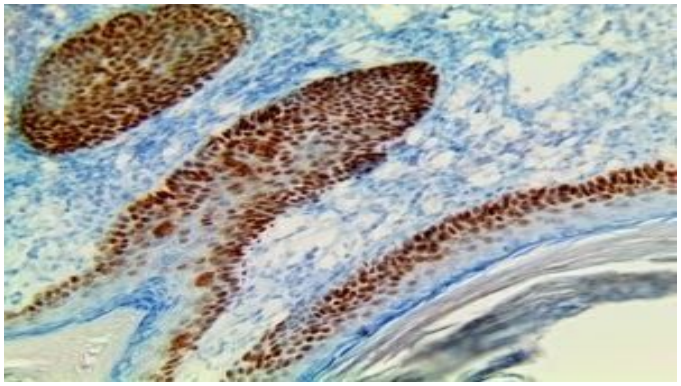
TintoFast p63

Clone: 4A4
Mouse Monoclonal

CE IVD



Inset: IHC of TintoFast p63 on a FFPE Basal Cell Carcinoma Tissue

Intended Use
For Mohs In Vitro Diagnostic Use.

This antibody is intended for the fast immunohistochemical detection of p63, a protein involved in apoptosis**,** during intraoperative Mohs surgery on frozen sections. Additionally, this antibody can also be used on FFPE specimens. Interpretation of results should be performed by a qualified medical professional.

Immunogen
Recombinant fragment corresponding to Human p63 aa 1-205.

Summary and Explanation
In addition to p53, mammalian cells contain two homologous genes, p63 and p73. These genes give rise to the expression of proteins that are highly similar to p53 in structure and function. In particular, p63 and p73 proteins can induce p53-responsive genes and elicit programmed cell death. p73 and p63 are important during development and differentiation. In particular, p63 appears to be primarily implicated in epithelial development.

Anti-p63 to human p63 protein labels an epitope common to all six p63 isotypes (TAp63 α , TAp63 β , TAp63 γ , ΔNp63 α , ΔNp63 β , ΔNp63 γ). p63 labels the nuclei of myoepithelial cells in the prostate gland as well as breast tissue, making it useful in differentiating benign vs. malignant prostate lesions and breast lesions.

Antibody Type	Mouse Monoclonal	**Clone**	4A4
Isotype	IgG2a/K	**Reactivity**	Paraffin, Frozen
Localization	Nuclear	**Control**	Prostate, Breast, Skin, Salivary Gland
Species Reactivity		Human, Mouse, Rat, Turtle	

Presentation
Anti-TintoFast p63 is a Mouse Monoclonal antibody derived from cell culture supernatant that is concentrated, dialyzed, filter sterilized and diluted in buffer pH 7.5, containing BSA and sodium azide as a preservative.

Catalog No.	*Antibody Type*	*Dilution*	*Volume/Qty*
BSB-3698-3	TintoFast Predilute	Ready-to-Use	3.0 mL
BSB-3698-7	TintoFast Predilute	Ready-to-Use	7.0 mL
BSB-3698-15	TintoFast Predilute	Ready-to-Use	15.0 mL

Control Slides Available

Catalog No.	*Quantity*
BSB-3698-CS	5 slides

Storage Store at 2-8°C (Control Slides: Store at 20-25°C)

Precautions
1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN_3) as a preservative. 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. 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 of the heating device used for epitope retrieval (TintoRetriever Pressure Cooker (BSB 7008), TintoDetector Incubator (BSB 7002) 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 after expiration date listed on the package label. Temperature fluctuations should be avoided. Store appropriately when not in use and avoid prolonged exposure to room temperature conditions.

Mohs IHC Protocol
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) or TintoDetector Cap Gap slides (BSB 7006).
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 or 10% NBF for 2 minutes at room temperature. Choice of fixation solution depends on antibody.
5. If tissue is fixed in 100% acetone, let the slide air dry. If the tissue is fixed in 10% NBF, rinse with distilled water and then dry dry the slides for 2 minutes at room temperature.

