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Anti-RAGE/AGER Antibody Picoband™ (monoclonal, 5C6C1)

Catalog # ABO16627

Product Information

Application	WB, IHC, FC
Primary Accession	Q15109
Host	Mouse
Isotype	IgG2b
Reactivity	Rat, Human, Mouse
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-RAGE/AGER Antibody Picoband™ (monoclonal, 5C6C1) . Tested in Flow Cytometry, IHC, WB applications. This antibody reacts with Human, Mouse, Rat.
Reconstitution	Adding 0.2 ml of distilled water will yield a concentration of 500 □g/ml.

Additional Information

Gene ID	177
Other Names	Advanced glycosylation end product-specific receptor, Receptor for advanced glycosylation end products, AGER, RAGE
Calculated MW	42803
Application Details	Western blot, 0.25-0.5 □g/ml, Mouse, Rat Immunohistochemistry(Paraffin-embedded Section), 2-5 □g/ml, Mouse, Rat Flow Cytometry, 1-3 □g/1x10^6 cells, Human
Source	Eukaryota
Contents	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na2HPO4.
Clone Names	Clone: 5C6C1
Immunogen	A synthetic peptide corresponding to a sequence at the N-terminus of human RAGE, different from the related mouse and rat sequences by six amino acids.
Purification	Immunogen affinity purified.
Storage	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 freezing and thawing.

Protein Information

Name	AGER
Synonyms	RAGE
Function	Cell surface pattern recognition receptor that senses endogenous stress signals with a broad ligand repertoire including advanced glycation end products, S100 proteins, high-mobility group box 1 protein/HMGB1, amyloid beta/APP oligomers, nucleic acids, histones, phospholipids and glycosaminoglycans (PubMed:27572515, PubMed:28515150, PubMed:34743181, PubMed:35974093, PubMed:24081950). Advanced glycation end products (AGEs) are nonenzymatically glycosylated proteins which accumulate in vascular tissue in aging and at an accelerated rate in diabetes (PubMed:21565706). These ligands accumulate at inflammatory sites during the pathogenesis of various diseases including diabetes, vascular complications, neurodegenerative disorders and cancers, and RAGE transduces their binding into pro-inflammatory responses. Upon ligand binding, uses TIRAP and MYD88 as adapters to transduce the signal ultimately leading to the induction of inflammatory cytokines IL6, IL8 and TNFalpha through activation of NF-kappa-B (PubMed:21829704, PubMed:33436632). Interaction with S100A12 on endothelium, mononuclear phagocytes, and lymphocytes triggers cellular activation, with generation of key pro-inflammatory mediators (PubMed:19386136). Interaction with S100B after myocardial infarction may play a role in myocyte apoptosis by activating ERK1/2 and p53/TP53 signaling (By similarity). Contributes to the translocation of amyloid- beta peptide (ABPP) across the cell membrane from the extracellular to the intracellular space in cortical neurons (PubMed:19906677). ABPP- initiated RAGE signaling, especially stimulation of p38 mitogen- activated protein kinase (MAPK), has the capacity to drive a transport system delivering ABPP as a complex with RAGE to the intraneuronal space. Participates in endothelial albumin transcytosis together with HMGB1 through the RAGE/SRC/Caveolin-1 pathway, leading to endothelial hyperpermeability (PubMed:27572515). Mediates the loading of HMGB1 in extracellular vesicles (EVs) that shuttle HMGB1 to hepatocytes by transferrin-mediated endocytosis and subsequently promote hepatocyte pyroptosis by activating the NLRP3 inflammasome (PubMed:34743181). Binds to DNA and promotes extracellular hypomethylated DNA (CpG DNA) uptake by cells via the endosomal route to activate inflammatory responses (PubMed:24081950, PubMed:28515150). Mediates phagocytosis by non-professional phagocytes (NPP) and this is enhanced by binding to ligands including RNA, DNA, HMGB1 and histones (PubMed:35974093). Promotes NPP-mediated phagocytosis of Saccharomyces cerevisiae spores by binding to RNA attached to the spore wall (PubMed:35974093). Also promotes NPP-mediated phagocytosis of apoptotic cells (PubMed:35974093). Following DNA damage, recruited to DNA double-strand break sites where it colocalizes with the MRN repair complex via interaction with double-strand break repair protein MRE11 (By similarity). Enhances the endonuclease activity of MRE11, promoting the end resection of damaged DNA (By similarity). Promotes DNA damage repair in trophoblasts which enhances trophoblast invasion and contributes to placental development and maintenance (PubMed:33918759). Protects cells from DNA replication stress by localizing to damaged replication forks where it stabilizes the MCM2-7 complex and promotes faithful progression of the replication fork (PubMed:36807739). Mediates the production of reactive oxygen species (ROS) in human endothelial cells (PubMed:25401185).
Cellular Location	Cell membrane; Single-pass type I membrane protein. Cell projection, phagocytic cup. Early endosome. Nucleus. Note=Detected on the surface of CD11c+ peripheral blood mononuclear cells under basal conditions and after

activation (PubMed:22509345). No surface expression is observed on resting T cells (PubMed:22509345). Localizes intracellularly in early endosomes in activated T cells of healthy controls and in resting T cells of patients with type I diabetes (PubMed:22509345). Nuclear translocation is enhanced by irradiation, hypoxia and reperfusion injury to brain or kidney (By similarity). Nuclear localization is enhanced by DNA damage in trophoblasts and increases in pre-term labor and preeclampsia placentas compared to control placentas (PubMed:33918759). {ECO:0000250|UniProtKB:Q62151, ECO:0000269|PubMed:22509345, ECO:0000269|PubMed:33918759} [Isoform 2]: Secreted.

