### Overview

<table>
<thead>
<tr>
<th><strong>Product name</strong></th>
<th>Anti-PTEN antibody [Y184]</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Description</strong></td>
<td>Rabbit monoclonal [Y184] to PTEN</td>
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<tr>
<td><strong>Host species</strong></td>
<td>Rabbit</td>
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<tr>
<td><strong>Specificity</strong></td>
<td>A 42kDa band is seen for some samples in addition to 50-54kDa band. We do not know the specificity of this band. For example, rat kidney, heart, spleen have bands around 50kDa but rat PC-12 cells have single band at ~42kDa.</td>
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**Tested applications**

- Suitable for: Flow Cyt, ICC/IF, WB

**Species reactivity**

- Reacts with: Mouse, Rat, Human

**Immunogen**

- Synthetic peptide within Human PTEN aa 350 to the C-terminus (C terminal). The exact sequence is proprietary.
  - (Peptide available as ab157804)

**Positive control**

- WB: HAP1, MCF7 and HEK-293 cell lysate; Human brain lysate; Mouse primary bone marrow derived macrophage whole cell lysate.

**General notes**

- Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

**We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.

This product is a recombinant rabbit monoclonal antibody.

### Properties

<table>
<thead>
<tr>
<th><strong>Form</strong></th>
<th>Liquid</th>
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<tbody>
<tr>
<td><strong>Storage instructions</strong></td>
<td>Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.</td>
</tr>
</tbody>
</table>
Storage buffer

- pH: 7.20
- Preservative: 0.01% Sodium azide
- Constituents: 49% PBS, 50% Glycerol, 0.05% BSA

Purity

- Protein A purified

Clonality

- Monoclonal

Clone number

- Y184

Isotype

- IgG

Applications

Our Abpromise guarantee covers the use of ab32199 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
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| Flow Cyt    | Use at an assay dependent concentration. PubMed: 20008304  
ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |
| ICC/IF      | ⭐⭐⭐⭐⭐ | Use at an assay dependent concentration. |
| WB          | 1/10000. Detects a band of approximately 54 kDa (predicted molecular weight: 47 kDa). Can be blocked with PTEN peptide (ab157804).  
For unpurified, use 1/500. |

Target

Function

- Tumor suppressor. Acts as a dual-specificity protein phosphatase, dephosphorylating tyrosine-, serine- and threonine-phosphorylated proteins. Also acts as a lipid phosphatase, removing the phosphate in the D3 position of the inositol ring from phosphatidylinositol 3,4,5-trisphosphate, phosphatidylinositol 3,4-diphosphate, phosphatidylinositol 3-phosphate and inositol 1,3,4,5-tetrakisphosphate with order of substrate preference in vitro PtIns(3,4,5)P3 > PtIns(3,4)P2 > PtIns3P > Ins(1,3,4,5)P4. The lipid phosphatase activity is critical for its tumor suppressor function. Antagonizes the PI3K-AKT/PKB signaling pathway by dephosphorylating phosphoinositides and thereby modulating cell cycle progression and cell survival. The unphosphorylated form cooperates with AIP1 to suppress AKT1 activation. Dephosphorylates tyrosine-phosphorylated focal adhesion kinase and inhibits cell migration and integrin-mediated cell spreading and focal adhesion formation. Plays a role as a key modulator of the AKT-mTOR signaling pathway controlling the tempo of the process of newborn neurons integration during adult neurogenesis, including correct neuron positioning, dendritic development and synapse formation. May be a negative regulator of insulin signaling and glucose metabolism in adipose tissue. The nuclear monoubiquitinated form possesses greater apoptotic potential, whereas the cytoplasmic nonubiquitinated form induces less tumor suppressive ability. In motile cells, suppresses the formation of lateral pseudopods and thereby promotes cell polarization and directed movement.  
Isoform alpha: Functional kinase, like isoform 1 it antagonizes the PI3K-AKT/PKB signaling pathway. Plays a role in mitochondrial energetic metabolism by promoting COX activity and ATP production, via collaboration with isoform 1 in increasing protein levels of PINK1. |

Tissue specificity

- Expressed at a relatively high level in all adult tissues, including heart, brain, placenta, lung, liver,
Involvement in disease
- Cowden syndrome 1
- Lhermitte-Duclos disease
- Bannayan-Riley-Ruvalcaba syndrome
- Squamous cell carcinoma of the head and neck
- Endometrial cancer

PTEN mutations are found in a subset of patients with Proteus syndrome, a genetically heterogeneous condition. The molecular diagnosis of PTEN mutation positive cases classifies Proteus syndrome patients as part of the PTEN hamartoma syndrome spectrum. As such, patients surviving the early years of Proteus syndrome are likely at a greater risk of developing malignancies.

