abcam

Product datasheet

Anti-Hsp90 antibody [D7a] ab59459

KO VALIDATED

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Overview

Product name	Anti-Hsp90 antibody [D7a]
Description	Mouse monoclonal [D7a] to Hsp90
Host species	Mouse
Specificity	Can isolate complexes of HSP90, Src kinase and cec37
Tested applications	**Suitable for:** WB, IHC-P, IHC-Fr, IP, ICC/IF, Flow Cyt
Species reactivity	**Reacts with:** Mouse, Rat, Rabbit, Chicken, Cow, Human, Pig
Immunogen	Full length native protein (purified) corresponding to Chicken Hsp90 alpha aa 1-728. Full length protein HSP90 purified from chicken brain Database link: **P11501**
Positive control	WB: HEK-293T, HEK-293, HAP1 and HeLa cell lysates. IHC-P: Human colon cancer tissue. Mouse inflamed colon tissue.
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.09% Sodium azide Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine)
Purity	Protein G purified
Clonality	Monoclonal

Clone number	D7a
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab59459 in the following tested applications.

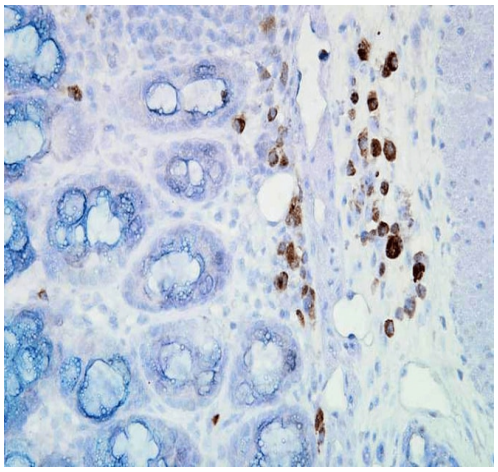
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ **(4)**	1/500. Predicted molecular weight: 95 kDa.
IHC-P		1/100000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
IHC-Fr	★★★★★ **(1)**	Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
ICC/IF	★★★★★ **(4)**	Use at an assay dependent concentration.
Flow Cyt		Use at an assay dependent concentration. **ab170190** - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Target

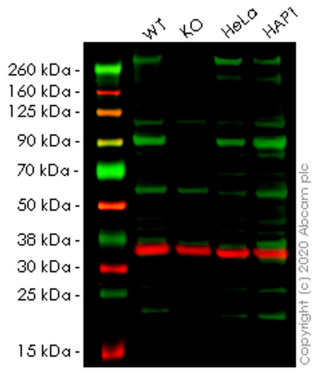
Function	Molecular chaperone that promotes the maturation, structural maintenance and proper regulation of specific target proteins involved for instance in cell cycle control and signal transduction. Undergoes a functional cycle that is linked to its ATPase activity. This cycle probably induces conformational changes in the client proteins, thereby causing their activation. Interacts dynamically with various co-chaperones that modulate its substrate recognition, ATPase cycle and chaperone function.
Sequence similarities	Belongs to the heat shock protein 90 family.
Domain	The TPR repeat-binding motif mediates interaction with TPR repeat-containing proteins like the co-chaperone STUB1.
Post-translational modifications	ISGylated. S-nitrosylated; negatively regulates the ATPase activity and the activation of eNOS by HSP90AA1.
Cellular localization	Cytoplasm. Melanosome. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hsp90 antibody [D7a] (ab59459)

Immunohistochemistry analysis of ab59459 staining Hsp90 alpha in mouse inflamed colon tissue section at 1/100000 dilution for 12 hours at 4°C using antibody amplifier™ system. Biotin Goat Anti-Mouse antibody was used at 1:2000 for 1 hour at room tempertature. Mayer Hematoxylin (purple/blue) nuclear stain at 200 µl for 2 minutes at room temperature was used as a counter stain. Localization: Inflammatory cells. Magnification: 40x.



