

Datasheet for 600-401-H16**RIP3 Antibody****Overview**

Description:	Anti-RIP3 (RABBIT) Antibody - 600-401-H16
Item No.:	600-401-H16
Size:	100 µg
Applications:	ELISA, IF, IHC, IP, WB
Reactivity:	Mouse, Rat
Host Species:	Rabbit

Product Details

Background:	RIP3 antibody detects murine RIP3. Certain serine/threonine protein kinases, such as ASK1, RIP, DAP, and ZIP kinases, are mediators of apoptosis. Receptor interacting proteins including RIP and RIP2/RICK mediate apoptosis induced by TNFR1 and Fas, two prototype members in the death receptor family. A novel member in the RIP kinase family was recently identified and designated RIP3. RIP3 contains N-terminal kinase domain but, unlike RIP or RIP2, lacks the C-terminal death or CARD domain. RIP3 binds to RIP and TNFR1, mediates TNFR1 induced apoptosis, and attenuates RIP and TNFR1 induced NF-κB activation. Overexpression of RIP3 induces apoptosis and NF-κB activation. The messenger RNA of RIP3 is expressed in a subset of adult tissues. Anti-RIP3 antibodies are ideal for investigators involved in NFκB and Kinase and Phosphatase research.
Synonyms:	Receptor-interacting serine/threonine-protein kinase 3, RIP-like protein kinase 3, Receptor-interacting protein 3, RIP-3, mRIP3, Ripk3, Rip3
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	Ripk3
Reactivity:	Mouse, Rat
Immunogen Type:	Conjugated Peptide

Immunogen:	RIP3 Antibody was produced from whole rabbit serum prepared by repeated immunizations with a peptide corresponding to 14 amino acids near the carboxy terminus of murine RIP3. The immunogen is located within the last 50 amino acids of RIP3.
Purity/Specificity:	Anti-RIP3 Antibody was affinity purified from monospecific antiserum by immunoaffinity purification. A BLAST analysis was used to suggest cross-reactivity with RIP3 with Rat and Mouse based on 100% homology with the immunizing sequence. Cross-reactivity with RIP3 from other sources has not been determined. Mouse RIP3 has one isoform (486aa, 53kD). Rat RIP3 also has one isoform (478aa, 52kD). Human RIP3 has 3 isoforms, including isoform 1 (518aa, 57kD), isoform 2 (252aa, 28kD) and isoform 3 (231aa, 25kD). Anti-RIP3 Antibody can detect can detect isoforms of mouse and rat.
Relevant Links:	• UniProtKB - Q9QZL0 • GeneID - 56532

Application Details

Tested Applications:	ELISA, IF, IHC, IP, WB
Application Note:	Anti-Rip3 Antibody is tested for use in E, WB, IF, IP, and IHC-P. Expect a band approximately ~53 kDa for mouse on specific lysates or tissues. Specific conditions for reactivity should be optimized by the end user. Positive controls used are A431 cells, NIH3T3 cells, C2C12 cells, and 3T3/Balb cells.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
IF:	20 µg/mL
IHC:	5 µg/mL
IP:	20µg/mL
WB:	0.5-1 ug/mL

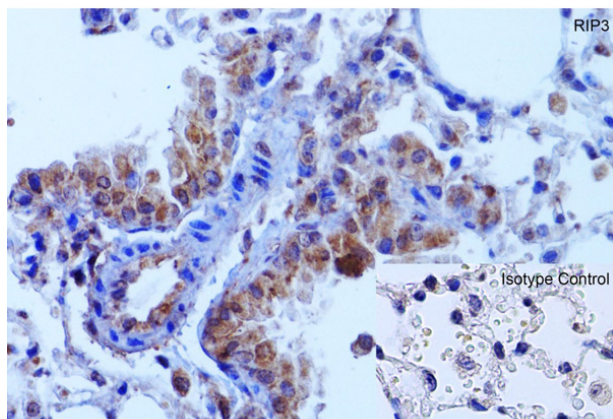
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.0 mg/ml by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.02% (w/v) Sodium Azide
Stabilizer:	None

