

Product Information

Anti-ADAM-17, Cytoplasmic Domain

Developed in Rabbit
Affinity Isolated Antibody

Product Number **A 4351**

Product Description

Anti-ADAM-17, Cytoplasmic Domain is developed in rabbit using a synthetic peptide corresponding to the cytoplasmic domain of full length human ADAM17 (A Disintegrin And Metalloproteinase-17) as immunogen. Affinity isolated antigen specific antibody is obtained from rabbit anti-ADAM-17 antiserum by immuno-specific purification which removes essentially all rabbit serum proteins, including immunoglobulins, which do not specifically bind to the peptide.

Anti-ADAM-17, Cytoplasmic Domain may be used for the detection and localization of ADAM17. The antibody reacts with human, porcine, and rat ADAM17 and does not react with other ADAMs. By immunoblotting against the reduced protein, the antibody recognizes a band of 110 kDa from conditioned media or cell lysates.

ADAM17, also known as TNF- α (tumor necrosis factor α) convertase or TACE (TNF- α Converting Enzyme), is a member of the ADAM (a disintegrin and metalloprotease-like domain) family. It was first cloned from human epithelial cells. Since then it has been purified from human and mouse by several groups.¹⁻³ The ADAM proteins are structurally similar, possessing a signal sequence, metalloprotease domain (inactive in some ADAMs), disintegrin domain, cystein-rich domain, EGF-like repeat, type-I transmembrane domain, and cytoplasmic domain.

ADAM-17 is known to process TNF- α trimer from the membrane-attached precursor form to the soluble form. It contains the canonical HEXxHxxxxxH zinc metalloproteinase motif and has been shown to be proteolytically active, cleaving the TNF precursor as well as TNF p75 receptor, myeloid precursor protein, α -cleavage of amyloid protein precursor (APP),⁴ L-selectin adhesion molecule, and TGF- α , making ADAM17 a "shedase."^{5, 6}

ADAM17 contains a cleavage site for prohormone convertases (furin). The majority of ADAM17 is secreted as activated enzyme and cleaved at the furin site. ADAM17 cleaves the extracellular part of the Notch1 receptor and plays a prominent role in the

activation of the Notch pathway.⁷ ADAM17 also associates with MAD2 via the cytoplasmic domain, implying cellular regulation pathways.⁸

Three human ADAM17 sequences encode proteins with different cytoplasmic domains. The 824 amino acid sequence has a predicted molecular mass of 92,960 Daltons. The 807 amino acid sequence encodes a 91,003 Dalton protein, and the 694 amino acid sequence encodes a 78,543 Dalton protein. Only the full-length ADAM17 contains the cytoplasmic epitope. Glycosylation and other posttranslational modifications increase the apparent molecular weight on PAGE gels. Mouse and rat ADAM17 sequences are both 827 amino acids, with predicted molecular masses of 93,073 and 93,017 Daltons, respectively.

Reagent

Anti-ADAM-17, Cytoplasmic Domain is supplied in phosphate buffered saline containing 50% glycerol and 0.05% sodium azide. The protein concentration is approximately 1 mg/ml.

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

For continuous use, store at 2-8 °C for up to six months. For extended storage, the solution may be stored -20 °C. Do not store below -22 °C. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use.

Product Profile

A minimum working antibody dilution of 1:1,000 is determined by immunoblotting tissue or cell lysates with an alkaline phosphatase conjugated secondary antibody and BCIP/NBT as the substrate. A starting dilution of 1:5,000 of the antibody is recommended for chemiluminescent substrates

Note: Higher antibody dilutions may be necessary for non-human samples. EDTA/EGTA treatment of tissues or lysates is necessary to see latent zymogen.

In order to obtain the best results and assay sensitivity in various techniques and preparations we recommend determining optimum working dilutions by titration.

References

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4. Slack, B.E., et al., Constitutive shedding of the amyloid precursor protein ectodomain is up-regulated by tumour necrosis factor- α converting enzyme. *Biochem. J.*, **357**, 787-794 (2001).
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8. Nelson, K.K., et al., Evidence for an interaction of the metalloprotease-disintegrin tumour necrosis factor- α convertase (TACE) with mitotic arrest deficient 2 (MAD2), and of the metalloprotease-disintegrin MDC9 with a novel MAD2-related protein, MAD2 β . *Biochem. J.*, **343**, 673-680 (1999).

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