abcam

Product datasheet

Anti-STK3/MST-2 antibody [EP1466Y] ab52641

KO VALIDATED Recombinant RabMAb

33 References 9 Images

Overview

Product name	Anti-STK3/MST-2 antibody [EP1466Y]
Description	Rabbit monoclonal [EP1466Y] to STK3/MST-2
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC/IF, WB, IP, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human STK3/MST-2 aa 1-100 (N terminal). The exact sequence is proprietary.
Positive control	ICC/IF: Wildtype HAP1 cells, NIH/3T3 (Mouse embryonic fibroblast) cells; WB: NIH/3T3, Hek293, HeLa, C6 cell lysate; IHC-P: Human lymphoma tissue; Flow Cyt (intra): NIH/3T3 (Mouse embryonic fibroblast).
General notes	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**.

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. Stable for 12 months at -20°C.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP1466Y

Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab52641 in the following tested applications.

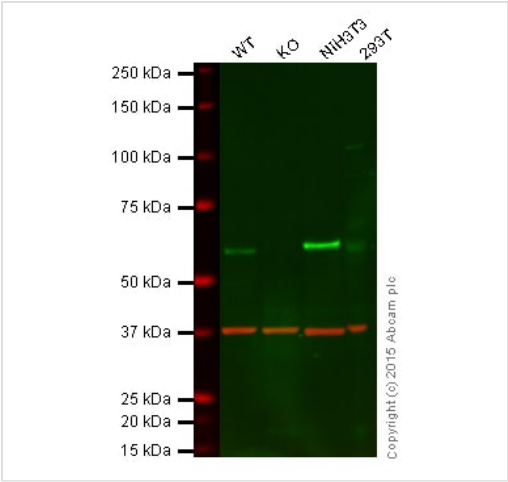
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/70.
ICC/IF		Use a concentration of 5 µg/ml.
WB		1/10000 - 1/50000. Detects a band of approximately 56 kDa (predicted molecular weight: 56 kDa).
IP		1/50 - 1/100.
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Antigen retrieval step strongly recommended for enhanced signal.

Target

Function	Stress-activated, pro-apoptotic kinase which, following caspase-cleavage, enters the nucleus and induces chromatin condensation followed by internucleosomal DNA fragmentation. Key component of the Hippo signaling pathway which plays a pivotal role in organ size control and tumor suppression by restricting proliferation and promoting apoptosis. The core of this pathway is composed of a kinase cascade wherein MST1/MST2, in complex with its regulatory protein SAV1, phosphorylates and activates LATS1/2 in complex with its regulatory protein MOB1, which in turn phosphorylates and inactivates YAP1 oncoprotein and WWTR1/TAZ. Phosphorylation of YAP1 by LATS2 inhibits its translocation into the nucleus to regulate cellular genes important for cell proliferation, cell death, and cell migration. MST1/MST2 are required to repress proliferation of mature hepatocytes, to prevent activation of facultative adult liver stem cells (oval cells), and to inhibit tumor formation. Phosphorylates NKX2-1.
Tissue specificity	Expressed at high levels in adult kidney, skeletal and placenta tissues and at very low levels in adult heart, lung and brain tissues.
Sequence similarities	Belongs to the protein kinase superfamily. STE Ser/Thr protein kinase family. STE20 subfamily. Contains 1 protein kinase domain. Contains 1 SARAH domain.
Cellular localization	Cytoplasm. Nucleus. The caspase-cleaved form cycles between nucleus and cytoplasm.

