APA547Hu61 2mg

Active Vascular Cell Adhesion Molecule 1 (VCAM1)

Organism Species: Homo sapiens (Human)

Instruction manual

FOR RESEARCH USE ONLY
NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

1st Edition (Apr, 2016)

[PROPERTIES]

Source: Eukaryotic expression.

Host: 293F cell

Residues: Gln109~Ser318 Tags: N-terminal His-tag

Purity: >92%

Endotoxin Level: <1.0EU per 1µg (determined by the LAL method).

Buffer Formulation: PBS, pH7.4, containing 5% trehalose.

Original Concentration: 650µg/mL

Applications: Cell culture; Activity Assays.

(May be suitable for use in other assays to be determined by the end user.)

Predicted isoelectric point: 5.5

Predicted Molecular Mass: 25.0kDa

Accurate Molecular Mass: 30kDa as determined by SDS-PAGE reducing conditions.

Phenomenon explanation:

The possible reasons that the actual band size differs from the predicted are as follows:

- 1. Splice variants: Alternative splicing may create different sized proteins from the same gene.
- 2. Relative charge: The composition of amino acids may affects the charge of the protein.
- 3. Post-translational modification: Phosphorylation, glycosylation, methylation etc.
- 4. Post-translation cleavage: Many proteins are synthesized as pro-proteins, and then cleaved to give the active form.
- 5. Polymerization of the target protein: Dimerization, multimerization etc.

[USAGE]

Reconstitute in 10mM PBS (pH7.6) to a concentration of 0.1-1.0 mg/mL. Do not vortex.

[STORAGE AND STABILITY]

Storage: Avoid repeated freeze/thaw cycles.

Store at 2-8°C for one month.

Aliquot and store at -80°C for 12 months.

Stability Test: The thermal stability is described by the loss rate. The loss rate was determined by accelerated thermal degradation test, that is, incubate the protein at 37°C for 48h, and no obvious degradation and precipitation were observed. The loss rate is less than 5% within the expiration date under appropriate storage condition.

[SEQUENCE]

QV EIYSFPKDPE IHLSGPLEAG KPITVKCSVA DVYPFDRLEI DLLKGDHLMK SQEFLEDADR KSLETKSLEV TFTPVIEDIG KVLVCRAKLH IDEMDSVPTV RQAVKELQVY ISPKNTVISV NPSTKLQEGG SVTMTCSSEG LPAPEIFWSK KLDNGNLQHL SGNATLTLIA MRMEDSGIYV CEGVNLIGKN RKEVELIVOE KPFTVEIS

[ACTIVITY]

Vascular cell adhesion protein 1 (VCAM-1) is a cell adhesion molecule and a member of the immunoglobulin superfamily. Alternatively spliced forms are known to occur, but the most common form is a type I transmembrane protein with a 674 amino acid extracellular segment that includes seven C2-type immunoglobulin domains. VCAM-1 is expressed by activated endothelial cells and certain other cell types including macrophages, dendritic cells, neurons, smooth muscle cells, fibroblasts, and oocytes. Besides, Integrin Alpha 4 (ITGa4) has been identified as an interactor of VCAM-1, thus a binding ELISA assay was conducted to detect the interaction of recombinant human VCAM-1 and recombinant human ITGa4. Briefly, biotin-linked VCAM-1 were diluted serially in PBS, with 0.01% BSA (pH 7.4).

Duplicate samples of 100 μ I were then transferred to ITGa4-coated microtiter wells and incubated for 1h at 37 $^{\circ}$ C. Wells were washed with PBST 3 times and incubation with Streptavidin-HRP for 30min, then wells were aspirated and washed 5 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37 $^{\circ}$ C. Finally, add 50 μ I stop solution to the wells and read at 450nm immediately. The binding activity of recombinant human VCAM-1 and recombinant human ITGa4 was shown in Figure 1, the EC50 for this effect is 0.261 ug/mL.

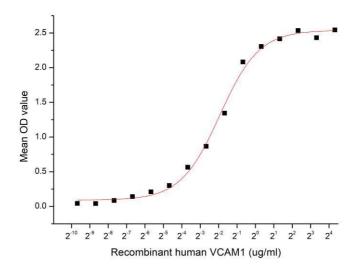


Figure 1. The binding activity of recombinant human VCAM-1 and recombinant human ITGa4

[IDENTIFICATION]

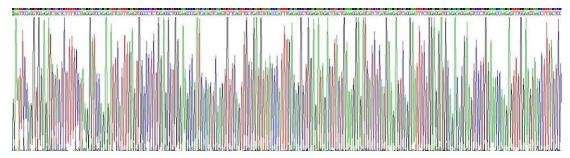


Figure 2. Gene Sequencing (extract)

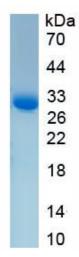


Figure 3. SDS-PAGE

Sample: Active recombinant VCAM1, Human

[IMPORTANT NOTE]

The kit is designed for research use only, we will not be responsible for any issue if the kit was used in clinical diagnostic or any other procedures.