Revision 1

Store at -20C

#14865

Phospho-Skp2 (Ser64) Antibody

Cell Signaling TECHNOLOGY®

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:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W, IP	H	Endogenous	48	Rabbit	#Q13309	6502

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:50

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-Skp2 (Ser64) Antibody recognizes endogenous levels of Skp2 protein only when phosphorylated at Ser64. This antibody also cross-reacts with an unidentified 160 kDa phosphoprotein in some cells.

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser64 of human Skp2 protein. Antibodies are purified by protein A and peptide affinity chromatography.

Background

Members of the F-box family of proteins are characterized by the approximate 40 amino acid F-box motif named after cyclin F (1,2). F-box proteins constitute one of the four subunits of the Skp1-Cullin-F-box (SCF) ubiquitin ligase complex. The substrate specificity of SCF complexes is determined by the interchangeable F-box proteins, which act as adaptors by associating with phosphorylated substrate proteins and recruiting them to the SCF core. F-box proteins contain two fundamental domains: the F-box motif mediates binding to Skp1 and a leucine rich repeat (LRR) domain mediates substrate interactions.
S phase kinase-associated protein 2 (Skp2) interacts with cyclin A/CDK2 and mediates G1 to S and G2 to M phase transitions by targeting the cyclin-dependent kinase (CDK) inhibitors p27, p21, and p130 for ubiquitination and subsequent proteolysis (3-6). Overexpression of Skp2 results in deregulated proliferation and genetic instabilities typical of cancer cells (7). Research studies have shown that increased Skp2/decreased p27 levels are associated with many aggressive lymphomas and human carcinomas such as colon, breast, prostate and lung cancers (7). Several recent research studies have demonstrated that Skp2 is subject to phosphorylation-dependent regulation by a network of pro-proliferative Ser/Thr kinases. It appears as though phosphorylation of Skp2 at Ser64 by CDK2 (8), Ser72 by Akt1 (9), and Thr417 by PIM1 (10) promotes stabilization of Skp2, possibly constituting an additional mechanism for Skp2 oncogenicity.

Background References

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2. Reed, S.I. (2003) *Nat Rev Mol Cell Biol* 4, 855-64.
3. Zhang, H. et al. (1995) *Cell* 82, 915-25.
4. Nakayama, K. et al. (2004) *Dev Cell* 6, 661-72.
5. Bornstein, G. et al. (2003) *J Biol Chem* 278, 25752-7.
6. Tedesco, D. et al. (2002) *Genes Dev* 16, 2946-57.
7. Bloom, J. and Pagano, M. (2003) *Semin Cancer Biol* 13, 41-7.
8. Rodier, G. et al. (2008) *EMBO J* 27, 679-91.
9. Gao, D. et al. (2009) *Nat Cell Biol* 11, 397-408.
10. Cen, B. et al. (2010) *J Biol Chem* 285, 29128-37.

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 **IP:** Immunoprecipitation

Cross-Reactivity Key

H: Human

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