Glioma 2
- VACTERL association with hydrocephalus
- Prostate cancer
- Macrocephaly/autism syndrome

A microdeletion of chromosome 10q23 involving BMPR1A and PTEN is a cause of chromosome 10q23 deletion syndrome, which shows overlapping features of the following three disorders: Bannayan-Zonana syndrome, Cowden disease and juvenile polyposis syndrome.

Sequence similarities
- Contains 1 C2 tensin-type domain.
- Contains 1 phosphatase tensin-type domain.

Domain
- The C2 domain binds phospholipid membranes in vitro in a Ca(2+)-independent manner; this binding is important for its tumor suppressor function.

Post-translational modifications
- Constitutively phosphorylated by CK2 under normal conditions. Phosphorylated in vitro by MAST1, MAST2, MAST3 and STK11. Phosphorylation results in an inhibited activity towards PIP3. Phosphorylation can both inhibit or promote PDZ-binding. Phosphorylation at Tyr-336 by FRK/PTK5 protects this protein from ubiquitin-mediated degradation probably by inhibiting its binding to NEDD4. Phosphorylation by ROCK1 is essential for its stability and activity. Phosphorylation by PLK3 promotes its stability and prevents its degradation by the proteasome. Monoubiquitinated; monoubiquitination is increased in presence of retinoic acid. Deubiquitinated by USP7; leading to its nuclear exclusion. Monoubiquitination of one of either Lys-13 and Lys-289 amino acid is sufficient to modulate PTEN compartmentalization. Ubiquitinated by XIAP/BIRC4.

Cellular localization
- Secreted. May be secreted via a classical signal peptide and reenter into cells with the help of a poly-Arg motif and Cytoplasm. Nucleus. Nuclear PML body. Monoubiquitinated form is nuclear. Nonubiquitinated form is cytoplasmic. Colocalized with PML and USP7 in PML nuclear bodies. XIAP/BIRC4 promotes its nuclear localization.
Lanes 1 and 5: PTEN knockout HAP1 cell lysate (20 µg)
Lanes 2 and 6: Wild-type HAP1 cell lysate (20 µg)
Lane 2: Green signal from target – ab32199 observed at 47 kDa
Lanes 3 and 4: Red signal from loading control – ab8245 observed at 37 kDa
Lanes 5 and 6: Merged (red and green) signal
ab32199 was shown to specifically recognize PTEN in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when PTEN knockout samples were used. Wild-type and PTEN knockout samples were subjected to SDS-PAGE, ab32199 and ab8245 (loading control to GAPDH) were diluted to 1/500 and 1/2000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1/10,000 dilution for 1 h at room temperature before imaging.

All lanes: Anti-PTEN antibody [Y184] (ab32199) at 1/10000 dilution (purified)

All lanes: brain lysate
Lysates/proteins at 10 µg per lane.

Secondary
All lanes: Anti-rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 47 kDa
Observed band size: 54 kDa
why is the actual band size different from the predicted?

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

**Lanes 1**: PTEN knockout HAP1 cell lysate (20 µg)
**Lanes 2**: Wild-type HAP1 cell lysate (20 µg)

Green signal from target
Red signal from loading control – ab8245 observed at 37 kDa

This western blot image is a comparison between ab32199 and a competitor's top cited mouse monoclonal antibody.

**All lanes**: Anti-PTEN antibody [Y184] (ab32199) at 1/10000 dilution (purified)

**Lane 1**: MCF7 whole cell lysate
**Lane 2**: HEK293 whole cell lysate

Lysates/proteins at 10 µg per lane.

**Secondary**
**All lanes**: Anti-rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

**Predicted band size**: 47 kDa
**Observed band size**: 54 kDa why is the actual band size different from the predicted?

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST
Western blot - Anti-PTEN antibody [Y184] (ab32199) at 1/500 dilution (Unpurified) + MCF7 cell lysate

**Predicted band size:** 47 kDa
**Observed band size:** 54 kDa *why is the actual band size different from the predicted?*

All lanes: Anti-PTEN antibody [Y184] (ab32199) at 1/500 dilution (Unpurified)

All lanes: Mouse primary bone marrow derived macrophage whole cell lysate

Lysates/proteins at 50 µg per lane.

Secondary

All lanes: HRP-conjugated goat anti-rabbit IgG polyclonal

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 47 kDa
**Observed band size:** 50 kDa *why is the actual band size different from the predicted?*

**Exposure time:** 30 seconds

Treatment:

Lane 1 - control siRNA 24 hours
Lane 2 - PTEN siRNA 24 hours
Lane 3 - control siRNA 48 hours
Lane 4 - PTEN siRNA 48 hours
Anti-PTEN antibody [Y184] (ab32199) at 1/500 dilution + RBL-2H3 whole cell lysate

Secondary
Goat Anti-rabbit HRP at 1/1000 dilution

Developed using the ECL technique.

Predicted band size: 47 kDa

Please note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE"

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