

3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

Product Information

3,3',5,5'-Tetramethylbenzidine

Catalog Number **T2885** Storage Temperature 2–8 °C

CAS RN 54827-17-7 Synonym: TMB

Product Description

Molecular Formula: C₁₆H₂₀N₂ Formula Weight: 240.34

3,3',5,5'-Tetramethylbenzidine (TMB) is a colorimetric substrate used with peroxidase and peroxidase-coupled systems. It produces a soluble end product, which is blue in color and can be read spectrophotometrically at 370 nm or 655 nm. The reaction can be stopped with 2 M sulfuric acid and read at 450 nm. This results in a 2–4 fold enhancement of the sensitivity of the reaction.

This product is also been used in the quantitative determination of hemoglobin and cytochemical staining for peroxidase.²

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

A TMB stock solution can be prepared by dissolving 1 mg/ml in DMSO. The working solution is made by diluting 1 ml of the DMSO stock with 9 ml phosphate-citrate buffer, pH 5.0, and then adding 2 μ l of fresh 30% hydrogen peroxide (Catalog Number H1009) per 10 ml of substrate/buffer solution immediately prior to use. The hydrochloride salt can be directly dissolved in aqueous media. For this reason, the hydrochloride salt is the preferred form of TMB; however, both forms will give identical results.

Stock solutions of TMB in DMSO can be frozen in 1–2 ml aliquots at –20 °C. Solutions appear to remain active for at least two years when stored in this manner. Any oxidation would be revealed as a color change in the working solution.

Storage/Stability

Store the product at 2–8 $^{\circ}$ C. It remains active for a minimum of two years as supplied.

References

 Liem, H.H., et al., Quantitative Determination of Hemoglobin and Cytochemical Staining for Peroxidase Using 3,3',5,5'-tetramethylbenzidine Dihydrochloride, a Safe Substitute for Benzidine. Anal. Biochem., 98, 388-393 (1979).

CS.CMH.MAM 03/09-1