

Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free ab220821

KO 評価済 リコンビナント RabMAb

画像数 9

製品の概要

製品名	Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free
製品の詳細	Rabbit monoclonal [EP1531Y] to Hsc70 - BSA and Azide free
由来種	Rabbit
アプリケーション	適用あり: Flow Cyt (Intra), ICC/IF, IP, WB, IHC-P
種交差性	交差種: Mouse, Rat, 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	WB: A431, MCF7, and HeLa cell lysates. IHC-P: Human cerebral cortex tissue. ICC/IF: MCF7 cells. Flow Cyt (intra): MCF7 cells. IP: HeLa cell lysate.
特記事項	<p>ab220821 is the carrier-free version of ab51052.</p> <p>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.</p> <p>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.</p> <p>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.</p> <p>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.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

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: 7.20 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	EP1531Y
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ab220821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。

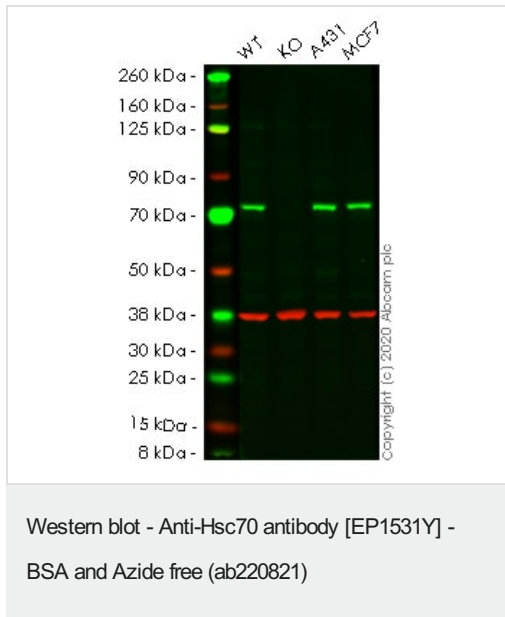
アプリケーション	Abreviews	特記事項
Flow Cyt (Intra)		Use at an assay dependent concentration. ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 71 kDa (predicted molecular weight: 71 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See <u>IHC antigen retrieval protocols</u> .

ターゲット情報

機能	Acts as a repressor of transcriptional activation. Inhibits the transcriptional coactivator activity of CITED1 on Smad-mediated transcription. Chaperone. Isoform 2 may function as an endogenous inhibitory regulator of HSC70 by competing the co-chaperones.
組織特異性	Ubiquitous.
配列類似性	Belongs to the heat shock protein 70 family.
ドメイン	The N-terminal 1-386 residues constitute the ATPase domain, while residues 387-646 form the peptide-binding domain.
翻訳後修飾	Phosphorylated upon DNA damage, probably by ATM or ATR. ISGylated.
細胞内局在	Cytoplasm. Melanosome. Localized in cytoplasmic mRNP granules containing untranslated

mRNAs. Translocates rapidly from the cytoplasm to the nuclei, and especially to the nucleoli, upon heat shock. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

画像



All lanes : Anti-Hsc70 antibody [EP1531Y] ([ab51052](#)) at 1/500 dilution

Lane 1 : Wild-type HeLa cell lysate

Lane 2 : HSPA8 knockout HeLa cell lysate

Lane 3 : A431 cell lysate

Lane 4 : MCF7 cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

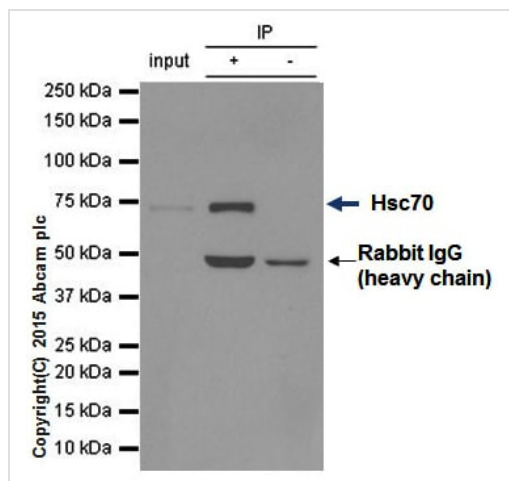
Predicted band size: 71 kDa

Observed band size: 71 kDa

This data was developed using the same antibody clone in a different buffer formulation ([ab51052](#)).

