Anti-TMS1 Antibody [SN07-10]

ET1611-62

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 22 kDa
Clone number:	SN07-10

Description: The death domain (DD) superfamily of proteins share one or more of the following domains: the DD, DED (death-effector domain), CARD (caspase-recruitment domain) and PYD (Pyrin domain). Each of these domains is characterized by a canonical death domain fold, which consists of a bundle of five or six antiparallel α-helices. As their names suggest, these domains play prominent roles in programmed cell death. Caspase-associated recruitment domains (CARDs) mediate the interaction between adaptor proteins such as Apaf-1 and the proform of caspases (e.g., CASP9) participating in apoptosis. ASC (apoptosis-associated speck-like protein containing a CARD, also known as TMS1or PYCARD) is a member of the CARD-containing adaptor protein family. ASC is a 195 amino acid protein, containing a N-terminal Pyrin-like domain (PYD) and an 87 residue C-terminal CARD. This motif is characteristic of numerous proteins involved in apoptotic signaling. ASC2 (apoptosis-associated speck-like protein containing a CARD 2), also known as Pyrin-only protein 1 or PADD-only protein 1, is an 89 amino acid member of the DD superfamily that contains one Pyrin domain. Localized to the cytoplasm, ASC2 interacts with ASC to modulate NF-κB and pro-caspase-1 regulation.

Immunogen:	Synthetic peptide within Human TMS1 aa 121-170 / 195.
Positive control:	THP-1 cell lysates, THP-1, human skin tissue, human lymph nodes tissue, human spleen tissue.
Subcellular location:	Cytoplasm, Endoplasmic reticulum, Golgi apparatus, Inflammasome, Membrane, Mitochondrion, Nucleus.
Database links:	SwissProt: Q9ULZ3 Human
Recommended Dilutions:	
WB	1:500-1:1,000
IF-Cell	1:100
IF-Tissue	1:100-1:500
IHC-P	1:50-1:400
FC	1:1,000
Storage Buffer:	1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
Purity:	Protein A affinity purified.

Images

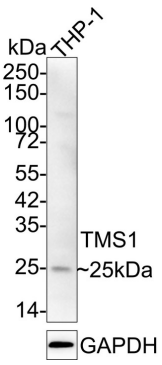

Fig1: Western blot analysis of TMS1 on THP-1 cell lysates with Rabbit anti-TMS1 antibody (ET1611-62) at 1/2,000 dilution.

Lysates/proteins at 10 µg/Lane.

Predicted band size: 22 kDa
Observed band size: 25 kDa

Exposure time: 3 minutes; ECL: K1802;
4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1611-62) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

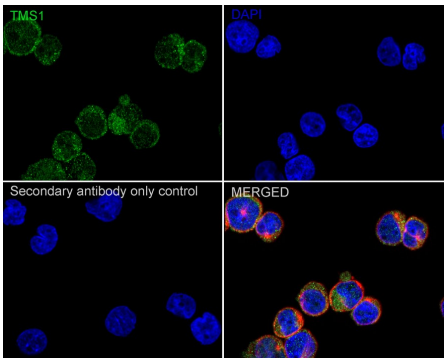

Fig2: Immunocytochemistry analysis of THP-1 cells labeling TMS1 with Rabbit anti-TMS1 antibody (ET1611-62) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-TMS1 antibody (ET1611-62) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

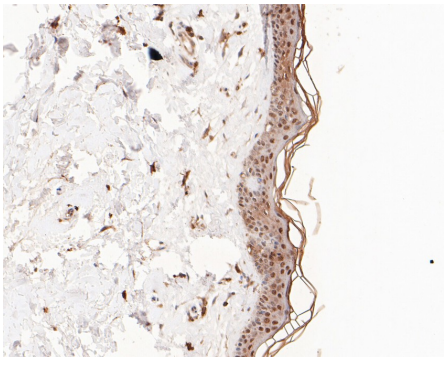

Fig3: Immunohistochemical analysis of paraffin-embedded human skin tissue using anti-TMS1 antibody. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1611-62, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

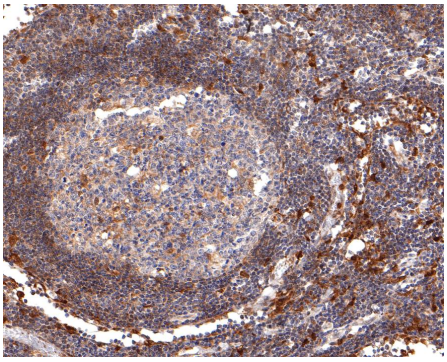

Fig4: Immunohistochemical analysis of paraffin-embedded human lymph nodes tissue with Rabbit anti-TMS1 antibody (ET1611-62) at 1/400 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1611-62) at 1/400 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

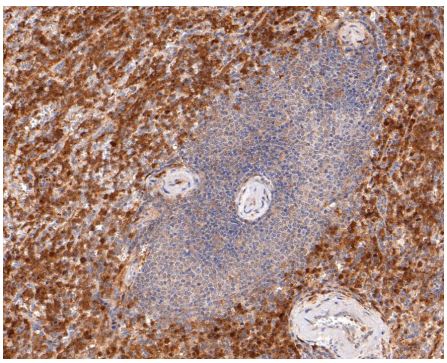

Fig5: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-TMS1 antibody (ET1611-62) at 1/400 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1611-62) at 1/400 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

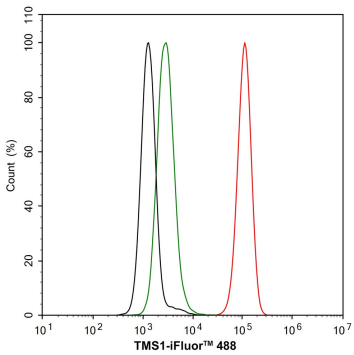

Fig6: Flow cytometric analysis of THP-1 cells labeling TMS1.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1611-62, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Background References

1. Ataide, MA. et al. Malaria-induced NLRP12/NLRP3-dependent caspase-1 activation mediates inflammation and hypersensitivity to bacterial superinfection. PLoS pathogens 10: e1003885 (2014).
2. Liu, D. et al. Activation of the Nlrp3 inflammasome by mitochondrial reactive oxygen species: a novel mechanism of albumin-induced tubulointerstitial inflammation. The international journal of biochemistry & cell biology 57: 7-19 (2014).