

VEGF-A splice variants bind VEGFRs with differential affinities

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Supplementary Tables

Supplementary Table S1. VEGF-A_{xxx}:VEGFR binding kinetics and affinities. Kinetic constants were obtained by performing global kinetic analysis using the BIAevaluation software across binding sensograms obtained at 10 nM, 20 nM, and 40 nM for each ligand-receptor pair. Affinity constants were calculated as the ratio of the association and dissociation rates. Rate constants where the χ^2 -to- $R_{\max}$ ratio exceeds 1.0 are shown *in italics*, indicating interactions that are dominated by non-specific interactions. Ligand-receptor pairs that produced negative association curves were not analyzed with global fitting (indicated by “NI” for no interaction). All values are presented as the arithmetic mean $\pm$ standard error of the mean (SEM). (*p < 0.05, **p < 0.01, ***p < 0.001; each concentration series was performed in triplicate).

	VEGFR1			VEGFR2		
	k_a (M ⁻¹ s ⁻¹)	k_d (s ⁻¹)	K_D (M)	k_a (M ⁻¹ s ⁻¹)	k_d (s ⁻¹)	K_D (M)
-A ₁₆₅	4.0 $\pm$ 0.04 * 10 ⁵	4.0 $\pm$ 0.1 * 10 ⁻⁷	1 $\pm$ 0.3 * 10 ⁻¹²	9.7 $\pm$ 0.3 * 10 ⁵	9.5 $\pm$ 0.2 * 10 ⁻⁶	9.8 $\pm$ 0.4 * 10 ⁻¹²
-A ₁₂₁	4.1 $\pm$ 0.3 * 10 ⁵	1.9 $\pm$ 0.07 * 10 ⁻³	3.7 $\pm$ 0.3 * 10 ⁻⁹	4.5 $\pm$ 1.4 * 10 ⁵	3.0 $\pm$ 0.5 * 10 ⁻⁴	6.6 $\pm$ 2.0 * 10 ⁻¹⁰
-A _{165b}	4.9 $\pm$ 0.8*10 ³	1.1 $\pm$ 0.3 * 10 ⁻⁵	2.3 $\pm$ 0.7 * 10 ⁻⁹	1.2 $\pm$ 0.9 * 10 ⁶	9.7 $\pm$ 0.1 * 10 ⁻⁷	8.1 $\pm$ 0.6 * 10 ⁻¹³

Supplementary Table S2. χ^2 -to- $R_{\max}$ quality-of-fit for VEGFA_{xxx}:VEGFR global kinetic fitting. The ratio reflects how well a global kinetic analysis of a multi-concentration sensogram series can fit to a simple 1:1 Langmuir interaction model can fit. True interactions were distinguished from those due to non-specific binding patterns where χ^2 -to- $R_{\max} < 1.0$ (indicated by green shading and polka dot texturing). Fitted curves where χ^2 -to- $R_{\max} > 1.0$ (in red-shaded, grid-pattern textured cells) were considered to be dominated by non-specific binding. Ligand-receptor pairs that produced negative association curves were not analyzed with global fitting (indicated by “NI” for no interaction).

	VEGFR1	VEGFR2
-A ₁₆₅	0.17	0.52
-A ₁₂₁	0.13	0.029
-A _{165b}	0.039	0.12