

K48-linkage Specific Polyubiquitin Antibody



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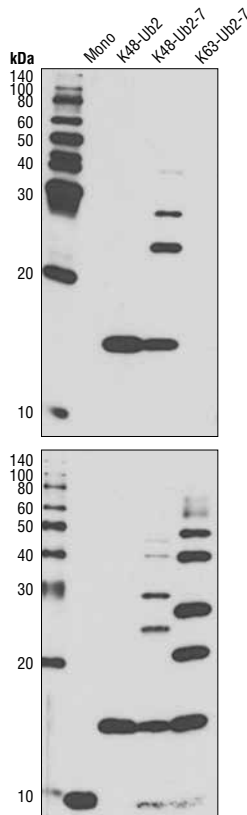
Applications	Species Cross-Reactivity*	Source
W Endogenous	All	Rabbit**

Background: Ubiquitin is a conserved polypeptide unit that plays an important role in the ubiquitin-proteasome pathway. 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 thioester complex with the activation component E1; the activated ubiquitin is subsequently transferred to the ubiquitin-carrier protein E2, then from E2 to ubiquitin ligase E3 for final delivery to the epsilon-NH2 of the target protein lysine residue (1-3). The ubiquitin-proteasome pathway has been implicated in a wide range of normal biological processes and in disease-related abnormalities. Several proteins such as IκB, p53, cdc25a and bcl-2 have been shown to be targets for the ubiquitin-proteasome process as part of regulation of cell cycle progression, differentiation, cell stress response and apoptosis (4-7).

Substrate proteins are linked to ubiquitin using seven distinct ubiquitin lysine residues (Lys6, Lys11, Lys27, Lys29, Lys33, Lys48 and Lys63). Formation of a polyubiquitin chain occurs when a lysine residue of ubiquitin is linked to the carboxy-terminal glycine of another ubiquitin. Proteins polyubiquitinated at specific lysine residues display a tendency to be targeted for different processes; K48-linked polyubiquitin chains mainly target proteins for proteasomal degradation while K63-linked polyubiquitin regulates protein function, subcellular localization, or protein-protein interactions (8).

Specificity/Sensitivity: K48-linkage Specific Polyubiquitin Antibody detects polyubiquitin chains formed by Lys48 residue linkage. Antibody demonstrates slight cross-reactivity with linear polyubiquitin chain. No cross-reactivity observed with monoubiquitin or polyubiquitin chains formed by specific linkage to different lysine residues.

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to the Lys48 branch the human diubiquitin chain. Antibodies are purified by protein A and peptide affinity chromatography.



Western blot analysis of recombinant monoubiquitin, K48-linked polyubiquitin, K48-linked polyubiquitin and K63-linked polyubiquitin using K48-linkage Specific Polyubiquitin Antibody (upper) and Ubiquitin (P4D1) Mouse mAb #3936 (lower).

Entrez-Gene ID #6233
Swiss-Prot Acc. #P62988

Storage: Supplied in 10 mM 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:

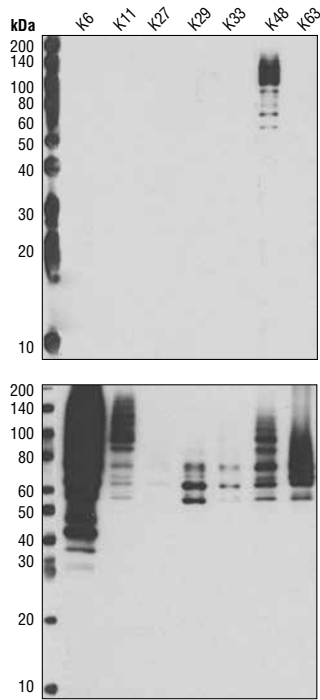
- (1) Ciechanover, A. (1998) *EMBO J.* 17, 7151-7160.
- (2) Hochstrasser, M. (2000) *Nat. Cell Biol.* 2, E153-E157.
- (3) Hochstrasser, M. (2000) *Science* 289, 563-564.
- (4) Bernardi, R. et al. (2000) *Oncogene* 19, 2447-2454.
- (5) Aberle, H. et al. (1997) *EMBO J.* 16, 3797-3804.
- (6) Salomoni, P. and Pandolfi, P.P. (2002) *Nat. Cell Biol.* 4, E152-E153.
- (7) Jesenberger, V. and Jentsch, S. (2002) *Nat. Rev. Mol. Cell Biol.* 3, 112-121.
- (8) Komander, D. (2009) *Biochem Soc Trans* 37, 937-53.

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.

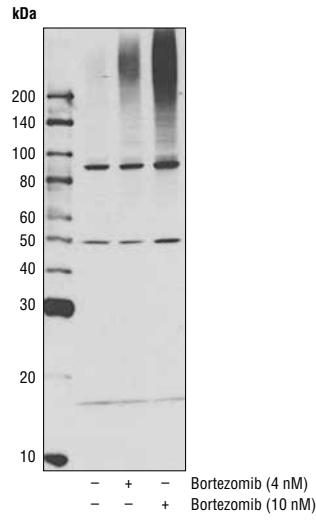
Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide

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 seven distinct recombinant polyubiquitin chains using K48-linkage Specific Polyubiquitin Antibody (upper) and Ubiquitin (P4D1) Mouse mAb #3936 (lower).



Western blot analysis of L363 cells, untreated or treated with bortezomib as indicated, using K48-linkage Specific Polyubiquitin Antibody.