abcam

Product datasheet

Anti-HSF1 antibody [EP1710Y] - BSA and Azide free ab232342

KO 評価済 リコンビナント RabMAb

画像数 12

製品の概要

製品名	Anti-HSF1 antibody [EP1710Y] - BSA and Azide free
製品の詳細	Rabbit monoclonal [EP1710Y] to HSF1 - BSA and Azide free
由来種	Rabbit
アプリケーション	**適用あり:** Flow Cyt (Intra), ChIP, IHC-P, ICC/IF, WB, IP
種交差性	**交差種:** Mouse, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	IHC-P: Human ovarian carcinoma tissue; Mouse testis and colon tissues. WB: K562, HAP1 and HeLa cell lysates. ICC/IF: MCF7 cells. Flow Cyt (intra): HeLa cells. IP: HeLa cell lysate. ChIP: HeLa cells.
特記事項	ab232342 is the carrier-free version of **ab52757**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C. Do Not Freeze.
バッファー	pH: 7.2 Constituent: PBS
キャリア・フリー	はい
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EP1710Y
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab232342の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ChIP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 85 kDa (predicted molecular weight: 57 kDa).
IP		Use at an assay dependent concentration.

ターゲット情報

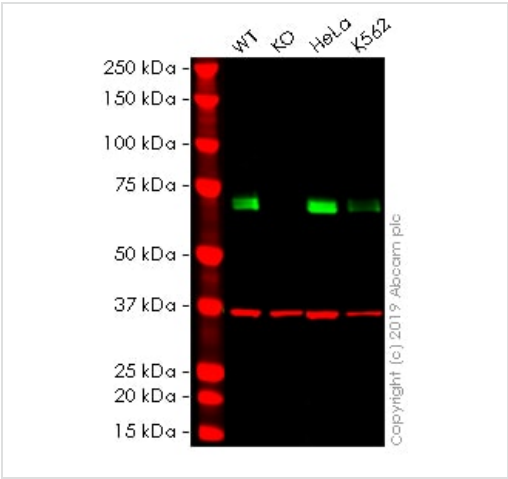
機能	DNA-binding protein that specifically binds heat shock promoter elements (HSE) and activates transcription. In higher eukaryotes, HSF is unable to bind to the HSE unless the cells are heat shocked.
配列類似性	Belongs to the HSF family.
ドメイン	the 9aaTAD motif is a transactivation domain present in a large number of yeast and animal transcription factors.
翻訳後修飾	Phosphorylated on multiple serine residues, a subset of which are involved in stress-related regulation of transcription activation. Constitutive phosphorylation represses transcriptional

activity at normal temperatures. Levels increase on specific residues heat-shock and enhance HSF1 transactivation activity. Phosphorylation on Ser-307 derepresses activation on heat-stress and in combination with Ser-303 phosphorylation appears to be involved in recovery after heat-stress. Phosphorylated on Ser-230 by CAMK2, in vitro. Cadmium also enhances phosphorylation at this site. Phosphorylation on Ser-303 is a prerequisite for HSF1 sumoylation. Phosphorylation on Ser-121 inhibits transactivation and promotes HSP90 binding. Phosphorylation on Thr-142 also mediates transcriptional activity induced by heat. Phosphorylation on Ser-326 plays an important role in heat activation of HSF1 transcriptional activity.
Sumoylated with SUMO1 and SUMO2 on heat-shock. Heat-inducible sumoylation occurs after 15 min of heat-shock, after which levels decrease and at 4 hours, levels return to control levels. Sumoylation has no effect on HSE binding nor on transcriptional activity. Phosphorylation on Ser-303 is a prerequisite for sumoylation.

細胞内局在

Cytoplasm. Nucleus. Cytoplasmic during normal growth. On activation, translocates to nuclear stress granules. Colocalizes with SUMO1 in nuclear stress granules.

画像



Western blot - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

All lanes : Anti-HSF1 antibody [EP1710Y] - ChIP Grade (**ab52757**) at 1/100000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : Hsf1 knockout HAP1 whole cell lysate
Lane 3 : HeLa whole cell lysate
Lane 4 : K562 whole cell lysate

Lysates/proteins at 20 µg per lane.