Tissue Location

Endothelial cells. Increased expression in pre-term labor and preeclampsia placentas compared to controls (PubMed:33918759).

Background

The receptor for advanced glycation end products (RAGE) is a multi-ligand member of the immunoglobulin superfamily of cell surface molecules. It interacts with distinct molecules implicated in homeostasis, development and inflammation, and certain diseases such as diabetes and Alzheimer's disease. RAGE is also a central cell surface receptor for amphoterin and EN-RAGE. And RAGE is associated with sustained NF-kappaB activation in the diabetic microenvironment and has a central role in sensory neuronal dysfunction. Moreover, RAGE propagates cellular dysfunction in several inflammatory disorders and diabetes, and it also functions as an endothelial adhesion receptor promoting leukocyte recruitment.

Images

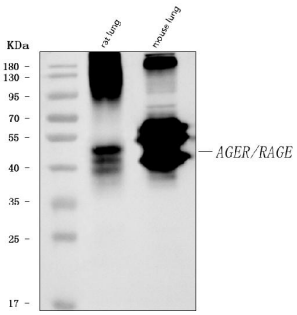
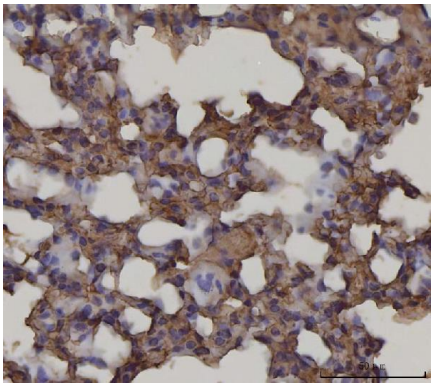


Figure 1. Western blot analysis of RAGE/AGER using anti-RAGE/AGER antibody (M03438-2).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.
Lane 1: rat lung tissue lysates,
Lane 2: mouse lung tissue lysates.
After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA 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-RAGE/AGER antigen affinity purified monoclonal antibody (Catalog # M03438-2) 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. A specific band was detected for RAGE/AGER at approximately 43 kDa. The expected band size for RAGE/AGER is at 43 kDa.

Figure 2. IHC analysis of RAGE/AGER using anti-RAGE/AGER antibody (M03438-2).
RAGE/AGER was detected in a paraffin-embedded section of mouse lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-RAGE/AGER Antibody (M03438-2)



overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

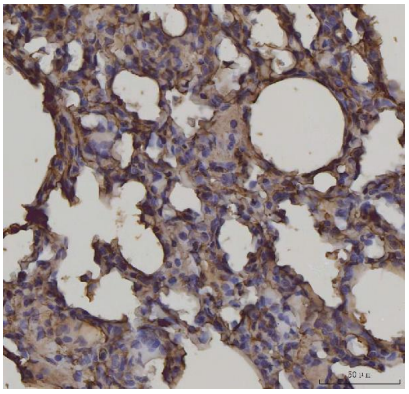


Figure 3. IHC analysis of RAGE/AGER using anti-RAGE/AGER antibody (M03438-2).
RAGE/AGER was detected in a paraffin-embedded section of rat lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-RAGE/AGER Antibody (M03438-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

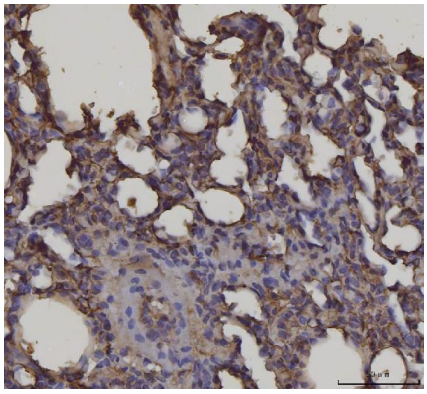


Figure 4. IHC analysis of RAGE/AGER using anti-RAGE/AGER antibody (M03438-2).
RAGE/AGER was detected in a paraffin-embedded section of rat lung tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-RAGE/AGER Antibody (M03438-2) overnight at 4°C. Peroxidase Conjugated Goat Anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Mouse IgG Super Vision Assay Kit (Catalog # SV0001) with DAB as the chromogen.

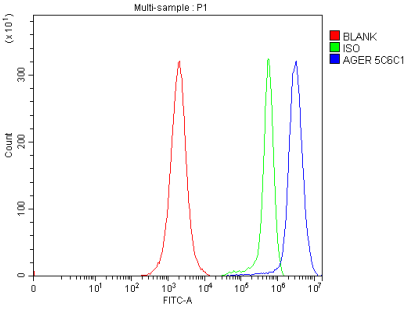


Figure 5. Flow Cytometry analysis of Jurkat cells using anti-RAGE/AGER antibody (M03438-2).
Overlay histogram showing Jurkat cells stained with M03438-2 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-RAGE/AGER Antibody (M03438-2, 1 µg/1x10^6 cells) 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.

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