

HSF1 Antibody

100 µl
 (10 western blots)

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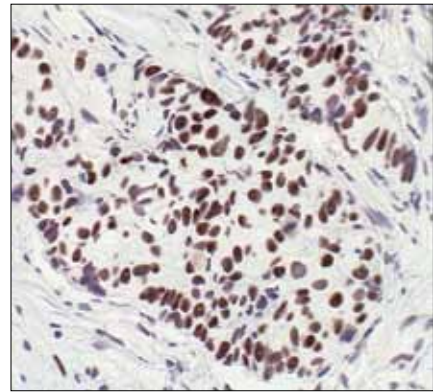
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W, IP, IHC-P, IF-IC, ChIP, F Endogenous	H, M, R, Mk	82 kDa	Rabbit**

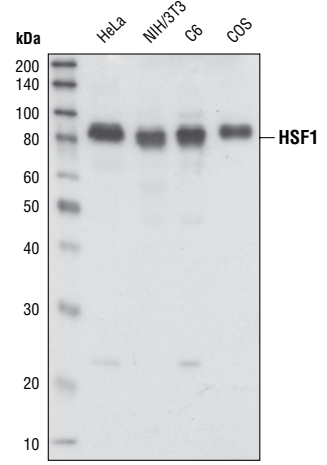
Background: All organisms respond to increased temperatures and other environmental stresses by rapidly inducing the expression of highly conserved heat shock proteins (HSPs) that serve as molecular chaperones to refold denatured proteins and promote the degradation of damaged proteins. Heat shock gene transcription is regulated by a family of heat shock factors (HSFs), transcriptional activators that bind to heat shock response elements (HSEs) located upstream of all heat shock genes (1). HSEs are highly conserved among organisms and contain multiple adjacent and inverse iterations of the pentanucleotide motif 5'-nGAAn-3'. HSFs are less conserved and share only 40% sequence identity. Vertebrate cells contain four HSF proteins: HSF1, 2 and 4 are ubiquitous, while HSF3 has only been characterized in avian species. HSF1 induces heat shock gene transcription in response to heat, heavy metals, and oxidative agents, while HSF2 is involved in spermatogenesis and erythroid cell development. HSF3 and HSF4 show overlapping functions with HSF1 and HSF2. The inactive form of HSF1 exists as a monomer and localizes to both the cytoplasm and nucleus, but does not bind DNA (1,2). In response to stress, HSF1 becomes phosphorylated, forms homotrimerers, binds DNA and activates heat shock gene transcription (1,2). HSF1 activity is positively regulated by phosphorylation of serine 419 by PLK1, which enhances nuclear translocation, and phosphorylation of serine 230 by CaMKII, which enhances transactivation (3,4). Alternatively, HSF1 activity is repressed by phosphorylation of serines 303 and 307 by GSK3 and ERK1, respectively, which leads to binding of 14-3-3 protein and sequestration of HSF1 in the cytoplasm (5,6). In addition, during attenuation from the heat shock response, HSF1 is repressed by direct binding of Hsp70, HSP40/Hdj-1, and HSF binding protein 1 (HSBP1) (7).

Specificity/Sensitivity: This antibody detects endogenous levels of total HSF1 protein. The antibody does not cross-react with other HSF proteins.

Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic peptide corresponding to amino acids at the carboxy-terminus of human HSF1 protein. Antibodies are purified by protein A and peptide affinity chromatography.



Immunohistochemical analysis of paraffin-embedded human breast carcinoma showing nuclear localization, using HSF1 Antibody.



Western blot analysis of extracts from HeLa, NIH/3T3, C6 and COS cells, using HSF1 antibody.

Storage: Supplied in 10 mM sodium 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
Immunoprecipitation	1:50
Immunohistochemistry (Paraffin)	1:250
Unmasking buffer:	Citrate
Antibody diluent:	SignalStain® Antibody Diluent #8112
Immunofluorescence (IF-IC)	1:500
Chromatin IP	1:50
Flow Cytometry	1:50

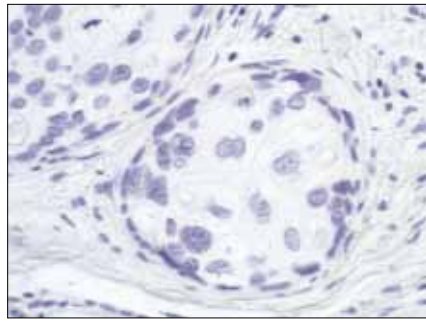
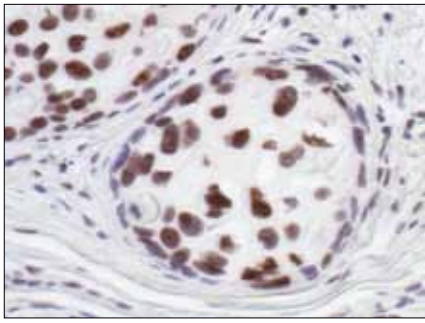
For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

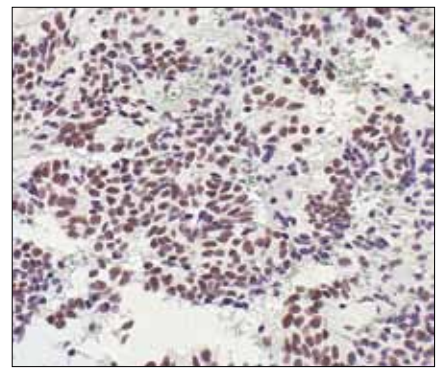
Background References:

