abcam

Product datasheet

# Anti-BACE1 antibody [EPR19523] ab183612

KO VALIDATED Recombinant RabMAb

9 Abreviews 28 References 15 Images

## Overview

| | |
|---|---|
| **Product name** | Anti-BACE1 antibody [EPR19523] |
| **Description** | Rabbit monoclonal [EPR19523] to BACE1 |
| **Host species** | Rabbit |
| **Tested applications** | **Suitable for:** IHC-P, WB, IP, IHC-Fr |
| **Species reactivity** | **Reacts with:** Mouse, Rat, Human |
| **Immunogen** | Recombinant fragment. This information is proprietary to Abcam and/or its suppliers. |
| **Positive control** | WB: Mouse and rat hippocampus and brain lysates. IHC-P: Mouse hippocampus and cerebrum tissues; rat cerebrum tissue. IHC-Fr: Mouse hippocampus tissue. IP: Rat and mouse hippocampus whole cell lysates. |
| **General notes** | This product is a recombinant monoclonal antibody, which offers several advantages including:<br>- High batch-to-batch consistency and reproducibility<br>- Improved sensitivity and specificity<br>- Long-term security of supply<br>- Animal-free production<br>For more information **see here**.<br>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. |

## Properties

| | |
|---|---|
| **Form** | Liquid |
| **Storage instructions** | Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle. |
| **Storage buffer** | pH: 7.2<br>Preservative: 0.01% Sodium azide<br>Constituents: 59% PBS, 40% Glycerol, 0.05% BSA |
| **Purity** | Protein A purified |
| **Clonality** | Monoclonal |
| **Clone number** | EPR19523 |

| | |
|---|---|
| **Isotype** | IgG |

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab183612 in the following tested applications.

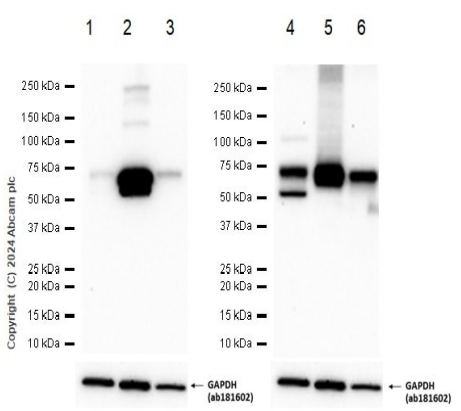
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

| Application | Abreviews | Notes |
|---|---|---|
| **IHC-P** | ★★★★★ (8) | 1/50. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. IHC-P is recommended for mouse only. Binding in rat is weak under our experimental conditions and requires further optimization. |
| **WB** | ★★★★☆ (1) | 1/1000. Detects a band of approximately 68 kDa (predicted molecular weight: 56 kDa). |
| **IP** | | 1/50. |
| **IHC-Fr** | | 1/250. IHC-Fr is recommended for mouse only. |

## Target

| | |
|---|---|
| **Function** | Responsible for the proteolytic processing of the amyloid precursor protein (APP). Cleaves at the N-terminus of the A-beta peptide sequence, between residues 671 and 672 of APP, leads to the generation and extracellular release of beta-cleaved soluble APP, and a corresponding cell-associated C-terminal fragment which is later released by gamma-secretase. |
| **Tissue specificity** | Expressed at high levels in the brain and pancreas. In the brain, expression is highest in the substantia nigra, locus coruleus and medulla oblongata. |
| **Sequence similarities** | Belongs to the peptidase A1 family. |
| **Domain** | The transmembrane domain is necessary for its activity. It determines its late Golgi localization and access to its substrate, APP. |
| **Post-translational modifications** | Glycosylated. |
| **Cellular localization** | Membrane. Golgi apparatus > trans-Golgi network. Endoplasmic reticulum. Endosome. Cell surface. Predominantly localized to the later Golgi/trans-Golgi network (TGN) and minimally detectable in the early Golgi compartments. A small portion is also found in the endoplasmic reticulum, endosomes and on the cell surface. |

