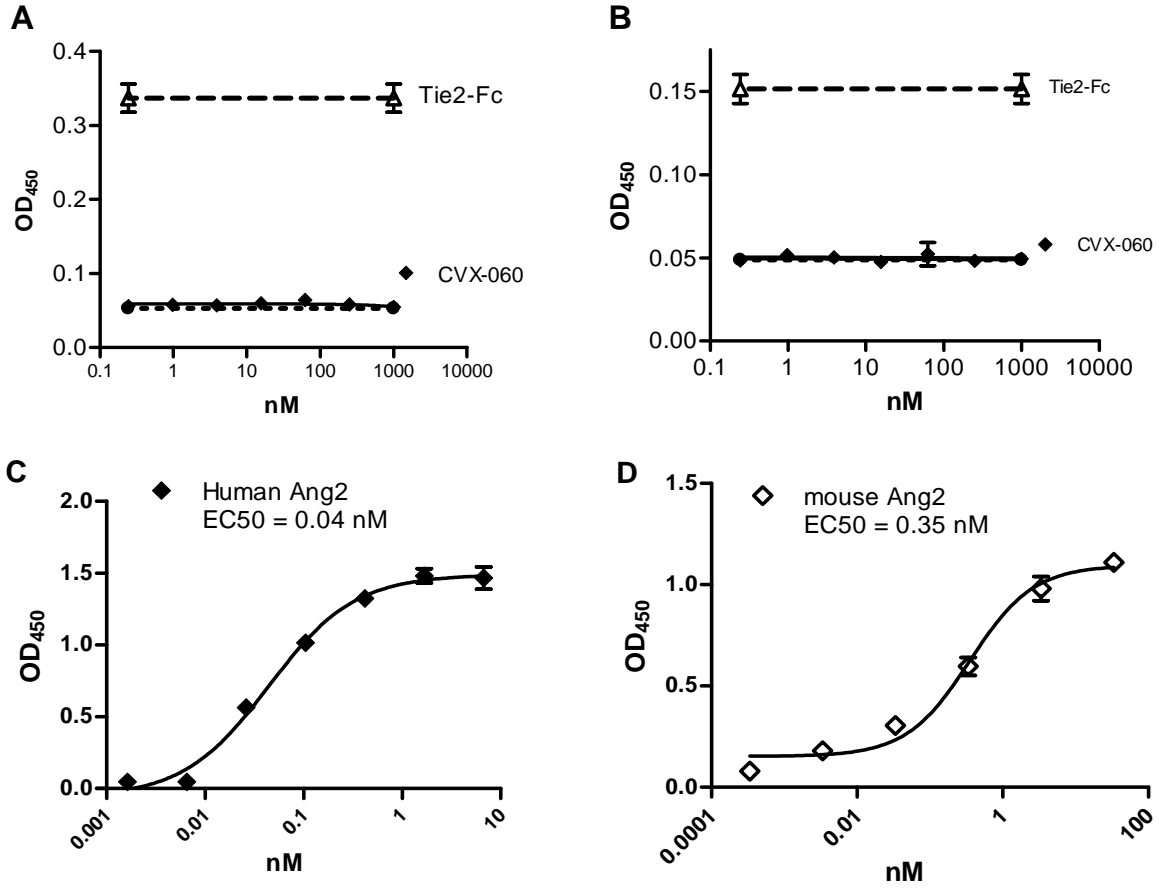
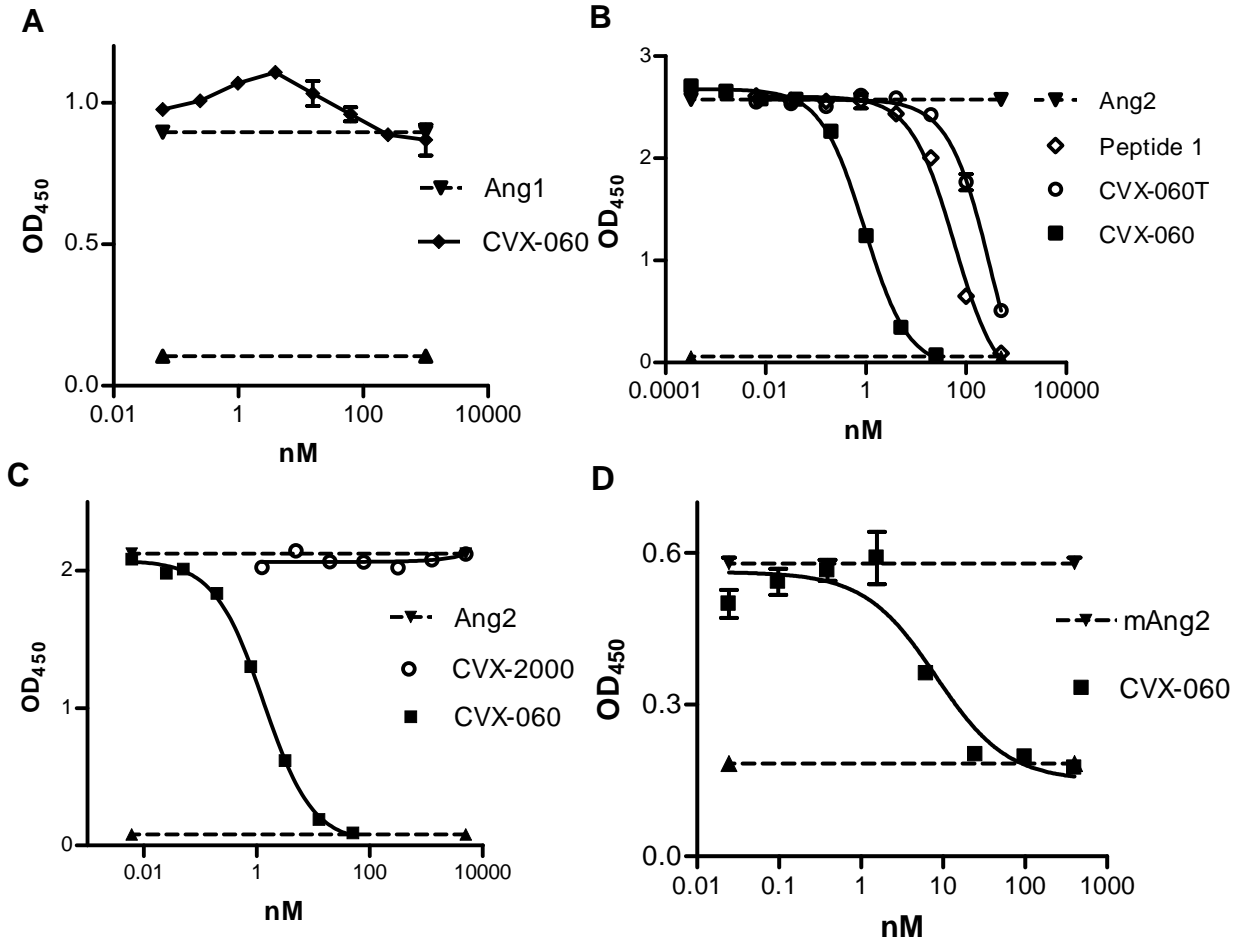


