Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb

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**Background:** β-Catenin is a key downstream effector in the Wnt signaling pathway (1). It is implicated in two major biological processes in vertebrates: early embryonic development (2) and tumorigenesis (3). CK1 phosphorylates β-catenin at Ser45. This phosphorylation event primes β-catenin for subsequent phosphorylation by GSK-3β (4–6). GSK-3β destabilizes β-catenin by phosphorylating it at Ser33, Ser37, and Thr41 (7). Mutations at these sites result in the stabilization of β-catenin protein levels and have been found in many tumor cell lines (8).

Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb specifically recognizes β-catenin that is non-phosphorylated at Ser45. Non-phospho β-Catenin (Ser45) protein is useful as readout of stabilized β-catenin protein, and therefore functionally active in cell-cell adhesion and mediating transcriptional activity via the canonical Wnt signaling pathway (9).

**Specificity/Sensitivity:** Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb recognizes endogenous levels of β-catenin protein only when Ser45 is not phosphorylated.

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Ser45 of human β-catenin protein.

**Background References:**

Western blot analysis of extracts from various cell lines using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb (upper) and β-Actin (D6A8) Rabbit mAb #8457 (lower).

**Applications**
- Western Blotting: 1:1000
- Immunoprecipitation: 1:50
- Immunofluorescence: 1:1000
- Immunohistochemistry (Paraffin): 1:1000†
- Flow Cytometry: 1:200
- ELISA-Peptide: 1:50,000
- Chromatin Immunoprecipitation (ChIP): 1:50
- Immunohistochemistry (Frozen): 1:1000

**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**
- Human
- Mouse
- Rat
- Canine
- Equine
- Hamster
- Other (all species expected to react based on 100% homology)

**Recommended Dilutions**
- Western Blotting: 1:1000
- Immunoprecipitation: 1:50
- Immunohistochemistry (Paraffin): 1:1000†
- Immunofluorescence: 1:1000
- Flow Cytometry: 1:200

**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.

**For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com**

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Entrez-Gene ID #1149
UniProt ID #P35222

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Immunohistochemical analysis of paraffin-embedded HeLa (left) or NCI-H28 (right) cell pellets using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb.

Immunohistochemical analysis of paraffin-embedded Apc (Min+/+) mouse intestinal adenoma using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb.

Immunohistochemical analysis of paraffin-embedded human ovarian carcinoma using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb in the presence of phospho-β-Catenin (Ser45) peptide (left) or non-phospho β-Catenin (Ser45) peptide (right).

Immunohistochemical analysis of paraffin-embedded human colon carcinoma using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb.

Immunoprecipitation of non-phospho β-catenin (Ser45) protein from COLO 205 cell extracts. Lane 1 is 10% input, lane 2 is Rabbit (DA1E) mAb IgG XP® Isotype Control #3900, and lane 3 is Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb. Western blot analysis was performed using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb.

Flow cytometric analysis of SW480 (green) and NCI-H28 (blue) cells using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb. Anti-rabbit IgG (H+L), F(ab’)2 Fragment (Alexa Fluor® 488 Conjugate) #4412 was used as a secondary antibody.

Confocal immunofluorescent analysis of SW480 (left and lower right; positive) or NCI-H28 (upper right; negative) cells using Non-phospho (Active) β-Catenin (Ser45) (D2U8Y) XP® Rabbit mAb (green) in the presence of phospho-β-Catenin (Ser45) peptide (lower left) or non-phospho β-Catenin peptide (lower right). Actin filaments were labeled with DyLight® 554 Phalloidin #13054 (red). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).

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