

Phospho-RIP (Ser321) Antibody



Orders: 877-616-CELL (2355)
orders@cellsignal.com

Support: 877-678-TECH (8324)

Web: info@cellsignal.com
cellsignal.com

3 Trask Lane | Danvers | Massachusetts | 01923 | USA

For Research Use Only. Not for Use in Diagnostic Procedures.

Applications: W	Reactivity: M	Sensitivity: Endogenous	MW (kDa): 78	Source/Isotype: Rabbit	UniProt ID: #Q60855	Entrez-Gene Id: 19766
---------------------------	-------------------------	-----------------------------------	------------------------	----------------------------------	-------------------------------	---------------------------------

Product Usage Information

Application

Western Blotting

Dilution

1:1000

Storage

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA and 50% glycerol. Store at -20°C. Do not aliquot the antibody.

Specificity/Sensitivity

Phospho-RIP (Ser321) Antibody recognizes endogenous levels of RIP protein only when phosphorylated at Ser321.

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phospho-peptide corresponding to residues surrounding Ser321 of mouse RIP protein. Antibodies are purified by protein A and peptide affinity chromatography.

Background

The receptor-interacting protein (RIP) family of serine-threonine kinases (RIP, RIP2, RIP3, and RIP4) are important regulators of cellular stress that trigger pro-survival and inflammatory responses through the activation of NF-κB, as well as pro-apoptotic pathways (1). In addition to the kinase domain, RIP contains a death domain responsible for interaction with the death domain receptor Fas and recruitment to TNF-R1 through interaction with TRADD (2,3). RIP-deficient cells show a failure in TNF-mediated NF-κB activation, making the cells more sensitive to apoptosis (4,5). RIP also interacts with TNF-receptor-associated factors (TRAFs) and can recruit IKKs to the TNF-R1 signaling complex via interaction with NEMO, leading to IκB phosphorylation and degradation (6,7). Overexpression of RIP induces both NF-κB activation and apoptosis (2,3). Caspase-8-dependent cleavage of the RIP death domain can trigger the apoptotic activity of RIP (8).

Necroptosis, a regulated pathway for necrotic cell death, is triggered by a number of inflammatory signals, including cytokines in the tumor necrosis factor (TNF) family, pathogen sensors such as toll-like receptors (TLRs), and ischemic injury (9,10). The process is negatively regulated by caspases and is initiated through a complex containing the RIP and RIP3 kinases, typically referred to as the necrosome. Necroptosis is inhibited by a small molecule inhibitor of RIP, necrostatin-1 (Nec-1) (11). Research studies show that necroptosis contributes to a number of pathological conditions, and Nec-1 has been shown to provide neuroprotection in models such as ischemic brain injury (12). RIP is phosphorylated at several sites within the kinase domain that are sensitive to Nec-1, including Ser14, Ser15, Ser161, and Ser166 (13).

RIP is also phosphorylated at Ser321(mouse)/Ser320(human) by MAPKAPK-2 (MK-2) and TAK1 in response to inflammatory signals such as TNF-α and LPS (14-17). Phosphorylation at this site suppresses RIP mediated apoptosis by inhibiting its interaction with FADD and caspase-8 (14-17).

Background References

1. Meylan, E. and Tschopp, J. (2005) *Trends Biochem Sci* 30, 151-9.
2. Hsu, H. et al. (1996) *Immunity* 4, 387-96.
3. Stanger, B.Z. et al. (1995) *Cell* 81, 513-23.
4. Ting, A.T. et al. (1996) *EMBO J* 15, 6189-96.
5. Kelliher, M.A. et al. (1998) *Immunity* 8, 297-303.
6. Devin, A. et al. (2000) *Immunity* 12, 419-29.
7. Zhang, S.Q. et al. (2000) *Immunity* 12, 301-11.
8. Lin, Y. et al. (1999) *Genes Dev* 13, 2514-26.
9. Christofferson, D.E. and Yuan, J. (2010) *Curr Opin Cell Biol* 22, 263-8.
10. Kaczmarek, A. et al. (2013) *Immunity* 38, 209-23.
11. Degterev, A. et al. (2008) *Nat Chem Biol* 4, 313-21.
12. Degterev, A. et al. (2005) *Nat Chem Biol* 1, 112-9.
13. Ofengeim, D. and Yuan, J. (2013) *Nat Rev Mol Cell Biol* 14, 727-36.
14. Jaco, I. et al. (2017) *Mol Cell* 66, 698-710.e5.
15. Geng, J. et al. (2017) *Nat Commun* 8, 359.
16. Dondelinger, Y. et al. (2017) *Nat Cell Biol* 19, 1237-1247.
17. Menon, M.B. et al. (2017) *Nat Cell Biol* 19, 1248-1259.

Species Reactivity	Species reactivity is determined by testing in at least one approved application (e.g., western blot).
Western Blot Buffer	IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.
Applications Key	W: Western Blotting
Cross-Reactivity Key	M: Mouse
Trademarks and Patents	<p>Cell Signaling Technology is a trademark of Cell Signaling Technology, Inc.</p> <p>XP is a registered trademark of Cell Signaling Technology, Inc.</p> <p>All other trademarks are the property of their respective owners. Visit cellsignal.com/trademarks for more information.</p>
Limited Uses	<p>Except as otherwise expressly agreed in a writing signed by a legally authorized representative of CST, the following terms apply to Products provided by CST, its affiliates or its distributors. Any Customer's terms and conditions that are in addition to, or different from, those contained herein, unless separately accepted in writing by a legally authorized representative of CST, are rejected and are of no force or effect.</p> <p>Products are labeled with For Research Use Only or a similar labeling statement and have not been approved, cleared, or licensed by the FDA or other regulatory foreign or domestic entity, for any purpose. Customer shall not use any Product for any diagnostic or therapeutic purpose, or otherwise in any manner that conflicts with its labeling statement. Products sold or licensed by CST are provided for Customer as the end-user and solely for research and development uses. Any use of Product for diagnostic, prophylactic or therapeutic purposes, or any purchase of Product for resale (alone or as a component) or other commercial purpose, requires a separate license from CST. Customer shall (a) not sell, license, loan, donate or otherwise transfer or make available any Product to any third party, whether alone or in combination with other materials, or use the Products to manufacture any commercial products, (b) not copy, modify, reverse engineer, decompile, disassemble or otherwise attempt to discover the underlying structure or technology of the Products, or use the Products for the purpose of developing any products or services that would compete with CST products or services, (c) not alter or remove from the Products any trademarks, trade names, logos, patent or copyright notices or markings, (d) use the Products solely in accordance with CST Product Terms of Sale and any applicable documentation, and (e) comply with any license, terms of service or similar agreement with respect to any third party products or services used by Customer in connection with the Products.</p>