Supplementary Data

Supplementary Figure S1. The binding of CVX-060 to Ang3 (A), Ang4 (B), human Ang2 (C) and mouse Ang2 (D).



Supplementary Figure S2. CovX-Bodies in competition ELISA. A. CVX-060 had no effect on Ang1-Tie2 interaction. B. CVX-060, CVX-060T (the pharmacophore of CVX-060) and peptide 1 in Ang2-Tie2 competition ELISA. C. CVX-2000 had no effect on Ang2-Tie2 interaction. D. CVX-060 blocked the interaction of mouse Ang2 and mouse Tie2. Background without Ang1 or Ang2 was included.



Supplementary Figure S3.

A Series-S CM5 chip was activated with a mixture of NHS/EDC for 7 minutes followed by an injection of the Ang-2 protein (original conc. 1 mg/ml) which was diluted 100 fold into 10 mM sodium acetate pH 5.0 as the coupling buffer. The protein was injected for different lengths of time until the desired coupling densities were achieved. Then the surfaces were blocked with a 7 minute injection of 1 M ethanolamine pH 8.2. Final coupling levels were 3100, 280, and 65 RU. 1 RU = 1 pg/mm² at the surface. Running buffer was 10 mM HEPES pH 7.4, 150 mM NaCl, 0.005% p20, and 0.1 mg/ml BSA. Experimental temperature was 25°C. The peptide CVX-85T was tested at 10 μ M as the highest concentration in a 2-fold dilution series with 12 concentrations in total. The entire concentration series was repeated 4 times. Data were collected at 25 degrees C. The binding of CVX-85 was tested at 4 μ M as the highest concentration of a 3-fold dilution series. Each sample was tested in triplicate. The peptide CVX-85T bound to the Ang-2 surfaces in a simple 1:1 interaction manner with an affinity of \sim 4 μ M. CVX-85 binding responses were complex and surface density dependent, consistent with a bivalent system. The monovalent affinity of CVX-060 for Ang-2 was \sim 2 μ M and similar to the peptide affinity, while the bivalent affinity of CVX-85 for Ang-2 was \sim 5 nM. These data demonstrate the effect of increased avidity of CVX-85 for Ang-2 in comparison with the peptide pharmacophore element.

	k_a ($M^{-1}s^{-1}$)	k_d (s^{-1})	K_D
CVX-85T	136000	0.54	4 μ M
CVX-85, site I	203000	0.001	4.9 nM
CVX-85, site II	8697	0.0155	1.8 μ M

