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β-Actin (8H10D10) Mouse mAb

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Entrez-Gene ID #60
Swiss-Prot Acc. #P60709

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IHC-P, IF-IC, F Endogenous	H, M, R, Mk, Hm, Dg	45 kDa	Mouse IgG2b**

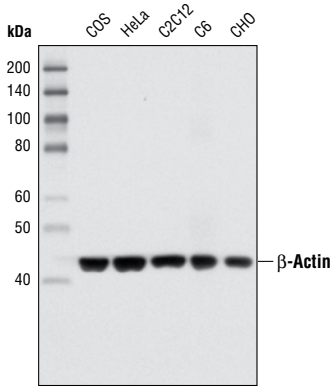
Background: Actin, a ubiquitous eukaryotic protein, is the major component of the cytoskeleton. At least six isoforms are known in mammals. Nonmuscle β- and γ-actin, also known as cytoplasmic actin, are predominantly expressed in nonmuscle cells, controlling cell structure and motility (1). α-cardiac 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 muscle cells (1). Actin exists mainly as a fibrous 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 hyperphosphorylated in primary breast tumors (3). Cleavage of actin under apoptotic conditions has been observed *in vitro* and in cardiac and skeletal muscle (4-6). Actin cleavage by caspase-3 may accelerate ubiquitin/proteosome dependent muscle proteolysis (6).

Specificity/Sensitivity: β-Actin (8H10D10) Mouse mAb detects endogenous levels of total β-actin protein.

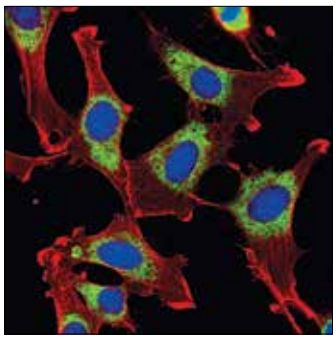
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to amino-terminal residues of human β-actin.

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 extracts from various cell types using β-Actin (8H10D10) Mouse mAb.



Confocal immunofluorescent analysis of NIH/3T3 cells using β-Actin (8H10D10) Mouse mAb (red) and PDI (C81H6) Rabbit mAb #3501 (green). Blue pseudocolor = DRAQ5® #4084 (fluorescent DNA dye).

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-mouse secondary antibodies must be used to detect this antibody**.

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunohistochemistry (Paraffin)	1:16000
Unmasking buffer:	Citrate
Antibody diluent:	SignalStain® Antibody Diluent #8112
Detection reagent:	SignalStain® Boost (HRP, Mouse) #8125

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

Immunofluorescence (IF-IC)	1:5000
IF Protocol:	Methanol Permeabilization required
Flow Cytometry	1:400

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 nonfat dry milk, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

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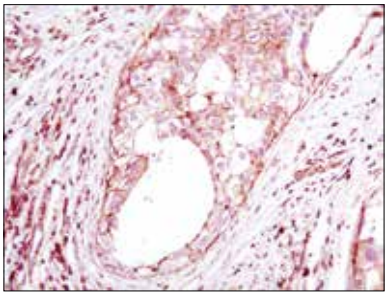
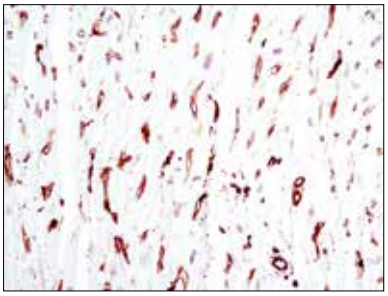
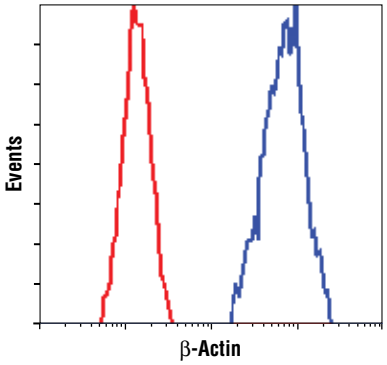
Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **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 breast carcinoma using β-Actin (8H10D10) Mouse mAb.



Immunohistochemical analysis of paraffin-embedded human heart using β-Actin (8H10D10) Mouse mAb. Note the lack of staining of cardiac muscle.



Flow cytometric analysis of HeLa cells using β-Actin (8H10D10) Mouse mAb (blue) compared to a nonspecific negative control antibody (red).