

Stable DAB/Plus

K047

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Intended Use

For In Vitro Diagnostic Use

Summary and Explanation

DAB, a widely used chromogen for immunoperoxidase staining, is well accepted among pathologists because of its increased sensitivity and ability to give cleaner background as compared to amino ethylcarbazole (AEC). Specimens stained in DAB can be dehydrated, cleared, and mounted for permanent record keeping. Stable DAB/Plus is more sensitive and stable than traditional working DAB solutions.

Principles of the Procedures

Substrate/chromogen in conjunction with peroxidase-based immunostaining systems.

Stable DAB/Plus offers several noteworthy improvements and benefits as compared with traditional working DAB solutions. Stable DAB/Plus is much more sensitive, providing the cost-effective option of diluting the primary antibody. Being stable for 5 days (as opposed to 6 hours for traditional DAB working solutions), Stable DAB/Plus allows the user the convenience of making one working solution for the entire work week. Hazardous waste generation from spent DAB solution is also significantly reduced. Stable DAB/Plus is ideal for high volume labs and automated stainers.

Peroxidase from the antibody detection system reacts with H₂O₂ substrate to degrade it, which then reacts with DAB, precipitating it at positive sites yielding a dark brown color.

Reagents Provided

Kits Contents	Volume
Concentrated Amber-Colored DAB Chromogen Solution	5 mL
Clear Stable DAB/Plus Substrate Buffer	200 mL
Empty Mixing Dropper Bottle	1

Prepare the Following Solutions Before Use

1. Aliquot 1mL of Stable DAB/Plus Buffer in mixing bottle.
2. Add 20µL (one drop) of concentrated Stable DAB/Plus Chromogen. Replace tip and mix.
3. The working Stable DAB/Plus solution is stable for at least 5 days and should be prepared in an opaque bottle.
4. Store at 2-8°C when not in use.
5. Any solution not used after this period should be discarded.

Materials Required But Not Provided

Some of the reagents and materials required for IHC are not provided. Pretreatment reagents, detection systems, control reagents and other

ancillary reagents are available from Diagnostic BioSystems. Please refer to the Diagnostic BioSystems website at www.dbiosys.com.

Storage and Handling

Store at 2°-8°C away from light. Do not use product after the expiration date printed on vial. If reagents are stored under conditions other than those specified here, they must be verified by the user. Diluted reagents should be used promptly.

Staining Procedure

1. After peroxidase incubation, wash tissue sections with wash buffer.
2. Wipe slides removing excess buffer. Add enough drops of working Stable DAB/Plus solution to cover tissue sections.
3. Incubate for 5-10 minutes at room temperature. For optimal results, observe reaction under the microscope for signal development.
4. Once the desired signal to noise ratio is achieved, stop the reaction by washing slides in buffer.

Precautions

1. Consult local and/or state authorities with regard to recommended method of disposal.
2. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
3. Avoid microbial contamination of reagents. Contamination could produce erroneous results.
4. This reagent may cause irritation. Avoid contact with eyes and mucous membranes.
5. If reagent contacts these areas, rinse with copious amounts of water.
6. Do not ingest or inhale any reagents.

Troubleshooting

If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem is suspected, contact Diagnostic BioSystems Technical Support at (925) 484-3350, extension 2 or techsupport@dbiosys.com.

