

Human VEGF Receptor 2 protein, His tag (active)

Cat. No. GTX00258-pro

Applications	Functional Assay	Package 10 μg
Species	Human	13

Applications

Application Note

Vascular Endothelial Growth Factor Receptor 2 (VEGFR2) also known as kinase insert domain receptor is a VEGF receptor. There are three main subtypes of VEGFR, numbered 1, 2 and 3. All members of the VEGF family stimulate cellular responses by binding to VEGF receptors on the cell surface, causing them to dimerize and become activated through transphosphorylation. VEGFR2 appears to mediate almost all of the known cellular responses to VEGF. Besides, vascular Endothelial Growth Factor C (VEGFC) has been identified as an interactor of VEGFR2, thus a binding ELISA assay was conducted to detect the interaction of recombinant human VEGFR2 and recombinant human VEGFC. Briefly, VEGFR2 were diluted serially in PBS, with 0. 01% BSA (pH 7. 4). Duplicate samples of 100 μ l were then transferred to VEGFC-coated microtiter wells and incubated for 2h at 37°C. Wells were washed with PBST and incubated for 1h with anti-VEGFR2 pAb, then aspirated and washed 3 times. After incubation with HRP labelled secondary antibody, wells were aspirated and washed 3 times. With the addition of substrate solution, wells were incubated 15-25 minutes at 37°C. Finally, add 50 μ l stop solution to the wells and read at 450nm immediately. The binding activity of VEGFR2 and VEGFC was in a dose dependent manner.

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 5000 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°C. Proliferation of ECV-304 cells after incubation with VEGFR2 for 96h observed by inverted microscope. Cell viability was assessed by CCK-8 (Cell Counting Kit-8) assay after incubation with recombinant VEGFR2 for 96h. And VEGFR2 significantly inhibit cell viability of ECV-304.

Observed MW (kDa) 33 kDa.

Properties		
Form	Lyophilized powder	
Buffer	Reconstitute with 20mM Tris and 150mM NaCl to 0.1-1.0mg/ml. Do not vortex. Lyophilized from 20mM Tris, 150mM NaCl, 1mM EDTA, 1mM DTT, 0.01% SKL, 5% Trehalose.	
Preservative	ProClin 300	
Storage	For short-term storage (1-2 weeks), store at 4°C. For long-term storage, store at -20°C or below. After reconstitution, keep as concentrated solution. Avoid freeze-thaw cycles.	
Region/Sequence	N-terminal His-Tag; Asn46~Thr320 (NP_002244.1)	
Expression System	E. coli	
Purity	> 97%	
Endotoxin	< 1 EU/μg	



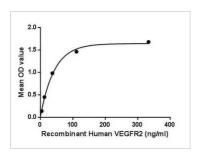
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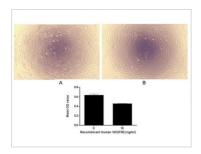
Conjugation	Unconjugated
Note	For laboratory use only. Not for any clinical, therapeutic, or diagnostic use in humans or animals. Not for animal or human consumption.

DATA IMAGES



GTX00258-pro Functional Assay Image

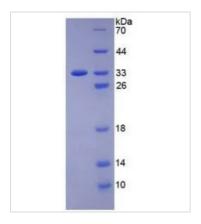
Functional ELISA analysis of GTX00258-pro Human VEGF Receptor 2 protein (active) which can bind immobilized VEGFC protein.



GTX00258-pro Functional Assay Image

The inhibition effect of GTX00258-pro Human VEGF Receptor 2 protein (active) on VEGF-dependent proliferation. Cell viability was measured by Cell Counting Kit-8 (CCK-8).

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



GTX00258-pro Image

SDS-PAGE analysis of GTX00258-pro Human VEGF Receptor 2 protein (active).



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