## SANTA CRUZ BIOTECHNOLOGY, INC.

# RhAG (D-5): sc-390045



### BACKGROUND

The Rh proteins in the red blood cell form a complex made up of one D-subunit, one CE-subunit, and two Rh-associated glycoprotein (RhAG) subunits. Along with its antigenic properties, this complex functions as a major interaction site between the membrane lipid bilayer and the cytoskeleton of the red cell, via ankyrin-R interaction with the C-terminal cytoplasmic domain of the Rh and RhAG proteins. Furthermore, studies comparing ammonium concentration in normal and Rh(null) red cells show that the complex also contributes to ammonium export from the cells. Rh(null) is a rare autosomal recessive disorder characterized by an absence of Rh antigens and a varying degree of hemolytic anemia and spherostomatocytosis. The associated genetic mutations effect the transmembrane domain of the protein, correlating the structural defect with the loss of transport function characteristic in these cells. RhAG is also known as CD241.

#### REFERENCES

- Huang, C.H., et al. 1999. Molecular basis for Rh(null) syndrome: identification of three new missense mutations in the Rh50 glycoprotein gene. Am. J. Hematol. 62: 25-32.
- Suyama, K., et al. 2000. Surface expression of Rh-associated glycoprotein (RhAG) in nonerythroid COS-1 cells. Blood 95: 336-341.
- Mouro-Chanteloup, I., et al. 2002. Cell-surface expression of RhD blood group polypeptide is posttranscriptionally regulated by the RhAG glycoprotein. Blood 100: 1038-1047.
- Nicolas, V., et al. 2003. Rh-RhAG/ankyrin-R, a new interaction site between the membrane bilayer and the red cell skeleton, is impaired by Rh(null)associated mutation. J. Biol. Chem. 278: 25526-25533.
- 5. Nakhoul, N.L., et al. 2004. Non-erythroid Rh glycoproteins: a putative new family of mammalian ammonium transporters. Pflugers Arch. 447: 807-812.

#### **CHROMOSOMAL LOCATION**

Genetic locus: RHAG (human) mapping to 6p12.3; Rhag (mouse) mapping to 17 B2.

#### SOURCE

RhAG (D-5) is a mouse monoclonal antibody raised against amino acids 1-56 mapping at the N-terminus of RhAG of human origin.

#### PRODUCT

Each vial contains 200  $\mu g$  IgG\_{2b} kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

RhAG (D-5) is available conjugated to agarose (sc-390045 AC), 500 μg/0.25 ml agarose in 1 ml, for IP; to HRP (sc-390045 HRP), 200 μg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-390045 PE), fluorescein (sc-390045 FITC), Alexa Fluor<sup>®</sup> 488 (sc-390045 AF488), Alexa Fluor<sup>®</sup> 546 (sc-390045 AF546), Alexa Fluor<sup>®</sup> 594 (sc-390045 AF594) or Alexa Fluor<sup>®</sup> 647 (sc-390045 AF647), 200 μg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor<sup>®</sup> 680 (sc-390045 AF680) or Alexa Fluor<sup>®</sup> 790 (sc-390045 AF790), 200 μg/ml, for Near-Infrared (NIR) WB, IF and FCM.

#### **APPLICATIONS**

RhAG (D-5) is recommended for detection of RhAG of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2  $\mu$ g per 100-500  $\mu$ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Suitable for use as control antibody for RhAG siRNA (h): sc-106869, RhAG siRNA (m): sc-152843, RhAG shRNA Plasmid (h): sc-106869-SH, RhAG shRNA Plasmid (m): sc-152843-SH, RhAG shRNA (h) Lentiviral Particles: sc-106869-V and RhAG shRNA (m) Lentiviral Particles: sc-152843-V.

Molecular Weight of RhAG: 50 kDa.

Positive Controls: C6 whole cell lysate: sc-364373, MEG-01 cell lysate: sc-2283 or TF-1 cell lysate: sc-2412.

## **RECOMMENDED SUPPORT REAGENTS**

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-lgG $\kappa$  BP-HRP: sc-516102 or m-lgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>TM</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-lgG $\kappa$  BP-FITC: sc-516140 or m-lgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

#### DATA





RhAG (D-5): sc-390045. Western blot analysis of RhAG expression in 3T3-L1 (**A**), C6 (**B**), M1 (**C**), c4 (**D**) and WEHI-231 (**E**) whole cell lysates.

RhAG (D-5): sc-390045. Western blot analysis of RhAG expression in TF-1 ( $\bf{A}$ ) and MEG-01 ( $\bf{B}$ ) whole cell lysates.

#### **STORAGE**

Store at 4° C, \*\*DO NOT FREEZE\*\*. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

#### **RESEARCH USE**

For research use only, not for use in diagnostic procedures.

#### PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

Alexa Fluor® is a trademark of Molecular Probes, Inc., Oregon, USA