

Datasheet for 600-401-AE1**Caspase-8 Antibody****Overview**

Description:	Anti-Caspase-8 (RABBIT) Antibody - 600-401-AE1
Item No.:	600-401-AE1
Size:	100 µg
Applications:	ELISA, IF, IHC, WB
Reactivity:	Human, Mouse, Rat
Host Species:	Rabbit

Product Details

Background:	Caspases are a family of cysteine proteases that can be divided into the apoptotic and inflammatory caspase subfamilies. Unlike the apoptotic caspases, members of the inflammatory subfamily are generally not involved in cell death but are associated with the immune response to microbial pathogens. The apoptotic subfamily can be further divided into initiator caspases, which are activated in response to death signals, and executioner caspases, which are activated by the initiator caspases and are responsible for cleavage of cellular substrates that ultimately lead to cell death. Caspase-8 is an initiator caspase that was identified as a member of the Fas/APO-1 death-inducing signaling complex. The adaptor molecule FADD couples procaspase-8 to the Fas receptor death domain; subsequent oligomerization promotes procaspase-8 autoactivation. FLIP, a catalytically inactive caspase-8-like molecule inhibits these interactions and thus can inhibit apoptosis.
Synonyms:	Caspase-8 Antibody, CAP4, MACH, MCH5, FLICE, ALPS2B, Casp-8, Caspase-8, Apoptotic cysteine protease, CASP-8
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	CASP8
Reactivity:	Human, Mouse, Rat

Immunogen Type:	Conjugated Peptide
Immunogen:	Anti-Caspase-8 antibody was prepared from whole rabbit serum produced by repeated immunizations with a 16 amino acid synthetic peptide from near the C-terminus of human Caspase-8 isoform A. The immunogen is located within the last 50 amino acids of Caspase-8.
Purity/Specificity:	Anti-Caspase-8 Antibody was affinity purified from monospecific antiserum by immunoaffinity chromatography. Depending on cell lines or tissues used, either full-length or other cleavage products may be observed.
Relevant Links:	<ul style="list-style-type: none">• UniProtKB - Q14790• GenelD - 841• NCBI - NP_001219

Application Details

Tested Applications:	ELISA, IF, IHC, WB
Application Note:	Anti-Caspase-8 Antibody has been tested for use in ELISA, Western Blotting, Immunocytochemistry, immunohistochemistry-P, and Immunofluorescence. Specific conditions for reactivity should be optimized by the end user. Expect a band at approximately 55 kDa in Western Blots of specific cell lysates and tissues. Positive controls: Jurkat cells, human spleen tissue, mouse spleen tissue, rat spleen tissue.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:5000 - 1:20,000
IF:	20 µg/mL
IHC:	5 µg/mL
WB:	0.5-2 µg/mL

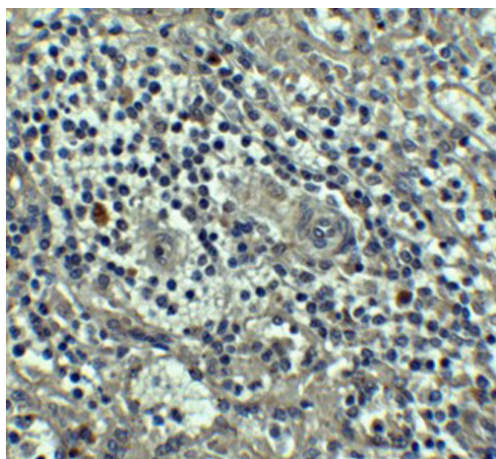
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1 mg/mL by UV absorbance at 280 nm
Buffer:	0.01 M Sodium Phosphate, 0.25 M Sodium Chloride, pH 7.2
Preservative:	0.02% (w/v) Sodium Azide
Stabilizer:	None

Shipping & Handling

Shipping Condition:	Wet Ice
Storage Condition:	Antibody can be stored at 4°C up to one year. Antibodies should not be exposed to prolonged high temperatures.
Expiration:	Expiration date is one (1) year from date of receipt.

Images

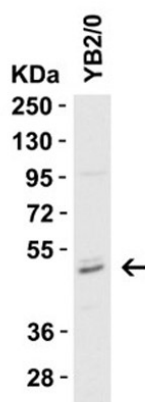


Immunohistochemistry

Immunohistochemistry of Caspase-8.

Tissue: human spleen tissue.