Western blot - Anti-Hsp90 antibody [D7a] (ab59459)

All lanes : Anti-Hsp90 antibody [D7a] (ab59459) at 1/1000 dilution

Lane 1 : Wild-type HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate
Lane 2 : HSP90AA1 knockout HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate
Lane 3 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 4 : HAP1 whole cell lyate

Lysates/proteins at 20 µg per lane.

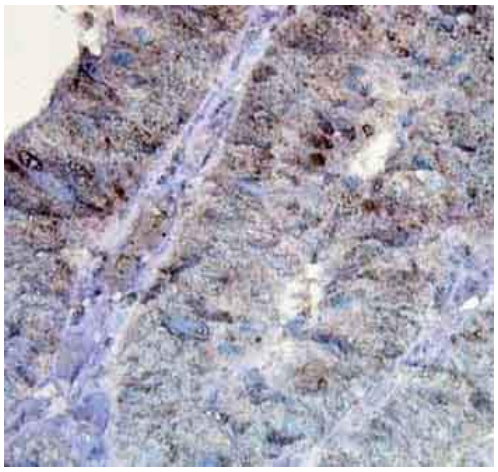
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (**ab216777**) at 1/10000 dilution

Predicted band size: 95 kDa
Observed band size: 90 kDa

Lanes 1-4: Merged signal (red and green). Green - ab59459 observed at 90 kDa. Red - loading control **ab181602** observed at 36 kDa.

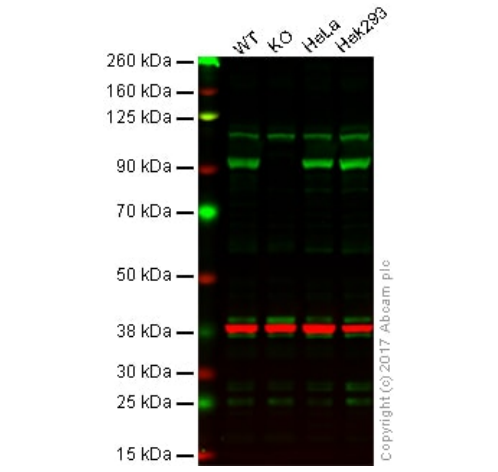
ab59459 Anti-Hsp90 antibody [D7a] was shown to specifically

react with Hsp90 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line **ab266591** (knockout cell lysate **ab258458**) was used. Wild-type and Hsp90 knockout samples were subjected to SDS-PAGE. ab59459 and Anti-GAPDH antibody[EPR16891] - Loading Control (**ab181602**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 680RD) preadsorbed (**ab216777**) and Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (**ab216772**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hsp90 antibody [D7a] (ab59459)

ab59459 staining Hsp90 alpha in human colon cancer tissue section by immunohistochemistry (Bouin's fixed paraffin embedded tissue sections). Tissue underwent heat mediated antigen retrieval in microwave in citrate buffer. The primary antibody was used at dilution of 1/100,000 dilution using antibody amplifier™ system.



Western blot - Anti-Hsp90 antibody [D7a] (ab59459)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: Hsp90 knockout HAP1 whole cell lysate (20 µg)
Lane 3: HeLa whole cell lysate (20 µg)
Lane 4: Hek 293 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab59459 observed at 90 kDa. Red - loading control, **ab181602**, observed at 37 kDa.

ab59459 was shown to specifically recognize Hsp90 in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when Hsp90 knockout cells were examined. Wild-type and Hsp90 knockout samples were subjected to SDS-PAGE. Ab59459 and **ab181602** (Rabbit anti GAPDH loading control) were incubated overnight at 4°C at 1/500 dilution and 1/20,000 dilution respectively. Blots were developed with Goat anti-Mouse IgG H&L (IRDye® 800CW) preabsorbed (**ab216772**) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (**ab216777**) secondary

antibodies at 1/20,000 dilution for 1 hour at room temperature before imaging.

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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