abcam

Product datasheet

Anti-MLK3 antibody [EP1460Y] ab51068

KO 評価済 リコンビナント RabMAb

13 References 画像数 5

製品の概要

製品名	Anti-MLK3 antibody [EP1460Y]
製品の詳細	Rabbit monoclonal [EP1460Y] to MLK3
由来種	Rabbit
アプリケーション	**適用あり:** WB, ICC/IF **適用なし:** Flow Cyt
種交差性	**交差種:** Human
免疫原	Synthetic peptide within Human MLK3 aa 800 to the C-terminus (C terminal). The exact sequence is proprietary. Database link: **Q16584**
ポジティブ・コントロール	WB: HepG2, A549, HAP1, A431 and HeLa cell lysates. ICC/IF: HeLa cells.
特記事項	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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
バッファー	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.01% BSA, 40% Glycerol (glycerin, glycerine), 59% PBS
精製度	Protein A purified

ポリ/モノ	モノクローナル
クローン名	EP1460Y
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ab51068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
WB		1/1000. Detects a band of approximately 100 kDa (predicted molecular weight: 93 kDa). **For unpurified use at 1/5000.**
ICC/IF		1/50.

追加情報 Is unsuitable for Flow Cyt.

ターゲット情報

機能 Activates the JUN N-terminal pathway. Required for serum-stimulated cell proliferation and for mitogen and cytokine activation of MAPK14 (p38), MAPK3 (ERK) and MAPK8 (JNK1). Plays a role in mitogen-stimulated phosphorylation and activation of BRAF, but does not phosphorylate BRAF directly. Influences microtubule organization during the cell cycle.

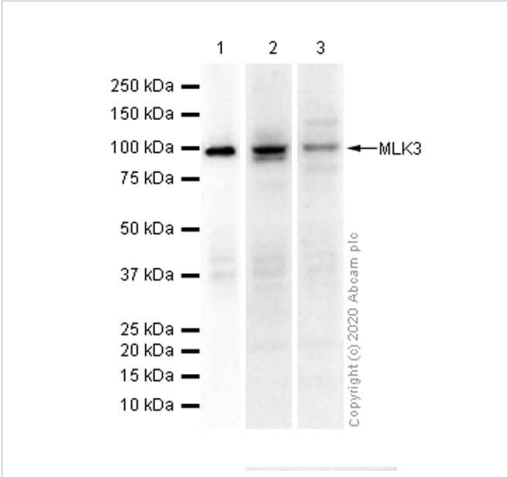
組織特異性 Expressed in a wide variety of normal and neoplastic tissues including fetal lung, liver, heart and kidney, and adult lung, liver, heart, kidney, placenta, skeletal muscle, pancreas and brain.

配列類似性 Belongs to the protein kinase superfamily. STE Ser/Thr protein kinase family. MAP kinase kinase kinase subfamily.
Contains 1 protein kinase domain.
Contains 1 SH3 domain.

翻訳後修飾 Autophosphorylation on serine and threonine residues within the activation loop plays a role in enzyme activation. Thr-277 is likely to be the main autophosphorylation site. Phosphorylation of Ser-555 and Ser-556 is induced by CDC42.

細胞内局在 Cytoplasm > cytoskeleton > centrosome. Location is cell cycle dependent.

画像



Western blot - Anti-MLK3 antibody [EP1460Y] (ab51068)

All lanes : Anti-MLK3 antibody [EP1460Y] (ab51068) at 1/1000 dilution (Purified)

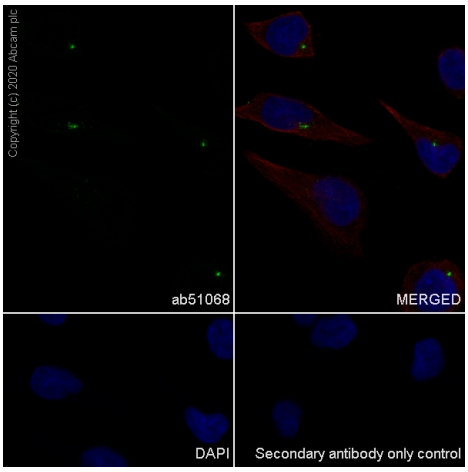
Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 2 : HepG2 (Human hepatocellular carcinoma epithelial cell) whole cell lysate
Lane 3 : A431 (Human epidermoid carcinoma epithelial cell) whole cell lysate

Lysates/proteins at 15 µg per lane.

