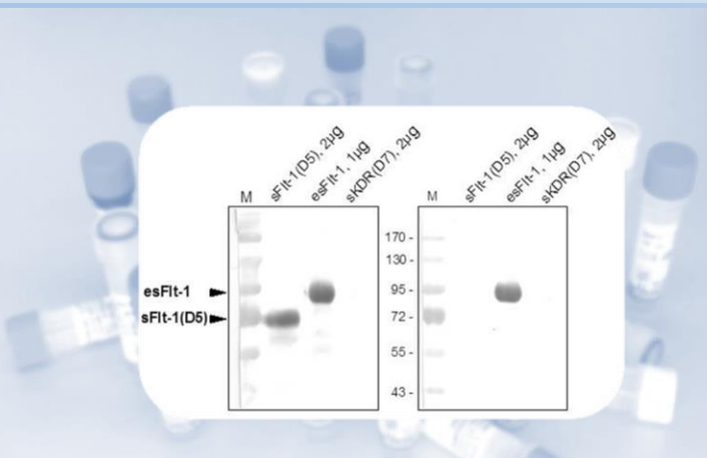

Western Blot Analysis with recombinant human sFlt-1(D5) **(#S01-011)**, sFlt-1 **(#S01-010)** and sKDR(D7) **(#S01-001)**. Our rabbit anti-human sFlt-1 **(#102-PA21,** right) only detects endogenous sFlt-1 whereas mouse anti-human Flt-1 (**#101-M30,** left) also detects sFlt-1(D5). There is no cross reactivity with sKDR(D7).

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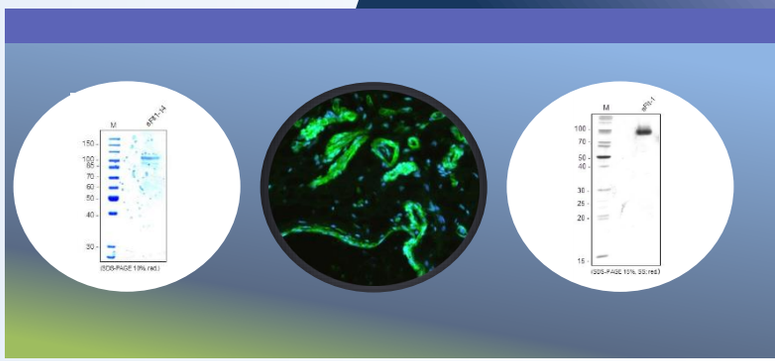
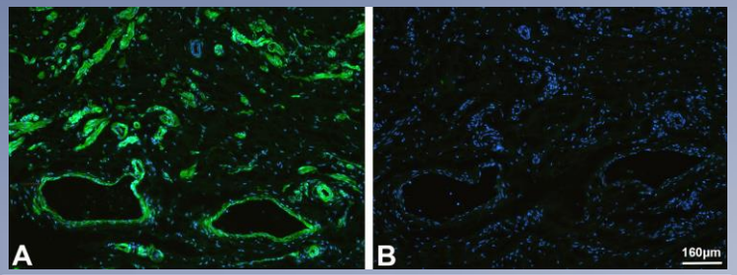


Human sFlt-1/sFlt1-14 and antibodies

Human sFlt-1/sFlt1-14

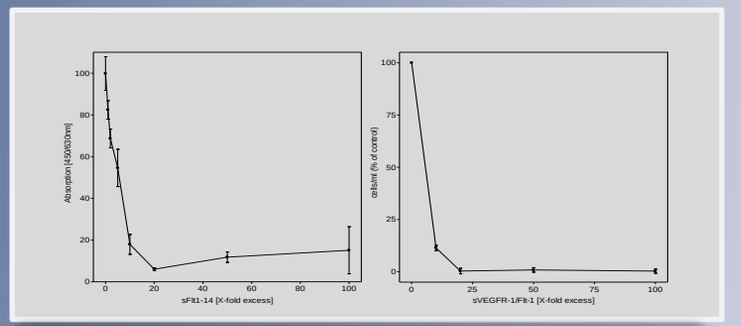
VEGFR-1 is the dominant VEGF receptor on monocytes and responsible for chemotaxis and tissue factor activation. It also plays an important role for signaling and recruitment of stem cells and endothelial precursor cells either alone or in combination with other VEGF receptors. The existence of a naturally occurring soluble Flt-1 (sFlt-1) is known since 1993. This soluble splice product is involved in the fine tuning of VEGF activity during the angiogenic cascade and a prognostic marker for tumor progression in breast cancer. In 2008 a second soluble Flt-1 form was identified, sFlt1-14. Both are differentially expressed and distributed in human tissue. sFlt1-14 is highly placenta-specific and seems to play a role in preeclampsia, whereas sFlt-1 is expressed in several tissues. The therapeutic potential of sFlt-1 as an anti-angiogenic agent has been validated by an increasing number of preclinical studies. Anti-angiogenic therapy changes the concentration of circulating VEGF, PlGF, sFlt-1, sKDR and even sFLT-4. Inactivation of the VEGFR-1 by homologous recombination in mouse has shown that VEGFR-1 is essential for vessel differentiation and maturation, as embryos die around E10 and lack functional blood vessel. Ligands for VEGFR-1 include several isoforms of PlGF, VEGF-A and VEGF-B.



