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# S100A9 Polyclonal Antibody

## **Product Details**

Size	100 µg
Species Reactivity	Human, Mouse, Rat
Host/Isotype	Rabbit / IgG
Class	Polyclonal
Туре	Antibody
Conjugate	Unconjugated
Immunogen	E. coli-derived human S100A9 recombinant protein (Position: T2-P114).
Form	Lyophilized
Concentration	500 μg/mL
Purification	Antigen affinity chromatography
Storage buffer	PBS with 5mg BSA
Contains	0.05mg sodium azide
Storage conditions	-20°C
RRID	AB_2747064

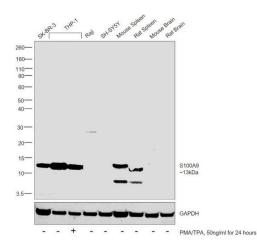
Applications	Tested Dilution	Publications
Western Blot (WB)	0.1-0.5 μg/mL	-
Immunohistochemistry (Paraffin) (IHC (P))	0.5-1 μg/mL	-
Immunocytochemistry (ICC/IF)	5 μg/mL	-
Flow Cytometry (Flow)	1-3 µg/1x10^6 cells	-
ELISA (ELISA)	0.1-0.5 μg/mL	-

## **Product Specific Information**

Reconstitute with 0.2 mL of distilled water to yield a concentration of 500  $\mu\text{g/mL}.$ 

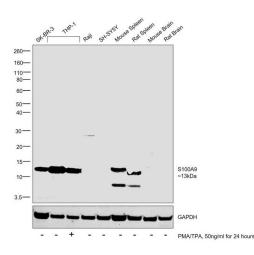
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## Product Images For S100A9 Polyclonal Antibody



#### S100A9 Antibody (PA5-79949)

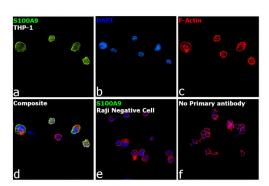
Antibody specificity was demonstrated by detection of differential basal expression of the target across cell lysates tested owing to their inherent genetic constitution. Expression of Calprotectin (S100A9) was observed in SK-BR-3 and THP-1 as compared to Raji and SH-SY5Y using Anti-S100A9 Polyclonal Antibody (Product # PA5-79949) in Western Blot. {RE}



#### S100A9 Antibody (PA5-79949) in WB

Western blot was performed using Anti-S100A9 Polyclonal Antibody (Product # PA5-79949) and a 13 kDa band corresponding to S100A9 was observed in SK-BR-3, THP-1, Mouse Spleen and Rat Spleen but was absent in Raji, SH-SY5Y, Mouse Brain and Rat Brain which are reported to be negative. Expression of Calprotectin S100A9 was downregulated upon treatment of THP-1 with PMA /TPA (50 ng/mL for 24 hours) [10.1073/pnas.0709958105]. Membrane enriched cell extracts (30 µg lysate) of SK-BR-3 (Lane 1), THP-1 (Lane 2), THP-1 treated with PMA/TPA (50 ng/mL for 24 hours) (Lane 3), Raji (Lane 4), SH-SY5Y (Lane 5), Mouse Spleen (Lane 6), Rat Spleen (Lane 7), Mouse Brain (Lane 8) and Rat Brain (Lane 9) were electrophoresed using Novex® NuPAGE® 4-12 % Bis-Tris gel (Product # NP0322BOX). Resolved proteins were then transferred onto a nitrocellulose membrane (Product # IB23001) by iBlot® 2 Dry Blotting System (Product # IB21001). The blot was probed with the primary antibody  $(0.5 \,\mu g/mL)$ and detected by chemiluminescence with Goat anti-Rabbit IgG (Heavy Chain), Superclonal<sup>™</sup> Recombinant Secondary Antibody, HRP (Product # A27036, 1: 4,000 dilution) using the iBright FL 1000 (Product # A32752). Chemiluminescent detection was performed using Novex® ECL Chemiluminescent Substrate Reagent Kit (Product # WP20005).

#### S100A9 Antibody (PA5-79949) in ICC/IF



Immunofluorescence analysis of S100A9 was performed using 70% confluent log phase THP-1 and Raji cells. The cells were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton<sup>™</sup> X-100 for 15 minutes, and blocked with 2% BSA for 1 hour at room temperature. The cells were labeled with S100A9 Polyclonal Antibody (Product # PA5-79949) at 1:250 dilution in 0.1% BSA, incubated at 4 degree Celsius overnight and then labeled with Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor Plus 488 (Product # A32790) at a dilution of 1:2000 for 45 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with SlowFade® Gold Antifade Mountant with DAPI (Product # S36938). F-actin (Panel c: red) was stained with Rhodamine Phalloidin (Product # R415, 1:300). Panel d represents the merged image of THP-1 showing localization to plasma membrane, nucleus and cytoplasm. Panel e shows Raji cells with no expression of S100A9. Panel f represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.

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