

Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System



Intended Use

For In Vitro Diagnostic Use.

This detection system is intended for the fast immunohistochemical detection of antibodies used for Mohs surgery on frozen or FFPE tissue sections. Interpretation of results should be performed by a qualified medical professional.

Summary and Explanation

The **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** is a non-biotin 2-step Fab micropolymeric detection system that allows for the demonstration of antigens in cryostat sections, formalin-fixed paraffin-embedded tissues, blood smears, cytospins, and cell preparations. The Mohs PolyDetector Plus kit has been developed using a biopolymer conjugated to monomeric Fab' immunoglobulin fractions targeting the Fc region of Mouse and Rabbit antibodies. Additionally, the biopolymer is labeled with high quality HRP for maximum sensitivity. This ensures excellent cellular penetration which generates consistent, reproducible sensitive and specific immunostainings for all types of nuclear, cytoplasmic and membranous localization in frozen and FFPE tissues.

The increased sensitivity of the **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** allows for rapid staining procedures without compromising stain quality. The **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** is suitable for use with mouse IgG and IgM and rabbit primary antibodies, both monoclonal and polyclonal. The **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** kits are optimized for use with the **Bio SB TintoFast primary antibodies** and the **Bio SB TintoDetector ImmunoDNA System**; however, they are universal kits and therefore should work equally well with antibodies from different vendors as long as they are properly optimized.

Precautions

- 1 For professional users only. Results should be interpreted by a qualified medical professional.
2. Ensure proper handling procedures are used with reagent. Minimize microbial contamination of reagents.
3. Always wear proper personal protective equipment such as laboratory coat, goggles and gloves when handling reagents.
4. Dispose of unused solution according to local and federal regulations.
5. Do not ingest reagent. If reagent is ingested, seek medical advice immediately.
6. Avoid contact with eyes. If contact occurs, flush with large quantities of water.
7. Follow safety precautions for the heating device used for epitope retrieval (Tinto Detector ImmunoDNA System (BSB 7000) and TintoRetriever Pressure Cooker (BSB 7008).
8. For additional safety information refer to Safety Data Sheet for this product.
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).

Presentation

The **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** contains Anti-Mouse/Rabbit Horseradish Peroxidase micropolymer link and label, a DAB Buffer Substrate, and a DAB Chromogen solution. All components are buffered with stabilizers and a non-azide anti-microbial agent.

| Catalog No. | Volume/Qty |
|--------------|-------------|
| BSB 0355-5 | 5 mL Each |
| BSB 0355-15 | 15 mL Each |
| BSB 0355-50 | 50 mL Each |
| BSB 0355-100 | 100 mL Each |

Storage

Store at 2-8°C

Stability

The **Mohs Mouse/Rabbit PolyDetector Plus DAB HRP Brown Detection System** is stable up to the expiration date listed on the product label. Do not use this product after the expiration date listed on the product label. Temperature fluctuations should be avoided. Store appropriately when not in use and avoid prolonged exposure to room temperature conditions.

Preparation of Working Solution

The Anti-Mouse/Rabbit Horseradish Peroxidase Link and Label are ready-to-use working solutions and require no further preparation. The DAB Chromogen is concentrated and needs to be diluted with the DAB buffer solution (1 drop chromogen/1 ml buffer). Prepare the working DAB Substrate Chromogen Solution up to 6 hours before use.

Specimen Preparation

Frozen sections and cell preparations: This product can be used for labeling acetone-fixed frozen sections and acetone-fixed cell preparations.

Paraffin sections: This product can be used on formalin-fixed paraffin-embedded (FFPE) tissue sections. Ensure tissue undergoes appropriate fixation for best results. Pre-treatment of tissues with heat-induced epitope retrieval (HIER) is recommended using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020 - BSB 0023), ImmunoDNA Retriever with EDTA (BSB 0030-BSB 0033). Additionally, TintoDeparaffinator Citrate or EDTA (BSB 0175 - BSB 0178) can be used to deparaffinize, retrieve and hydrate FFPE Tissues. Tissue should remain hydrated using Bio SB Immuno/DNA Washer solutions (BSB 0029 & BSB 0042).

Mohs IHC Procedure

Specimen Preparation of Mohs Frozen Tissues

1. Embed the specimen in OCT inside a cryostat.
2. Cut sections at 4-5 µm and mount on a positively charged glass slide such as the Bio SB Hydrophilic Plus Slides (BSB 7028).
3. Air dry the slide at room temperature for 2 minutes and then incubate the slide at 60 °C for 3 minutes in an incubator or dry bath.
4. Fix in 100% acetone and air dry only or in 10% NBF and rinse with distilled water and air dry for 2 minutes at room temperature.

Insert the TintoDetector Slide handle with the Cap Gap slides and FFPE tissues inside the TintoDetector incubator at 110° C and incubate for 30 minutes.

c. TintoRetriever PT Module or Water Bath Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA at 95°-99° C. Incubate for 20-30 minutes.

d. Conventional Steamer Method

Place tissues/slides in a pre-warmed staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA in a steamer, cover and steam for 20-30 minutes.

