

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

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype	
W. IHC-P. IF-IC. F	H. M. R. Hm. Mk. X. Z. B.	60 kDa	Rabbit loG**	
Endogenous	Pq, (C, Dq, Hr)		······	

Background: In both prokaryotic and eukaryotic cells the misfolding and aggregation of proteins during biogenesis and under conditions of cellular stress are prevented by molecular chaperones (1-3). HSP60 has primarily been known as a mitochondrial protein that is important for folding key proteins after import into the mitochondria (4). Research studies have recently shown that a significant amount of HSP60 is also present in the cytosol of many cells and that it is induced by stress, inflammatory and immune responses, autoantibodies correlated with Alzheimer's, coronary artery diseases, MS, and diabetes (5-8).

Specificity/Sensitivity: HSP60 (D6F1) XP[®] Rabbit mAb recognizes endogenous levels of total HSP60 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Trp68 of human HSP60 protein.





Immunohistochemical analysis of paraffin-embedded human ovarian carcinoma using HSP60 (D6F1) XP[®] Rabbit mAb in the presence of control peptide (upper) or antigen-specific peptide (lower).



Immunohistochemical analysis of paraffin-embedded human breast carcinoma using HSP60 (D6F1) XP® Rabbit mAb.



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Entrez-Gene ID #3329 UniProt ID #P10809

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μ g/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)	1:400†			
Unmasking buffer:	Citrate			
Antibody diluent: SignalStain® Antibody	Diluent #8112			
Detection reagent: SignalStain [®] Boost (HRP, Rabbit) #8114				
†Optimal IHC dilutions determined using SignalStain® Boost IHC				
Detection Reagent.				
Immunofluorescence (IF-IC)	1:800			
Flow Cytometry	1:200			

For product specific protocols please see the web page for this product at www.cellsignal.com.

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A-204



Confocal immunofluorescent analysis of A-204 cells using HSP60 (D6F1) XP[®] Rabbit mAb (green). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red). Blue pseudocolor = DRAQ5[®] #4084 (fluorescent DNA dye).



Western blot analysis of extracts from various cell lines using HSP60 (D6F1) XP[®] 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.

Alexa Fluor is a registered trademark of Molecular Probes, Inc. DRAQ5 is a registered trademark of Biostatus Limited. DyLight is a trademark of Thermo Fisher Scientific Inc. and its subsidiaries. Tween is a registered trademark of ICI Americas, Inc.



Flow cytometric analysis of HeLa cells using HSP60 (D6F1) XP® Rabbit mAb (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control #3900 (red). Antirabbit IgG (H+L), F(ab'), Fragment (Alexa Fluor® 488 Conjugate)

#4412 was used as a secondary antibody.

Immunohistochemical analysis of paraffin-embedded mouse lung using HSP60 (D6F1) XP® Rabbit mAb.

Background References:

(1) Hartl, F.U. (1996) Nature 381, 571-579.

- (2) Bukau, B. and Horwich, A.L. (1998) Cell 92, 351-366.
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- (4) Jindal, S. et al. (1989) Mol. Cell Biol. 9, 2279-2283.
- (5) Itoh, H. et al. (2002) Eur. J. Biochem. 269, 5931-5938.
- (6) Gupta, S. and Knowlton, A.A. J. Cell Mol. Med. 9, 51-58.
- (7) Deocaris, C.C. et al. (2006) *Cell Stress Chaperones* 11, 116-128.
- (8) Lai, H.C. et al. (2007) *Am. J. Physiol. Endocrinol. Metab.* 292, E292-E297.