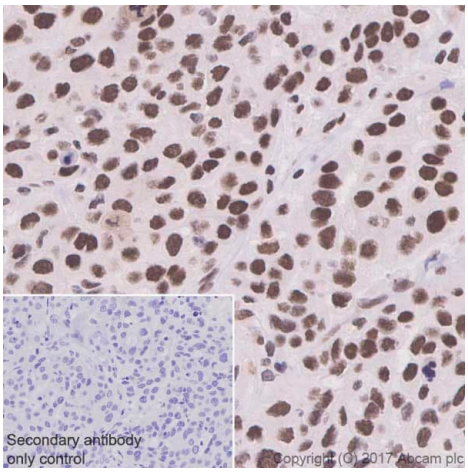
Predicted band size: 57 kDa

Lanes 1 - 4: Merged signal (red and green). Green - **ab52757** observed at 57 kDa. Red - loading control, **ab9484**, observed at 37 kDa.

ab52757 was shown to specifically react with in wild-type HAP1 cells as signal was lost in Hsf1 knockout cells. Wild-type and Hsf1 knockout samples were subjected to SDS-PAGE. Ab52757 and **ab9484** (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/100000 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed **ab216773** and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed **ab216776** secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

This data was developed using the same antibody clone in a

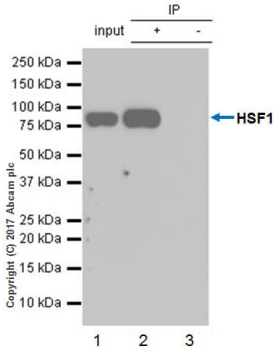
different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human ovarian carcinoma tissue sections labeling HSF1 with Purified **ab52757** at 1:250 dilution (1.06 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



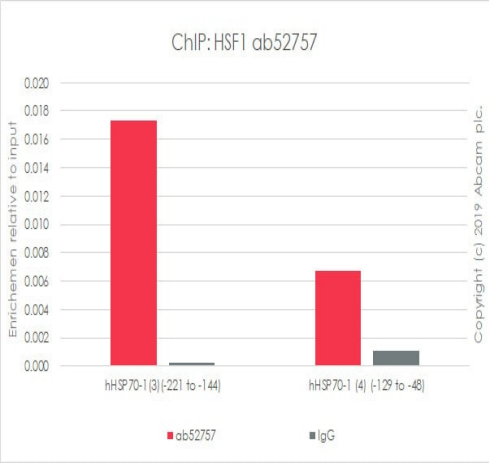
Immunoprecipitation - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

ab52757 (purified) at 1:20 dilution (2µg) immunoprecipitating HSF1 in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate.

Lane 1 (input): HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate 10ug
Lane 2 (+): **ab52757** & HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab52727** in HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



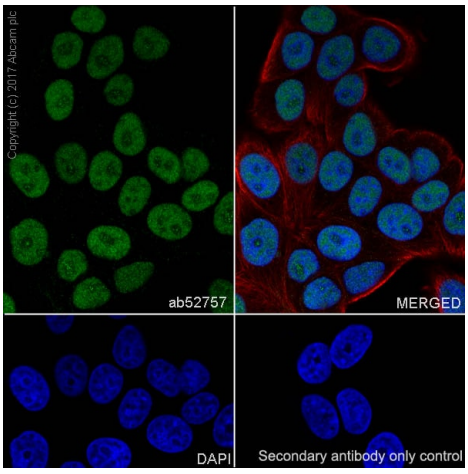
ChIP - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Chromatin was prepared from HeLa cells heat shocked (42^{o}C 30 minutes) according to the Abcam Dual X-ChIP protocol. Cells were fixed with EGS for 30 minutes, then formaldehyde for 10 minutes. The ChIP was performed with 25 µg of chromatin, 5 µg of **ab52757** (red), and 20 µl of Protein A/G sepharose beads. 5 µg of rabbit normal IgG was added to the beads control (gray). The immunoprecipitated DNA was quantified by real time PCR (sybr green approach).

Primers and probes are located in the first kb of the transcribed region.

*http://www.abcam.com/resources?keywords=X%20ChIP%20protocol

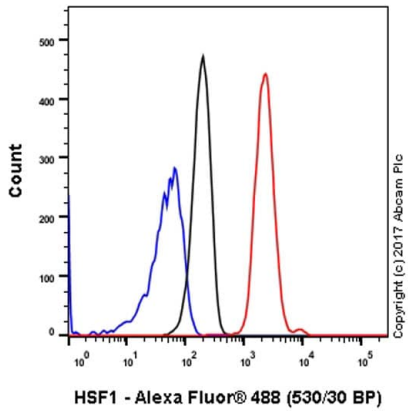
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Immunocytochemistry/ Immunofluorescence - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunocytochemistry/ Immunofluorescence analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling HSF1 with Purified **ab52757** at 1:100 dilution. Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200. **ab150077** Goat anti rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.

