# Biotinylated Human VEGF165 Protein, His,Avitag™ (HPLC verified)

Catalog # VE5-H82Q0



### Synonym

RP1-261G23.1,MGC70609,MVCD1,VEGFA,VPF

#### Source

Biotinylated Human VEGF165, His, Avitag(VE5-H82Q0) is expressed from human 293 cells (HEK293). It contains AA Ala 27 - Arg 191 (Accession # P15692-4).

Predicted N-terminus: His

#### **Molecular Characterization**



This protein carries a polyhistidine tag at the N-terminus, followed by an Avi tag (Avitag<sup>TM</sup>).

The protein has a calculated MW of 22.4 kDa (monomer). The protein migrates as 23-28 kDa (monomer) under reducing (R) condition, and 40-55 kDa (homodimer) when calibrated against <u>Star Ribbon Pre-stained Protein Marker</u> under non-reducing (NR) condition (SDS-PAGE) due to glycosylation.

#### Labeling

Biotinylation of this product is performed using Avitag<sup>TM</sup> technology. Briefly, the single lysine residue in the Avitag is enzymatically labeled with biotin.

#### **Protein Ratio**

Passed as determined by the HABA assay / binding ELISA.

## **Purity**

>95% as determined by SDS-PAGE.

>95% as determined by SEC-HPLC.

#### **Formulation**

Lyophilized from  $0.22~\mu m$  filtered solution in PBS, pH7.4 with trehalose as protectant.

Contact us for customized product form or formulation.

#### Reconstitution

Please see Certificate of Analysis for specific instructions.

For best performance, we strongly recommend you to follow the reconstitution protocol provided in the CoA.

#### **Storage**

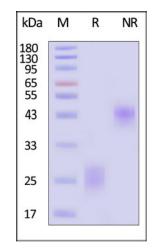
For long term storage, the product should be stored at lyophilized state at -20°C or lower.

Please avoid repeated freeze-thaw cycles.

This product is stable after storage at:

- -20°C to -70°C for 12 months in lyophilized state;
- -70°C for 12 months under sterile conditions after reconstitution.

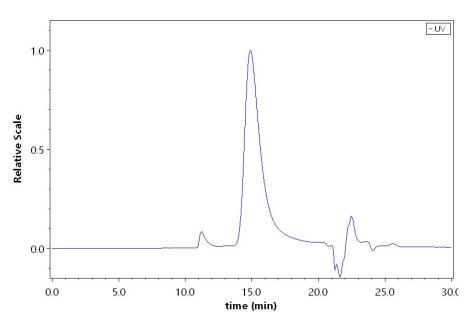
# SDS-PAGE



Biotinylated Human VEGF165, His, Avitag on SDS-PAGE under reducing (R) and non-reducing (NR) conditions. The gel was stained with Coomassie Blue. The purity of the protein is greater than 95% (With <u>Star Ribbon Pre-stained</u> <u>Protein Marker</u>).

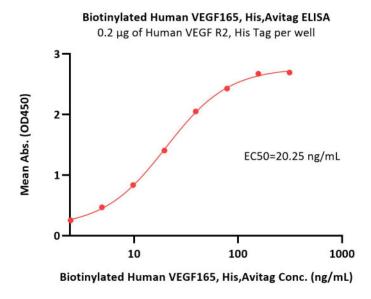
# **Bioactivity-ELISA**

### **SEC-HPLC**

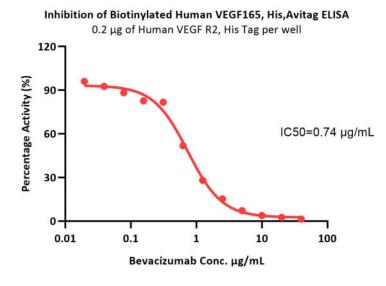


The purity of Biotinylated Human VEGF165, His, Avitag (Cat. No. VE5-H82Q0) was greater than 95% as determined by SEC-HPLC.





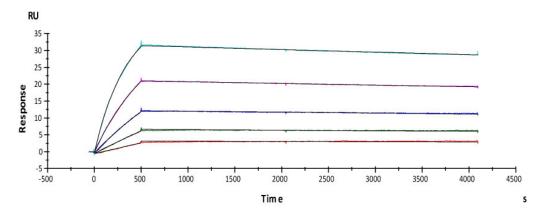
Immobilized Human VEGF R2, His Tag (Cat. No. KDR-H5227) at 2  $\mu$ g/mL (100  $\mu$ L/well) can bind Biotinylated Human VEGF165, His,Avitag (Cat. No. VE5-H82Q0) with a linear range of 2-39 ng/mL (QC tested).