## Images




Western blot - Anti-BACE1 antibody [EPR19523] (ab183612)

**All lanes :**

**Lanes 1 & 4 :** SH-SY5Y (human neuroblastoma epithelial cell) whole cell lysate
**Lanes 2 & 5 :** Mouse brain tissue lysate
**Lanes 3 & 6 :** Neuro-2a (Mouse neuroblastoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

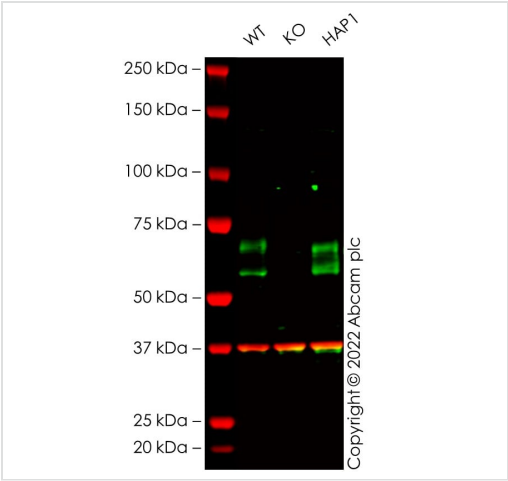
**Predicted band size:** 56 kDa

**Exposure time:** 180 seconds

Blocking and dluting buffer: 5% NFDM/TBST

We suggest optimizing experimental protocols (increasing lysate amount, using lower dilution or higher sensitivity ECL substrate) to improve results.

**ab263901** could be an alternative for getting stronger signal in testing cell lines




Western blot - Anti-BACE1 antibody [EPR19523] (ab183612)

**All lanes :** Anti-BACE1 antibody [EPR19523] (ab183612) at 1/1000 dilution

**Lane 1 :** Wild-type SH-SY5Y cell lysate
**Lane 2 :** Bace1 knockout SH-SY5Y cell lysate
**Lane 3 :** HAP1 cell lysate

Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

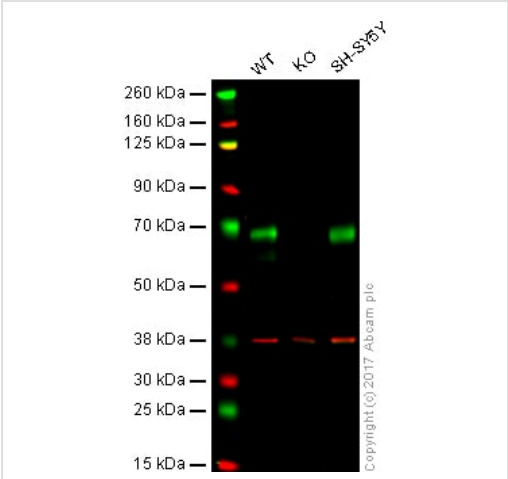
Performed under reducing conditions.

**Predicted band size:** 56 kDa
**Observed band size:** 60,70 kDa

False colour image of Western blot: Anti-BACE1 antibody [EPR19523] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in red.

In Western blot, ab183612 was shown to bind specifically to BACE1. A band was observed at 60/70 kDa in wild-type SH-SY5Y cell lysates with no signal observed at this size in Bace1 knockout cell line **ab280078** (knockout cell lysate **ab280137**).

