Anti-human VEGFR-2/KDR (#3 (4H3))

20210818DS

FOR RESEARCH ONLY! NOT FOR HUMAN USE!

Cat.-no.:	**101-M32**
Size:	100 µg
Lot. No.:	According to product label
Country of origin:	Germany

Preparation: Monoclonal antibodies were produced with the help of BALB/c mice using recombinant human soluble extracellular KDR (D7) as the immunizing antigen

Target Background

Synonyms:	Vascular endothelial growth factor receptor 2

Disruption of the precise balance of positive and negative molecular regulators of blood and lymphatic vessel growth can lead to myriad diseases. Although dozens of natural inhibitors of hemangiogenesis have been identified, an endogenous selective inhibitor of lymphatic vessel growth has not to our knowledge been previously described. A splice variant of the gene encoding vascular endothelial growth factor receptor-2 (VEGFR-2) that encodes a secreted form of the protein, designated endogenous soluble VEGFR-2 (esVEGFR-2/KDR) has been described. The endogenous soluble esKDR inhibits developmental and reparative lymphangiogenesis by blocking VEGF-C function. Tissue-specific loss of esKDR in mice induced, at birth, spontaneous lymphatic invasion of the normally alymphatic cornea and hyperplasia of skin lymphatics without affecting blood vasculature. Administration of esKDR inhibited lymphangiogenesis but not hemangiogenesis induced by corneal suture injury or transplantation, enhanced corneal allograft survival and suppressed lymphangioma cellular proliferation. Naturally occurring esKDR thus acts as a molecular uncoupler of blood and lymphatic vessels; modulation of esKDR might have therapeutic effects in treating lymphatic vascular malformations, transplantation rejection and, potentially, tumor lymphangiogenesis and lymphedema.

References

1. Shibata et al, BMC Medicine 8 (2010)
2. Albaquerque et al, Nature Med 2009
3. Ebos et al, Mol Cancer Res 2 (2004)
4. Ebos et al, Cancer res 68 (2008).
5. Benzinger et al., BBA 1466:71, 2000

Database References Antigen

Protein RefSeq:	NP_002241.1
Uniprot ID:	P22333
mRNA RefSeq:	NM_002253.2

Product Specifications

Species reactivity	human
Clone/Ab feature	IgG_1; #3 (4H3)
Cross reactivity	ND
Host	mouse
Clonality	monoclonal
Purification	Protein G purified
Immunogen	recombinant human soluble KDR (D7) (RT# S01-003)
Formulation	lyophilized
Buffer	PBS

Stability: The lyophilized antibody is stable for at least 2 years at -20°C. After sterile reconstitution the antibody is stable at 2-8°C for up to 6 months. Frozen aliquots are stable for at least 6 months when stored at -20°C. Addition of a carrier protein or 50% glycerol is recommended for frozen aliquots.

Reconstitution: Centrifuge vial prior to opening. Reconstitute in sterile water to a concentration of 0.1-1.0 mg/ml.

AVOID REPEATED FREEZE AND THAW CYCLES!

Specificity: The antibody will detect native human VEGFR-2/KDR on the surface of different human cell types.

Applications

Western Blot:	Use at 2-5 µg/ml
FACS:	Use at 2-5 µg/ml
ELISA:	Use at 1-5 µg/ml
IF/IHC	Use at 1-5 µg/ml

NOTE: OPTIMAL DILUTIONS SHOULD BE DETERMINED BY EACH LABORATORY FOR EACH APPLICATION!

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Handling/Applications

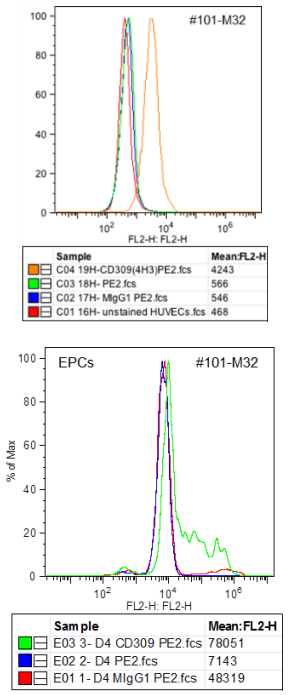

Fig. 1: FACS analysis of VEGFR-2/KDR expression in HUVE cells (upper level) and EPCs derived from PBMcs (lower level) [5µg/ml #101-M32; 5µg/ml PE goat anti-mouse IgG].
The experiment was performed by Trisha M. Westerhof, University of California, Irvine.

