



DAB Substrate System

Code	Description	Size
E885-200ML	DAB Substrate System	200 mL

General Information

In blotting, immunohistochemical and electron microscopy applications, 3,3'-diaminobenzidine (DAB) is frequently used for the chromogenic detection of horseradish peroxidase activity (HRP). An insoluble brown precipitate, resistant to alcohol and other organic solvents, as well as sunlight, forms upon the reaction of DAB with HRP in the presence of peroxide. The accumulation of the precipitate is visible without the need for specialized imaging equipment.

DAB is a suspected carcinogen and protocols based on DAB chromogenic detection require preparation of fresh working solution for each experiment. VWR Life Science AMRESCO's DAB Substrate System eliminates the need for weighing this hazardous material and also simplifies preparation and dispensing of a working solution. The kit includes twenty DAB substrate tablets, peroxidase solution, and a convenient mixing/dispensing bottle. Each DAB tablet yields 10 mL of substrate solution, enough to effectively detect 50 cm² of blotting membrane or about 20 slides.

Storage/Stability

Store cold (2 – 8°C). Prepared substrate solution can be stored cold for up to 6 hours.

Product Use Limitations

For research use only. Not for therapeutic or diagnostic use.

Materials Supplied

E733-20TABS	DAB Substrate Tablets, 5 mg/240 mg
E882-5ML	Hydrogen Peroxide, 30%
N/A	15 mL Mixing/Dispensing Bottle

Protocol/Procedure

HRP Detection with DAB Substrate System

1. Drop one DAB Substrate Tablet into the Mixing/Dispensing bottle and add 10 mL of deionized water. With the bottle sealed, shake gently until the tablet dissolves, about 2 – 3 minutes.
2. Dispense 1 drop of Hydrogen Peroxide, 30% into the Mixing/Dispensing bottle containing DAB solution. With the bottle sealed, mix the DAB/Hydrogen Peroxide mixture by gentle inversion.
3. Use the Mixing/Dispensing bottle to place the substrate solution onto HRP-labeled samples. Generally, 1 mL of substrate solution is sufficient for chromogenic detection of 5 – 10cm² of membrane and 0.5 mL is sufficient for most slides.
4. Unused substrate solution may be stored at 4°C for up to 6 hours.

For Technical Support

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