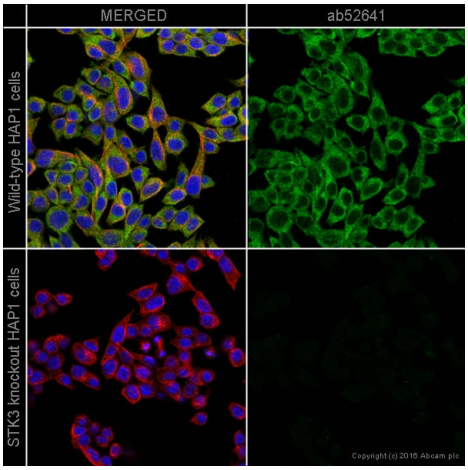
Images



Western blot - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

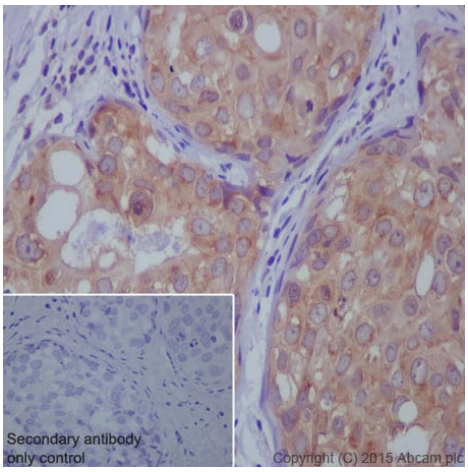
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: STK3/MST-2 knockout HAP1 cell lysate (20 µg)
Lane 3: NIH/3T3 cell lysate (20 µg)
Lane 4: 293T cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab52641 observed at 56 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab52641 was shown to specifically react with STK3/MST-2 when STK3/MST-2 knockout samples were used. Wild-type and STK3/MST-2 knockout samples were subjected to SDS-PAGE. ab52641 and **ab8245** (loading control to GAPDH) were diluted 1/10 000 and 1/2000 and incubated overnight at 4°C. 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 1/10 000 dilution for 1 h at room temperature before imaging.



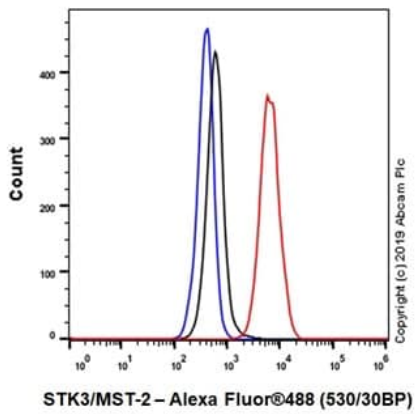
Immunocytochemistry/ Immunofluorescence - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

ab52641 staining STK3/MST-2 in wild-type HAP1 cells (top panel) and STK3/MST-2 knockout HAP1 cells (bottom panel). The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab52641 at 5µg/ml concentration and **ab195889** at 1/250 dilution (shown in pseudo colour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (**ab150081**) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.



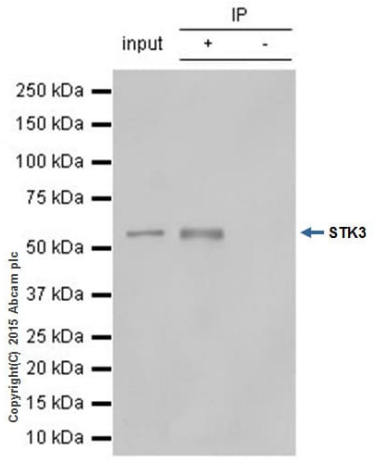
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

Immunohistochemical staining of paraffin embedded human breast carcinoma with purified ab52641 at a working dilution of 1/50. The secondary antibody used is **ab97051**, a goat anti-rabbit IgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed 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.



