

### Anti-MLKL antibody [EPR17514] ab184718

KO 評価済 リコンビナント RabMAb

★★★★★ 5 Abreviews 64 References 画像数 8

#### 製品の概要

製品名	Anti-MLKL antibody [EPR17514]
製品の詳細	Rabbit monoclonal [EPR17514] to MLKL
由来種	Rabbit
アプリケーション	適用あり: ICC/IF, WB, IHC-P
種交差性	交差種: Human
免疫原	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	WB: HUVEC, HT-29 and HeLa whole cell lysates; Human fetal kidney lysate. IHC-P: Human tonsil and colonic adenocarcinoma tissues.
特記事項	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> <li>- High batch-to-batch consistency and reproducibility</li> <li>- Improved sensitivity and specificity</li> <li>- Long-term security of supply</li> <li>- Animal-free production</li> </ul> <p>For more information <a href="#">see here</a>.</p> <p>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb® patents</a>.</p>

#### 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	<p>pH: 7.2</p> <p>Preservative: 0.01% Sodium azide</p> <p>Constituents: 59% PBS, 40% Glycerol, 0.05% BSA</p>
精製度	Protein A purified
ポリ/モノ	モノクローナル
クローン名	EPR17514
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ab184718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
ICC/IF	★★★★★ (1)	1/200.
WB	★★★★★ (4)	1/1000. Detects a band of approximately 54 kDa (predicted molecular weight: 54 kDa).
IHC-P		1/400. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

## ターゲット情報

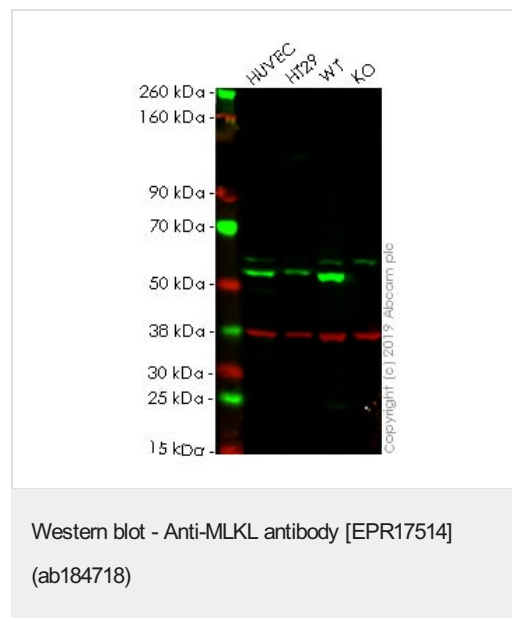
### 配列類似性

Belongs to the protein kinase superfamily.  
Contains 1 protein kinase domain.

### ドメイン

The protein kinase domain is predicted to be catalytically inactive.

## 画像



**All lanes :** Anti-MLKL antibody [EPR17514] (ab184718) at 1/1000 dilution

**Lane 1 :** HUVEC cell lysate

**Lane 2 :** HT-29 cell lysate

**Lane 3 :** Wild-type HeLa cell lysate

**Lane 4 :** MLKL knockout HeLa cell lysate

Lysates/proteins at 20 µg per lane.

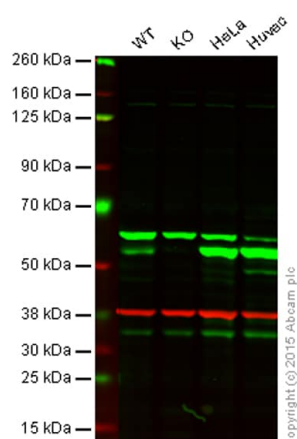
Performed under reducing conditions.

**Predicted band size:** 54 kDa

**Lanes 1 - 4:** Merged signal (red and green). Green - ab184718 observed at 54 kDa. Red - loading control, **ab8245** observed at 37 kDa.

ab184718 was shown to react with MLKL in wild-type HeLa cells.

Loss of signal was observed when knockout cell line **ab255408** (knockout cell lysate **ab263788**) was used. Wild-type and MLKL knockout samples were subjected to SDS-PAGE. ab184718 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) 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® 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-MLKL antibody [EPR17514] (ab184718)

**All lanes :** Anti-MLKL antibody [EPR17514] (ab184718) at 1/1000 dilution

**Lane 1 :** Wild-type HAP1 cell lysate

**Lane 2 :** MLKL knockout HAP1 cell lysate

**Lane 3 :** HeLa cell lysate

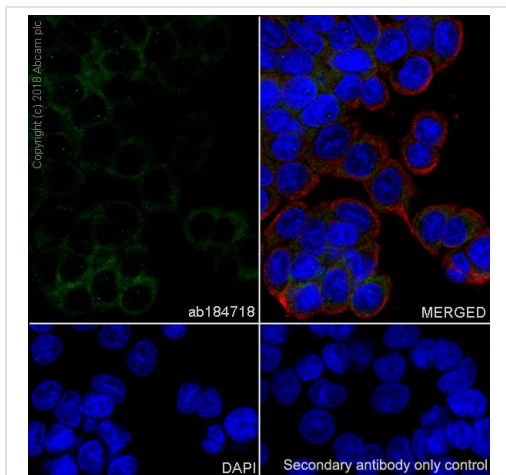
**Lane 4 :** Huvec cell lysate

Lysates/proteins at 20 µg per lane.