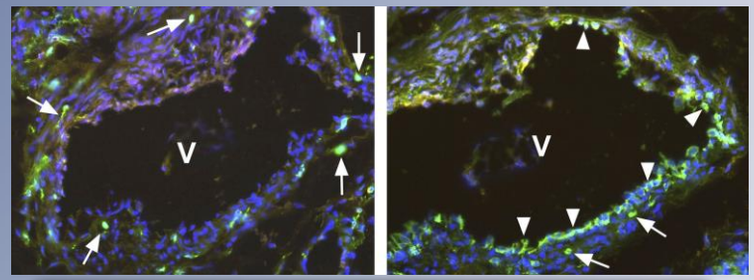
Immunofluorescence staining (green) of cryo-sections of human foreskin with mouse anti-human sFlt1-14 antibody **[#101-M29]** and counter staining of nuclei with Dapi (blue). (B) Control; Fixation: 4% PFA (app. 25min); Dilution: 1:500. Staining in smooth muscle cells, the perineurium of nerves and the Stratum basale of the epidermis as well as some scattered cells.

The experiment was performed by the research group of Prof. Dr. J. Wilting and M. Lohrberg, University Göttingen, Germany.

Functional ELISA: Recombinant human sFlt-1(D5) **[#S01-012]**, sFlt-1 [**#S01-010**] and sFlt1-14 **[#S01-072]** were coated with 1µg/ml in PBS. The sFlt1-14 specific mouse anti-human sFlt1-14 antibody **[#101-M29]** generated against a peptide from the unique C terminal end was used with 2µg/ml, the Flt-1 specific mouse anti-human Flt-1 #EWI [**#101-M30**] recognizing all Flt-1 proteins was used as control [1µg/ml].

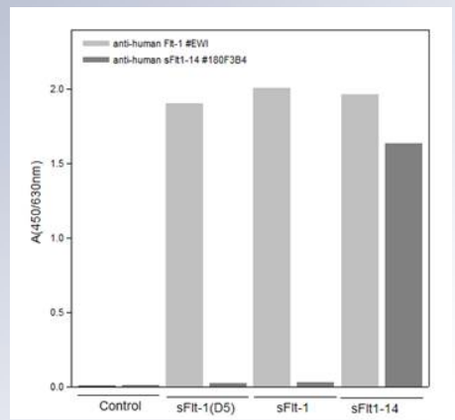


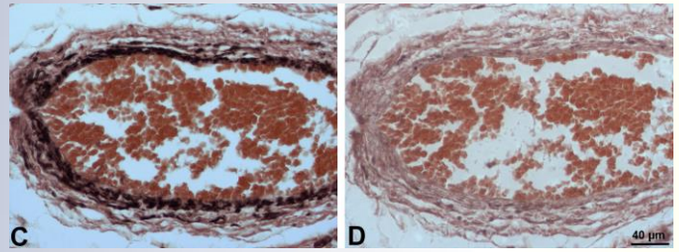
Blocking activity was shown for the two endogenously occurring soluble VEGFR-1 isoforms sFlt1-14 (**Cat# S01-072**) and sFlt-1 (**Cat# S01-010**) by the inhibition of $VEGF_{165}$-induced proliferation of primary human dermal lymphatic endothelial cells (HDLEC).



Immunofluorescence staining (green) of two neighboring sections of a human vein (V), located near a hemangioma. The antibody against the soluble Flt-1 marked single cells (arrows) within the media and adventitia of the vein. The antibody against the membrane-bound Flt-1 marked single cells (arrows) and the endothelium (arrowhead) of the vein. Cell nuclei are stained with DAPI (blue).

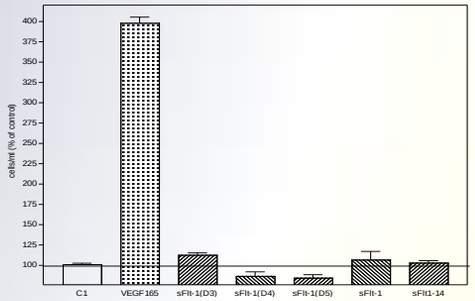
Soluble & endogenous



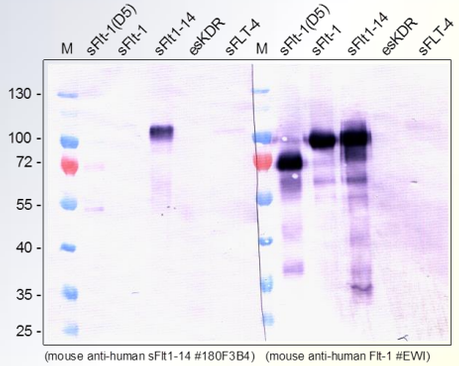


Immunoperoxidase staining of paraffin-embedded sections of human foreskin with mouse anti-human sFlt1-14 antibody. (D) Control; Fixation: 4% PFA (app. 25min); Dilution: 1:100-500. The staining in smooth muscle cells is visible.

The experiment was performed by the research group of Prof. Dr. J. Wilting and M. Lohrberg, University Göttingen, Germany.



Blocking activity of different forms of soluble Flt-1 receptor (all available under www.reliatech.de) as shown by the inhibition of $VEGF_{165}$-induced proliferation of primary HUVECs.



Western Blot Analysis of rec. human sFlt-1(D5) **[#S01-012]**, sFlt-1 [**#S01-010**], sFlt1-14 [**#S01-072**], sKDR(D7) [**#S01-002**] and sFLT-4 [**#S01-018**] using a mouse anti-human sFlt1-14 antibody [**#101-M29**] generated against a peptide from the unique C terminal end (left panel) and a mouse anti-human Flt-1 [**#101-M30**] recognizing all Flt-1 proteins.

Anti-human sFlt-1/sFlt1-14