abcam

Product datasheet

Anti-S100A8 + S100A9 antibody [RM1038] ab288715



重组 RabMAb

15 图像 1 References

概述

产品名称 Anti-S100A8 + S100A9抗体[RM1038]

描述 兔重组multiclonal [RM1038] to S100A8 + S100A9

宿主 Rabbit

经测试应用 适用于: Flow Cyt (Intra), IHC-Fr, IHC-P, ICC/IF, WB, IP

种属反应性 与反应: Mouse, Rat, Human

免疫原 This product was produced with the following immunogens:

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

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

Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Mouse spleen, Rat spleen, SK-BR-3, HL-60, Mouse PBMC and Mouse spleen lysates. IHC-

> P: Human spleen, Human stomach carcin, Mouse spleen and Rat spleen tissues. ICC: Mouse PBMC and HL-60 cells. Flow Cyt (intra): HL-60 and Mouse blood cells. IP: HL-60 and Mouse

spleen 10 cells. IHC-Fr: mouse spleen, rat spleen

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit 常规说明

monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

Recombinant multiclonals are a mixture of recombinant antibodies co-expressed from a library of

heavy and light chains.

Recombinant multiclonal antibodies offer the sensitivity of polyclonal antibodies by recognising

multiple epitopes, along with consistency of a recombinant antibody.

性能

形式

存放说明 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.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯**度** Protein A purified

克隆 Recombinant Multiclonal

克隆编号 RM1038

同种型 IgG

应用

The Abpromise guarantee Abpromi

Abpromise™承诺保证使用ab288715于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/500.
IHC-Fr		1/100.
IHC-P		1/5000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		1/500.
WB		1/1000. Predicted molecular weight: 13 kDa.
IP		1/30.

靶标

功能

S100A9 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, adhesion, can increase the bactericidal activity of neutrophils by promoting phagocytosis via activation of SYK, PI3K/AKT, and ERK1/2 and can induce degranulation of neutrophils by a MAPK-dependent mechanism. 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, transfering 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.

组织特异性

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.

序列相似性

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

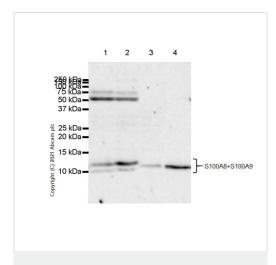
翻译后修饰

Phosphorylated. Phosphorylation inhibits activation of tubulin polymerization.

细胞定位

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.

图片



Western blot - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715) **All lanes**: Anti-S100A8 + S100A9 antibody [RM1038] (ab288715) at 1/1000 dilution

Lane 1: Mouse spleen lysate

Lane 2: Rat spleen lysate

Lane 3: SK-BR-3 (Human breast adenocarcinoma epithelial cell)

whole cell lysate

Lane 4: HL-60 (Human acute promyelocytic leukemia

promyeloblast) whole cell lysate

Secondary

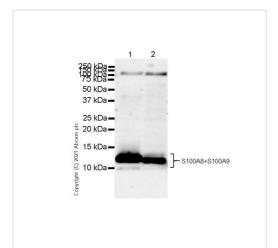
All lanes: Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated

(ab97051) at 1/100000 dilution

Predicted band size: 13 kDa

Observed band size: 11,14 kDa

Exposure time: 3 min



Western blot - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715) **All lanes :** Anti-S100A8 + S100A9 antibody [RM1038] (ab288715) at 1/1000 dilution

Lane 1 : Mouse PBMC lysate
Lane 2 : Mouse spleen lysate

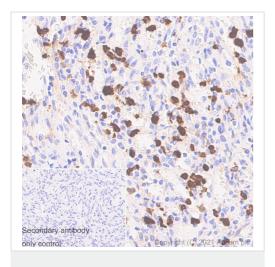
Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 13 kDa **Observed band size:** 11,14 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST

Exposure time: 3 min

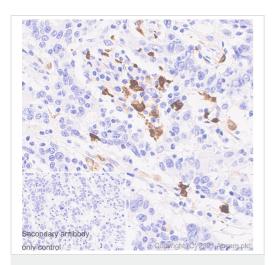


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling S100A8+S100A9 with ab288715 at 1/5000 (0.096 ug/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Positive staining on the human spleen. The section was incubated with ab288715 for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND™ RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond® Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

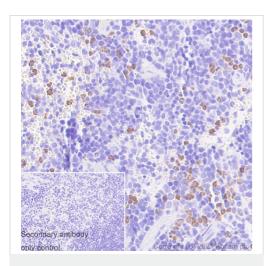


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Immunohistochemical analysis of paraffin-embedded Human stomach carcin tissue labeling S100A8+S100A9 with ab288715 at 1/5000 (0.096 ug/ml) followed by a ready to use LeicaDS9800 (Bond® Polymer Refine Detection). Positive staining on the stroma inflammatory cells in human stomach carcinoma. The section was incubated with ab288715 for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND™ RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond® Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

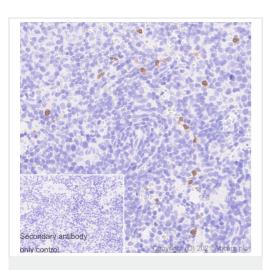


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Immunohistochemical analysis of paraffin-embedded Mouse spleen tissue labeling S100A8+S100A9 with ab288715 at 1/5000 (0.096 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on the mouse spleen. The section was incubated with ab288715 for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

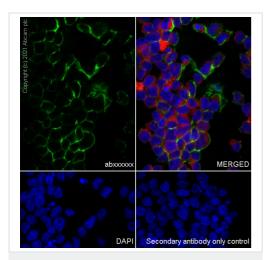


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Immunohistochemical analysis of paraffin-embedded Rat spleen tissue labeling S100A8+S100A9 with ab288715 at 1/5000 (0.096 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on the rat spleen. The section was incubated with ab288715 for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins

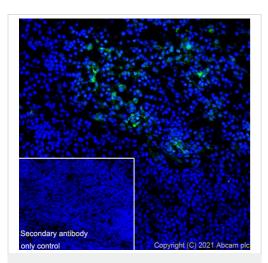


Immunocytochemistry/ Immunofluorescence - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HL-60 cells labelling S100A8+S100A9 with ab288715 at 1/500 (0.96 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing membranous staining in HL-60 cell line is observed.

ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

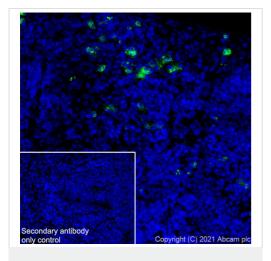
Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



Immunohistochemistry (Frozen sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Positive staining on the red pulp of mouse spleen.

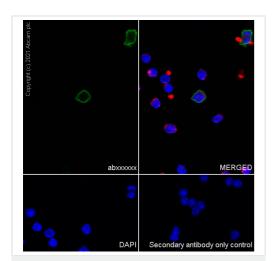
Fresh mouse spleen was fixed with 4% PFA and parmeabilised with 0.2 % Triton X100. ab288715 was used as a primary antibody at 1/100 dilution. **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) was preadsorbed and used as a sacondary antiobdy at 1/1000 dilution. DAPI was used as a nuclear counter stain.



Immunohistochemistry (Frozen sections) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Positive staining on the red pulp of rat spleen.

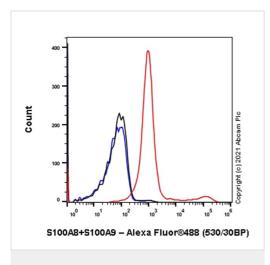
Fresh rat spleen was fixed with 4% PFA and parmeabilised with 0.2 % Triton X100. ab288715 was used as a primary antibody at 1/100 dilution. ab150081 Goat Anti-Rabbit lgG H&L (Alexa Fluor[®] 488) was preadsorbed and used as a sacondary antiobdy at 1/1000 dilution. DAPI was used as a nuclear counter stain.



Immunocytochemistry/ Immunofluorescence - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

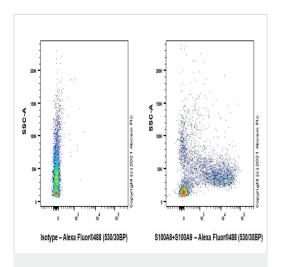
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized Mouse PBMC cells labelling S100A8+S100A9 with ab288715 at 1/500 (0.96 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing membranous staining in subsets of mouse PBMCs is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 (2 ug/ml) dilution.

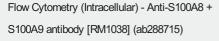


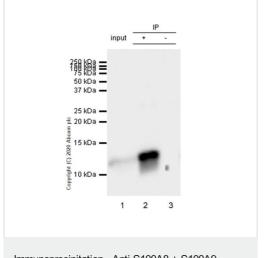
Flow Cytometry (Intracellular) - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

Flow cytometric analysis of 4% paraformaldehyde fixed 0.1% Tween-20 permeabilized HL-60 (Human acute promyelocytic leukemia promyeloblast) cells labelling S100A8+S100A9 with ab288715 at 1/500 dilution (0.1ug) (Red) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Flow cytometric analysis of 4% paraformaldehyde fixed 0.1% Tween-20 permeabilized Mouse blood cells cells labelling S100A8+S100A9 with ab288715 at 1/500 dilution (0.1ug)/ Right compared with a Rabbit monoclonal IgG (ab172730) / Left isotype control. A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.





Immunoprecipitation - Anti-S100A8 + S100A9 antibody [RM1038] (ab288715)

S100A8+S100A9 was immunoprecipitated from 0.35 mg HL-60 (Human acute promyelocytic leukemia promyeloblast) whole cell lysate 10ug with ab288715 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab288715 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

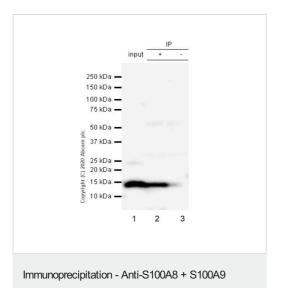
Lane 1: HL-60 (Human acute promyelocytic leukemia promyeloblast) whole cell lysate 10ug

Lane 2: ab288715 IP in HL-60 whole cell lysate

Lane 3:Rabbit monoclonal IgG ($\underline{ab172730}$) instead of ab288715 in HL-60 whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds



antibody [RM1038] (ab288715)

S100A8+S100A9 was immunoprecipitated from 0.35 mg Mouse spleen lysate 10ug with ab288715 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab288715 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution.

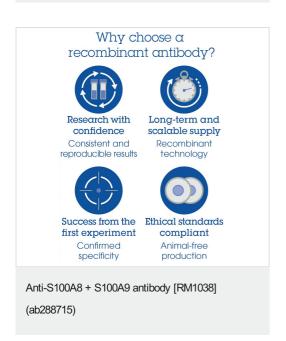
Lane 1: Mouse spleen lysate 10ug

Lane 2: ab288715 IP in Mouse spleen lysate

Lane 3: Rabbit monoclonal $\lg G$ ($\underline{ab172730}$) instead of ab288715 in Mouse spleen lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 10 seconds



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