To generate this image, wild-type and Bace1 knockout SH-SY5Y cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

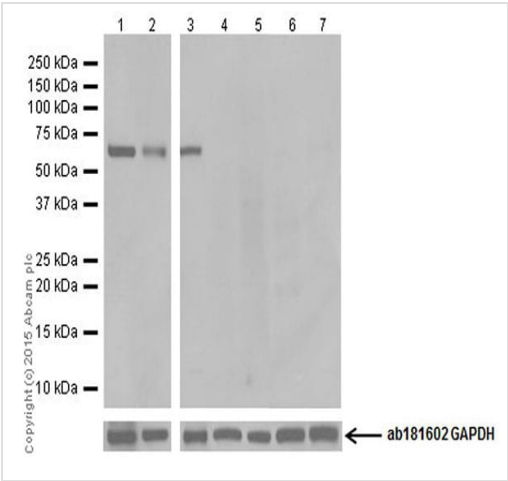



Western blot - Anti-BACE1 antibody [EPR19523] (ab183612)

**Lane 1:** Wild-type HAP1 whole cell lysate (20 µg)
**Lane 2:** BACE1 knockout HAP1 whole cell lysate (20 µg)
**Lane 3:** SHSY5Y whole cell lysate (20 µg)

**Lanes 1 - 3:** Merged signal (red and green). Green - ab183612 observed at 68 kDa. Red - loading control, **ab181602**, observed at 37 kDa.

ab183612 was shown to specifically react with BACE1 in wild-type HAP1 cells as signal was lost in BACE1 knockout cells. Wild-type and BACE1 knockout samples were subjected to SDS-PAGE. ab183612 and **ab181602** (Rabbit anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.




Western blot - Anti-BACE1 antibody [EPR19523] (ab183612)

**All lanes :** Anti-BACE1 antibody [EPR19523] (ab183612) at 1/1000 dilution

**Lane 1 :** Mouse hippocampus lysate
**Lane 2 :** Rat brain lysate
**Lane 3 :** Rat hippocampus lysate
**Lane 4 :** Mouse ovary lysate
**Lane 5 :** Neuro-2a (Mouse neuroblastoma cell line) whole cell lysate
**Lane 6 :** C6 (Rat glial tumor cell line) whole cell lysate
**Lane 7 :** SH-SY5Y (Human neuroblastoma cell line from bone marrow) whole cell lysate

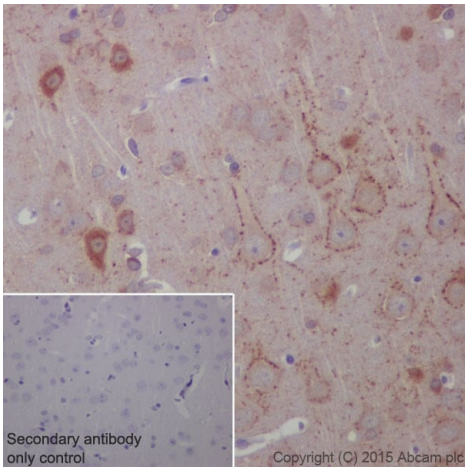
Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

**Predicted band size:** 56 kDa
**Observed band size:** 68 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1 and 2: 1 minute; Lane 3,4,5,6,7 and 8: 3 minutes.



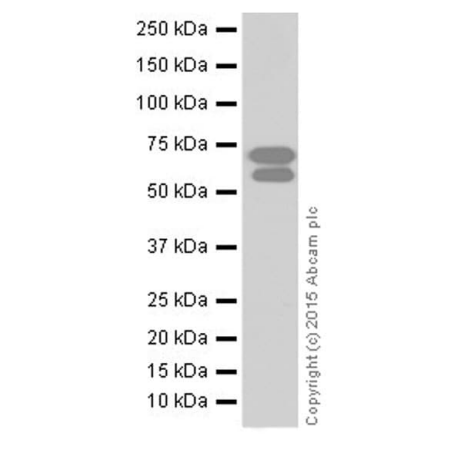

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labeling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasm staining on some neurons of the rat cerebrum is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Binding in rat was weak under our experimental conditions and requires further optimization.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.