Lanes 1- 4: Merged signal (red and green). Green - [ab51052](#) observed at 71 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) observed at 37 kDa.

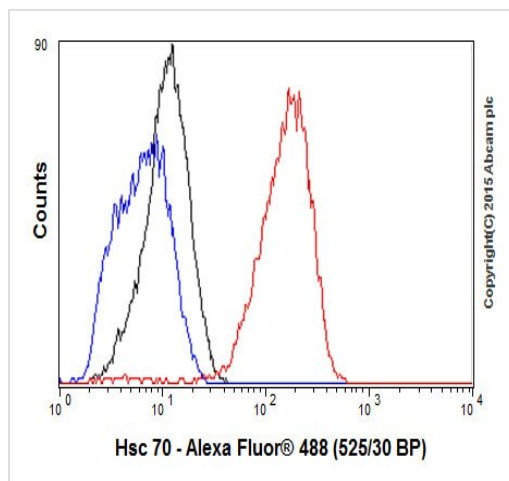
[ab51052](#) was shown to react with Hsc70 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line [ab265664](#) (knockout cell lysate [ab256944](#)) was used. Wild-type HeLa and HSPA8 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. [ab51052](#) and Anti-GAPDH antibody [6C5] - Loading Control ([ab8245](#)) overnight at 4°C at a 1 in 500 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed ([ab216776](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunoprecipitation - Anti-Hsc70 antibody
[EP1531Y] - BSA and Azide free (ab220821)

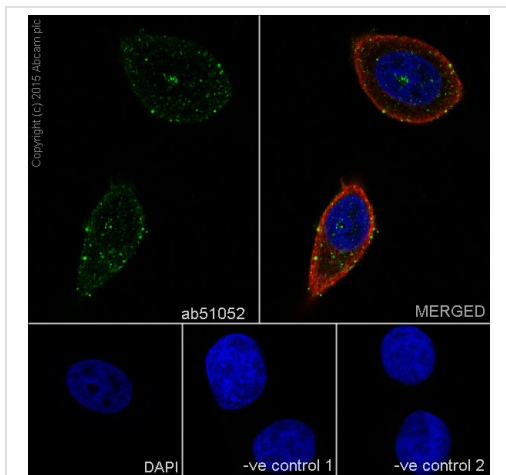
ab51052 (purified) at 1/20 immunoprecipitating Hsc70 in 10 µg HeLa (Lanes 1 and 2, observed at 71 kDa). Lane 3 - PBS. For western blotting, HRP Veriblot for IP (**ab131366**) was used for detection at 1/1000 dilution. Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).



Flow Cytometry (Intracellular) - Anti-Hsc70 antibody
[EP1531Y] - BSA and Azide free (ab220821)

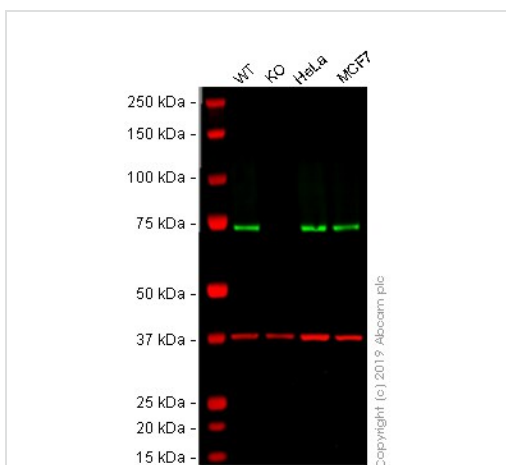
Overlay histogram showing MCF7 cells fixed in 4% PFA and stained with purified **ab51052** at a dilution of 1 in 70 (red line). The secondary antibody used was FITC goat anti-rabbit at a dilution of 1 in 500. Rabbit monoclonal IgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line). This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).



