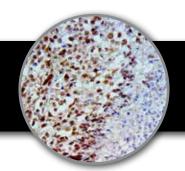
BAP1

Clone: BSB-109 Mouse Monoclonal C€ IVD





Inset: IHC of BAP1 on a FFPE Mesothelioma 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**

Synthetic peptide corresponding to the C-terminus of the human BAP1 protein.

## **Summary and Explanation**

BAP1 or BRCA1 associated protein-1 (ubiquitin carboxy-terminal hydrolase) is a deubiquitinating enzyme that in humans is encoded by the BAP1 gene. Recent studies have shown that BAP1 and its fruit fly homolog, Calypso, are members of the polycomb-group proteins (PcG) of highly conserved transcriptional repressors required for long-term silencing of genes that regulate cell fate determination, stem cell pluripotency, and other developmental processes.

In cancer, BAP1 can function both as a Tumor suppressor and as a metastasis suppressor. Exome sequencing identified inactivating mutations in BAP1 in 47% of Uveal melanomas, and BAP1 mutation have been found to be strongly associated with metastasis. The atypical melanocytic lesions resemble Spitz nevi and have been characterized as "atypical Spitz tumors" (ASTs), although they have a unique histology and exhibit both BRAF and BAP1 mutations.

BAP1 mutations have been identified in aggressive Mesotheliomas with similar mutations as seen in melanomas. Sequencing studies have been used to identify germline mutations in BAP1 in families with genetic predispositions to mesothelioma and melanocytic skin tumors. Mutations in the tumor suppressor gene BAP1 occur in approximately 15% of clear cell renal cell carcinoma cases. Sequencing efforts demonstrated worse outcomes in patients with BAP1 mutated clear cell renal cell carcinoma. Immunohistochemistry for BAP1 is a prognostic biomarker to predict poor oncologic outcomes and adverse clinicopathological features in patients with non-metastatic clear cell renal cell carcinoma. BAP1 assessment using immunohistochemistry on needle biopsy may benefit preoperative risk stratification and guide treatment planning

Antibody Type	Mouse Monoclonal	Clone	BSB-109 Paraffin, Frozen			
Isotype	lgG1	Reactivity				
Localization	Nuclear	Control	Testis, TCC, Mesothelioma			
Species Reactiv	ity	Human, Mouse, Rat				

# **Presentation**

BAP1 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.

### **Presentations**

Catalog Num.	Antibody Type	Dilution	Volume/Qty		
BSB 3300	Tinto Prediluted	Ready-to-Use	3.0 mL		
BSB 3301	Tinto Prediluted	Ready-to-Use	7.0 mL		
BSB 3302	Tinto Prediluted	Ready-to-Use	15.0 mL		
BSB 3303	Concentrated	1:50 - 1:200	0.1 mL		
BSB 3304	Concentrated	1:50 - 1:200	0.5 mL		
BSB 3305	Concentrated	1:50 - 1:200	1.0 mL		
BSB 3306	Control Slides	Not Applicable	5 slides		

## **Precautions**

- 1. For professional users only. Ensure results are interpreted by a medical professional.
- 2. This product contains sodium azide (NaN3), a toxic chemical which may react with plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent sodium azide build-up.
- 3. Ensure proper handling procedures are used with reagent. Always wear proper laboratory equipment such as laboratory coat and gloves when handling reagents.
- **4.** Unused solution should be disposed of according to local and federal regulations.
- 5. Do not ingest reagent. If reagent ingested, contact a poison control center immediately.
- 6. 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" (10).

## Storage

Store at 2-8 °C. 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 paraffinembedded (FFPE) tissue sections. Ensure tissue undergoes appropriate fixation to ensure 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**

- 1. Cut and mount 3-5 micron formalin-fixed paraffin-embedded tissues on positive 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 epitope retrieval 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 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 staining, perform antibody incubation at ambient temperature. For automated staining methods, perform antibody incubation according to instrument manufacturer's instructions.
- 8. Wash slides with IHC wash buffer or DI water.
- 9. Continue IHC staining protocol.

#### **Recommended IHC Protocol**

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

#### **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 medical professional.

## References

- Entrez Gene: BAP1 BRCA1 associated protein 1 [ Homo sapiens (human) ] https://www.ncbi.nlm.nih.gov/gene?Db=gene&Cmd=Show-DetailView&TermToSearch=8314
- 2. Jensen DE, et al. BAP1: a novel ubiquitin hydrolase which binds to the BRCA1 RING finger and enhances BRCA1-mediated cell growth suppression". Oncogene. 1998; 16 (9): 1097–112.
- 3. Gaytán AA, et al. A genetic screen identifies novel polycomb group genes in Drosophila. Genetics. 2007; 176(4): 2099–108.
- 4. Ventii KH, et al. BRCA1-associated protein-1 is a tumor suppressor that requires deubiquitinating activity and nuclear localization. Cancer Research. 2008; 68(17): 6953–62.
- 5. Harbour JW,et al. Frequent mutation of BAP1 in metastasizing uveal melanomas . Science. 2010; 330(6009): 1410–3.
- 6. Bott M, et al. The nuclear deubiquitinase BAP1 is commonly inactivated by somatic mutations and 3p21.1 losses in malignant pleural mesothelioma". Nature Genetics.2011; 43 (7): 668–72.
- 7. Peña-Llopis S, et al. BAP1 loss defines a new class of renal cell carcinoma". Nature Genetics. 2012; 44 (7): 751–9.
- 8. Testa JR, et al. Germline BAP1 mutations predispose to malignant mesothelioma". Nature Genetics. 2011; 43 (10): 1022–5.
- 9. Kapur P, eta al. BAP1 immunohistochemistry predicts outcomes in a multi-institutional cohort with clear cell renal cell carcinoma". The Journal of Urology. 2014; 191 (3): 603—10.
- 10. 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	V-8.C	Storage Temperature Limites de température Zulässiger Temperaturbereich	3	Manufacturer Fabricant Hersteller	REF	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	$\sum$	Expiration Date Utiliser jusque Verwendbar bis	LOT	Lot Number Code du lot Chargenbezeichnung