- (1) Morimoto, R.I. (1998) *Genes Dev.* 12, 3788–3796.
- (2) Mercier, P.A. et al. (1999) *J. Cell Science* 112, 2765–2774.
- (3) Kim, S.A. et al. (2005) *J. Biol. Chem.* 280, 1 2653–12657.
- (4) Holmberg, C.I. et al. (2001) *EMBO J.* 20, 3800–3810.
- (5) Chu, B. et al. (1996) *J. Biol. Chem.* 271, 30847–30857.
- (6) Wang, X.Z. et al. (2003) *Mol. Cell. Biol.* 23, 6013–6026.
- (7) Satyal, S.H. et al. (1998) *Genes Dev.* 12, 1962–1974.

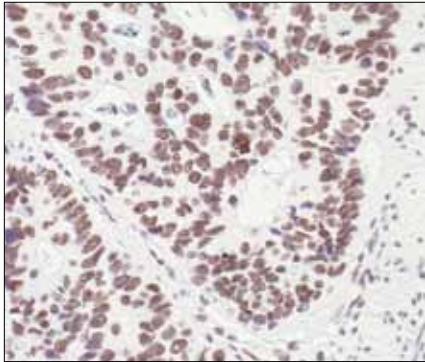
IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.



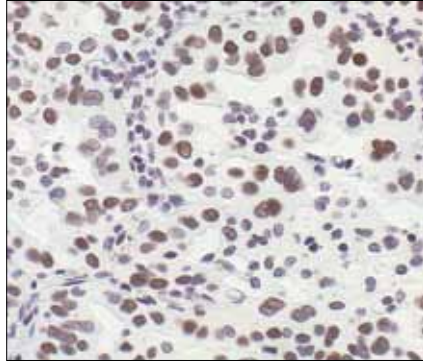
Immunohistochemical analysis of paraffin-embedded human breast carcinoma, using HSF1 Antibody in the presence of control peptide (left) or antigen-specific peptide (right).



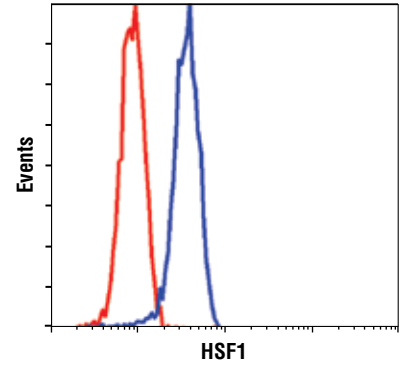
Immunohistochemical analysis of paraffin-embedded human pituitary adenoma, using HSF1 Antibody.



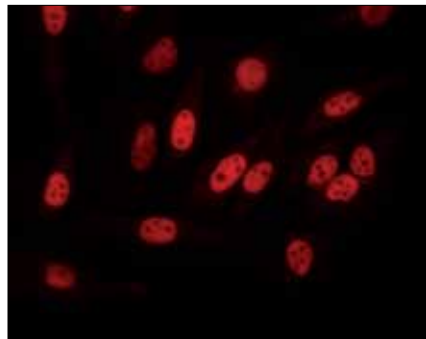
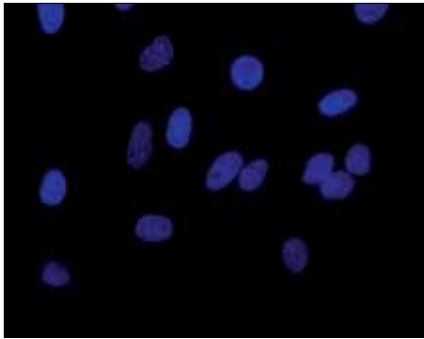
Immunohistochemical analysis of paraffin-embedded human colon carcinoma, using HSF1 Antibody.



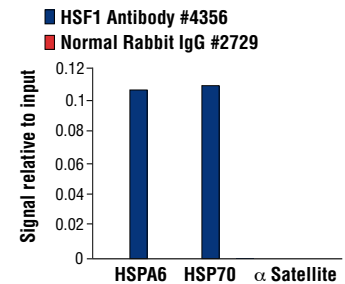
Immunohistochemical analysis of paraffin-embedded human lung carcinoma, using HSF1 Antibody.



Flow cytometric analysis of K562 cells, using HSF1 Antibody (blue) compared to a nonspecific negative control antibody (red).



DAPI staining (left) and immunofluorescent staining (right) of paraformaldehyde-fixed HeLa cells, using HSF1 antibody.



HeLa cells were either untreated (left panel) or heat shocked (right panel) for 1h. Chromatin immunoprecipitations were performed with cross-linked chromatin from 4×10^6 cells and either 10 μ l of HSF1 Antibody or 2 μ l of Normal Rabbit IgG #2729 using SimpleChIP[®] Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using SimpleChIP[®] Human HSPA6 Promoter Primers #5551, human HSP70 intron 1 primers, and SimpleChIP[®] Human α Satellite Repeat Primers #4486. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.