u> - <u>FLAMMABLE</u> - see below -

Provided as 100 mL or 50 mL Solutions ( see above) Part A and Part B. Store DARK at 4 Deg. C. KEEP AWAY FROM OPEN FLAME - HEAT SOURCE.

Prepare substrate solution no more than 15 minutes before last incubation of assay: Mix one part TMB Solution A with one part TMB Soultion B in a clean container. If, upon mixing, TMB solution turns blue - TMB solution is CONTAMINATED - DO NOT USE. Use mixed substrate solution WITHIN 2 HOURS, AND AVOID DIRECT LIGHT.

Apply TMB soultion to wells of assay as substrate. For 96-well microplates, usually 100 uL per well is recommended. Be sure the ELISA plate is **thoroughly washed (5 times)** before adding TMB solution. (Wash steps are critical!)

For most ELISA applications, incubate TMB substrate solution for 15-20 minutes at 37 Deg. C. Then add 100 uL stop solution (2N Sulphuric Acid) and mix by gently tapping plate, then read O.D. at 450 nm within 30 minutes. - or follow customer's usual protocol.

CAUTION: Substrate solution B contains 20% acetone - FLAMMABLE - keep away from sources of heat or flame.