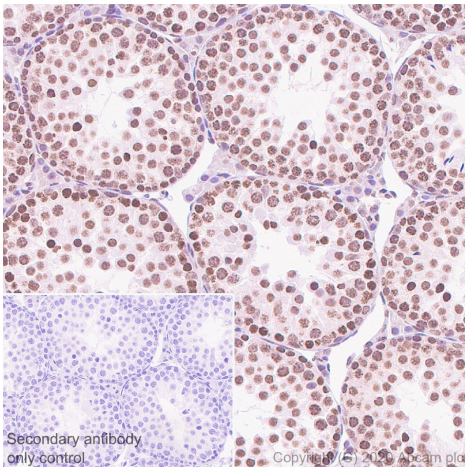
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Flow Cytometry (Intracellular) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling HSF1 with purified **ab52757** at 1/20 dilution (10 µg/ml) (red). Cells were fixed with 80% Methanol and permeabilised with 0.1% Tween-20. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



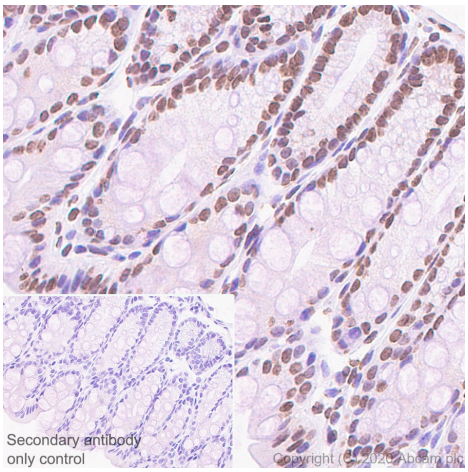
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunohistochemical analysis of paraffin-embedded Mouse testis tissue labeling HSF1 with **ab52757** at 1/1000 followed by a Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) Ready to use. Nuclear staining on mouse testis. The section was incubated with **ab52757** for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) Ready to use.

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



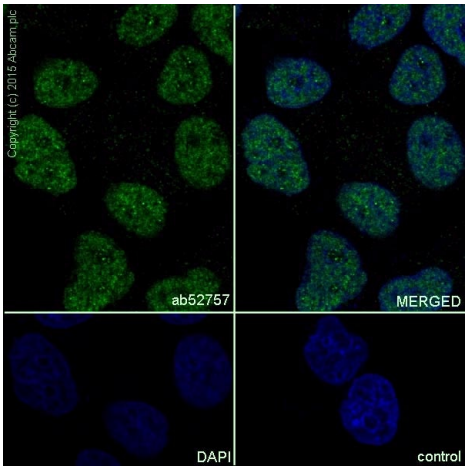
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labeling HSF1 with **ab52757** at 1/1000 followed by a Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) Ready to use. Nuclear staining on mouse colon. The section was incubated with **ab52757** for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**) Ready to use.

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

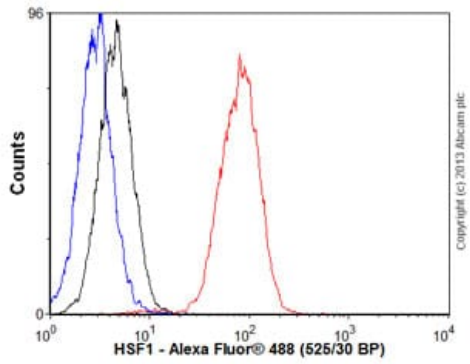
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Immunocytochemistry/ Immunofluorescence - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling HSF1 with unpurified **ab52757** at 1/1000. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor® 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody.
Control: PBS only.
Nuclear counter stain: DAPI.

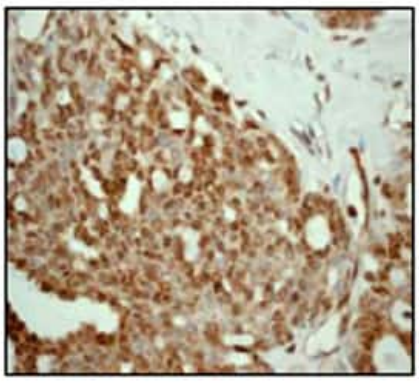
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Flow Cytometry (Intracellular) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Overlay histogram showing HeLa cells stained with unpurified **ab52757** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (unpurified **ab52757**, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

Immunohistochemical staining of paraffin-embedded human ovarian carcinoma using unpurified **ab52757** at a 1:100 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab52757**).

Anti-HSF1 antibody [EP1710Y] - BSA and Azide free (ab232342)

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