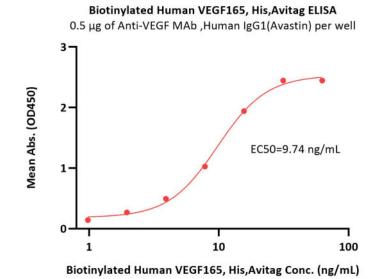
Immobilized Human VEGF R2, His Tag (Cat. No. KDR-H5227) at 2  $\mu$ g/mL (100  $\mu$ L/well), can bind pre-mixed increasing concentrations of Bevacizumab and 0.05  $\mu$ g/mL (100  $\mu$ L/well) Biotinylated Human VEGF165, His,Avitag (Cat. No. VE5-H82Q0) with a half maximal inhibitory concentration (IC50) of 0.74  $\mu$ g/mL (Routinely tested).

# **Bioactivity-SPR**

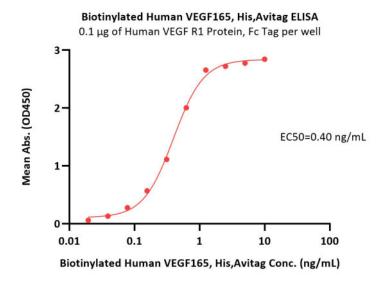




Immobilized Biotinylated Human VEGF165, His, Avitag (Cat. No. VE5-H82Q0) on CM5 Chip via Streptavidin, can bind Avastin with an affinity constant of 0.417 nM as determined in SPR assay (Biacore T200) (Routinely tested).



Immobilized Anti-VEGF MAb, Human IgG1 (Avastin) at 5  $\mu$ g/mL (100  $\mu$ L/well) can bind Biotinylated Human VEGF165, His,Avitag (Cat. No. VE5-H82Q0) with a linear range of 1-15.6  $\mu$ g/mL (Routinely tested).



Immobilized Human VEGF R1 Protein, Fc Tag (Cat. No. VE1-H5253) at 1  $\mu$ g/mL (100  $\mu$ L/well) can bind Biotinylated Human VEGF165, His,Avitag (Cat. No. VE5-H82Q0) with a linear range of 0.02-1 ng/mL (Routinely tested).

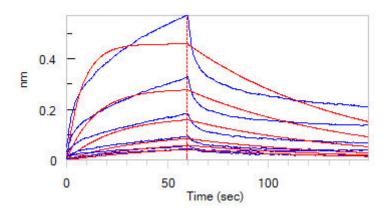


# Biotinylated Human VEGF165 Protein, His,Avitag™ (HPLC verified)

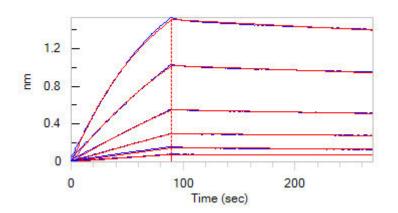
Catalog # VE5-H82Q0



## **Bioactivity-BLI**



Loaded Biotinylated Human VEGF165, His, Avitag (Cat. No. VE5-H82Q0) on SA Biosensor, can bind Human Neuropilin-1, His Tag (Cat. No. NR1-H5228) with an affinity constant of 21.1 nM as determined in BLI assay (ForteBio Octet Red96e) (Routinely tested).



Loaded Biotinylated Human VEGF165, His, Avitag (Cat. No. VE5-H82Q0) on SA Biosensor, can bind Human VEGF R1, His Tag with an affinity constant of 3.36 nM as determined in BLI assay (ForteBio Octet Red96e) (Routinely tested).

## **Background**

VEGF165 is the most abundant splice variant of VEGF-A. VEGF165 is produced by a number of cells including endothelial cells, macrophages and T cells. VEGF165 is involved in angiogenesis, vascular endothelial cell survival, growth, migration and vascular permeability. VEGF gene expression is induced by hypoxia, inflammatory cytokines and oncogenes. VEGF165 binds to heparan sulfate and is retained on the cell surface and in the extracellular matrix. VEGF165 binds to the receptor tyrosine kinases, VEGFR1 and VEGFR2. VEGF165 is the only splice variant that binds to co-receptors NRP-1 and NRP-2 that function to enhance VEGFR2 signaling. Binding of VEGF165 to VEGFR1 and VEGFR2 leads to activation of the PI3K/AKT, p38 MAPK, FAK and paxillin. VEGF plays a key role in tumor angiogenesis in many cancers.

