

Product datasheet

# Anti-pro Caspase-3 antibody [E61] (HRP) ab205733

**KO VALIDATED** Recombinant RabMAb<sup>®</sup>

3 References 2 Images

Overview

<b>Product name</b>	Anti-pro Caspase-3 antibody [E61] (HRP)
<b>Description</b>	Rabbit monoclonal [E61] to pro Caspase-3 (HRP)
<b>Host species</b>	Rabbit
<b>Conjugation</b>	HRP
<b>Tested applications</b>	<b>Suitable for:</b> IHC-P, WB
<b>Species reactivity</b>	<b>Reacts with:</b> Human <b>Predicted to work with:</b> Mouse 
<b>Immunogen</b>	Synthetic peptide within Human pro Caspase-3 (N terminal). The exact sequence is proprietary. Database link: <a href="#">P42574</a>
<b>Positive control</b>	WB: HeLa and wildtype HAP1 (untreated and Stuarosporine treated) whole cell lysates. IHC-P: normal human tonsil tissue sections
<b>General notes</b>	Our RabMAb <sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMab<sup>®</sup> patents</a> .

Properties

<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Stable for 12 months at -20°C. Store In the Dark.
<b>Storage buffer</b>	pH: 7.4 Preservative: 0.1% Proclin Constituents: PBS, 30% Glycerol, 1% BSA
<b>Purity</b>	Protein A purified
<b>Clonality</b>	Monoclonal
<b>Clone number</b>	E61
<b>Isotype</b>	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab205733** 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		1/2500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB		1/5000. Detects a band of approximately 35 kDa (predicted molecular weight: 31 kDa).

## Target

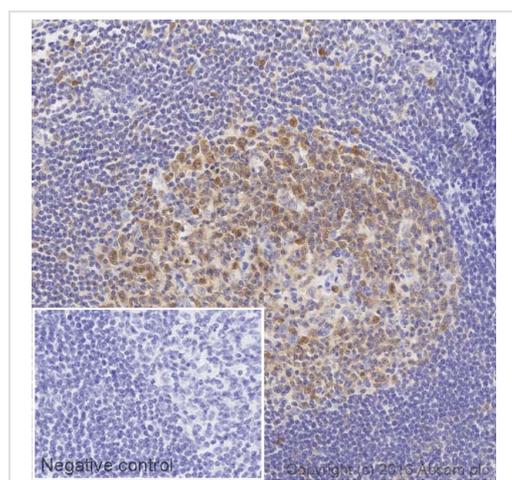
### Relevance

Caspases are a family of cysteine proteases that are key mediators of programmed cell death or apoptosis. The precursor form of all caspases is composed of a prodomain, and large and small catalytic subunits. The active forms of caspases are generated by several stimuli including ligand-receptor interactions, growth factor deprivation and inhibitors of cellular functions. All known caspases require cleavage adjacent to aspartates to liberate one large and one small subunit, which associate into  $\alpha_2\beta_2$  tetramer to form the active enzyme. Gene for Caspase 3 also known as Yama, CPP32, and apopain codes for a 32-kDa protein. Caspase 3 cleaves the death substrate poly(ADP-ribose) polymerase (PARP) to a specific 85 kDa form observed during apoptosis and is inhibitable by the CrmA protein. Other Caspase 3 substrates include DNA-PK, actin, GAS2, and procaspase-6, etc. Caspase 3 is activated by cleavage events at Asp-28/Ser-29 (between N-terminal pro-domain) and Asp-175/Ser-176 (between large and small subunits) to generate a large subunit of 17-kDa and a small subunit of 12-kDa.

### Cellular localization

Cytoplasmic

## Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-pro Caspase-3 antibody [E61] (HRP) (ab205733)

IHC image of pro Caspase-3 staining in a section of formalin-fixed paraffin-embedded normal human tonsil\*, performed on a Leica BOND™. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab205733, 1/2500 dilution, for 15 mins at room temperature. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

\*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Western blot - Anti-pro Caspase-3 antibody [E61]  
(HRP) (ab205733)

**All lanes** : Anti-pro Caspase-3 antibody [E61] (HRP) (ab205733)  
at 1/5000 dilution

**Lane 1** : HeLa whole cell lysate (ab150035) at 10 µg

**Lane 2** : Wild-type HAP1 cell lysate at 20 µg

**Lane 3** : Wild-type HAP1 cell lysate Stuarosporine Treated (1µM  
for 4h) at 20 µg

**Lane 4** : Caspase-3 knockout HAP1 cell lysate at 20 µg

**Lane 5** : Caspase-3 knockout HAP1 cell lysate Stuarosporine  
Treated (1µM for 4h) at 20 µg

Developed using the ECL technique.

Performed under reducing conditions.

**Predicted band size:** 31 kDa

**Observed band size:** 35 kDa

[why is the actual band size different from the predicted?](#)

**Exposure time:** 3 minutes

This blot was produced using a 4-12% Bis-tris gel under the MOPS buffer system. The gel was run at 200V for 50 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 3% milk before being incubated with ab205733 overnight at 4°C. Antibody binding was visualised using ECL development solution [ab133406](#).

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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