Product Datasheet

PTEN Antibody (Y184) NB110-57441

Unit Size: 0.1 ml

Store at -20C. Avoid freeze-thaw cycles.

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NB110-57441

PTEN Antibody (Y184)

Product Information	
Unit Size	0.1 ml
Concentration	Please see the vial label for concentration. If unlisted please contact technical services.
Storage	Store at -20C. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	Y184
Preservative	0.01% Sodium Azide
Isotype	IgG
Purity	Protein A or G purified
Buffer	49% PBS, 0.05% BSA and 50% Glycerol
Product Description	
Host	Rabbit
Gene ID	5728
Gene Symbol	PTEN
Species	Human
Reactivity Notes	Human. Mouse and Rat cross reactivity tested by western blot.
Immunogen	A synthetic peptide corresponding to residues in the C-term of human PTEN was used as immunogen.
Notes	Produced using Abcam's RabMab® technology. RabMab® technology is covered by the following U.S. Patents, No. 5,675,063 and/or 7,429,487.
Product Application Details	
Applications	Western Blot, Flow Cytometry, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 1:1000-10000, Flow Cytometry 1:20, Immunohistochemistry 1:10- 1:500, Immunocytochemistry/Immunofluorescence 1:50-1:100, Immunoprecipitation 1:50, Immunohistochemistry-Paraffin
Application Notes	This product is useful for: Western Blot, Immunocytochemistry, Immunoprecipitation, Flow Cytometry. In Western blot this antibody detects a band at approximately 54kDa. Use in IHC on paraffin section was reported in scientific literature (PMID: 23746542).



Images

Western Blot: PTEN Antibody (Y184) [NB110-57441] MCF7 cell lysate.



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Immunohistochemistry: PTEN Antibody (Y184) [NB110-57441] -Immunohistochemical analysis of paraffin-embedded human thyroid gland carcinoma using anti-PTEN (C-term) (catalog #110-57441).



Publications

Carvalho Katia C, Maia Beatriz M, Omae Samantha V et al. Best practice for PTEN gene and protein assessment in anatomic pathology. Acta Histochem. 2014 Jan 01 [PMID: 23746542]

Eritja N, Santacana M, Maiques O et al. Modeling glands with PTEn deficient cells and microscopic methods for assessing PTEn loss: Endometrial cancer as a model. Methods. 2014 Nov 18 [PMID: 25461816]

Liu L, Jiang Y, Zhang H et al. miR-22 functions as a micro-oncogene in transformed human bronchial epithelial cells induced by anti-benzo[a]pyrene-7,8-diol-9,10-epoxide. Toxicol In Vitro 2010 Jun [PMID: 20170724]

Liu L, Jiang Y, Zhang H et al. Overexpressed miR-494 down-regulates PTEN gene expression in cells transformed by anti-benzo(a)pyrene-trans-7,8-dihydrodiol-9,10-epoxide. Life Sci 2010 Jan [PMID: 20006626] (WB)

Sangale Z, Prass C, Carlson A et al. A robust immunohistochemical assay for detecting PTEN expression in human tumors Appl Immunohistochem Mol Morphol 2011 Mar [PMID: 20930614] (IHC, Human)



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Immunohistochemistry Protocol for PTEN Antibody (NB110-57441)

Immunohistochemistry Protocol for Paraffin-embedded Tissues

- 1. Solutions and reagents
- 1.1. Xylene
- 1.2. Ethanol, anhydrous denatured, histological grade (100%, 95%, 70%)
- 1.3. Washing buffer:
- TBST washing buffer: 1XTBS/0.1% Tween-20

To prepare stock solution of 10X TBS: add 24.2 g Trizma base and 80 g sodium chloride to 1L of dH2O. Adjust pH to 7.6.

Working solution. 1XTBST/0.1% Tween-20: add 100ml 10XTBS to 900 ml dH2O. Add 1 ml Tween-20 and mix well. 1.4. Distilled water (dH2O)

1.5. Antigen Retrieval Solution:

0.01M Sodium Citrate Buffer, pH 6.0

To prepare stock solutions:

Solution A. 0.1 M citric acid solution: dissolve 21.0 g of citric acid, monohydrate (C6H8O7.H2O) in 1 liter of dH2O. Solution B. 0.1M sodium citrate solution: dissolve 29.4 g trisodium citrate dihydrate (C6H5Na3O7.2H2O) in 1 liter of dH2O.

Working solution: Add 9 ml of Stock solution A and 41 ml of stock solution B to 450 ml of dH2O. Adjust pH to 6.0. 1.6. 3% Hydrogene Peroxide

1.7. Blocking buffer:

PBS (Dulbeccos Phosphate Buffered Salts, 1X, catalog #21-031-CV from Mediatech, Inc.) + 10% serum (serum origin depends on the host of the secondary antibody)

1.8. Hematoxylin QS (catalog #H-3404 from Vector Laboratories, Inc.)

1.9. Permanent Mounting medium (VectaMount, catalog# H-5000 Vector Laboratories, Inc.)

2. Protocol

2.1. Deparaffinization/Rehydration

2.1.1. Heat slides in an oven at 65C for 1 hour.

2.1.2. De-paraffinize/hydrate using the following series of washes: two Xylene washes (5 min each), followed by two 100% ethanol rinses (5 min each), followed by 95% ethanol, 70% ethanol, 50% ethanol, 30% ethanol, followed by H2O and a TBST wash for 5 min on a shaker.

2.2. Antigen Retrieval

2.2.1. Immerse slides into staining dish containing Antigen Retrieval Solution.

2.2.2. Place covered staining dish into the rice cooker. Add 120 ml of dH2O.

2.2.3. When cook is turned to warm (about 20 to 30 min), unplug the cooker and remove the staining dish to the bench top.

2.2.4. Allow to cool down, without cover, for 20 min.

2.3. Staining

- 2.3.1. Wash slides with TBST for 5 min on a shaker.
- 2.3.2. Inactivate endogenous peroxidase by covering tissue with 3% hydrogen peroxide for 10 min.
- 2.3.3. Wash slides three times with TBST (3 min each on a shaker).
- 2.3.4. Block slides with the blocking solution for 1 hour.
- 2.3.5. Dilute primary antibody in the blocking buffer per recommendation on the data sheet.

2.3.6. Apply primary antibody to each section and incubate overnight in the humidified chamber (4C).

2.3.7. Wash slides three times with TBST (3 min each on a shaker).

2.3.8. Apply to each section secondary HRP-conjugated anti-rabbit antibody diluted in the blocking solution per manufacturers recommendation; incubate for 1 hour at room temperature.

2.3.9. Wash slides three times with TBST (3 min each on a shaker).

2.3.10. Add freshly prepared DAB substrate to the sections.

2.3.11. Incubate tissue sections with the substrate at room temperature until suitable staining develops (generally 2 to 5 min).

2.3.12. Rinse sections with water.

2.3.13. Counterstain with Hematoxylin.

2.3.14. Rinse sections with water.

2.3.15. Dehydrate samples using two rinses with 100% Ethanol (20 dips per rinse) followed by two rinses with Xylene (30 dips per rinse).

2.3.16. Mount coverslips on slides using Permount medium.





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Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

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