## Skp1 (D3J4N) Rabbit mAb

**✓** 100 µl (10 western blots)



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

**Applications** Species Cross-Reactivity\* Molecular Wt. Isotype W. IP H, M, R, Mk, (Z) 19 kDa Rabbit IgG\*\* Endogenous

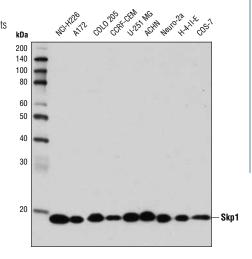
Background: Ubiquitin can be covalently linked to many cellular proteins by the ubiquitination process, which targets proteins for degradation by the 26S proteasome. Three components are involved in the target protein-ubiquitin conjugation process. Ubiquitin is first activated by forming a thiolester complex with the activation component E1; the activated ubiquitin is subsequently transferred to the ubiquitin-carrier protein E2 and then from E2 to ubiquitin ligase E3 for final delivery to the epsilon-NH<sub>a</sub> of the target protein lysine residue (1-3). Combinatorial interactions of different E2 and E3 proteins result in substrate specificity (4). Recent data suggest that activated E2 associates transiently with E3, and the dissociation is a critical step for ubiquitination (5). S phase kinase-associated protein 1 (Skp1) is a critical scaffold protein of the Skp1/CUL1/Fbox (SCF) E3 ubiquitin ligase protein complex. Various F-box proteins (e.g., β-TrCP, Skp2) mediate an interaction with Skp1, via their defining and conserved domain of 40 amino acids, and with substrates to be ubiquitinated (e.g., β-catenin, p27) (4).

Specificity/Sensitivity: Skp1 (D3J4N) Rabbit mAb recognizes endogenous levels of total Skp1 protein.

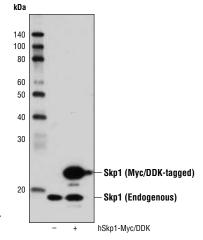
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues near the carboxy terminus of human Skp1 protein.

## **Background References:**

- (1) Ciechanover, A. (1998) EMBO J. 17, 7151-60.
- (2) Hochstrasser, M. (2000) Nat. Cell Biol. 2, E153-7.
- (3) Hochstrasser, M. (2000) Science 289, 563-4.
- (4) DeSalle, L.M. and Pagano, M. (2001) FEBS Lett. 490, 179-89
- (5) Deffenbaugh, A.E. et al. (2003) Cell 114, 611-22.



Western blot analysis of extracts from various cell lines using Skp1 (D3J4N) Rabbit mAb.



Entrez-Gene ID #6500 Swiss-Prot Acc. #P63208

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

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

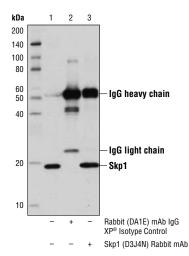
\*\*Anti-rabbit secondary antibodies must be used to detect this antibody.

## **Recommended Antibody Dilutions:**

1:1000 Western blotting Immunoprecipitation 1:100

For product 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 complementary products.



Immunoprecipitation of Skp1 from 293T cell extracts using Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (lane 2) or Skp1 (D3J4N) Rabbit mAb (lane 3). Lane 1 is 10% input. Western blot analysis was performed using Skp1 (D3J4N) Rabbit mAb.

Western blot analysis of extracts from 293T cells, mock transfected ▶ (-) or transfected with a construct expressing Myc/DDK-tagged fulllength human Skp1 isoform b (hSkp1-Myc/DDK; +), using Skp1 (D3J4N) Rabbit mAb.

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.