

Mouse Carbonic Anhydrase IX Monoclonal Antibody | anti-CA9 antibody

Carbonic Anhydrase IX (CA9) (PE)

CATALOG # AAA14680	HOST Mouse	APPLICATIONS Flow Cytometry, Functional Assay
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PRODUCT SYNONYMS

Carbonic Anhydrase IX; Monoclonal Antibody; Carbonic Anhydrase IX (CA9) (PE); Anti -Carbonic Anhydrase IX (CA9) (PE); anti-CA9 antibody

PRODUCT SPECIFICATIONS

Host	Mouse
Clonality	Monoclonal
Isotype	IgG2a
Clone Number	7H20
Specificity	Recognizes human Carbonic Anhydrase IX (CA9). Does not crossreact with recombinant mouse CA9 or with recombinant human CA1, 2, 3, 4, 5A, 6, 7, 8, 10, 12, 13, or 14.
Purity/Purification	Affinity Purified Purified by Protein A affinity chromatography from hybridoma culture supernant.
Form/Format	Supplied as a liquid in saline, 0.5% BSA, 0.09% sodium azide. Labeled with R-Phycoerythrin (PE).
Concentration	0.025mg/ml (varies by lot)
Applicable Applications for anti-CA9 antibody	FCM/FACS (Flow Cytometry)
Application Notes	Suitable for use in Flow Cytometry. Dilution: Flow Cytometry: Neat. 10ul labels 1x10e6 cells. Tested using U-87 MG human glioblastoma/astrocytoma cell line. Optimal dilutions to be dertermined by the researcher.
Immunogen	Recombinant protein corresponding to Pro59-Asp414 of human Carbonic Anhydrase IX (CA9) expressed in NSO cells.

Flow Cytometry Protocol

Reagents Not Provided:
-PBS (Dulbecco's PBS) -BSA
Intended Use:

Designed to quantitatively determine the percentage of cells bearing Carbonic Anhydrase IX (CA9) within a population and qualitatively determine the density of CA9 on cell surfaces by flow cytometry.

Principle of the Test:
Washed cells are incubated with AAA14680, which binds to cells expressing CA9. Unbound phycoerythrin-conjugated antibody is then washed from the cells. Cells expressing CA9 are fluorescently stained: with the intensity of staining directly proportional to the density of expression of CA9. Cell surface expression of CA9 is determined by flow cytometric analysis using 488nm wavelength laser excitation and monitoring emitted fluorescence with a detector optimized to collect peak emissions at 565-605nm.

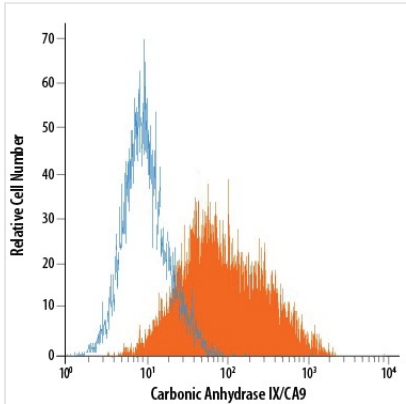
Sample Preparation:
Peripheral Blood Cells:
Whole blood should be collected in evacuated tubes containing EDTA or heparin as the anticoagulant. Contaminating serum components should be removed by washing the cells three times in an isotonic phosphate buffer (supplemented With 0.50/0 BSA) by centrifugation at 500 x g for 5 minutes. Transfer 50ul of packed cells to a 5ml tube for staining with the monoclonal antibody. Whole blood will require lysis of RBC following the staining procedure.
Cell Cultures: Continuous cell lines or activated cell cultures should be centrifuged at 500 x g for 5 minutes and washed three times in an isotonic PBS buffer (supplemented with 0.50/0 BSA), as described above, to remove any residual growth factors that may be present in the culture medium. Cells should then be resuspended in the same buffer to a final concentration of 4xl Oe6 cells/ml and 25ul of cells (1 xl Oe5) transferred to a Sml tube for staining. Note: Adherent cell lines may require pretreatment with 0.5mM EDTA to facilitate removal from substrate. Cells that require trypsinization to enable removal from substrate should be further incubated in medium for 6-10 hours on a rocker platform to enable regeneration of the receptors. The use of the rocker platform will prevent reattachment to the substrate.

Sample Staining:
1. Cells should be Fe-blocked by treatment with 1 ug of human IgG/ IOe5 cells for 15 minutes at RT prior to staining. Do not wash excess blocking IgG from this reaction.
2. Transfer 25ul of the Fe-blocked cells (1 xl Oe5 cells) or SOul of packed whole blood to a 5ml tube.
3. Add 1 Oul of CII 05-80A. Incubate for 30-45 minutes at 4°C.
4. Following this incubation, remove unreacted CA9 reagent by washing the cells twice in 4ml of the same PBS buffer (note: whole blood will require an RBC lysis step at this point using any commercially available lysing reagent).
5. Finally, resuspend the cells in 200-400ul of PBS buffer for final flow cytometric analysis.
6. As a control for analysis, cells in a separate tube should be treated with PE-labeled mouse IgG2a antibody. Optimal conditions should be determined by each laboratory.

Preparation and Storage

May be stored at 4°C in the dark. DO NOT FREEZE! Stable for 12 months at 4°C after received as an undiluted liquid. Further dilutions can be made in assay buffer.
Caution: PE conjugates are sensitive to light. Centrifuge before opening to ensure complete recovery of vial contents.

IMAGES



FCM (Flow Cytometry)
Detection of Carbonic Anhydrase IX/CA9 in U937/MG Human Cell Line
by Flow Cytometry using AAA14680.

ADDITIONAL INFORMATION

Related Product Information for anti-CA9 antibody	Carbonic anhydrase (CA) catalyzes the reversible reaction of $\text{CO}_2 + \text{H}_2\text{O} = \text{HCO}_3^- + \text{H}^+$, which is fundamental to many processes such as respiration, renal tubular acidification and bone resorption. CA9, also known as membrane antigen MN and renal cell carcinoma (RCC)-associated antigen G250, is a transmembrane enzyme expressed primarily in carcinoma cells. It is one of the best markers for hypoxia and for RCC.
Product Categories/Family for anti-CA9 antibody	Antibodies; Abs to Enzymes

NCBI AND UNIPROT INFORMATION

NCBI GI #	119578763
NCBI GeneID	768
UniProt Accession #	Q16790; Q5T4R1
NCBI Official Full Name	carbonic anhydrase IX
NCBI Official Synonym Full Names	carbonic anhydrase IX
NCBI Official Symbol	CA9
NCBI Official Synonym Symbols	MN; CAIX
NCBI Protein Information	carbonic anhydrase 9; pMW1; CA-IX; P54/58N; OTTHUMP00000022773; membrane antigen MN; carbonic dehydratase; carbonate dehydratase IX; RCC-associated antigen G250; RCC-associated protein G250; renal cell carcinoma-associated antigen G250
UniProt Protein Name	Carbonic anhydrase 9
UniProt Gene Name	CA9
UniProt Synonym Gene Names	G250; MN
UniProt Entry Name	CAH9_HUMAN

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Small volumes of product may become entrapped in the seal of the vial during shipment and storage; if necessary, briefly centrifuge the vial on a tabletop centrifuge to dislodge any liquid in the cap.

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Online datasheet: <https://stage.aaabiotech.com/monoclonal/carbonic-anhydrase-ix-antibody-clone-7h20-flow-cytometry-elisa/14680>