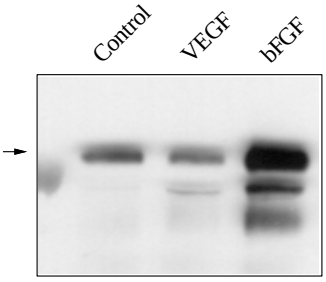

Fig. 2: Up-regulation of VEGFR-2 in primary HUVECs by bFGF. Freshly isolated HUVECs (passage 1) were cultured in EBM. Subconfluent cultures were stimulated with VEGF (5 ng/ml) or bFGF (10 ng/ml) for 3 days. Total lysate was prepared and subjected to immunoprecipitation (anti-human VEGFR-2 **[#101-M32**] followed by Western blotting (anti-human VEGFR-2 **[#101-M34**]. (Bernhard Barleon et. al., unpublished data!)

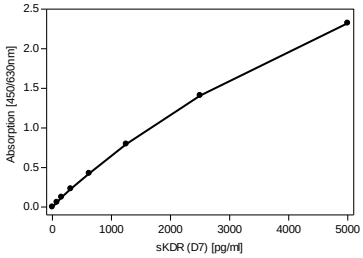

Fig. 3: VEGFR-2/KDR Sandwich-ELISA using soluble KDR (D7) [Cat# S01-002] as standard. Mouse anti-human VEGFR-2 [Cat# 101-M32] was used as capture antibody, Biotinylated rabbit anti-human VEGFR-2 [Cat# 102-PABi18] was used for detection.

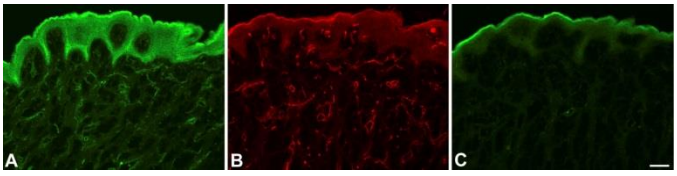

Fig. 4: Consecutive sections of unfixed, human foreskin. A) Staining with anti-soluble VEGFR2/KDR antibodies (#102-PA19). Note signal in epidermis and vessels. B) Staining with anti-membrane-bound VEGFR-2/KDR (#101-M32). Note staining in vessels. C) Negative control. Note non-specific fluorescence in the hornified layer of the epithelium.
Provided by Prof. J. Wilting, Göttingen, Germany.

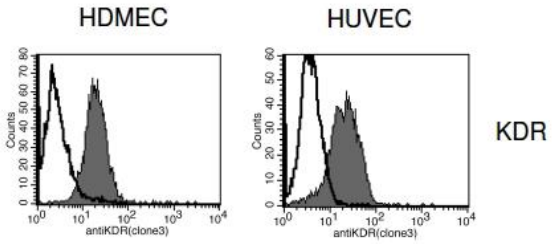

Fig. 5: FACS analysis of VEGFR-2/KDR expression in human primary dermal microvascular and umbilical vein endothelial cells.

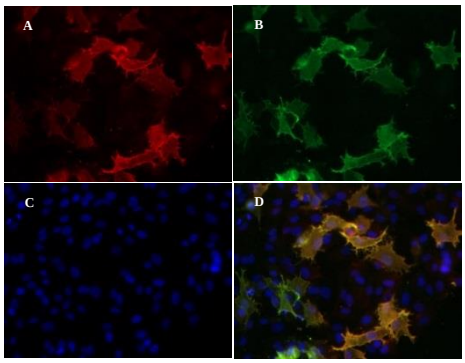

Fig. 6: IF double staining of human KDR in a co-culture of PAE-Flt-1, PAE-KDR and PAE-FLT-4 with (A) mouse anti-human KDR [#101-M32], (B) rabbit anti-human KDR [B, #102-PA18AG], (C) DAPI and (D) merged.
Conjugated secondary antibody: goat anti-rabbit ALEXA Flour (1:600) [Dianova] and goat anti-mouse PE [Santa Cruz].

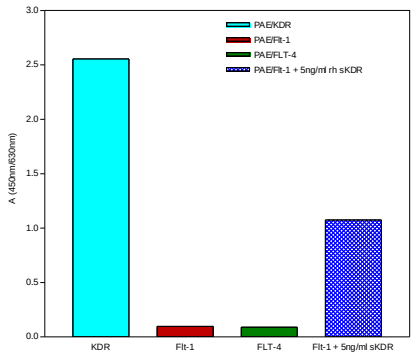

Fig. 7: Total lysate from PAE/KDR cells was measured for expression of VEGFR-1/KDR by a standard Sandwich ELISA using a mouse monoclonal anti-human VEGFR-1 antibody (#101-M32) for capturing and a Biotin conjugated rabbit polyclonal anti-human VEGFR-1 antibody (#102-PABi18) for detection. Total lysate from PAE/Flt-1 and PAE/FLT-4 was used as negative control. As positive control total lysate from PAE/Flt-1 was spiked with recombinant human sKDR.