Tissue Pretreatment Procedure for Mohs Frozen Tissues
1. Subject tissues to HIER (heat-induced epitope retrieval) using Bio SB ImmunoDNA Retriever with Citrate (BSB 0020-BSB 0023). Optionally, Mohs Immuno Digestor (BSB 0108-0112) can be used for cytokeratin targets instead of HIER.

a. For Mohs PolyDetector HRP Green or DAB protocol use the TintoRetriever Pressure Cooker (BSB 7008) or Equivalent. Place tissues/slides in a staining dish or coplin jar containing the ImmunoDNA Retriever with Citrate , and place on trivet or staining dish support in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high (110-121° C). Incubate for 5 minutes. Open and immediately transfer slides to room temperature. Cool off for 5 -10 min

b. For Mohs PolyDetector Plus HRP Green or DAB protocol use the TintoDetector Incubator (BSB 7002). Preheat the TintoDetector Incubator to 110 °C. Place TintoDetector Cap Gap slides (BSB 7006) face to face and insert them into the TintoDetector Slide Holder (BSB 7003). Submerge slides in ImmunoDNA Retriever with Citrate to draw up enough solution by capillary action to cover the tissues. Heat the slides in a preheated TintoDetector Incubator for 3 minutes. Transfer slides to room temperature and cool off for 1 min.

c. For targets that are compatible with Mohs Immuno Digestor, Incubate with Mohs ImmunoDigestor at room temperature for 1 min and rinse the slides with ImmunoDNA washer (BSB 0029 & BSB 0042) after 1 min.

Mohs IHC Detection

1. After HIER, transfer slides to ImmunoDNA washer and let it 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 IHC detection protocol. Wash slides between each step with ImmunoDNA washer solution.

Abbreviated Mohs Immunohistochemical Protocol

Step	Mohs PolyDetector HRP Green or DAB 10 min* Protocol	Mohs PolyDetecto Plus HRP Green or DAB 20 min Protocol
HIER	5 min**	3 min
Primary Antibody	4 min.	5 min.
1st Step Detection	3 min.	4 mn.
2nd Step Detection	NA.	4 min.
Substrate-Chromogen	2 min.	1-2 min.
Counterstain / Coverslip	Varies	Varies

*instrument setup and HIER time not included
**1 min PIER for compatible targets

IHC Protocol 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.
2. Air dry for 2 hours at 58° C.

Tissue Pretreatment Procedure for FFPE Tissues

1. Deparaffinize, dehydrate, and rehydrate tissues.
2. Subject tissues to HIER using a suitable retrieval solution such as ImmunoDNA Retriever with Citrate or EDTA.
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 trivet in the pressure cooker. Add 1-2 inches of distilled water to the pressure cooker and turn heat to high. Incubate for 10- 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 20-30 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 20-30 minutes.

4. After HIER, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 10 minutes.
5. For manual IHC, perform antibody incubation at ambient temperature. For automated IHC methods, perform antibody incubation according to instrument manufacturer's instructions.
6. Wash slides with ImmunoDNA washer or DI water.
7. Continue IHC protocol. Wash slides between each step with ImmunoDNA washer solution.

Mounting Protocols

Mount with aqueous media 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. Yang A, et al. Mol Cell. 1998;2:305-16
2. Signoretti S, et al. Am J Pathol. 2000;157:1769-75 3.
3. Yang A, et al. Nature. 1999;398:714-18
4. Barbareschi M, et al. Am J Surg Pathol. 2001;Aug;25(8);1054-60
5. Werling RW, et al. Am J Surg Pathol. 2003;Jan;27(1):82-90
6. Rajal B Shah, et al. Am J Surg Pathol. 2002;26(9):1161-1168
7. 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.

Symbol Key / Légende des symboles/Erläuterung der Symbole

EC REP EMERGO EUROPE Prinsessegracht 20 2514 AP The Hague The Netherlands	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

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