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

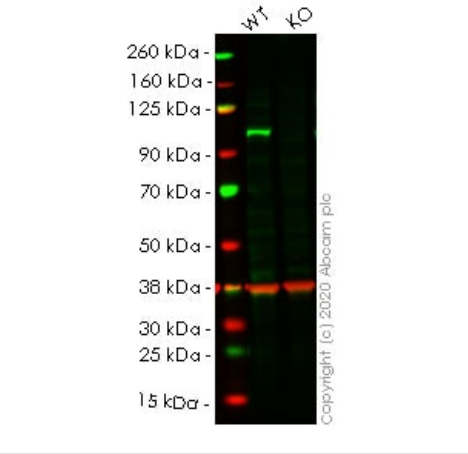
Predicted band size: 93 kDa
Observed band size: 100 kDa

Blocking buffer: 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-MLK3 antibody [EP1460Y] (ab51068)

Immunocytochemistry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling MLK3 with Purified ab51068 at 1:50 dilution (3.52 µg/ml). Cells were fixed in 100% Methanol and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488,**ab150077**) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Western blot - Anti-MLK3 antibody [EP1460Y] (ab51068)

All lanes : Anti-MLK3 antibody [EP1460Y] (ab51068) at 1/5000 dilution

Lane 1 : Wild-type A549 cell lysate
Lane 2 : MAP3K11 knockout A549 cell lysate

Lysates/proteins at 20 µg per lane.

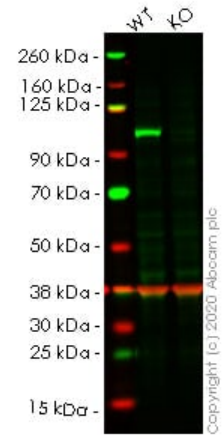
Performed under reducing conditions.

Predicted band size: 93 kDa
Observed band size: 105 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab51068 observed at 105 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab51068 was shown to react with MLK3 in wild-type A549 cells in western blot. Loss of signal was observed when knockout cell line **ab267169** (knockout cell lysate **ab257519**) was used. Wild-type A549 and MAP3K11 knockout A549 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. ab51068 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 5000 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.



Western blot - Anti-MLK3 antibody [EP1460Y] (ab51068)

All lanes : Anti-MLK3 antibody [EP1460Y] (ab51068) at 1/5000 dilution

Lane 1 : Wild-type A549 cell lysate
Lane 2 : MAP3K11 knockout A549 cell lysate

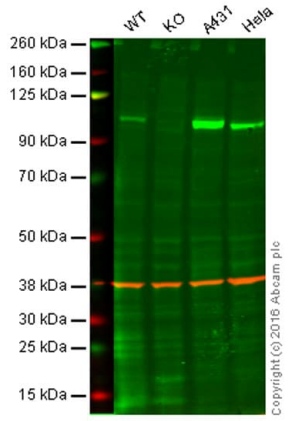
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 93 kDa
Observed band size: 105 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab51068 observed at 105 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab51068 was shown to react with MLK3 in wild-type A549 cells in western blot. Loss of signal was observed when knockout cell line **ab267168** (knockout cell lysate **ab257518**) was used. Wild-type A549 and MAP3K11 knockout A549 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. ab51068 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) overnight at 4°C at a 1 in 5000 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.



Western blot - Anti-MLK3 antibody [EP1460Y] (ab51068)

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: MLK3 knockout HAP1 cell lysate (20 µg)
Lane 3: A431 cell lysate (20 µg)
Lane 4: HeLa cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab51068 observed at 105 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab51068 was shown to specifically react with MLK3 when MLK3 knockout samples were used. Wild-type and MLK3 knockout samples were subjected to SDS-PAGE. ab51068 and **ab8245** (loading control to GAPDH) were diluted 1/5000 and 1/1000 respectively 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 1/10000 dilution for 1 hour at room temperature before imaging.

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