

Product Information

3,3',5,5'-Tetramethylbenzidine Dihydrochloride

Tablet, 1 mg Substrate per Tablet

T3405

Product Description

3,3',5,5'-Tetramethylbenzidine (TMB) dihydrochloride is a peroxidase substrate suitable for use in ELISA procedures.¹⁻⁵ This substrate produces a soluble end product that is blue in color and can be read spectrophotometrically at 370 or 655 nm. The reaction may be stopped with 2 M H₂SO₄, resulting in a yellow solution that is read at 450 nm.

Each tablet contains 1 mg of TMB dihydrochloride substrate. The product is available in packages of 5, 50, or 100 tablets per box, individually foil wrapped for ease of use, storage, and safety. Custom packaging and bulk purchase information are available upon request.

Precautions and Disclaimer

This product is for Research Use Only. Not for Use in Diagnostic Procedures. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

0.05 M Phosphate-Citrate Buffer

- Dissolve one phosphate-citrate buffer tablet (for example, P4809) in 100 mL of ultrapure water with stirring, to yield a 0.05 M phosphate-citrate buffer, pH 5.0.

Or

- Add 25.7 mL of 0.2 M dibasic sodium phosphate (for example, S0876), 24.3 mL of 0.1 M citric acid (for example, C7129) and 50 mL of ultrapure water. Adjust the pH to 5.0, if necessary.

TMB Substrate Solution

- Dissolve one TMB dihydrochloride tablet in 1 mL of DMSO and add to 9 mL of 0.05 M Phosphate-Citrate Buffer, pH 5.0. Add 2 µL of fresh 30% hydrogen peroxide (for example, H1009) per 10 mL of substrate buffer solution, immediately prior to use.

Or

- Dissolve one TMB dihydrochloride tablet in 1 mL of DMSO and add to 9 mL of 0.05 M Phosphate-Citrate buffer, pH 5.0, containing 0.03% sodium perborate (capsules, P4922).

Stop Solution

The reaction may be stopped by the addition of 50 µL of 2 M H₂SO₄ per 200 µL of reaction mixture.

Storage/Stability

Store the TMB dihydrochloride tablets at 2–8 °C. Protect from heat, light, and moisture. Allow tablets to reach room temperature prior to use.

References

1. Liem, H.H. et al., *Anal. Biochem.*, 98(2), 388-393 (1979).
2. Rosenqvist, E. et al., "Determination of Antibody Responses to Meningococcal Antigens by ELISA", in *Meningococcal Vaccines: Methods and Protocols* (A.J. Pollard and M.C.J. Maiden, eds.). *Methods in Molecular Medicine*, Springer/Humana Press, pp. 255-273 (2001).
3. Fulton, S.A. et al., *J. Infect. Dis.*, 178(4), 1105-1114 (1998).
4. Slocombe, L.L., and Colditz, I.G., *Food Agr. Immunol.*, 22(2), 135-143 (2011).
5. Ucisik, M.H. et al., *Colloids Surf. B Biointerfaces*, 128, 132-139 (2015).

Notice

We provide information and advice to our customers on application technologies and regulatory matters to the best of our knowledge and ability, but without obligation or liability. Existing laws and regulations are to be observed in all cases by our customers. This also applies in respect to any rights of third parties. Our information and advice do not relieve our customers of their own responsibility for checking the suitability of our products for the envisaged purpose.

The information in this document is subject to change without notice and should not be construed as a commitment by the manufacturing or selling entity, or an affiliate. We assume no responsibility for any errors that may appear in this document.

Technical Assistance

Visit the tech service page at SigmaAldrich.com/techservice.

Terms and Conditions of Sale

Warranty, use restrictions, and other conditions of sale may be found at SigmaAldrich.com/terms.

Contact Information

For the location of the office nearest you, go to SigmaAldrich.com/offices.

The life science business of Merck KGaA, Darmstadt, Germany operates as MilliporeSigma in the U.S. and Canada.

MilliporeSigma, and Sigma-Aldrich are trademarks of Merck KGaA, Darmstadt, Germany or its affiliates. All other trademarks are the property of their respective owners. Detailed information on trademarks is available via publicly accessible resources.

© 2024 Merck KGaA, Darmstadt, Germany and/or its affiliates. All Rights Reserved.

T3405pis Rev0124

The logo for MilliporeSigma, featuring the word "Millipore" in a red, sans-serif font above the word "SIGMA" in a bold, red, sans-serif font.