Primary antibody: Caspase-8 antibody at 5 µg/mL.



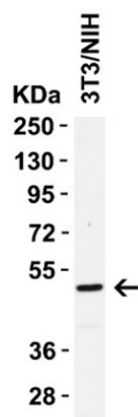
Western Blot

Western Blot Validation of Caspase 8.

Load: 15 µg of rat YB2/0 cell lysate.

Primary antibody: Caspase 8 at 1 µg/mL for 1 h incubation at RT in 5% NFDm/TBST.

Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000 dilution.



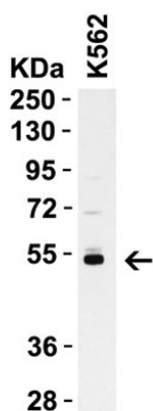
Western Blot

Western Blot Validation of Caspase 8.

Load: 15 μ g of 3T3/NIH cell lysate.

Primary antibody: Caspase-8 at 1 μ g/mL for 1 h incubation at RT in 5% NFDM/TBST.

Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000 dilution.



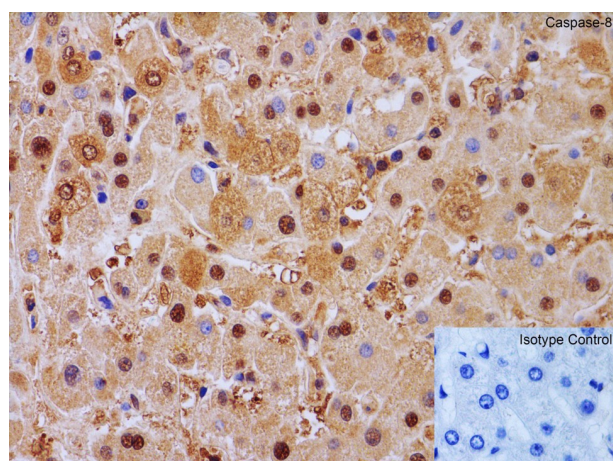
Western Blot

Western Blot Validation of Caspase 8.

Load: 15 μ g of K562 cell lysate.

Primary antibody: Caspase 8 at 1 μ g/mL for 1 h incubation at RT in 5% NFDM/TBST.

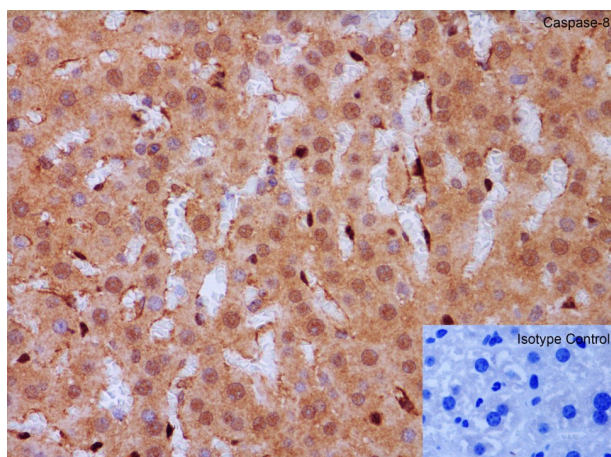
Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000 dilution.



Immunohistochemistry

Immunohistochemistry Validation of Caspase-8.

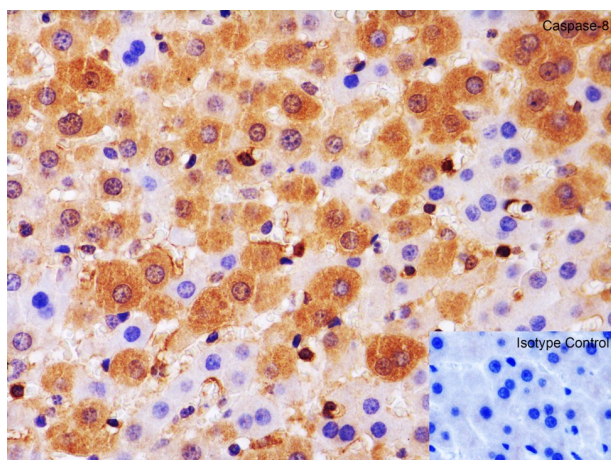
Immunohistochemical analysis of paraffin-embedded human liver tissue using anti-Caspase-8 antibody at 2 μ g/mL. Tissue was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4°C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.



Immunohistochemistry

Immunohistochemistry Validation of Caspase-8.