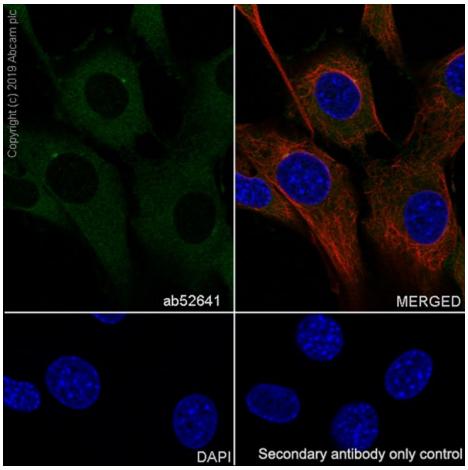
Flow Cytometry (Intracellular) - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

NIH/3T3 (Mouse embryonic fibroblast) cells were fixed with4% paraformaldehyde and permeabilised with 90% methanol. The primary antibody (ab52641) was used at a 1/70 dilution (1 µg) (red). A Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at a 1/2000 dilution. A Rabbit monoclonal IgG (**ab172730**) (black) was used as an isotype control.Cells without incubation with primary antibody and secondary antibody were used as an unlabelled control (blue).



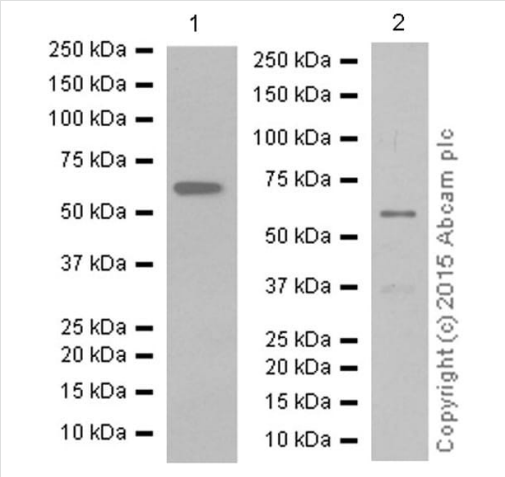
Immunoprecipitation - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

ab52641 (purified) at 1/50 immunoprecipitating STK3/MST-2 in 10 µg C6 cell lysate (Lanes 1 and 2, observed at 56 kDa). Lane 3 - Rabbit monoclonal IgG (**ab172730**). For western blotting, HRP Veriblot for IP (**ab131366**) was used for detection (1/10 000). Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

Confocal image showing cytoplasmic staining of STK3/MST-2 in NIH/3T3 (mouse embryonic fibroblast) cells using ab52641 . The cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. The cells were then incubated with ab52641 at 1/70 dilution and counterstained with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (**ab195889**) at 1/100 dilution (red). Goat anti Rabbit IgG (Alexa Fluor® 488) (**ab150077**) was used as the secondary antibody at 1/1000 dilution (green). Nuclei counterstained with DAPI (blue).



Western blot - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

All lanes : Anti-STK3/MST-2 antibody [EP1466Y] (ab52641) at 1/50000 dilution (purified)

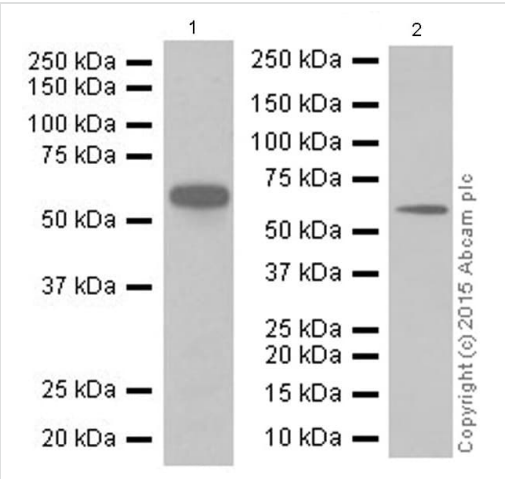
Lane 1 : C6 whole cell lysate
Lane 2 : NIH/3T3 whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/50000 dilution

Predicted band size: 56 kDa
Observed band size: 56 kDa

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST



Western blot - Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

All lanes : Anti-STK3/MST-2 antibody [EP1466Y] (ab52641) at 1/50000 dilution (purified)

Lane 1 : HEK293 whole cell lysate
Lane 2 : HeLa whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 56 kDa
Observed band size: 56 kDa

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST

Anti-STK3/MST-2 antibody [EP1466Y] (ab52641)

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