Doc #: PI2375 Version #: 5

BIOSCIENCE FOR THE WORLD

Clone: BSB-131 Mouse Monoclonal





Inset: IHC of SDHB on a FFPE Hepatocellular Carcinoma 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

A Recombinant full length of the human succinate dehydrogenase iron-sulfur protein.

Summary and Explanation

Succinate dehydrogenase [ubiquinone] iron-sulfur subunit, mitochondrial (SDHB) also known as iron-sulfur subunit of complex II (Ip) is a protein that in humans is encoded by the SDHB gene. The gene that codes for the SDHB protein is nuclear, not mitochondrial DNA. However, the expressed protein is located in the inner membrane of the mitochondria. Four subunits comprise the SDH protein complex: a flavochrome subunit (SDHA), an iron-sulfur protein (SDHB) and two membrane-bound subunits (SDHC and SDHD) anchored to the inner mitochondrial membrane. Mutation in this protein is associated with wide range of diseases such as Renal Cell Carcinoma, Paraganglioma, Gastrointestinal Stromal Tumors (GISTs), Pituitary Adenoma, and many others.

Mutations in the tumor suppressor genes SDHB, SDHC, and SDHD (or collectively SDHx) cause the inherited paraganglioma syndromes, characterized by pheochromocytomas and paragangliomas. The IHC for SDHB is negative in all SDH mutated paragangliomas regardless of whether the B, C or D subunit is involved. However, other tumors have been associated with SDHx mutations, such as Gastrointestinal Stromal Tumors, specifically in the context of Carney-Stratakis syndrome. It has been shown that SDHB immunohistochemistry is a reliable technique for the identification of pheochromocytomas and paragangliomas caused by SDHx mutations. It's been shown that Carney-Stratakis syndrome- and Carney-triad-associated GISTs are negative by immunohistochemistry for SDHB in contrast to KIT- or PDGFRA-mutated GISTs and a majority of sporadic GISTs, and it has been suggested that GISTs of epithelioid cell morphology are tested for SDHB immunohistochemically.

Antibody Type	Mouse Monoclonal	Clone	BSB-131	
lsotype	lgG1/K	Reactivity	Paraffin, Frozen	
Localization	Cytoplasmic, Membranous	Species Reactivity	Human, Mouse, Rat	
Control	Breast, Adrenal, Prostate, Kidney, Spleen, Tonsil, Breast Carcinoma, Hepatocellular Carcinoma, Lung Adenocarcinoma, Prostate Carcinoma, Papillary Thyroid Carcinoma			
Application	Neural & Neuroendocrine Cancer, Pituitary, Kidney & Urothelial Cancer			

Presentation

Anti-SDHB 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.	Presentation	Dilution	Volume	
BSB-2375-3	Predilute	Ready-to-Use	3.0 mL	
BSB-2375-7	Predilute	Ready-to-Use	7.0 mL	
BSB-2375-15	Predilute	Ready-to-Use	15.0 mL	
BSB-2375-01	Concentrate	1:50-1:200	0.1 mL	
BSB-2375-05	Concentrate	1:50-1:200	0.5 mL	
BSB-2375-1	Concentrate	1:50-1:200	1.0 mL	

Control Slides Available

Catalog No.	Quantity
BSB-9376-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 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 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 on acetone-fixed frozen sections and acetone-fixed cell preparations.

IHC Protocol

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). 2. Air dry for 2 hours at 58° C.

3. Deparaffinize, dehydrate, and rehydrate tissues.

4. 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).

5. 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 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.

6. After heat treatment, transfer slides in ImmunoDNA Retriever with Citrate or EDTA to room temperature and let stand for 15-20 minutes.

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

8. Wash slides with ImmunoDNA washer or DI water.

9. Continue IHC 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	

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. "Entrez Gene: succinate dehydrogenase complex":

https://www.ncbi.nlm.nih.gov/gene?Db=gene&Cmd=ShowDetailView&TermToSe arch=6390

 Au HC, et al. Structural organization of the gene encoding the human iron-sulfur subunit of succinate dehydrogenase". Gene. 1995; 159 (2): 249–53.
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Brouwers FM, et al. High frequency of SDHB germline mutations in patients with malignant catecholamine-producing paragangliomas: implications for genetic testing. J. Clin. Endocrinol. Metab. 2006; 91 (11): 4505–9.
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Gaal J et al, SDHB immunohistochemistry: a useful tool in the diagnosis of Carney-Stratakis and Carney triad gastrointestinal stromal tumors. Mod Pathol. 2011 Jan;24(1):147-51.

8. 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. https://www.cdc.gov/mmwr/pdf/other/su6101.pdf

Symbol Key/Légende des symboles/Erläuterung der Symbole

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IVD	In Vitro Diagnostic Medical Device Dispositif médical de diagnostic in vitro In-Vitro-Diagnostikum	[]i	Read Instructions for Use Consulter les instructions d'utilisation Gebrauchsanweisung beachten	\square	Expiration Date Utiliser jusque Verwendbar bis	LOT	Lot Number Code du lot Chargenbezeichnung
Bio SB ??							



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