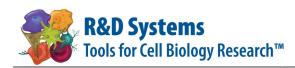
Human Phospho-VEGF R2/KDR/Flk-1 (Y1214) Antibody



Antigen Affinity-purified Polyclonal Rabbit IgG Catalog Number: AF1766

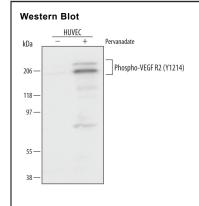
DESCRIPTION			
Species Reactivity	Human		
Specificity	Detects human VEGF R2/KDR/Flk-1 in Western blots when phosphorylated at Y1214.		
Source	Polyclonal Rabbit IgG		
Purification	Antigen Affinity-purified		
Immunogen	Phosphopeptide containing human VEGF R2/KDR/Flk-1 Y1214 site		
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 µm filtered solution in PBS.		

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

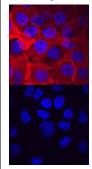
	Recommended Concentration	Sample
Western Blot	0.5 μg/mL	See Below
Immunocytochemistry	5-15 μg/mL	See Below
Intracellular Staining by Flow Cytometry	2.5 μg/10 ⁶ cells	See Below

DATA



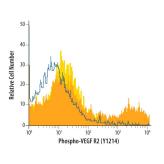
Detection of Human Phospho-VEGF R2/KDR/Flk-1 (Y1214) by Western Blot. Western blot shows lysates of HUVEC human umbilical vein endothelial cells untreated (-) or treated (+) with 100 µM pervanadate (PV) for 10 minutes. PVDF membrane was probed with 0.5 μg/mL of Rabbit Anti-Human Phospho-VEGF R2/KDR/Flk-1 (Y1214) Antigen Affinity-purified Polyclonal Antibody, followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). Specific bands were detected for Phospho-VEGF R2/KDR/ Flk-1 (Y1214) at approximately 210 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry



Phospho-VEGF R2/KDR/FIk-1 (Y1214) in A431 Human Cell Line. VEGF R2/KDR/FIk-1 phosphorylated at Y1214 was detected in immersion fixed A431 human epithelial carcinoma cell line untreated (lower panel) or treated (upper panel) with pervanadate using Rabbit Anti-Human Phospho-VEGF R2/KDR/FIk-1 (Y1214) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1766) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Rabbit IgG Secondary Antibody (red; Catalog # NL004) and counterstained with DAPI (blue). View our protocol for Fluorescent ICC Staining of Cells on Coverslips.

Intracellular Staining by Flow Cytometry



Detection of Phospho-VEGF R2/ KDR/Flk-1 in pervanadate-treated HUVEC by Flow Cytometry.

HUVEC human umbilical vein endothelial cells were unstimulated (yellow filled histogram) or treated with 100 µM pervanadate for 15 minutes (orange filled histogram), then stained with Rabbit Anti-Human Phospho-VEGF R2/KDR/Fik-1

(Y1214) Antigen Affinity-purified Polyclonal Antibody (Catalog # AF-1766), or control antibody (Catalog # AB-105-C, open histogram), followed by Phycoerythrin-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # F0110). To facilitate intracellular staining, cells were fixed with paraformaldehyde and permeabilized with methanol.

PREPARATION AND STORAGE

Reconstitution Reconstitute at 0.2 mg/mL in sterile PBS.

Shipping The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

*Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C

- 12 months from date of receipt, -20 to -70 °C as supplied
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 6 months, -20 to -70 °C under sterile conditions after reconstitution.

RED

Rev. 3/13/2015 Page 1 of 2



Human Phospho-VEGF R2/KDR/Flk-1 (Y1214) Antibody

Antigen Affinity-purified Polyclonal Rabbit IgG Catalog Number: AF1766

BACKGROUND

VEGF R2 (KDR/Flk-1), VEGF R1 (Flt-1) and VEGF R3 (Flt-4) belong to the class III subfamily of receptor tyrosine kinases (RTKs). All three receptors contain seven immunoglobulin-like repeats in their extracellular domains and kinase insert domains in their intracellular regions. The expression of VEGF R1, 2, and 3 is almost exclusively restricted to the endothelial cells. These receptors are likely to play essential roles in vasculogenesis and angiogenesis.

VEGF R2 cDNA encodes a 1356 amino acid (aa) residue precursor protein with a 19 aa residue signal peptide. Mature VEGF R2 is composed of a 745 aa residue extracellular domain, a 25 aa residue transmembrane domain and a 567 aa residue cytoplasmic domain. In contrast to VEGF R1 which binds both P/GF and VEGF with high affinity, VEGF R2 binds VEGF but not P/GF with high affinity. The recombinant soluble VEGF R2/Fc chimera binds VEGF with high affinity and is a potent VEGF antagonist.

References:

1. Ferra, N. and R. Davis-Smyth (1997) Endocrine Reviews 18:4



Rev. 3/13/2015 Page 2 of 2