Western blot - Anti-BACE1 antibody [EPR19523] (ab183612)

Anti-BACE1 antibody [EPR19523] (ab183612) at 1/1000 dilution + Mouse brain lysate at 10 µg

**Secondary**

Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

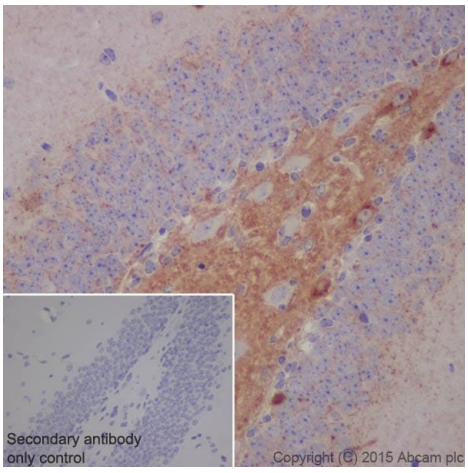
**Predicted band size:** 56 kDa

**Observed band size:** 68 kDa

**Exposure time:** 30 seconds

An additional band was observed at 70 kD. The expression profile is consistent with what has been described in the literature (PMID: 22741101).

Blocking/Dilution buffer: 5% NFDM/TBST.

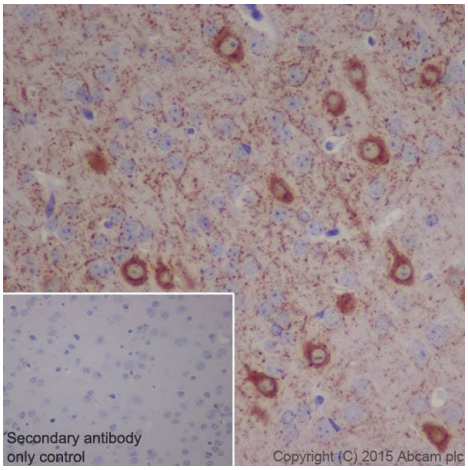



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded mouse hippocampus tissue labeling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasm staining on mouse Hilar region of the dentate gyrus is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

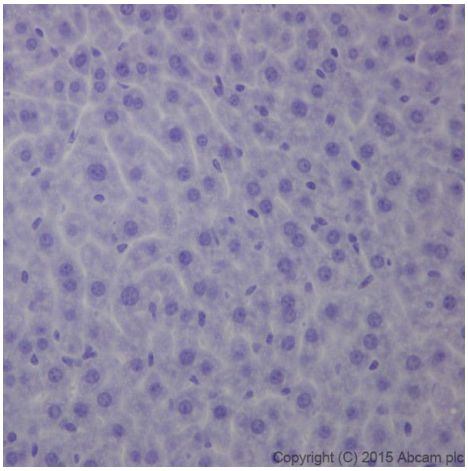



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labeling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Cytoplasm staining on neurons of the mouse cerebrum is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

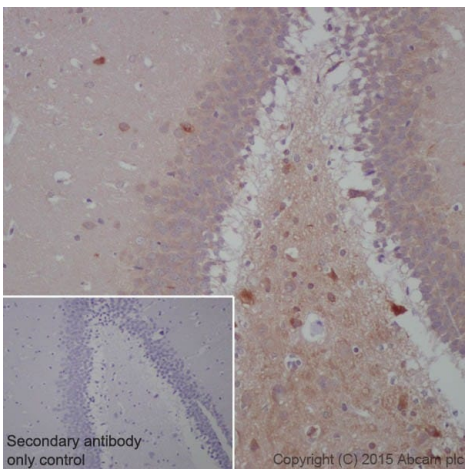



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labeling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Negative staining on mouse liver. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is **ab97051** at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

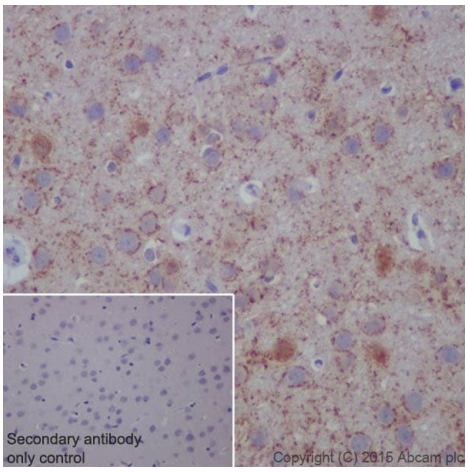



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded rat hippocampus tissue labelling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. The sample was counterstained with hematoxylin. Antigen retrieval was performed using Tris/EDTA buffer; pH 9.0.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Binding in rat was weak under our experimental conditions and requires further optimization.