Shipping & Handling

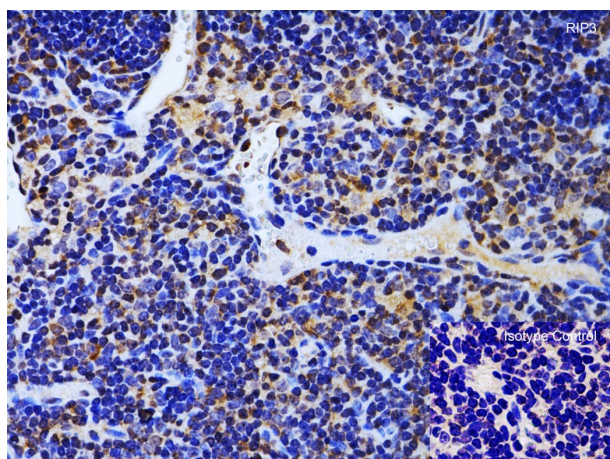
Shipping Condition:	Wet Ice
Storage Condition:	Antibody can be stored at 4°C up to one year. Antibodies should not be exposed to prolonged high temperatures.
Expiration:	Expiration date is one (1) year from date of receipt.

Images



Immunohistochemistry

Immunohistochemistry Validation of RIP3 in Mouse Lung.



Immunohistochemistry

Immunohistochemistry Validation of RIP3.

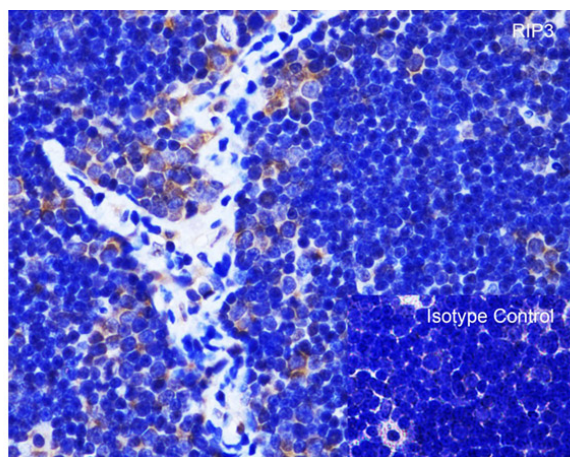
Tissue: Mouse Thymus.

Fixation: paraffin-embedded, formaldehyde and blocked with 10% serum for 1 h at RT.

Antigen retrieval: heat mediation with a citrate buffer (pH6).

Primary Antibody: anti-RIP3 antibody at 1 µg/ml overnight at 4°C.

Secondary: goat anti-rabbit IgG H&L (HRP) at 1:250. Counter stained with Hematoxylin.



Immunohistochemistry

Immunohistochemistry Validation of RIP3.

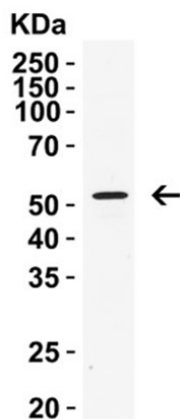
Tissue: Rat Thymus.

Fixation: paraffin-embedded, formaldehyde and blocked with 10% serum for 1 h at RT.

Antigen retrieval: heat mediation with a citrate buffer (pH6).

Primary Antibody: anti-RIP3 antibody at 1 µg/ml overnight at 4°C.

Secondary: goat anti-rabbit IgG H&L (HRP) at 1:250. Counter stained with Hematoxylin.



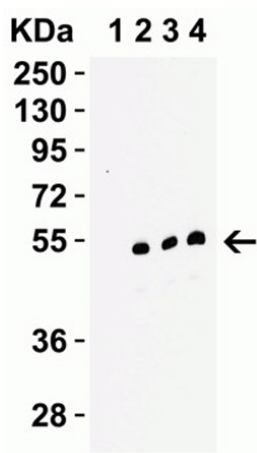
Western Blot

Western Blot Validation of RIP3.

Load: 15 µg of Rat Thymus lysate.

Primary antibody: RIP3 at 0.5 µg/mL for 1h incubation at RT in 5% NFDm/TBST.

Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



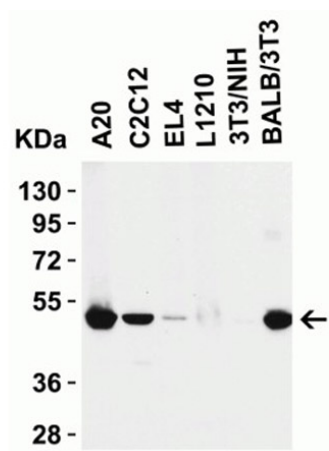
Western Blot

Western Blot Validation of RIP3.

