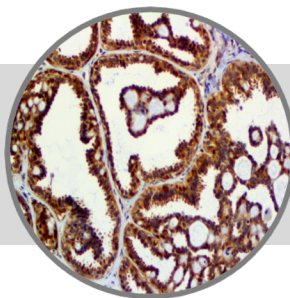


**PTEN, RMAb**

Clone: RBT-PTEN

Rabbit Monoclonal


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Inset: IHC of PTEN on a FFPE Breast 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**

Synthetic peptide corresponding to the N-terminus of the human PTEN protein.

**Summary and Explanation**

Phosphatase and tensin homolog (PTEN) is a protein that, in humans, is encoded by the PTEN gene. Unlike most of the protein tyrosine phosphatases, this protein preferentially dephosphorylates phosphoinositide substrates. It negatively regulates intracellular levels of phosphatidylinositol-3,4,5-trisphosphate in cells and functions as a tumor suppressor by negatively regulating Akt/PKB signaling pathway.

PTEN is one of the most commonly lost tumor suppressors in human cancer; in fact, up to 70% of men with prostate cancer are estimated to have lost a copy of the PTEN gene at the time of diagnosis. During tumor development, mutations and deletions of PTEN occur that inactivate its enzymatic activity leading to increased cell proliferation and reduced cell death. Frequent genetic inactivation of PTEN occurs in glioblastoma, endometrial cancer, and prostate cancer; and reduced expression is found in many other tumor types such as lung and breast cancer. In breast and prostate cancer, loss of PTEN expression has been shown to correlate positively with advanced stage. Furthermore, PTEN mutation also causes a variety of inherited predispositions to cancer.

|                           |                      |                   |  |
|---------------------------|----------------------|-------------------|--|
| <b>Antibody Type</b>      | Rabbit Monoclonal    | <b>Clone</b>      | RBT-PTEN   |
| <b>Isotype</b>            | IgG                  | <b>Reactivity</b> | Paraffin, Frozen   |
| <b>Localization</b>       | Nuclear, Cytoplasmic | <b>Control</b>    | Colon, Thymus, Skin, Kidney, Breast, Prostate, Breast Carcinoma, Prostatic Carcinoma |
| <b>Species Reactivity</b> |                      | Human             |  |

**Presentation**

PTEN is a rabbit 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.

| <b>Catalog No.</b> | <b>Antibody Type</b> | <b>Dilution</b> | <b>Volume/Qty</b> |
|--------------------|----------------------|-----------------|-------------------|
| BSB 2643           | Tinto Prediluted     | Ready-to-Use    | 3.0 mL            |
| BSB 2644           | Tinto Prediluted     | Ready-to-Use    | 7.0 mL            |
| BSB 2645           | Tinto Prediluted     | Ready-to-Use    | 15.0 mL           |
| BSB 2646           | Concentrated         | 1:50 - 1:200    | 0.1 mL            |
| BSB 2647           | Concentrated         | 1:50 - 1:200    | 0.5 mL            |
| BSB 2648           | Concentrated         | 1:50 - 1:200    | 1.0 mL            |

**Control Slides Available**

| <b>Catalog No.</b> | <b>Quantity</b> |
|--------------------|-----------------|
| BSB 2648-1         | 5 slides        |

**Precautions**

1. For professional users only. Results should be interpreted by a qualified medical professional.
2. This product contains <0.1% sodium azide (NaN<sub>3</sub>) 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).

**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                |

## 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.

## Performance Characteristics

| Normal Tissues       |             |
|----------------------|-------------|
| Positive (+)         |             |
| Prostate             | 5/5 (100%)  |
| Kidney               | 3/3 (100%)  |
| Abnormal Tissues     |             |
| Positive (+)         |             |
| Breast Carcinoma     | 28/33 (84%) |
| Colorectal Carcinoma | 82/90 (91%) |
| Thyroid Carcinoma    | 83/91 (91%) |
| Endometrial Cancer   | 50/72 (69%) |









## 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

- Steck P, et al. "Identification of a candidate tumour suppressor gene, MMAC1, at chromosome 10q23.3 that is mutated in multiple advanced cancers". *Nature Genetics*. 1997;15 (4): 356-62.
- Chu E, Tarnawski A. "PTEN regulatory functions in tumor suppression and cell biology". *Medical Science Monitor*, 2004; 10 (10): RA235-41.
- Chen Z, et al. "Crucial role of p53-dependent cellular senescence in suppression of Pten-deficient tumorigenesis". *Nature*, 2005; 436 (7051): 725-30.
- 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<br>Prinsessegracht 20<br>2514 AP The Hague<br>The Netherlands                              |  | Storage Temperature<br>Limites de température<br>Zulässiger Temperaturbereich                        |  | Manufacturer<br>Fabricant<br>Hersteller              |  | Catalog Number<br>Référence du catalogue<br>Bestellnummer |
|  | In Vitro Diagnostic Medical Device<br>Dispositif médical de diagnostic in vitro<br>In-Vitro-Diagnostikum |  | Read Instructions for Use<br>Consulter les instructions d'utilisation<br>Gebrauchsanweisung beachten |  | Expiration Date<br>Utiliser jusque<br>Verwendbar bis |  | Lot Number<br>Code du lot<br>Chargenbezeichnung           |

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