Sterile

#8908

Human Vascular Endothelial Growth Factor-121 (hVEGF$_{121}$)

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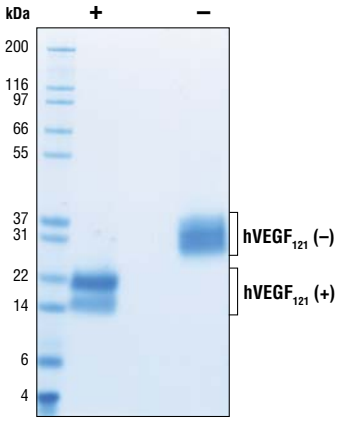
For Research Use Only. Not For Use In Diagnostic Procedures.

Source: Recombinant human VEGF$_{121}$ (hVEGF$_{121}$) Ala207-Arg327 (Accession #NP_001020541.2) was expressed in human 293 cells at Cell Signaling Technology.

Molecular Characterization: Recombinant hVEGF$_{121}$ contains no "tags" and the nonglycosylated protein has a calculated MW of 14,057. DTT-reduced protein migrates as a 14-22 kDa polypeptide. Heterogeneity in SDS-PAGE is due to glycosylation. The non-reduced cystine-linked homodimer migrates as a 30-36 kDa protein. The expected amino-terminal APMAE of recombinant hVEGF$_{121}$ was verified by amino acid sequencing.

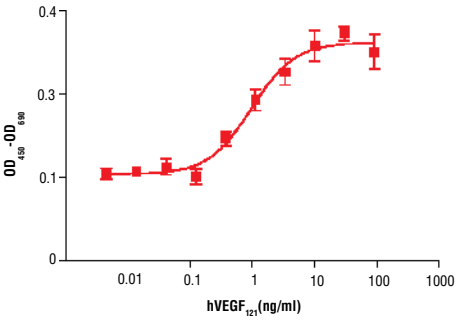
Endotoxin: Less than 0.01 ng endotoxin/1μg hVEGF$_{121}$.

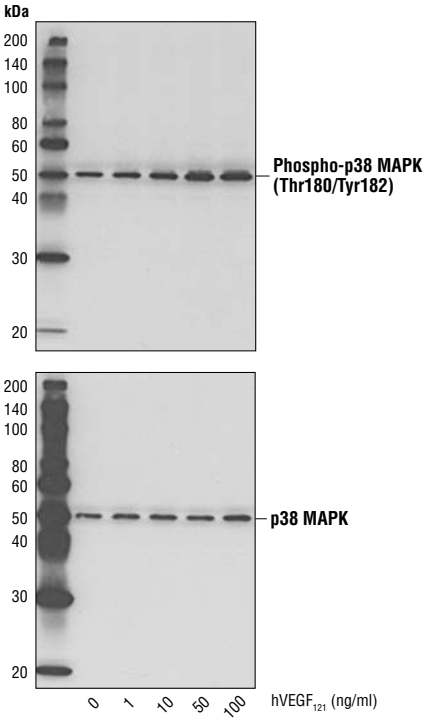
Purity: >98% as determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant hVEGF$_{121}$. All lots are greater than 98% pure.



The purity of recombinant hVEGF$_{121}$ was determined by SDS-PAGE of 6 μg reduced (+) and non-reduced (-) recombinant hVEGF$_{121}$ and staining overnight with Coomassie Blue.

Bioactivity: The bioactivity of recombinant hVEGF$_{121}$ was determined in a cell proliferation assay using HUVEC. The ED$_{50}$ of each lot is between 0.5-2 ng/ml.





Western blot analysis of extracts from HUVEC untreated or treated with hVEGF$_{121}$ for 15 minutes, using Phospho-p38 MAPK (Thr180/Tyr182) (3D7) Rabbit mAb #9215 (upper) and p38 MAPK Antibody #9212 (lower).

◀ *The proliferation of HUVEC treated with increasing concentrations of hVEGF$_{121}$ was assessed. After 72-hour treatment with hVEGF$_{121}$ cells were incubated with a tetrazolium salt and the OD$_{450}$ - OD$_{650}$ was determined.*

Formulation: With carrier: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2 containing 20 μg BSA per 1 μg hVEGF$_{121}$.

Carrier free: Lyophilized from a 0.22 μm filtered solution of PBS, pH 7.2.

Reconstitution:
With carrier: Add sterile PBS, or PBS containing 1% bovine or human serum albumin or 5-10% FBS to a final hVEGF$_{121}$ concentration of greater than 50 μg/ml. Solubilize for 30 minutes at room temperature with occasional gentle vortexing.

Carrier free: Add sterile PBS, or PBS containing protein to minimize absorption of hVEGF$_{121}$ to surfaces. Solubilize for 30 minutes at room temperature with occasional gentle vortexing. Stock hVEGF$_{121}$ should be greater than 50 μg/ml.

Storage: Stable in lyophilized state at -20°C for 1 year after receipt. Sterile stock solutions reconstituted with carrier protein are stable at 4°C for 2 months and at -20°C for 6 months. Avoid repeated freeze-thaw cycles.

Maintain sterility. Storage at -20°C should be in a manual defrost freezer.

Applications: Optimal concentration for the desired application should be determined by the user.

Background: VEGF$_{121}$ is the second most abundant splice variant of VEGF-A (1,2). VEGF$_{121}$ is produced by endothelial cells, macrophages, T-cells and other cell types. VEGF$_{121}$ is involved in angiogenesis, vascular endothelial cell survival, growth, migration and vascular permeability (1). VEGF$_{121}$ expression is induced by hypoxia, inflammatory cytokines and through oncogene products in tumors (1-3). VEGF$_{121}$ binds to VEGFR1 and VEGFR2 receptor tyrosine kinases (1). Binding of VEGF$_{121}$ to VEGFR1 and VEGFR2 leads to activation of pathways involving PI3K/AKT, P38 MAPK, and FAK (1). VEGF plays a key role in tumor angiogenesis in many cancers (2).

Background References:

(1) Takahashi, H. and Shibuya, M. (2005) *Clin Sci (Lond)* 109, 227-41.

(2) Neufeld, G. et al. (1999) *FASEB J* 13, 9-22.

(3) Robinson, C.J. and Stringer, S.E. (2001) *J Cell Sci* 114, 853-65.