

Cytokeratin 5 & 6

Clone: D5/16B4

Mouse Monoclonal


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Inset: IHC of Cytokeratin 5 & 6 on a FFPE Prostate Tissue

Intended Use

For In Vitro Diagnostic Use.

This antibody is intended for use in Immunohistochemical applications on formalin-fixed paraffin-embedded tissues (FFPE), frozen tissue sections and cell preparations. Interpretation of results should be performed by a qualified medical professional.

Immunogen

Purified human cytokeratin 5 & 6.

Summary and Explanation

Cytokeratin 5 (58 kDa) is a high-molecular weight, basic type of cytokeratin expressed in basal, intermediate and superficial-cell layers of stratified epithelia as well as transitional epithelia, complex epithelia, mesothelial cells and Mesothelioma. Cytokeratin 6 (56 kDa) is also a high-molecular weight, basic type cytokeratin expressed by proliferating squamous epithelium often paired with Cytokeratin 16.

CK 5 and 6 are positively seen in nearly 100% of Malignant Mesotheliomas and is rarely seen in Lung Adenocarcinomas. CK 5 and 6 can positively be seen in undifferentiated Large-cell Carcinoma as well as Squamous Carcinoma. Fewer than 10% of Carcinomas of the breast, colon, and prostate stain positively for this marker. CK 5 and 6 have also been used successfully as a myoepithelial cell marker in the prostate to determine.

Antibody Type	Mouse Monoclonal	Clone	D5/16B4
Isotype	IgG1	Reactivity	Paraffin, Frozen
Localization	Cytoplasmic	Control	Mesothelioma, Prostate
Species Reactivity		Human	

Precautions

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN₃) as a preservative. Ensure proper handling procedures are used with this reagent.
3. Always wear personal protective equipment such as laboratory coat, goggles and gloves when handling reagents.
4. Dispose of unused solution with copious amount 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 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).

Presentation

Anti-CK 5 & 6 is a cocktail of mouse monoclonal antibodies 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.

<i>Catalog No.</i>	<i>Antibody Type</i>	<i>Dilution</i>	<i>Volume/Qty</i>
BSB 5400	Tinto Prediluted	Ready-to-Use	3.0 mL
BSB 5401	Tinto Prediluted	Ready-to-Use	7.0 mL
BSB 5402	Tinto Prediluted	Ready-to-Use	15.0 mL
BSB 5403	Concentrated	1:25 - 1:100	0.1 mL
BSB 5404	Concentrated	1:25 - 1:100	0.5 mL
BSB 5405	Concentrated	1:25 - 1:100	1.0 mL

Control Slides Available

<i>Catalog No.</i>	<i>Quantity</i>
BSB 5406	5 slides

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

Stability

This product is stable up to the expiration date on the product label. Do not use after expiration date listed on package label. Temperature fluctuations should be avoided. Store appropriately when not in use, and avoid prolonged exposure to room temperature conditions.

Specimen Preparation

Paraffin sections: The antibody 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) or ImmunoDNA Digestor (BSB 0108-0112). See reverse side for complete protocol. Tissue should remain hydrated via use of Bio SB Immuno/DNA Washer solutions (BSB 0029 & BSB 0042).

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

Staining Procedure

- Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positively charged slides such as Bio SB Hydrophilic Plus Slides (BSB 7028).
- Air dry for 2 hours at 58° C.
- Deparaffinize, dehydrate and rehydrate tissues.
- 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).
- Any of three heating methods may be used:
 - 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 15 minutes. Open and immediately transfer slides to room temperature.
 - 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.
 - 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.
- After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.
- For manual staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
- Wash slides with ImmunoDNA washer or DI water.
- 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

Product Limitations

Normal Tissues	
Positive (+)	
stratified squamous epithelia	basal cells in complex epithelia
Negative (-)	
Simple epithelia	non-epithelial cells
Abnormal Tissues	
Positive (+)	
epithelioid pleural mesotheliomas (formalin-fixed, paraffin-embedded) 56/61 (92%)	
metastatic adenocarcinomas (formalin-fixed, paraffin-embedded) 9/63 (14%)	
Reactive mesothelium	
mesotheliomas with epithelioid differentiation (strongly labelled)	
ductal hyperplasia (High levels)	
Secondary adenocarcinomas of the pleura (1 focally positive)	
Negative (-)	
Mesotheliomas of desmoplastic type	Mesotheliomas of sarcomatoid type
atypical ductal hyperplasia in situ	ductal carcinoma in situ
sarcomatoid areas of tumours with a mixed morphology (weak or absent)	
secondary adenocarcinomas of the pleura 0/0	

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

- Ordonez NG, Am J Surg Pathol. 1998;22(10):1215-1221
- Ordonez NG, Am J Surg Pathol. 22(10):1203-1214
- Cury PM, Butcher DW, et al. Mod Pathol. 2000;13(2):107-12
- 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

	EMERGO EUROPE Prinsessegracht 20 2514 AP The Hague The Netherlands		Storage Temperature Limites de température Zulässiger Temperaturbereich		Manufacturer Fabricant Hersteller		Catalog Number Référence du catalogue Bestellnummer
	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 Number Code du lot Chargenbezeichnung



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