10320 Camino Santa Fe, Suite G
San Diego, CA 92121
Tel: 858.875.1900 Fax: 858.875.1999

Anti-ARSA Antibody Picoband™ (monoclonal, 4C10)

Catalog # ABO14785

Product Information

Application	WB, IHC, FC
Primary Accession	P15289
Host	Mouse
Isotype	Mouse IgG2a
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-ARSA Antibody Picoband™ (monoclonal, 4C10) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	410
Other Names	Arylsulfatase A, ASA, 3.1.6.8, Cerebroside-sulfatase, Arylsulfatase A component B, Arylsulfatase A component C, ARSA
Calculated MW	53588
Application Details	Western blot, 0.1-0.5 □g/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 0.5-1 □g/ml, Human, By Heat Flow Cytometry, 1-3 □g/1x10^6 cells, Human, Mouse
Subcellular Localization	Lysosome.
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Clone Names	Clone: 4C10
Immunogen	A synthetic peptide corresponding to a sequence at the C-terminus of human ARSA, different from the related mouse sequence by six amino acids.
Cross Reactivity	No cross-reactivity with other proteins.
Storage	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.

Protein Information

Name	ARSA (HGNC:713)
Function	Lysosomal enzyme that catalyzes the hydrolysis of cerebroside-3-sulfate (N-acyl-1-beta-D-(3-O-sulfo)-galactosyl-sphing-4- enine) into cerebroside and sulfate, a reaction that requires the activator protein saposin B (PubMed:10751093, PubMed:19224915, PubMed:24294900). Also involved in the catabolism of other sulfoglycosphingolipids such as SB1a and SM3 (beta-D-3-sulfogalactosyl- (1->4)-beta-D-glucosyl-(11')-N-acylsphing-4-enine) (By similarity).
Cellular Location	Endoplasmic reticulum. Lysosome

Background

Arylsulfatase A (ARSA) is an enzyme that breaks down sulfatides, namely cerebroside 3-sulfate intocerebroside and sulfate. In humans, arylsulfatase A is encoded by the ARSA gene. ARSA is mapped to 22q13.33. The protein encoded by this gene hydrolyzes cerebroside sulfate to cerebroside and sulfate. Defects in this gene lead to metachromatic leucodystrophy (MLD), a progressive demyelination disease which results in a variety of neurological symptoms and ultimately death. Alternatively spliced transcript variants have been described for this gene.

Images

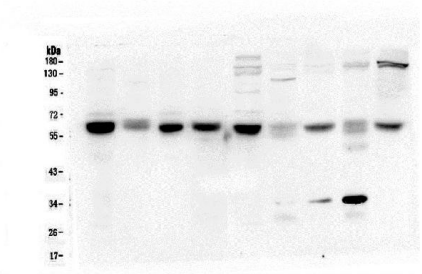
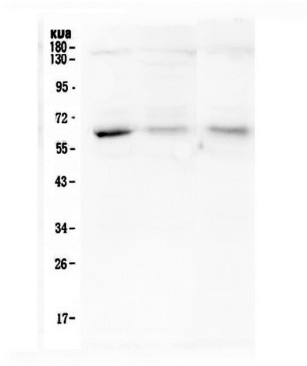


Figure 2. Western blot analysis of ARSA using anti-ARSA antibody (M02583).
Electrophoresis was performed on a 10% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: rat testis tissue lysate,
Lane 2: rat liver tissue lysate,
Lane 3: rat brain tissue lysate,
Lane 4: rat lung tissue lysate,
Lane 5: mouse testis tissue lysate,
Lane 6: mouse liver tissue lysate,
Lane 7: mouse brain tissue lysate,
Lane 8: mouse lung tissue lysate,
Lane 9: mouse HEPA1-6 whole cell lysate.
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-ARSA antigen affinity purified monoclonal antibody (Catalog # M02583) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system.

Figure 3. Western blot analysis of ARSA using anti-ARSA antibody (M02583).



Electrophoresis was performed on a 10% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human A375 whole cell lysate,
Lane 2: human A549 whole cell lysate,
Lane 3: human SMMC-7721 whole cell lysate.
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-ARSA antigen affinity purified monoclonal antibody (Catalog # M02583) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system.

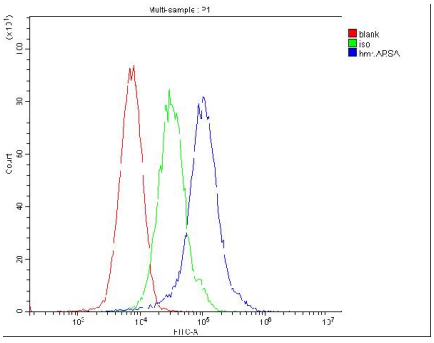


Figure 1. Flow Cytometry analysis of ANA-1 cells using anti-ARSA antibody (M02583).
Overlay histogram showing ANA-1 cells stained with M02583 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-ARSA Antibody (M02583,1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

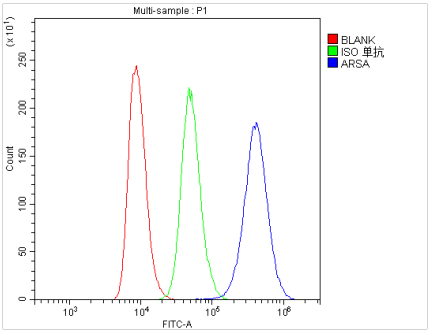


Figure 4. Flow Cytometry analysis of Raji cells using anti-ARSA antibody (M02583).
Overlay histogram showing Raji cells stained with M02583 (Blue line).The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-ARSA Antibody (M02583, 1 µg/1x10^6 cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 µg/1x10^6 cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 µg/1x10^6) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

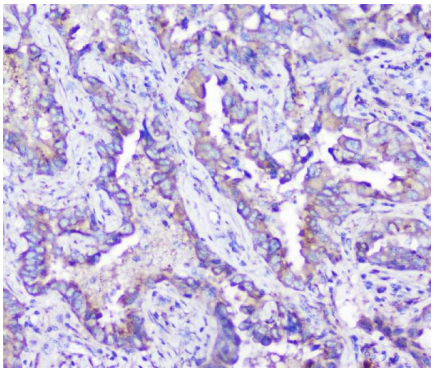


Figure 5. IHC analysis of ARSA using anti-ARSA antibody (M02583).
ARSA was detected in paraffin-embedded section of human lung cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1 µg/ml rabbit anti-ARSA Antibody (M02583) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.