abcam

Product datasheet

Anti-S100A8 antibody [EPR3554] ab92331

Recombinant RabMAb

★★★★ 7 Abreviews 43 References 10 Images

Overview

Product name Anti-S100A8 antibody [EPR3554]

Description Rabbit monoclonal [EPR3554] to S100A8

Host species Rabbit

Tested applications Suitable for: WB, IP, IHC-P

Unsuitable for: Flow Cyt or ICC/IF

Species reactivity Reacts with: Mouse, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Human (adult and fetal) and mouse spleen tissue lysates and PMBC and HL-60 cell lysates.

IHC-P: Human cervical carcinoma, breast carcinoma, tonsil and spleen tissues. IP: HL-60 cell

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.

Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number **EPR3554**

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab92331 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
WB	★★★★ ★ (4)	1/1000. Predicted molecular weight: 11 kDa. For unpurified use at 1/2000 - 1/10000.
IP		1/30. For unpurified use at 1/20.
IHC-P	**** (1)	1/500 - 1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/100 - 1/250.

Application notes

Is unsuitable for Flow Cyt or ICC/IF.

Target

Function

S100A8 is a calcium- and zinc-binding protein which plays a prominent role in the regulation of inflammatory processes and immune response. It can induce neutrophil chemotaxis and adhesion. Predominantly found as calprotectin (S100A8/A9) which has a wide plethora of intraand extracellular functions. The intracellular functions include: facilitating leukocyte arachidonic acid trafficking and metabolism, modulation of the tubulin-dependent cytoskeleton during migration of phagocytes and activation of the neutrophilic NADPH-oxidase. Activates NADPHoxidase by facilitating the enzyme complex assembly at the cell membrane, transferring arachidonic acid, an essential cofactor, to the enzyme complex and S100A8 contributes to the enzyme assembly by directly binding to NCF2/P67PHOX. The extracellular functions involve proinfammatory, antimicrobial, oxidant-scavenging and apoptosis-inducing activities. Its proinflammatory activity includes recruitment of leukocytes, promotion of cytokine and chemokine production, and regulation of leukocyte adhesion and migration. Acts as an alarmin or a danger associated molecular pattern (DAMP) molecule and stimulates innate immune cells via binding to pattern recognition receptors such as Toll-like receptor 4 (TLR4) and receptor for advanced glycation endproducts (AGER). Binding to TLR4 and AGER activates the MAP-kinase and NFkappa-B signaling pathways resulting in the amplification of the proinflammatory cascade. Has antimicrobial activity towards bacteria and fungi and exerts its antimicrobial activity probably via chelation of Zn(2+) which is essential for microbial growth. Can induce cell death via autophagy and apoptosis and this occurs through the cross-talk of mitochondria and lysosomes via reactive oxygen species (ROS) and the process involves BNIP3. Can regulate neutrophil number and apoptosis by an anti-apoptotic effect; regulates cell survival via ITGAM/ITGB and TLR4 and a signaling mechanism involving MEK-ERK. Its role as an oxidant scavenger has a protective role in preventing exaggerated tissue damage by scavenging oxidants. Can act as a potent amplifier of inflammation in autoimmunity as well as in cancer development and tumor spread. The iNOS-S100A8/A9 transnitrosylase complex directs selective inflammatory stimulus-dependent S-

nitrosylation of GAPDH and probably multiple targets such as ANXA5, EZR, MSN and VIM by recognizing a [IL]-x-C-x-x-[DE] motif; S100A8 seems to contribute to S-nitrosylation site selectivity.

Tissue specificity

Calprotectin (S100A8/9) is predominantly expressed in myeloid cells. Except for inflammatory conditions, the expression is restricted to a specific stage of myeloid differentiation since both proteins are expressed in circulating neutrophils and monocytes but are absent in normal tissue macrophages and lymphocytes. Under chronic inflammatory conditions, such as psoriasis and malignant disorders, also expressed in the epidermis. Found in high concentrations at local sites of inflammation or in the serum of patients with inflammatory diseases such as rheumatoid, cystic fibrosis, inflammatory bowel disease, Crohn's disease, giant cell arteritis, cystic fibrosis, Sjogren's syndrome, systemic lupus erythematosus, and progressive systemic sclerosis. Involved in the formation and deposition of amyloids in the aging prostate known as corpora amylacea inclusions. Strongly up-regulated in many tumors, including gastric, esophageal, colon, pancreatic, bladder, ovarian, thyroid, breast and skin cancers.

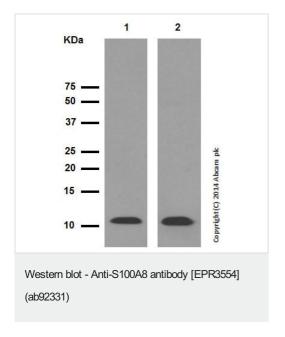
Sequence similarities

Belongs to the S-100 family. Contains 2 EF-hand domains.

Cellular localization

Secreted. Cytoplasm. Cytoplasm, cytoskeleton. Cell membrane. Predominantly localized in the cytoplasm. Upon elevation of the intracellular calcium level, translocated from the cytoplasm to the cytoskeleton and the cell membrane. Upon neutrophil activation or endothelial adhesion of monocytes, is secreted via a microtubule-mediated, alternative pathway.

