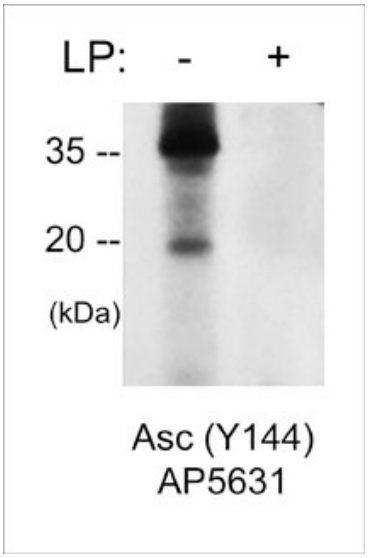
Asc (Tyr-144), phospho-specific

Rabbit Polyclonal

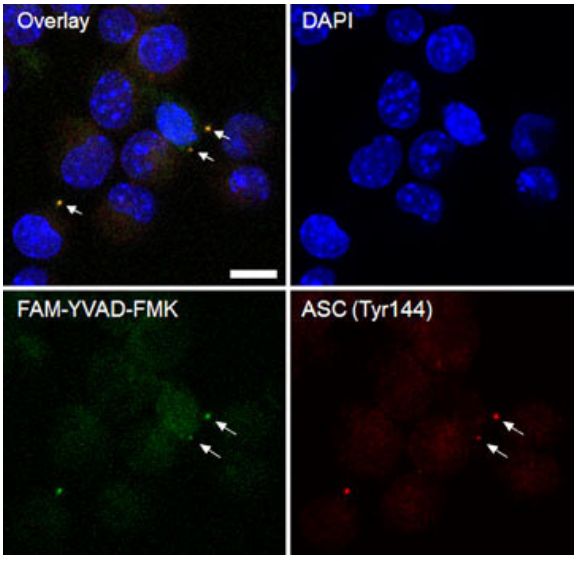
Cat. # AP5631
Size 100 µl

Background

Host- and pathogen-associated cytoplasmic double-stranded DNA triggers the activation of a NALP3-independent inflammasome, which activates caspase-1, leading to maturation of pro-interleukin-1beta and inflammation. Several studies have isolated AIM2 (absent in melanoma 2) as a candidate cytoplasmic-DNA-sensing protein that contains an N-terminal pyrin domain and C-terminal oligonucleotide binding domain. A screen for transcripts induced by interferon-beta identified AIM2 gene expression. AIM2 protein bound double-stranded DNA, recruited the inflammasome adaptor ASC, and localized to ASC containing speckles. AIM2 and ASC form a pyroptosome, which induces pyroptotic cell death mediated by caspase-1. Asc can be phosphorylated at Tyr-144 in a Syk and JNK-dependent manner. This phosphorylation is critical for Asc speck formation and Caspase-1 activation.



Western blot analysis of mouse macrophage J774A.1 cells stimulated with pervanadate (1 mM for 30 min.), then untreated (-) or treated (+) with alkaline phosphatase. The blot was probed with rabbit polyclonal anti-Asc (Tyr-144) phospho-specific antibody (AP5631) at 1:500.



Immunocytochemical labeling of Asc (Tyr-144) in inflammasomes. Paraformaldehyde fixed J774 cells were primed with LPS and treated with nigericin. Cells were co-labeled with DAPI, a caspase-1 inhibitor (FAM-YVAD-FMK), and anti-Asc (Tyr-144) phosphospecific antibody detected with AlexaFluor 568 secondary. (Image provided by Jordan Yaron, Center for Biosignatures Discovery Automation, Arizona State University)

Background References

Roberts, T.L. et al. (2009). Science. 323(5917):1057.
Bürckstümmer, T. et al. (2009). Nat Immunol. 10(3):266.
Hara, H. et al. (2013) Nature Immunol. 14(12):1247.

Product Citations

Yu, SH et al. (2019) J Ethnopharmacol. 239:111917.
WB: mouse bone marrow macrophages

Dubois, EC et al. (2019) PLoS Pathog. 15(4):e1007709.
WB: mouse macrophage

Furuya, MY et al. (2018) Arthritis Res Ther. 20(1):196.
WB: human neutrophils

Kwak, SB et al. (2018) Mediators Inflamm. 2018:6054069.
WB: mouse J774A.1, BMDMs

Hoyt, L.R. et al. (2016) J Immunol. 197:1322.
WB: mouse macrophage, LPS

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Rev 9/19/2019

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Rabbit Polyclonal

Cat. # AP5631
Size 100 µl

Immunogen

Uniprot ID: Q9EPB4

Asc (Tyr-144) phospho-peptide (coupled to KLH) corresponding to amino acid residues surrounding Tyr-144 in mouse Asc. This peptide sequence is highly conserved in human and rat Asc.

Buffer and Storage

Rabbit polyclonal, affinity-purified antibody is supplied in 100 µl phosphate-buffered saline, 50% glycerol, 1 mg/ml BSA, and 0.05% sodium azide. Store at –20°C. Stable for 1 year.

Applications

WB	1:1000
ELISA	1:1000
ICC	1:100

Species Reactivity

Hu, Rt, Ms

End user should determine optimal dilution for their particular applications and experiments.
Western blot membranes were incubated with diluted antibody in 5% non-fat milk, Tris buffer, 0.04% Tween20 for 1 hour at room temperature.
Abbreviations: E = ELISA, ICC = immunocytochemistry, IHC = immunohistochemistry, IP = immunoprecipitation, MS = mass spectrometry, WB = western blot
Hu = Human, Ms = Mouse, Rt = Rat, Ck = Chicken, F = Frog, B = Bovine

Specificity

This antibody was cross-adsorbed to unphosphorylated Asc (Tyr-144) peptide before affinity purification using phospho-Asc (Tyr-144) peptide (without carrier). The antibody detects phosphorylated Asc (Tyr-144) in the inflammasome. In J774 macrophage cells primed with LPS and treated with nigericin, the antibody colocalized with an inflammasome marker, caspase-1 inhibitor (FAM-YVAD-FMK). In addition, the antibody detects a 20 kDa Asc protein in western blots of J774A.1 mouse macrophages treated with pervanadate.

*All molecular weights (MW) are confirmed by comparison to MW standards and to western blot mobilities of known proteins with similar MW.
"Native" western blot utilizes non-reducing sample buffer (no mercaptoethanol or SDS), normal SDS-PAGE gel electrophoresis, and no methanol in transfer buffers.

Related Products

AP3851 AIM2 (N-terminal region) Rabbit Polyclonal

CM4891 Caspase-1 (C-terminal region) Mouse Monoclonal

CM4911 Caspase-3 (p17 subunit) Mouse Monoclonal

CM3771 Caspase-3 (N-terminal region) Mouse Monoclonal

CK6360 Caspase Family Antibody Sampler Kit

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