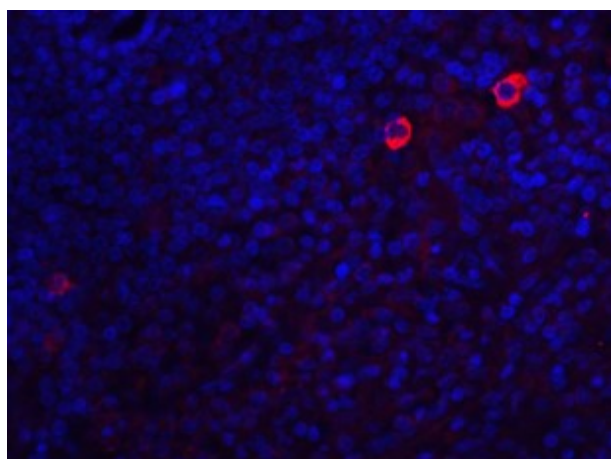
Immunohistochemical analysis of paraffin-embedded mouse liver tissue using anti-Caspase-8 antibody at 2 $\mu\text{g}/\text{ml}$. Tissue was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4°C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.



Immunohistochemistry

Immunohistochemistry Validation of Caspase-8.

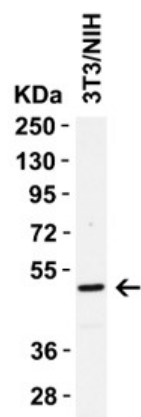
Immunohistochemical analysis of paraffin-embedded rat liver tissue using anti-Caspase-8 antibody at 2 $\mu\text{g}/\text{ml}$. Tissue was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4°C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.



Immunofluorescence Microscopy

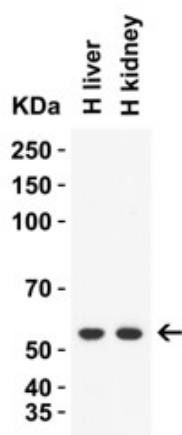
Immunofluorescence Validation of Caspase-8.

Immunofluorescent analysis of 4% paraformaldehyde-fixed human spleen tissue labeling Caspase-8 with at 20 $\mu\text{g}/\text{mL}$, followed by goat anti-rabbit IgG secondary antibody at 1/500 dilution (red) and DAPI staining (blue).



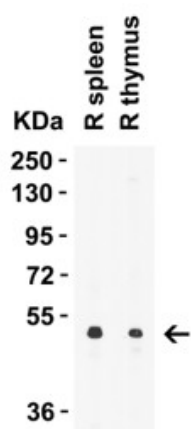
Western Blot

WB Validation. Loading: 15 µg of 3T3 lysate Antibodies:
 Caspase-8, 1 µg/mL, 1 h incubation at RT in 5% NFDm/TBST.
 Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000
 dilution.



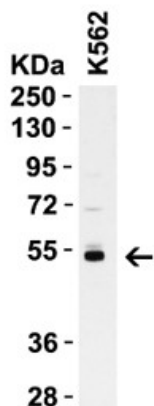
Western Blot

WB Validation. Loading: 15 µg of human tissue lysate.
 Antibodies: Caspase-8, 1 µg/mL, 1 h incubation at RT in 5%
 NFDm/TBST. Secondary: Goat Anti-Rabbit IgG HRP conjugate
 at 1:10000 dilution.



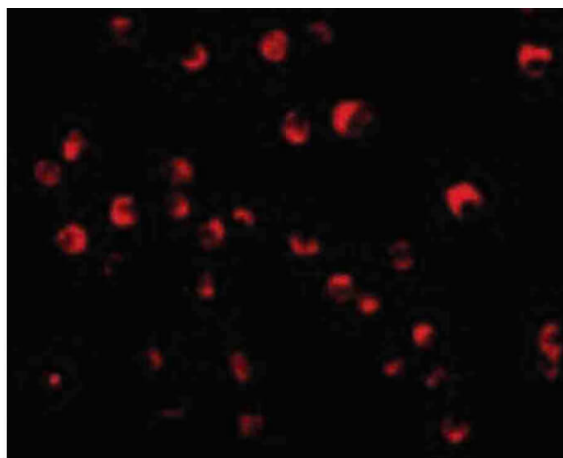
Western Blot

WB Validation. Loading: 15 µg of rat testis lysate. Antibodies:
 Caspase-8, 2 µg/mL, 1 h incubation at RT in 5% NFDm/TBST.
 Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000
 dilution.



