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Phospho-ULK1 (Ser757) Antibody

Cell Signaling TECHNOLOGY®

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For Research Use Only. Not For Use In Diagnostic Procedures.

Entrez-Gene ID #8408
UniProt ID #O75385

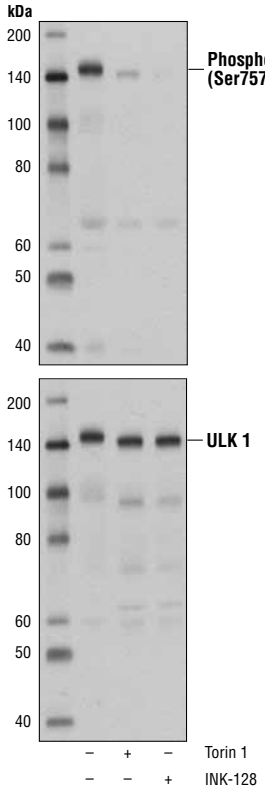
Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W Endogenous	H, M, Mk	140-150 kDa	Rabbit**

Background: Two related serine/threonine kinases, UNC-51-like kinase -1 and -2 (ULK1, ULK2), were discovered as mammalian homologs of the *C. elegans* gene UNC-51 in which mutants exhibited abnormal axonal extension and growth (1-4). Both proteins are widely expressed and contain an amino terminal kinase domain followed by a central proline/serine rich domain and a highly conserved carboxy-terminal domain (CTD). The roles of ULK1 and ULK2 in axon growth have been linked to studies showing that the kinases are localized to neuronal growth cones and are involved in endocytosis of critical growth factors such as NGF (5). Yeast two-hybrid studies found ULK1/2 associated with modulators of the endocytic pathway, SynGap, and syntenin (6). Structural similarity of ULK1/2 has also been recognized with the yeast autophagy protein Atg1/Apg1 (7). Knockdown experiments using siRNA demonstrated that ULK1 is essential for autophagy (8), a catabolic process for the degradation of bulk cytoplasmic contents (9,10). It appears that Atg1/ULK1 can act as a convergence point for multiple signals that control autophagy (11), and can bind to several autophagy-related (Atg) proteins, regulating phosphorylation states and protein trafficking (12-16).

AMPK, activated during low nutrient conditions, directly phophorylates ULK1 at muliptle sites including Ser317, Ser555, and Ser777 (17, 18). Conversely, mTOR, which is a regulator of cell growth and is an inhibitor of autophagy, phosphorylates ULK1 at Ser757 and disrupts the interaction between ULK1 and AMPK (17).

Specificity/Sensitivity: Phospho-ULK1 (Ser757) Antibody recognizes endogenous levels of ULK1 protein only when phosphorylated at Ser757 of mouse ULK1 (equivalent to Ser758 of human ULK1).

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Ser757 of mouse ULK1 protein (equivalent to Ser758 of human ULK1). Antibodies are purified by protein A and peptide affinity chromatography.



Western blot analysis of extracts from A172 cells, untreated (-), Torin 1-treated (+; 250 nM; 5 hrs), or INK-128-treated (+; 250 nM; 5 hrs) using Phospho-ULK1 (Ser757) Antibody (upper) or total ULK1 (D8H5) Rabbit mAb #8054 (lower).

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.*

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Background References:

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(7) Matsuura, A. et al. (1997) *Gene* 192, 245-50.
(8) Chan, E.Y. et al. (2007) *J Biol Chem* 282, 25464-74.
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(10) Codogno, P. and Meijer, A.J. (2005) *Cell Death Differ* 12 Suppl 2, 1509-18.
(11) Stephan, J.S. and Herman, P.K. (2006) *Autophagy* 2, 146-8.
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(13) Young, A.R. et al. (2006) *J Cell Sci* 119, 3888-900.
(14) Kamada, Y. et al. (2000) *J Cell Biol* 150, 1507-13.
(15) Lee, S.B. et al. (2007) *EMBO Rep* 8, 360-5.
(16) Hara, T. et al. (2008) *J Cell Biol* 181, 497-510.
(17) Kim, J. et al. (2011) *Nat Cell Biol* 13, 132-41.
(18) Egan, D.F. et al. (2011) *Science* 331, 456-61.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

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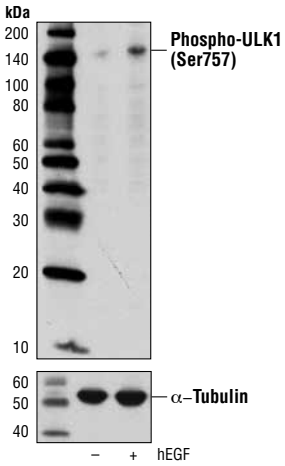
Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



Western blot analysis of extracts from A-431 cells, untreated or treated with Human Epidermal Growth Factor (hEGF) #8916 (100 ng/ml, 30 min) using Phospho-ULK1 (Ser757) Antibody (upper), or α-Tubulin (11H10) Rabbit mAb #2125 (lower).