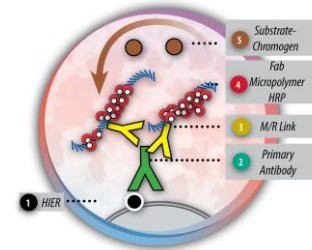
4. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15 minutes.

5. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.

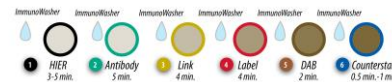
6. Wash slides with ImmunoDNA washer or DI water.

7. Continue IHC staining protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Immunohistochemical Protocol



| Step | Mohs PolyDetector Plus DAB 20 min Protocol |
|--------------------------|--|
| HIER | 3-5 min |
| Primary Antibody | 5 min |
| 1st Step Detection | 4 min |
| 2nd Step Detection | 4 min |
| Substrate-Chromogen | 2 min |
| Counterstain / Coverslip | 0.5 min – 1 min |



Mounting Protocols

Mount with aqueous mounting such as AquaMounter (BSB-0090- BSB 0093) or permanent mounting media such as XyGreen PermaMounter (BSB 0169-0174) or organic solvent-based resin such as PermaMounter (BSB 0094-0097).

Product Limitations

Due to inherent variability present in immunohistochemical procedures (including fixation time of tissues, dilution factor of antibody, retrieval method utilized and incubation time), optimal performance should be established through the use of positive and negative controls. Results should be interpreted by a qualified medical professional.

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.

Tissue Pretreatment Procedure for Mohs Frozen Tissues

1. Subject tissues to epitope retrieval using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023), ImmunoDNA Retriever with EDTA (BSB 0030-BSB 0033) or Mohs ImmunoDigester (BSB 0324- 0326) based on antibody pretreatment recommendations. It is recommended that a user choose either a 3-5 minutes heat-induced epitope retrieval (HIER) method using a TintoDetector ImmunoDNA System (BSB 7000) or a pressure cooker (BSB TintoRetriever Pressure Cooker) at 110 °C, or a 1-minute proteolytic induced epitope retrieval (PIER) method (for Cytokeratins using 10% NBF fixed tissues). Morphology is better with 10% NBF fixed tissues, especially when using proteolytic induced epitope retrieval method, however, antigen maybe easier to detect on the 100% acetone fixed tissue. 2. Two different HIER methods may be used:

a. TintoDetector ImmunoDNA System (BSB 7000).

Pair the TintoDetector Cap Gap Plus slides (BSB 7006) with Mohs Frozen Tissues face to face and dip slides into a retrieving solution. Make sure the retrieving solution covers the tissues. Insert the TintoDetector Slide handle with the slides inside the TintoDetector incubator at 110° C and incubate the slides for 3- 5 minutes.

b. TintoRetriever Pressure Cooker (BSB 7008). Please note that this process will require additional 15 minutes for the pressure cooker to warm up and additional 5- 10 minutes for cooling down the slides. Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA, and place on staining dish support in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high (110° C). Incubate for 5 minutes. Open and immediately transfer slides to room temperature. Cool off for 5 -10 min.

3. For PIER, apply ImmunoDNA Digester (BSB 0108-0112) at room temperature for 1 min then wash.

IHC Detection Procedure

1. After HIER or PIER, transfer slides to ImmunoDNA washer and let stand for 1-2 minutes.

2. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.

3. Wash slides with ImmunoDNA washer or DI water.

4. Continue Mohs IHC detection protocol. Wash slides between each step with ImmunoDNA washer solution.

Specimen Preparation for FFPE Tissues

1. Cut and mount 3-5-micron formalin-fixed paraffin-embedded tissues on positively charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028) or Cap Gap Plus slides (BSB 7006). 2. Air dry for 2 hours at 58° C.

Tissue Pretreatment Procedure for FFPE Tissues

1. Deparaffinize and rehydrate tissues.

2. Subject tissues to heat induced epitope retrieval (HIER) using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023) or EDTA (BSB 0030-BSB 0033).

3. Any of three heating methods may be used:

a. TintoRetriever Pressure Cooker or Equivalent

Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate or EDTA, and place on staining dish support in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high (110° C). Incubate for 10-15 minutes. Open and immediately transfer slides to room temperature. Cool off the slides for 5-10 min.

b. TintoDetector ImmunoDNA System (BSB 7000).

Symbol Key / Légende des symboles/Erläuterung der Symbole

| | | | |
|--|--|--|---|
| Storage Temperature Limites de température Zulässiger Temperaturbereich | Manufacturer Fabricant Hersteller | REF | Catalog Number Référence du catalogue Bestellnummer |
| IVD In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro In-Vitro-Diagnostikum | Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten | Expiration Date Utiliser jusque Verwendbar bis | LOT Lot Number Code du lot Chargenbezeichnung |