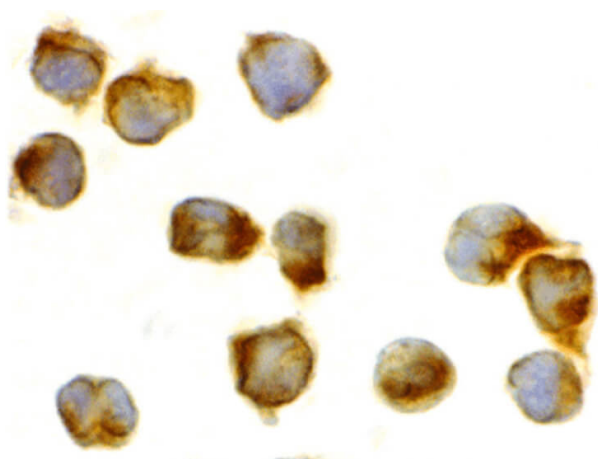
Western Blot

WB Validation. Loading: 15 µg of K562 lysate. Antibodies: Caspase-8, 1 µg/mL, 1 h incubation at RT in 5% NFDM/TBST. Secondary: Goat Anti-Rabbit IgG HRP conjugate at 1:10000 dilution.



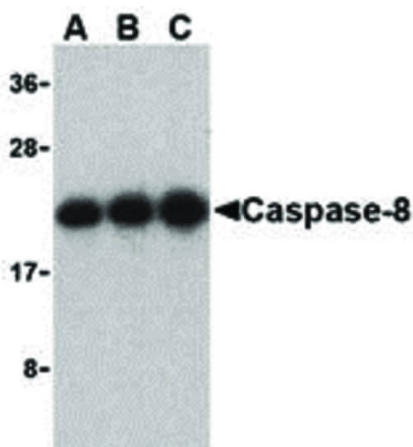
Immunofluorescence Microscopy

Immunofluorescence Microscopy of Caspase-8 antibody. Cell Type: Jurkat cells. Fixation: 0.5% PFA. Antigen retrieval: not required. Primary antibody: Caspase-8 antibody at 20 µg/mL for 1 h at RT. Secondary antibody: Fluorescein rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: Caspase-8 is cytoplasmic. Staining: Caspase-8 as red fluorescent signal.



Immunohistochemistry

Immunocytochemistry of Caspase-8 antibody. Cell Type: Jurkat cells. Fixation: formalin fixed paraffin embedded. Antigen retrieval: not required. Primary antibody: Caspase-8 antibody at 2 µg/mL for 1 h at RT. Secondary antibody: Peroxidase rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: Caspase-8 is cytoplasmic. Staining: Caspase-8 as is stained brown with hematoxylin purple nuclear counterstain.



Western Blot

Western Blot of Caspase-8 antibody in Jurkat cell lysate.

Lane A: Caspase-8 antibody at 0.5 µg/mL. Lane B: Caspase-8 antibody at 1 µg/mL. Lane C: Caspase-8 antibody at 2 µg/mL Load: 35 µg per lane. Primary antibody: Caspase-8 antibody at designated concentrations for overnight at 4°C.

Secondary antibody: Peroxidase rabbit secondary antibody at 1:10,000 for 45 min at RT. Block: 5% BLOTTO overnight at 4°C. Predicted/Observed size: 55 kDa, 21 kDa for Caspase-8. Other band(s): Caspase-8 splice variants and isoforms.

Disclaimer

This product is for research use only and is not intended for therapeutic or diagnostic applications. Please contact a technical service representative for more information. All products of animal origin manufactured by Rockland Immunochemicals are derived from starting materials of North American origin. Collection was performed in United States Department of Agriculture (USDA) inspected facilities and all materials have been inspected and certified to be free of disease and suitable for exportation. All properties listed are typical characteristics and are not specifications. All suggestions and data are offered in good faith but without guarantee as conditions and methods of use of our products are beyond our control. All claims must be made within 30 days following the date of delivery. The prospective user must determine the suitability of our materials before adopting them on a commercial scale. Suggested uses of our products are not recommendations to use our products in violation of any patent or as a license under any patent of Rockland Immunochemicals, Inc. If you require a commercial license to use this material and do not have one, then return this material, unopened to: Rockland Inc., P.O. BOX 5199, Limerick, Pennsylvania, USA.