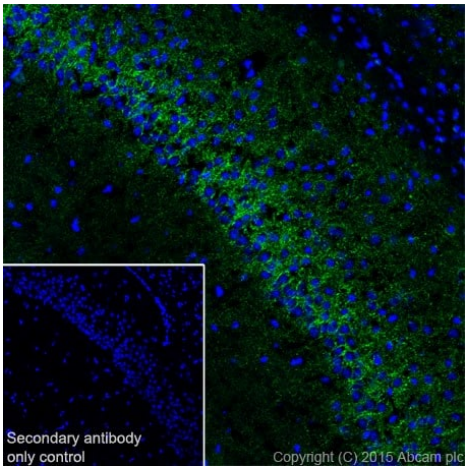



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labelling BACE1 with ab183612 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. The sample was counterstained with hematoxylin. Antigen retrieval was performed using Tris/EDTA buffer; pH 9.0.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

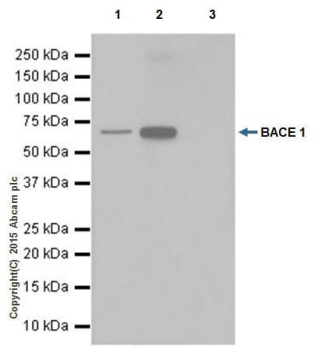
Binding in rat was weak under our experimental conditions and requires further optimization.




Immunohistochemistry (Frozen sections) - Anti-BACE1 antibody [EPR19523] (ab183612)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse hippocampus tissue labeling BACE1 with ab183612 at 1/250 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). The result showed mainly cytoplasmic staining on mouse hippocampus. The nuclear counterstain is DAPI (blue).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution.




Immunoprecipitation - Anti-BACE1 antibody [EPR19523] (ab183612)

BACE1 was immunoprecipitated from 1mg of rat hippocampus whole cell lysate with ab183612 at 1/50 dilution. Western blot was performed from the immunoprecipitate using ab183612 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

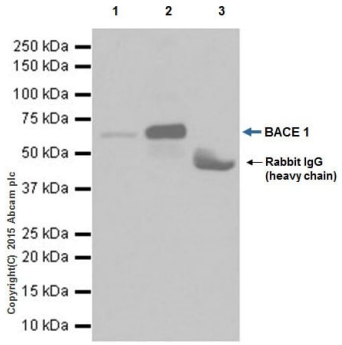
Lane 1: Rat hippocampus whole cell lysate, 10µg (Input).

Lane 2: ab183612 IP in Rat hippocampus whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR25A] - Isotype Control (**ab172730**) instead of ab183612 in rat hippocampus whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 5 seconds.




Immunoprecipitation - Anti-BACE1 antibody [EPR19523] (ab183612)

BACE1 was immunoprecipitated from 1mg of mouse hippocampus whole cell lysate with ab183612 at 1/50 dilution. Western blot was performed from the immunoprecipitate using ab183612 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution.

Lane 1: Mouse hippocampus whole cell lysate, 10µg (Input).

Lane 2: ab183612 IP in mouse hippocampus whole cell lysate.

Lane 3: Rabbit IgG,monoclonal [EPR25A] - Isotype Control (**ab172730**) instead of ab183612 in Mouse hippocampus whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second.


Anti-BACE1 antibody [EPR19523] (ab183612)

## Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

## Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors