Product Information

3,3'-Diaminobenzidine tetrahydrochloride hydrate

Catalog Number D5637
Storage Temperature –20 °C

CAS RN 868272-85-9
Synonyms: DAB HCl, 3,3',4,4'-Tetraaminobiphenyl tetrahydrochloride

Product Description

![Chemical Structure](image)

Molecular formula: C₁₂H₁₄N₄·4HCl·xH₂O
Molecular weight: 360.11 (anhydrous basis)

Diaminobenzidine is utilized in many applications for visualization of peroxidase activity. In the peroxidase reaction DAB serves as a hydrogen donor in the presence of peroxide; the oxidized DAB forms an insoluble brown end-product.

Precautions and Disclaimer

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

Storage/Stability

This product is shipped at ambient temperature. Storage at –20 °C, protected from light and moisture, is recommended.

Solution Stability

Diaminobenzidine HCl is soluble in water at 10 mg/mL.

Solutions of DAB should be made immediately prior to use for the best results. If absolutely necessary, solutions can be kept in the dark and refrigerated for overnight storage. Storing solutions overnight reduces reproducibility.

Literature indicates that it is possible to store solutions of DAB at –20 °C.¹ DAB dissolved in Tris buffered saline, pH 7.6, at 0.6 mg/mL was reported to be stable for several weeks at 4 °C.²

Procedure

**Immunoblotting and Immunohistological Staining**

1. Dissolve 10 mg of Diaminobenzidine HCl in 15 mL of buffer (Tris-buffered saline tablet, Catalog Number T5030).

2. Add 12 µL of fresh 30% hydrogen peroxide solution (Catalog Number H1009) prior to use. The substrate solution may be filtered through a 0.2 µm filter immediately prior to use if necessary.

Notes: The addition of nickel, cobalt, or silver ions has been found to increase the intensity of DAB staining.³,⁴ Several tablets and preformulated liquid substrates of DAB are offered, including a tableted product which contains a metal enhancer (see Related Products).

DAB and peroxide concentrations, as well as buffer composition can be varied in order to optimize specific applications.⁵,⁶,⁷

DAB has been used for staining and oxidative detection of mitochondria.⁸,⁹,¹⁰

Silver-gold enhancement has been utilized for electron microscopy applications.¹¹

DAB oxidation can be measured spectrophotometrically as a clear colloidal suspension in gelatin at 465 nm.¹²

Counterstaining methods for DAB include methylene blue, methyl green, and hematoxylin.¹³
References


Related Products

D9015 3,3’-Diaminobenzidine tetrahydrochloride hydrate, 100 mg ISOPAC®
D8001 3,3’-Diaminobenzidine Free Base
D5905 3,3’-Diaminobenzidine tetrahydrochloride 10 mg tablets (makes 15 mL)
D0426 SIGMAFAST™ DAB with metal enhancer Tablet Set (makes 5 mL)
D4168 SIGMAFAST 3,3’-Diaminobenzidine Tablet Set (makes 1 mL)
D4293 SIGMAFAST 3,3’-Diaminobenzidine Tablet Set (makes 5 mL)
D4418 SIGMAFAST 3,3’-Diaminobenzidine Tablet Set (makes 15 mL)
D7304 3,3’-Diaminobenzidine (DAB) Liquid Substrate System tetrahydrochloride
D7679 3,3’-Diaminobenzidine (DAB) Liquid Substrate Dropper System
SRE0079 3,3’-Diaminobenzidine tetrahydrochloride hydrate, Suitable for use in diagnostic manufacturing

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