Human S100A11 Mouse Monoclonal Antibody (Clone YK-1A4)

Cat# CY-M1037

100 µg (0.8 mg/mL x 125 µL)

Clone Name	Applications	Species Cross-Reactivity	Molecular Wt.	Source Isotype
YK-1A4	WB, E	H	1.2-1.3 kDa	Mouse IgG1

Background

S100A11 (S100C, calgizarrin) is a member of the S100 family of EF-hand Ca2+-binding proteins, which is expressed in smooth muscle and other tissues. It is also localized in the cytoplasm in resting cells and moves to the cell periphery in cultured epidermal keratinocytes following calcium challenge. This movement requires the presence of intact microtubules. S100A11 was shown to bind to annexin A1 and the S100A11/annexin A1 complex is a heterotetramer consisting of two S100A11 and two annexin I proteins. Ca2+ binding to S100A11 induces a conformational change that exposes a hydrophobic surface for interaction with target proteins. S100A11 was also shown to interact with annexin A6 in Ca2+-dependent manner.

Specificity/Sensitivity: The Human S100A11 Monoclonal Antibody detects endogenous S100A11 by western blotting and sandwich ELISA.

Source/Purification: The antibody is produced by immunizing mice with a recombinant full length human S100A11. IgG is purified by protein A-Sepharose chromatography.

Recommended Antibody Dilutions: Western blotting: 0.5-1 µg/mL

Storage: Supplied in 20 mM phosphate buffer (pH 7.5), 300 mM NaCl, 50 % glycerol. Store at -20°C.

Applications Key: WB: Western **IP:** Immunoprecipitation **IHC:** Immunohistochemistry **IC:** Immunocytochemistry **F:** Flow cytometry **E:** ELISA **FP:** Fluorescence Polarization assay
Species Cross-Reactivity Key: H: human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Mi:** mink **C:** chicken **X:** *Xenopus* **Z:** zebra fish **All:** all species expected Species enclosed in parentheses are predicted to react based on 100% sequence homology.

General References:

1. Todoroki, H. et al. J. Biol. Chem. 266: 18668-18673, 1991.

2. Tanaka, M. et al. Cancer Lett. 89: 195-200, 1995.

3. Moog-Lutz, C. et al. Int. J. Cancer 63: 297-303, 1995.

4. Mailliard, W. S. Et al. J. Biol. Chem. 271: 719–725, 1996

5. Rety, S. et al. Structure 8: 175–184, 2000

6. Sakaguchi, M. et al. J. Cell Biol. 163: 825–835, 2003

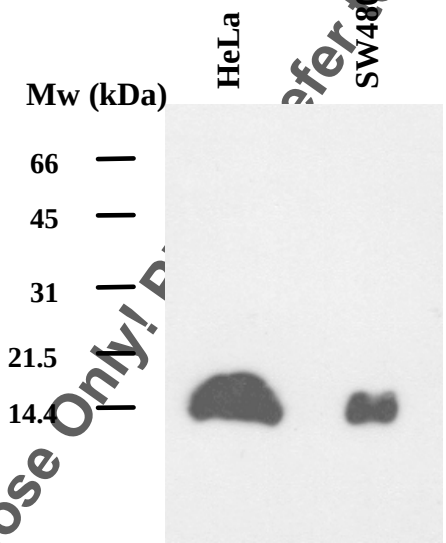
7. Dempsey, A. C. et al. Structure 11: 887–897, 2003

8. Sakaguchi, M. et al. J. Cell Biol. 164: 979–984, 2004

9. Ohuchida, K. et al. Clin. Cancer Res. 12: 5417–5422, 2006

10. Sakaguchi, M. et al. Mol. Biol. Cell 19: 78-85, 2008

Fig.1 Western blot analysis of Human S100A11



Western Immunoblotting Protocol

Solutions and Reagents

Note: Prepare solutions with Milli-Q or equivalently purified water.

Transfer Buffer: 25 mM Tris base, 0.2 M glycine, 20% methanol (pH 8.5)
SDS Sample Buffer (1X): 62.5 mM Tris-HCl (pH 6.8 at 25°C), 2% w/v SDS, 10% glycerol, 50 mM DTT, 0.01% w/v bromophenol blue or phenol red
Blocking Buffer: 1X TBS, 0.1% Tween-20 with 5% w/v nonfat dry milk; for 150 mL, add 15 mL 10X TBS to 135 mL water, mix. Add 7.5 g nonfat dry milk and mix well. While stirring, add 0.15 mL Tween-20 (100%).
10X TBS (Tris-buffered saline): To prepare 1 liter of 10X TBS: 24.2 g Tris base, 80 g NaCl; adjust pH to 7.6 with HCl (use at 1X).
Primary Antibody Dilution Buffer: 1X TBS, 0.1% Tween-20 with 5% blocking agent for 20 mL, add 2 mL 10X TBS to 18 ml water, mix. Add 1.0 g BSA and mix well. While stirring, add 20 µL Tween-20 (100%).
Chemiluminescent HRP Detection: secondary anti-rabbit antibody conjugated to horseradish peroxidase (HRP), ECL™ chemiluminescent reagent (Amersham Pharmacia)
Wash Buffer TBS/T: 1X TBS, 0.1% Tween-20
Blotting Membrane: This protocol has been optimized for nitrocellulose membranes, which we recommend. PVDF membranes may also be used.

