β-Actin (13E5) Rabbit mAb

#4970 Store at –20°C

☐ Small 100 µl (10 western blots)
☐ Large 300 µl (30 western blots)

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

Entrez-Gene ID #60
UniProt ID #P60709

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IHC-P, IHC-F, IF-IC, F Endogenous	H, M, R, Mk, Pg, B, (C, Dg, Hr, Hm)	45 kDa	Rabbit IgG**

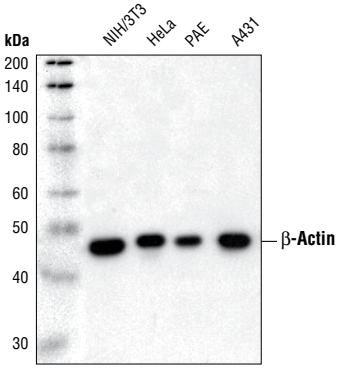
Background: Actin, a ubiquitous protein in eukaryotes, is the major component of the cytoskeleton. At least six isoforms are know in mammals. Nonmuscle β- and γ-actin, also known as cytoplasmic actin, are predominantly expressed in nonmuscle cells, controling cell structure and motility (1). α-cardiac and α-skeletal actin are expressed in striated cardiac and skeletal muscles, respectively; two smooth muscle actins, α- and γ-actin, are found primarily in vascular smooth muscle and enteric smooth muscle, respectively. These actin isoforms regulate contractile potentials for the muscle cells (1). Actin exists mainly as a fiberous polymer, F-actin. In response to cytoskeletal reorganizing signals during processes such as cytokinesis, endocytosis, or stress, cofilin promotes fragmentation and depolymerization of F-actin, resulting in an increase in the monomeric, globular form, G-actin (2). The Arp2/3 complex stabilizes F-actin fragments and promotes formation of new actin filaments (2). It has been reported that actin is hyper-phosphorylated in primary breast tumors (3). Cleavage of actin under apoptotic conditions has been observed *in vitro* and in cardiac and skeletal muscles (4-6). Actin cleavage by caspase-3 may accelerate ubiquitin/proteosome dependent muscle proteolysis (6).

Specificity/Sensitivity: β-Actin (13E5) Rabbit mAb detects endogenous levels of total β-actin protein. This antibody may cross-react with the γ-actin (cytoplasmic isoform). It does not cross-react with α-skeletal, α-cardiac, α-vascular smooth, or γ-enteric smooth muscle isoforms.

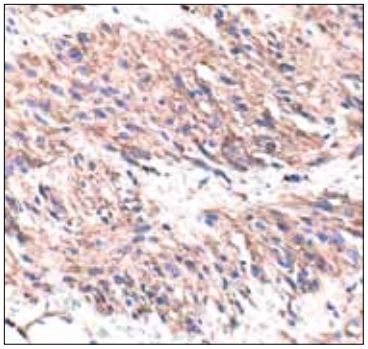
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding residues near the amino-terminus of human β-actin.

Background References:

(1) Herman, I.M. (1993) *Curr. Opin. Cell Biol.* 5, 48–55.
(2) Condeelis, J. (2001) *Trends Cell Biol.* 11, 288–293.
(3) Lim, Y. P. et al. (2004) *Clin. Cancer Res.* 10, 3980–3987.
(4) Kayalar, C. et al. (1996) *Proc. Natl. Acad. Sci. USA.* 93, 2234–2238.
(5) Communal, C. et al. (2002) *Proc. Natl. Acad. Sci. USA.* 99, 6252–6256.
(6) Du, J. et al. (2004) *J. Clin. Invest.* 113, 115–123.



Western blot analysis of cell extracts from various cell lines using β-Actin (13E5) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded human leiomyoma using β-Actin (13E5) Rabbit mAb.

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:

Western blotting	1:1000
Immunohistochemistry (Paraffin)	1:100†
Unmasking buffer:	Citrate
Antibody diluent:	SignalStain® Antibody Diluent #8112
Detection reagent:	SignalStain® Boost (HRP, Rabbit) #8114
Immunohistochemistry (Frozen)	1:50†
Fixative	10% Neutral buffered formalin
Detection reagent:	SignalStain® Boost (HRP, Rabbit) #8114
Immunofluorescence (IF-IC)	1:200
IF Protocol:	Methanol Permeabilization required
Flow Cytometry	1:200

†*Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.*

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.

