

**APB367Hu61 10µg**

**Active Vascular Endothelial Growth Factor Receptor 2 (VEGFR2)**

**Organism Species: *Homo sapiens* (Human)**

***Instruction manual***

FOR RESEARCH USE ONLY

NOT FOR USE IN CLINICAL DIAGNOSTIC PROCEDURES

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1st Edition (Apr, 2016)

## **[ PROPERTIES ]**

**Source:** Eukaryotic expression.

**Host:** 293F cell

**Residues:** Ala20~Glu764

**Tags:** N-terminal His-tag

**Purity:** >98%

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

**Buffer Formulation:** 10mM PBS, pH7.6, containing 5% trehalose.

**Applications:** Cell culture; Activity Assays.

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

**Predicted isoelectric point:** 6.5

**Predicted Molecular Mass:** 84.9kDa

**Accurate Molecular Mass:** 90kDa 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** ]

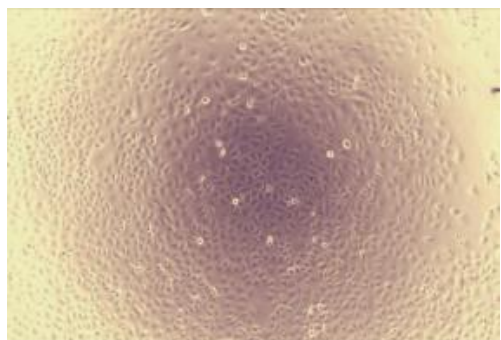
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          A SVGLPSVSLD LPRLSIQKDI LTIKANTTLQ
ITCRGQRDLG WLWPNNQSGS EQRVEVTECS DGLFCKTLTI PKVIGNDTGA
YKCFYRETDL ASVIYVYVQD YRSPFIASVS DQHGVDVYITE NKNKTVVIPC
LGSISNLNVS LCARYPEKRF VPDGNRISWD SKKGFTIPSY MISYAGMVFC
EAKINDESYQ SIMYIVVVVG YRIYDVVLSV SHGIELSVGE KLVLNCTART
ELNVGIDFNW EYPSSKHQHK KLVNRDLKTQ SGSEMKKFLS TLTIDGVTRS
DQGLYTCAAS SGLMTKKNST FVRVHEKPFV AFGSGMESLV EATVGERVRI
PAKYLGYPPP EIKWYKNGIP LESNHTIKAG HVLTIMEVSE RDTGNYTVIL
TNPISKEKQS HVVSLVVYVP PQIGEKSLIS PVDSYQYGTG QTLTCTVYAI
PPPHHIIHWYW QLEEECANEP SQAVSVTNPY PCEEWRVSD FQGGNKIEVN
KNQFALIEGK NKTVSTLVIQ AANVSALYKC EAVNKVGRGE RVISFHVTRG
PEITLQPDMQ PTEQESVSLW CTADRSTFEN LTWYKLGPPQ LPIHVGELPT
PVCKNLDTLW KLNATMFSNS TNDILIMELK NASLQDQGDY VCLAQDRKTK
KRHCVVRQLT VLERVAPTIT GNLENQTTSI GESIEVSCA SGNPPPQIMW
FKDNETLVED SGIVLKDGNR NLTIRVRKE DEGLYTCQAC SVLGCAKVEA
FFIIEGAQEK TNLE
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## [ **ACTIVITY** ]

Vascular Endothelial Growth Factor Receptor 2 (VEGFR2) also known as kinase insert domain receptor acts as a cell-surface receptor for VEGFA, VEGFC and VEGFD. VEGFR2 functions as the primary mediator of vascular endothelial growth factor activation in endothelial cells. Regulation of VEGFR-2 expression appears critical in mitogenesis, differentiation, and angiogenesis. To test the effect on inhibit the VEGF-dependent proliferation of endothelium cells, ECV-304 cells were seeded into triplicate wells of 96-well plates at a density of 5,000 cells/well and allowed to attach, replaced with serum-free overnight, then the medium was replaced with 2% serum standard DMEM including 1 $\mu$ g/mL Vascular Endothelial Growth Factor C (VEGFC) and various concentrations of recombinant human VEGFR2. After incubated for 96h, cells were observed by inverted microscope and cell proliferation was measured by Cell Counting Kit-8 (CCK-8). Briefly, 10 $\mu$ L of CCK-8 solution was added to each well of the plate, then the absorbance at 450nm was measured using a microplate reader after incubating the plate for 1-4 hours at 37 $^{\circ}$ C. Proliferation of ECV-304 cells after incubation with VEGFR2 for 96h observed by inverted microscope was shown in Figure 1. Cell viability was assessed by CCK-8 (Cell Counting Kit-8) assay after incubation with recombinant VEGFR2 for 96h. The result was shown in Figure 2. It was obvious that VEGFR2 significantly inhibit cell viability of ECV-304.



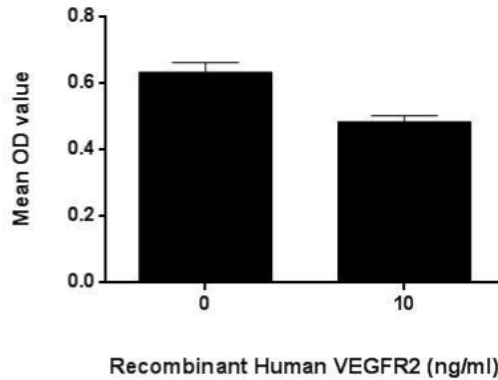
A



B

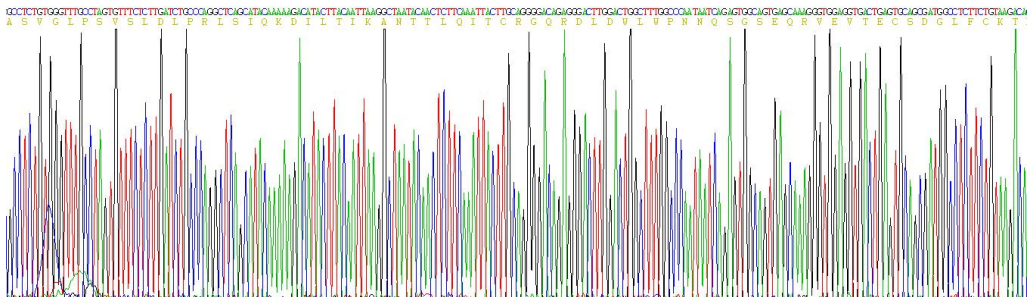
**Figure 1. Cell proliferation of ECV-304 cells inhibit by VEGFR2.**

- (A) ECV-304 cells cultured in DMEM, stimulated with 10ng/mL VEGFR2 for 96h;
- (B) Unstimulated ECV-304 cells cultured in DMEM for 96h.

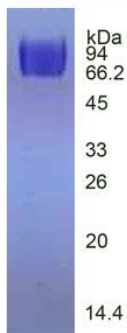


**Figure 2. VEGFR2 inhibit VEGF-dependent proliferation of ECV-304 cells.**

### [ IDENTIFICATION ]



**Figure 3. Gene Sequencing (extract)**



**Figure 4. SDS-PAGE****Sample: Active recombinant VEGFR2, Human****[ IMPORTANT NOTE ]**

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