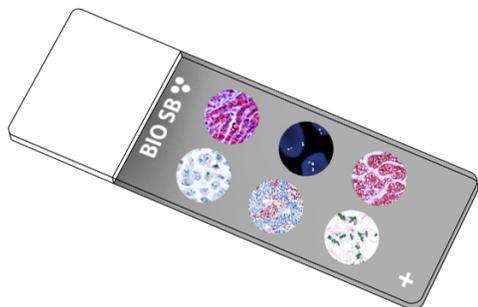


Tri-Methyl-Histone

H3(Lys27)/H3K27Me3  

Control Slides



Intended Use

For In Vitro Diagnostic Use.

Summary and Explanation

H3K27me3 is a downstream target of the polycomb repressive complex 2 (PRC2); PRC2 contains evolutionarily conserved proteins essential for regulating gene expression. Gain- or loss-of-function mutations in EZH2, a component of PRC2, have been found in a variety of cancer types such as Lymphoma, Melanoma, and Myelodysplastic Lymphoma; gain-of-function mutations can result in H3K27Me hyperactivity and chromatin dysregulation. KDMA6A and B demethylases can act on H3K27Me, and loss-of-function mutations in these proteins have been found in solid and non-solid tumors including types of Leukemia, Lymphoma, Melanoma, Renal and Bladder Cancers, Medulloblastoma and Prostate Cancer.

As a gene transcription repressor, H3K27me3 has diverse roles in embryogenesis and neoplasia. Loss of H3K27me3 occurs in a significant subset of Malignant Peripheral Nerve Sheath tumors. Other neoplasms may show loss of H3K27me3 expression, such as Meningioma, Radiation Associated Unclassified Sarcoma, Radiation-Associated Angiosarcoma, Dedifferentiated Chondrosarcoma, Melanoma and Merkel Cell Carcinoma, amongst others. Some tumors may exhibit heterogeneous H3K27me3 expression (mosaic pattern).

Presentation

Five slides of Tri-Methyl-Histone H3(Lys27)/H3K27Me3 positive tissues, each mounted on Hydrophilic Plus Slides, provided in a plastic mailer.

Catalog No.	Quantity
BSB-9444-CS	5 slides

Storage Store at 20-25°C

Precautions

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as a laboratory coat, goggles, and gloves when handling reagents.
4. Dispose of unused solution with copious amounts of water.
5. Follow safety precautions of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker or similar).
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" (see References in this document).

Stability

This product is stable up to the expiration date on the product label.

Do not use the after expiration date listed on the package label.

IHC Protocol

1. 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).

2. 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 the trivet in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 15 minutes. Open and immediately transfer slides to room temperature.

b. 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 30-60 minutes.

c. 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 30-60 minutes.

3. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
4. For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to the instrument manufacturer's instructions.
5. Wash slides with ImmunoDNA washer or DI water.
6. Continue IHC staining protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Immunohistochemical Protocol

Step	ImmunoDetector AP/HRP	PolyDetector AP/HRP	PolyDetector Plus HRP
Peroxidase/AP Blocker	5 min.	5 min.	5 min
Primary Antibody	30-60 min.	30-60 min.	30-60 min.
1st Step Detection	10 min.	30-45 min.	15 min.
2nd Step Detection	10 min.	Not Applicable	15 min.
Substrate- Chromogen	5-10 min.	5-10 min.	5-10 min.
Counterstain/Coverslip	Varies	Varies	Varies

Abbreviated IF Protocol

Step	Incubation Time
Rinse slides in IF wash buffer	5 minutes
Drain and wipe excess IF wash buffer off slide	
Conduct remaining steps in the dark	
Apply Antibody	30-60 minutes
Rinse with 3 changes of IF wash buffer	3x15 minutes each
Coverslip with IF mounting medium	

Mounting Protocols

For detailed instructions using biodegradable permanent mounting media such as XyGreen PermaMounter (BSB 0169-0174) or organic solvent based resin such as PermaMounter (BSB 0094-0097), refer to PI0174 or PI0097.

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. Laugesen A, et al. Role of the polycomb repressive complex 2 (PRC2) in transcriptional regulation and cancer. *Cold Spring Harb Perspect Med.* 2016;6.9: a026575
2. Das P, et al. Regulating methylation at H3K27: a trick or treat for cancer cell plasticity. *Cancers.* 2020;12.10: 2792.
3. Margueron R, et al. The Polycomb complex PRC2 and its mark in life. *Nature.* 2011;469.7330: 343-349.
4. Zhang M, et al. Somatic mutations of SUZ12 in malignant peripheral nerve sheath tumors. *Nat Gen.* 2014;46.11: 1170-1172.
5. Le Guellec S, et al. Loss of H3K27 trimethylation is not suitable for distinguishing malignant peripheral nerve sheath tumor from melanoma: a study of 387 cases including mimicking lesions. *Modern Pathol.* 2017;30.12: 1677-1687.

Symbol Key / Légende des symboles/Erläuterung der Symbole

EC REP	QAdvis EAR AB Ideon Science Park Scheelevägen 17 SE-223 70 Lund, Sweden	 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