

3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

## **ProductInformation**

# Anti-Endothelial Cell Differentiation Gene 8, (EDG-8), C-Terminal

Developed in Rabbit, Fractionated Antisera

Product Number E 7528

#### **Product Description**

Anti-EDG-8 (Endothelial Cell Differentiation Gene-8) C-terminal antibody is developed in rabbit using a synthetic peptide derived from the C-terminal region of human EDG-8 receptor as immunogen. The serum is purified by ammonium sulfate precipitation and then dialyzed to prevent exposure to extreme pH changes.

Anti-EDG-8 recognizes recombinant and native human EDG-8 receptor (42 kDa). It has been used in immunoblotting applications.

The lysosphingolipid sphingosine-1-phosphate (S1P) and the structurally related lipid lysophosphatidic acid (LPA) elicit a wide spectrum of biological responses in a variety of cell types, including calcium mobilization, mitogenesis, cell-shape changes, migration and contraction. S1P and LPA have been implicated in a variety of pathophysiological conditions, including blood clotting, subarachinoid hemorrhage, inflammation and colitis. S1P mediates some of its activities intracellularly by acting as a second messenger, while LPA may mediate mitogenesis and platelet aggregation via an EDG receptor-independent mechanism. LPA is also an antiapoptotic factor.

Recent studies have identified the existence of the G protein-coupled heptahelical receptor subfamily (Endothelial Cell Differentiation Genes) for the biologically active lysophospholipids, which consists of two receptor subgroups specific for S1P and LPA, respectively. The S1P receptor subgroup comprises five members: EDG-1, -3, -5/AGR16, -6, and -8, with considerable amino acid similarity among them. The LPA subgroup includes EDG-2, -4, and -7. EDG receptors are developmentally regulated and differ in their tissue expression. The amino acid sequence similarity between EDG receptors reflects the similarity of S1P and LPA.

EDG-8 receptor (400 amino acids) shares 42% to 49% amino acid identity with the other human S1P receptors EDG-1, -3 and -5. Expression of EDG-8 leads to high-

affinity binding for labeled S1P. EDG-8 transcript is widely expressed in tissues, with strongest expression in astrocytes in brain, spleen and white matter tracts of the brain.  $^{3,4}$  In Chinese hamster ovary cells heterologously expressing EDG-8, S1P inhibited forskolin-induced cAMP accumulation and activated c-Jun NH2-terminal kinase. Together the data demonstrate that EDG-8 is a high affinity S1P receptor that couples to  $G(i/o)\alpha$  proteins and is expressed predominantly in rat brain.

## Reagent

Anti-EDG-8 is supplied as approximately 100  $\mu g$  of antibody in 100  $\mu l$  of sterile phosphate buffered saline, pH 7.3, with 0.08% sodium azide as a preservative.

## **Precautions and Disclaimer**

Due to the sodium azide content, a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazards and safe handling practices.

## Storage/Stability

Store at -20 °C. For extended storage, freeze in working aliquots. Avoid repeated freezing and thawing to prevent denaturing of the antibody. Do not store in a frost-free freezer. The antibody is stable for at least 12 months when stored appropriately.

#### **Product Profile**

A recommended working concentration of 5 to 10  $\mu$ g/ml was determined by immunoblotting using RH7777 cells transfected with full length human EDG-8. Data show the specificity of Anti-EDG-8 for the EDG-8 receptor protein (42 kDa). Preincubation of the antibody with immunizing peptide blocks the detection of EDG-8. For the detection of natural EDG-5 in cells and tissues use higher concentrations of the antibody.

**Note**: In order to obtain best results in different techniques and preparations we recommend determining optimal working concentration by titration test

#### References

- Kupperman, E., et al., A sphingosine-1-phosphate receptor regulates cell migration during vertebrate heart development., Nature, 406, 192-195 (2000).
- Takuwa, Y., et al., Subtype-specific, differential activities of the EDG family receptor sphingosine-1phosphate, a novel lysophospholipid mediator. Mol. Cell Endocrinol., 177, 3-11 (2001).
- 3. Im, D.-S., et al., Characterization of a novel human G-protein coupled receptor, EDG7, for lysophosphatidic acid. J. Biol. Chem., **275**, 14281-14286 (2000).
- 4. Lee, D.K., et al., Discovery of a receptor related to the galanin receptors. FEBS Lett., **446**, 103-107 (1999).

AH/JK 12/2003