Immunocytochemistry/ Immunofluorescence - Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (ab220821)

Immunofluorescence staining of MCF7 cells with purified **ab51052** at a working dilution of 1/250, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti-rabbit (**ab150077**), used at a dilution of 1/1000. **ab7291**, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along with **ab150120** (Alexa Fluor® 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified **ab51052** was used at a dilution of 1/500 followed by an Alexa Fluor® 594 goat anti-mouse antibody (**ab150120**) at a dilution of 1/500. For negative control 2, **ab7291** (mouse anti-tubulin) was used at a dilution of 1/500 followed by an Alexa Fluor® 488 goat anti-rabbit antibody (**ab150077**) at a dilution of 1/400.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).



Western blot - Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (ab220821)

All lanes : Anti-Hsc70 antibody [EP1531Y] (**ab51052**) at 1/500 dilution

Lane 1 : Wild-type A431 whole cell lysate

Lane 2 : HSPA8 knockout A431 whole cell lysate

Lane 3 : HeLa whole cell lysate

Lane 4 : MCF7 whole cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 71 kDa

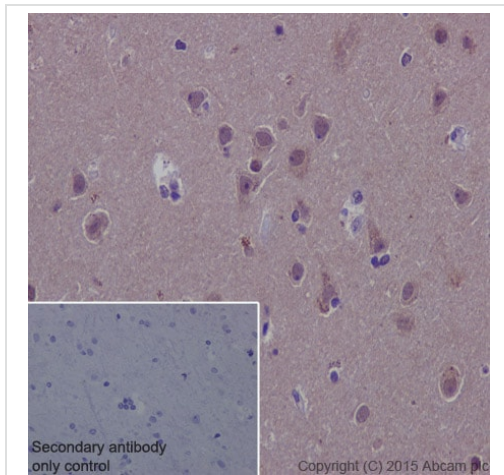
Observed band size: 73 kDa

This data was developed using the same antibody clone in a different buffer formulation (**ab51052**).

Lanes 1 - 4: Merged signal (red and green). Green - **ab51052** observed at 73 kDa. Red - loading control, **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab51052 was shown to react with HSPA8 in A431 wild-type cells in Western blot. Loss of signal was observed when HSPA8 knockout sample was used. A431 wild-type and HSPA8 knockout cell lysates

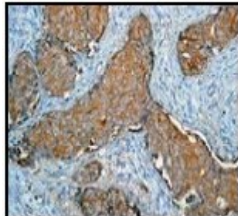
were subjected to SDS-PAGE. Membranes were blocked in 3% Milk in TBS-T (0.1% Tween®) before incubation with **ab51052** and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 500 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemical staining of paraffin embedded human cerebral cortex with purified **ab51052** at a working dilution of 1/500. The secondary antibody used is HRP goat anti-rabbit IgG H&L (**ab97051**) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).

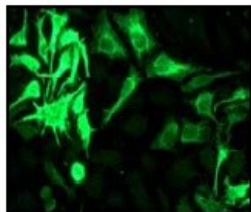
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (**ab220821**)



Unpurified **ab51052** (1/250) staining Hsc70 in paraffin embedded human breast carcinoma tissue by Immunohistochemistry.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (**ab220821**)



Immunofluorescent staining of HeLa cells using unpurified **ab51052** (1/100).

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab51052**).

Immunocytochemistry/ Immunofluorescence - Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (ab220821)

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-Hsc70 antibody [EP1531Y] - BSA and Azide free (ab220821)

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