**Predicted band size:** 54 kDa

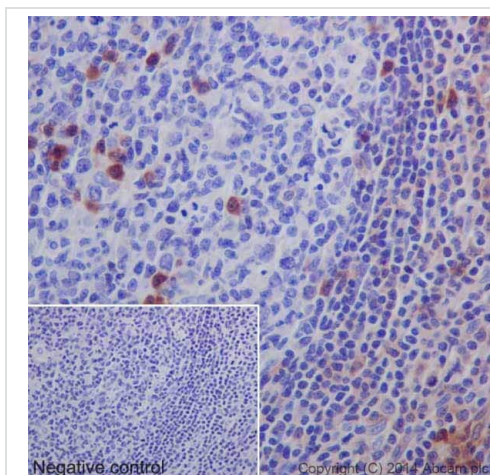
**Lanes 1 - 4:** Merged signal (red and green). Green - ab184718 observed at 55 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab184718 was shown to recognize MLKL in wild-type HAP1 cells along with additional cross-reactive bands. No band was observed when MLKL knockout samples were examined. Wild-type and MLKL knockout samples were subjected to SDS-PAGE. ab184718 and **ab8245** (loading control to GAPDH) were diluted to 1/1000 and 1/2000 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 1/10,000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-MLKL antibody [EPR17514] (ab184718)

Ab184718 staining MLKL in HT-29 (Human colorectal adenocarcinoma epithelial cell) cells by Immunocytochemistry (ICC). Cells were fixed with 100% Methanol. Samples were incubated with primary antibody at 1/200 dilution (6.5µg/ml). An AlexaFluor® 488 Goat anti-Rabbit (**ab150077**) was used as the secondary antibody at 1/1000 dilution (2µg/ml). Ab195889 , Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) was used as the counterstain antibody (1/200 dilution, 2.5 µg/ml) . DAPI was used as a nuclear counterstain. Confocal image showing cytoplasmic staining on HT-29 cell line.

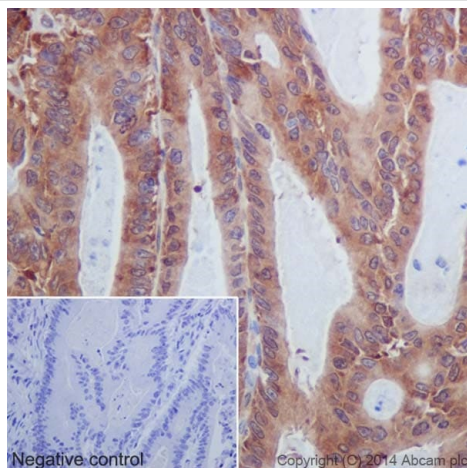


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MLKL antibody [EPR17514] (ab184718)

Immunohistochemical analysis of paraffin-embedded Human tonsil tissue labeling MLKL with ab184718 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) secondary antibody (**ab97051**) at 1/500 dilution. Cytoplasmic staining on the lymphocytes of human tonsil is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

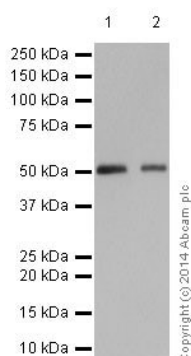


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MLKL antibody [EPR17514] (ab184718)

Immunohistochemical analysis of paraffin-embedded Human colonic adenocarcinoma tissue labeling MLKL with ab184718 at 1/400 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) secondary antibody (**ab97051**) at 1/500 dilution. Cytoplasmic staining on tumor cells of human colonic adenocarcinoma is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-MLKL antibody [EPR17514] (ab184718)

**All lanes** : Anti-MLKL antibody [EPR17514] (ab184718) at 1/20000 dilution

**Lane 1** : HUVEC (Human umbilical vein endothelial cell line) whole cell lysate

**Lane 2** : HT-29 (Human colorectal adenocarcinoma cells) whole cell lysate

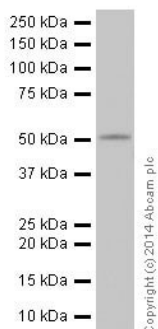
Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes** : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

**Predicted band size:** 54 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-MLKL antibody [EPR17514]  
(ab184718)

Anti-MLKL antibody [EPR17514] (ab184718) at 1/1000 dilution +  
Human fetal kidney lysate at 10 µg

#### Secondary

Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at  
1/1000 dilution

**Predicted band size:** 54 kDa

**Observed band size:** 54 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

#### 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-MLKL antibody [EPR17514] (ab184718)

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