Load: 15 µg of mouse C2C12 Cell lysates per lane.

Primary antibody: RIP3 at (Lane 1: 0.1 µg/mL in the presence of peptide blocking. Lane 2: 0.1 µg/mL. Lane 3: 0.2 µg/mL. Lane 4: 0.5 µg/mL) for 1h incubation at RT in 5% NFDm/TBST.

Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



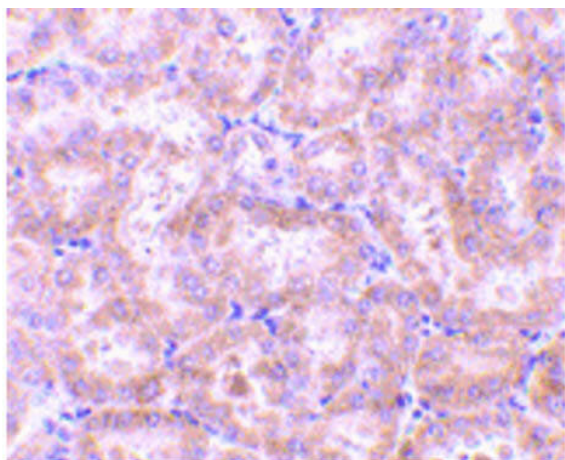
Western Blot

Western Blot Validation of RIP3.

Load: 15 µg of mouse cell lysates. Lane 1: A20, Lane 2: C2C12, Lane 3: EL4, Lane 4: L1210, Lane 5: 3T3/NIH, Lane 6: BALB/3T3.

Primary antibody: RIP3 at 0.5 µg/mL for 1h incubation at RT in 5% NFDN/TBST.

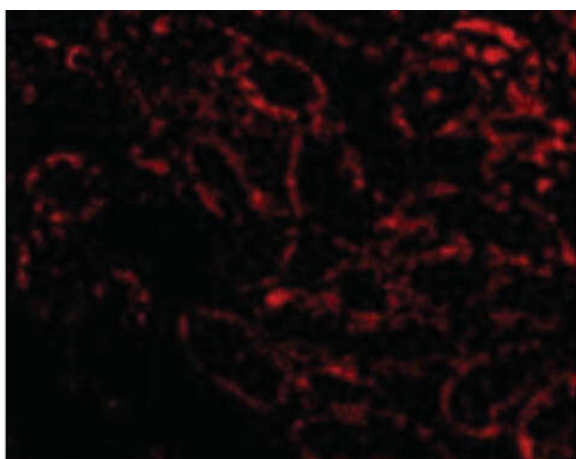
Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



Immunohistochemistry

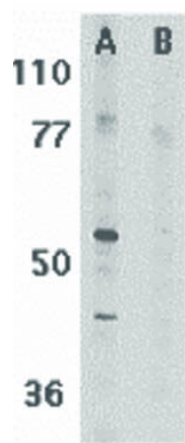
Immunohistochemistry of RIP3 antibody. Tissue: Rat kidney tissue. Fixation: formalin fixed paraffin embedded.

Antigen retrieval: not required. Primary antibody: RIP3 antibody at 5 µg/mL for 1 h at RT. Secondary antibody: Peroxidase rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: RIP3 is cytoplasmic and is also localized in the cell membrane and mitochondria. Staining: RIP3 as precipitated brown signal with hematoxylin purple nuclear counterstain.



Immunofluorescence Microscopy

Immunofluorescence Microscopy of RIP3 antibody. Tissue: Rat kidney cells. Fixation: 0.5% PFA. Antigen retrieval: not required. Primary antibody: RIP3 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: RIP3 is cytoplasmic and is also localized in the cell membrane and mitochondria. Staining: RIP3 as red fluorescent signal.

**Western Blot**

Western Blot of RIP3 antibody. Lane 1: Mouse 3T3 whole cell lysate. Lane 2: Mouse 3T3 whole cell lysate in the presence of blocking peptide. Load: 35 µg per lane. Primary antibody: RIP3 antibody at 1 µg/mL 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: 56.9 kDa, 62 kDa for RIP3. Other band(s): RIP3 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.