Protein Blotting

A general protocol for sample preparation is described below.

1. Treat cells by adding fresh media containing regulator for desired time.
2. Aspirate media from cultures; wash cells with 1X PBS, aspirate.
3. Lyse cells by adding 1X SDS Sample Buffer (100 µL per well of 6-well plate or 500 µL per plate of 10 cm2 plate). Immediately scrape the cells off the plate and transfer the extract to a microcentrifuge tube. Keep on ice.
4. Sonicate for 10–15 seconds to shear DNA and reduce sample viscosity.
5. Heat a 20 µL sample to 95–100°C for 5 minutes, cool on ice.
6. Microcentrifuge for 5 minutes.
7. Load 20 µL onto SDS-PAGE gel (10 cm x 10 cm).
8. Electrotransfer to nitrocellulose membrane.

Membrane Blocking and Antibody Incubations

Note: Volumes are for 10 cm x 10 cm (100 cm^2) of membrane; for different sized membranes, adjust volumes accordingly.

1. (Optional) After transfer, wash nitrocellulose membrane with 25 mL TBS for 5 minutes at room temperature.
2. Incubate membrane in 25 mL of Blocking Buffer for 1 hour at room temperature.
3. Wash 3 times for 5 minutes each with 15 ml of TBS/T.
4. Incubate membrane and primary antibody (at the appropriate dilution) in 10 mL Primary Antibody Dilution Buffer with gentle agitation overnight at 4°C.
5. Wash 3 times for 5 minutes each with 15 mL of TBS/T.
6. Incubate membrane with HRP-conjugated secondary antibody (1:3000 in 10 mL of Blocking Buffer with gentle agitation for 1 hour at room temperature.
7. Wash 3 times for 5 minutes each with 15 mL of TBS/T.

Detection of Proteins

1. Incubate membrane with 4 mL ECL™ with gentle agitation for 1 minute at room temperature.
2. Drain membrane of excess developing solution, do not let dry, wrap in plastic wrap and expose to x-ray film. An initial ten-second exposure should indicate the proper exposure time.

Related Products

*** CircuLex S100A13 ELISA Kit: Cat# CY-8057**
*** CircuLex S100A12 ELISA Kit: Cat# CY-8058**
*** CircuLex S100P ELISA Kit: Cat# CY-8060**
*** CircuLex S100A8-MRP8 ELISA Kit: Cat# CY-8061**
*** CircuLex S100A9-MRP14 ELISA Kit: Cat# CY-8062**
*** CircuLex S100A11 ELISA Kit: Cat# CY-8063**
*** CircuLex S100A14 ELISA Kit: Cat# CY-8064**
*** CircuLex S100A7/Psoriasin ELISA Kit: Cat# CY-8073**
*** CircuLex S100A4 ELISA Kit Ver.2: Cat# CY-8086**

*** Anti-Human S100A3 (Clone YK-3E3): Cat# CY-M1039**
*** Anti-Human S100A4 (p9Ka): Cat# CY-P1026**
*** Anti-Human S100P: Cat# CY-P1028**
*** Anti-Human S100A10: Cat# CY-P1033**
*** Anti-Human S100A16: Cat# CY-P1034**
*** Anti-Human S100A3: Cat# CY-P1039**
*** Anti-Human S100A2: Cat# CY-P1040**

*** Human S100B: Cat# CY-R2250**
*** Human S100A1: Cat# CY-R2251**
*** Human S100A2: Cat# CY-R2252**
*** Human S100A3: Cat# CY-R2253**
*** Human S100A4: Cat# CY-R2254**
*** Human S100A5: Cat# CY-R2255**
*** Human S100A6: Cat# CY-R2256**
*** Human S100A7: Cat# CY-R2257**
*** Human S100A8: Cat# CY-R2258**
*** Human S100A9: Cat# CY-R2259-G**
*** Human S100A9: Cat# CY-R2259-H**
*** Human S100A10: Cat# CY-R2260**
*** Human S100A12: Cat# CY-R2262-G**
*** Human S100A12: Cat# CY-R2262-H**
*** Human S100A13: Cat# CY-R2263**
*** Human S100A14: Cat# CY-R2264**
*** Human S100A16: Cat# CY-R2266**
*** Human S100P: Cat# CY-R2267**
*** Human S100A11: Cat# CY-R2269**

*** Human S100A1 Low Endotoxin: Cat# CY-R2451**
*** Human S100A3 Low Endotoxin: Cat# CY-R2453**
*** Human S100A4 Low Endotoxin: Cat# CY-R2454**
*** Human S100A7 Low Endotoxin: Cat# CY-R2457**
*** Human S100A8 Low Endotoxin: Cat# CY-R2458**
*** Human S100A9 Low Endotoxin: Cat# CY-R2459-G**

*** Human S100A11 Low Endotoxin: Cat# CY-R2461**
*** Human S100A12 Low Endotoxin: Cat# CY-R2462-G**
*** Human S100A14 Low Endotoxin: Cat# CY-R2464**
*** Human S100P Low Endotoxin: Cat# CY-R2467**

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