Diagnostic BioSystems

IHC Made Affordable

DAB Chromogen/Substrate Kit

K 007

Document #: DS-4012-A
Effective Date: 04/01/2015

Intended Use

For In Vitro Diagnostic Use

Summary and Explanation

DAB is a widely used chromogen for immunoperoxidase staining and immunoblotting. It has been well accepted amongst the pathologist because of its superior performance as compared to Amino Ethylcarbazole (AEC). DAB is much more sensitive and gives cleaner background as opposed to AEC. Specimens stained in DAB can be dehydrated in ethanol and cleared in Xylene and can be mounted for permanent record keeping. However, because of its carcinogenic nature, some labs avoid using DAB powder. To resolve this problem, we have designed DAB in liquid format to minimize the exposure of DAB to lab personnel.

Principles of the Procedures

As a substrate/chromogen in conjunction with peroxidase-based immunostaining systems. Peroxidase reacts with 3% Hydrogen Peroxide Substrate to degrade it, which in turn reacts with DAB to precipitate it at the positive sites giving dark brown color.

Reagents Provided

Kits Contents	Volume
DAB Substrate Buffer	200 mL
DAB Chromogen	10 mL
Empty Mixing Bottle	1

Prepare the Following Solutions Before Use

1. Transfer 1 mL of DAB Substrate Buffer in the mixing vial.
2. Add 50µL (two drops) of DAB Chromogen. Replace the tip and mix.
3. The working chromogen solution is stable for 6 hours. Any solution not used after this period should be properly 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. Once sections have been incubated with peroxidase, wash sections with wash buffer.
2. Wipe the glass to remove excess of buffer and add enough drops of the working DAB Substrate/Chromogen solution to cover the tissue sections.
3. Incubate for 5-15 minutes at room temperature.
4. For the best results, look under the microscope for the signal development.
5. Once desired signal to noise ratio is achieved, stop the reaction by washing slides in wash buffer.

Precautions

1. DAB has been classified as suspected carcinogen and can cause skin irritation upon contact.
2. Avoid contact with clothes and exposed skin.
3. If accidentally contacted, flush with tap water immediately.
4. Follow instructions provided by your local authorities for disposal.
5. Consult local and/or state authorities with regard to recommended method of disposal.
6. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
7. Avoid microbial contamination of reagents. Contamination could produce erroneous results.
8. This reagent may cause irritation. Avoid contact with eyes and mucous membranes.
9. If reagent contacts these areas, rinse with copious amounts of water.
10. 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.

Diagnostic BioSystems
6616 Owens Drive
Pleasanton, CA 94588
Tel: (925) 484 3350
www. dbiosys.com

CE

EC REP

Emergo Europe
Molenstraat 15
2513 BH, The Hague
The Netherlands
Tel: (31) (0) 70 345 8570