Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System

Intended Use

For In Vitro Diagnostic Use

Summary and Explanation

The Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System is a Biotin-Streptavidin-Horseradish Peroxidase Detection System that allows for the demonstration of antigens in paraffin-embedded tissue, cryostat sections, cytosmears, and cell preparations. The increased sensitivity of the Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System allows for rapid staining procedures without compromises in the quality of stains.

The Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System is suitable for use with mouse or rabbit primary antibodies. The Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System kits are optimized for use with Bio SB primary antibodies; however, they are universal kits and therefore work equally well with prediluted and concentrated antibodies from different vendors.

| Availability | | Presentation |
|----------------|--------------|---|
| Catalog Number | Volume | The Mouse/Rabbit ImmunoDetector AEC HRP Red Detection System |
| BSB 0002S | 5 mL Each | contains a Peroxidase Blocker solution, a Biotinylated Anti-Mouse and Anti- |
| BSB 0002 | 15 mL Each | Rabbit immunoglobulin solution, a Streptavidin conjugated to Horseradish |
| BSB 0004 | 50 mL Each | Peroxidase solution, and an AEC Chromogen Solution. All the components are |
| BSB 0006 | 100 mL Each | buffered with stabilizers and an anti-microbial. |
| BSB 0008 | 200 mL Each | |
| BSB 0010 | 1000 mL Each | Storage: Store at 2°C - 8°C |

Stability: Stable up to the expiration date listed on the label. Do not use this product after the expiration date listed on the product label.

Protocol

Preparation of Working Solutions

The ImmunoDetector Peroxidase Blocker, Anti-Mouse/Rabbit Biotinylated Link, Streptavidin Conjugated HRP Label, and AEC Chromogen are ready-to-use working solutions and require no further preparation.

Mounting Protocol

A. AquaMounter Protocol

- 1. After the histological, immunohistochemical or in situ hybridization staining procedure is completed, rinse slides in deionized water. Do not incubate tissue or cell specimens in solvents such as alcohol, toluene, or xylene.
- 2. After counterstaining, apply 1-3 drops of AquaMounter (BSB 0090- BSB0093) to each slide making sure the specimen is covered.
- Coverslip.
- 4. Observe under a light microscope.

B. ChromoProtector Permanent Mounting Protocol

- 1. After the histological, immunohistochemical or in situ hybridization staining procedure is completed, rinse slides in deionized water. Do not incubate tissue or cell specimens in solvents such as alcohol, toluene, or xylene.
- 2. Using a Coplin jar or a staining dish, immerse slides with tissues in ChromoProtector (BSB 0151 BSB 0156) or lay wet slides horizontally and apply sufficient drops of ChromoProtector (BSB 0151 BSB 0156) to completely cover the tissue. Carefully spread ChromoProtector if needed, but avoid contacting the tissue.
- 3. Incubate slides for ten minutes at 60 °C to allow ChromoProtector to penetrate tissues.
- 4. Remove excess ChromoProtector by placing slides vertically over an absorbent material and let excess drain off into absorbent material. Do not rinse slides.
- 5. Allow slides to COMPLETELY air dry.
- 6. NOTE: The ChromoProtector will protect tissue from drying artifacts during the air-drying process.
- 7. When slides are completely dried, they can be mounted using most standard mounting methods such as aqueous or permanent.
- 8. Organic solvent permanent mounting medium such as PermaMounter (Cat# BSB 0094-0097) or similar permanent mounting media, or the organic solvent-free XyGreen PermaMounter (Cat # BSB 0169-0174), can be added directly to the slide until the tissue or cell specimen is covered.
- 9. If the permanent mounting medium does not spread evenly on the dry slide, the slide can be dipped in toluene or xylene for 1-2 seconds to aid spreading of the mounting medium. Use a minimum amount of mounting medium so that slides dry rapidly.
- 10. Apply coverslip and air dry before microscopic observation.

Recommended Immunohistochemical Protocol

- 1. Cut and mount 3-4 micron formalin-fixed paraffin-embedded tissues on positive charged slides.
- 2. Air dry for 2 hours at 58° C.
- 3. Deparaffinize, dehydrate and rehydrate tissues.
- Subject tissues to heat epitope retrieval using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).
- 5. Wash with 5 changes of IHC Wash buffer.
- 6. Place slides in ImmunoDetector Peroxidase Blocker for 5 min.
- 7. Wash with 3 changes of IHC wash buffer.
- 8. Cover tissue with the **Primary Antibody** following manufacturer's recommended protocol. If using concentrated antibodies, we suggest using our **ImmunoDetector Protein Blocker/Antibody Diluent** to dilute antibodies.
- 9. Wash with 3 changes of IHC wash buffer.
- 10. Cover tissue with ImmunoDetector Biotin Link, incubate for 10 min.
- 11. Rinse with 3 changes of IHC wash buffer.
- 12. Cover tissue with **ImmunoDetector HRP Label**, incubate for 10 min.
- 13. Rinse with 5 changes of DI water.
- 14. Cover Tissue with the **AEC Chromogen** solution, incubate for 5 10 min.
- 15. Rinse with 5 changes of DI water.
- 16. Counterstain.
- 17. Coverslip with a water-based mounting medium.

| Abbreviated Immunohistochemical Protocol | | |
|--|-------------------------------|--|
| Step | ImmunoDetector HRP | |
| Peroxidase Blocker | 5 minutes | |
| Primary Antibody | 30 – 60 minutes | |
| Biotin Link | 10 minutes | |
| HRP Label | 10 minutes | |
| AEC Chromogen | 5 - 10 minutes | |
| Counterstaining | Time varies with counterstain | |

Precautions

- 1. For professional users only. Results should be interpreted by a medical professional.
- 2. Ensure proper handling procedures are used with reagent. Always wear proper personal protective equipment such as laboratory coat, goggles and gloves when handling reagents.
- 3. This product contains components with sodium azide (NaN₃), a toxic chemical. At product concentrations it is not classified as hazardous due to its low concentration. Sodium azide may react with plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build up.
- 4. Minimize microbial contamination of reagents.
- 5. Dispose of unused solution according to local and federal regulations.
- 6. Do not ingest reagent. If reagent ingested, seek medical advice immediately.
- 7. Avoid contact with eyes. Flush with large quantities of water if contact occurs.
- 8. Follow safety precautions for the heating device (TintoRetriever Pressure Cooker or similar).
- 9. For complete recommendations for handling biological specimens please refer to the CDC document, "Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories" (1).

References

1. U.S. Department of Health and Human Services: Centers for Disease Control and Prevention. Guidelines for Safe Work Practices in Human and Animal Medical Diagnostic Laboratories. Supplement / Vol. 61, January 6, 2012.