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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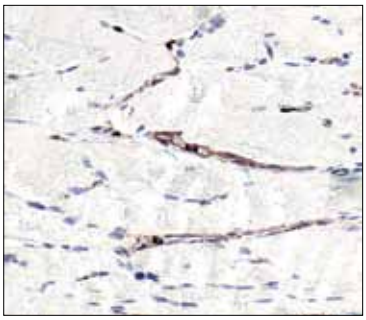
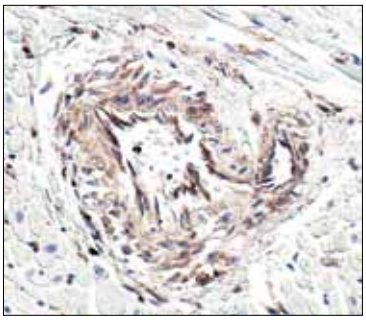
Tween®20 is a registered trademark of ICI Americas, Inc.

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

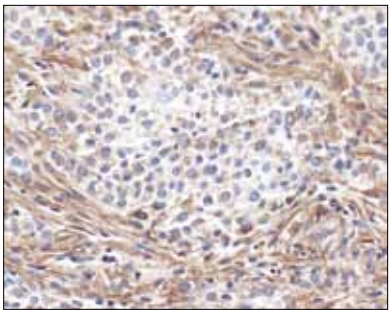
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



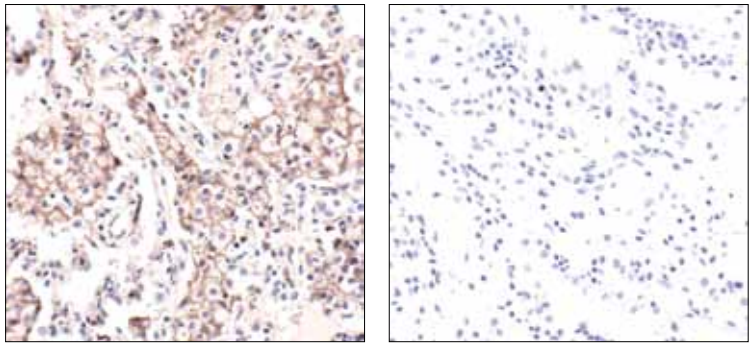
Immunohistochemical analysis of paraffin-embedded human skeletal muscle using β-Actin (13E5) Rabbit mAb. Note the lack of staining of skeletal muscle actin.



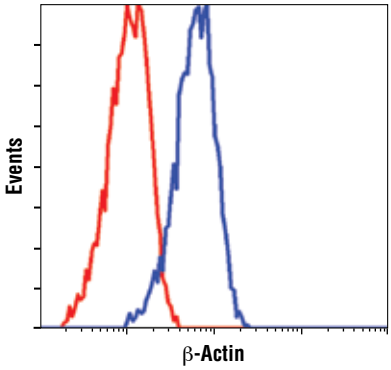
Immunohistochemical analysis of paraffin-embedded human heart using β-Actin (13E5) Rabbit mAb. Note the lack of staining of cardiac actin.



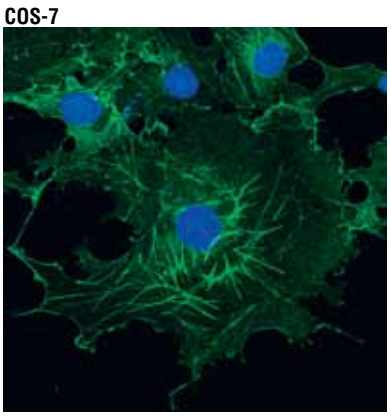
Immunohistochemical analysis of frozen H1650 xenograft, showing membrane and cytoplasmic localization using β-Actin (13E5) Rabbit mAb.



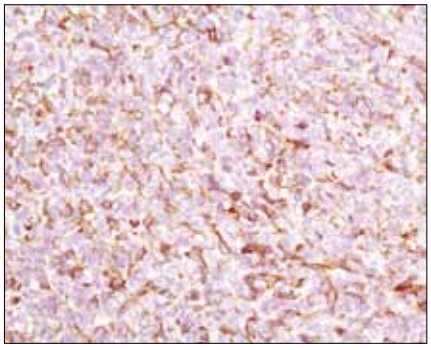
Immunohistochemical analysis of paraffin-embedded human lung carcinoma using β-Actin (13E5) Rabbit mAb (#4970) in the presence of control peptide (left) or β-Actin Blocking Peptide #1025 (right).



Flow cytometric analysis of NIH/3T3 cells using β-Actin (13E5) Rabbit mAb (blue) compared to a nonspecific negative control antibody (red).



Confocal immunofluorescent analysis of COS-7 cells using β-Actin (13E5) Rabbit mAb (green). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).



Immunohistochemical analysis of paraffin-embedded 4T1 syngeneic mouse tumor using β-actin (13E5) Rabbit mAb.