Images



All lanes : Anti-S100A8 antibody [EPR3554] (ab92331) at 1/20000 dilution (purified)

Lane 1: HL-60 (Human promyelocytic leukemia cell line) cell lysate

Lane 2: Human fetal spleen tissue lysate

Lysates/proteins at 20 µg per lane.

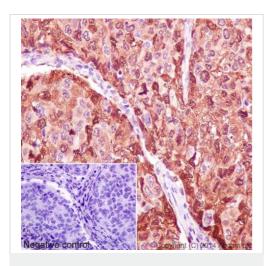
Secondary

All lanes : Peroxidase-conjugated goat anti-rabbit lgG (H+L) at

1/1000 dilution

Predicted band size: 11 kDa **Observed band size:** 11 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

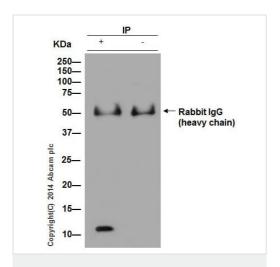


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 antibody
[EPR3554] (ab92331)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervix carcinoma tissue labeling S100A8 with purified ab92331 at 1/500.

Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0. <u>ab97051</u>, an HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500).

Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

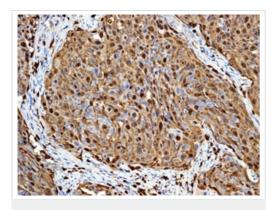


Immunoprecipitation - Anti-S100A8 antibody [EPR3554] (ab92331)

ab92331 (purified) at 1/30 immunoprecipitating S100A8 in HL-60 (Human promyelocytic leukemia cell line) cell lysate (**Lane 1**).

Lane 2 - rabbit monoclonal lgG instead of ab92331 in HL-60 lysates. For western blotting, a peroxidase-conjugated goat antirabbit lgG (H+L) was used as the secondary antibody (1/1500).

Blocking/Dilution buffer: 5% NFDM/TBST.

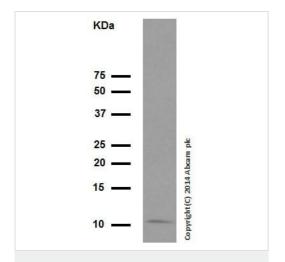


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 antibody
[EPR3554] (ab92331)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cervical carcinoma tissue labeling S100A8 with unpurified ab92331.

Detection: DAB staining.

Heat mediated antigen retrieval was performed via the pressure cooker method before commencing with IHC staining protocol.



Western blot - Anti-S100A8 antibody [EPR3554] (ab92331)

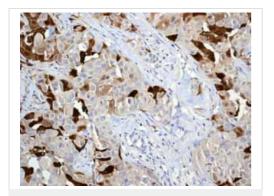
Anti-S100A8 antibody [EPR3554] (ab92331) at 1/2000 dilution (purified) + Mouse spleen tissue lysate at 20 µg

Secondary

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

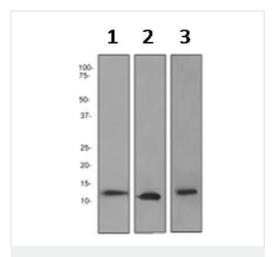
Predicted band size: 11 kDa **Observed band size:** 11 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

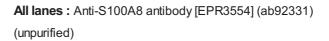


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 antibody
[EPR3554] (ab92331)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human breast carcinoma tissue labeling S100A8 with unpurified ab92331. Heat mediated antigen retrieval was performed via the pressure cooker method before commencing with IHC staining protocol.



Western blot - Anti-S100A8 antibody [EPR3554] (ab92331)



Lane 1: Human spleen tissue lysate

Lane 2: PBMC cell lysate

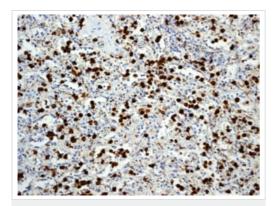
Lane 3: HL-60 (Human promyelocytic leukemia cell line) cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP-conjugated goat anti-rabbit lgG at 1/2000 dilution

Predicted band size: 11 kDa

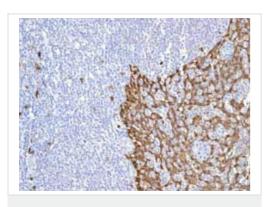


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 antibody
[EPR3554] (ab92331)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human spleen tissue labeling S100A8 with unpurified ab92331.

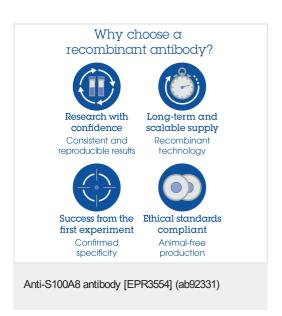
Detection: DAB staining.

Heat mediated antigen retrieval was performed via the pressure cooker method before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 antibody
[EPR3554] (ab92331)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labeling S100A8 with unpurified ab92331. Heat mediated antigen retrieval was performed via the pressure cooker method before commencing with IHC staining protocol.



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