Supporting Information for:

Nuclear Magnetic Resonance Structural Mapping Reveals Promiscuous Interactions between Clathrin-Box Motif Sequences and the N-Terminal Domain of the Clathrin Heavy Chain

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(a)

Approximate [Peptide]/[TD]	AP2 peptide expt. [TD] uM	AP2 peptide expt. [Peptide] uM	AP2 peptide expt. Exact [Peptide]/[TD]
0	765.00	0.00	0.00
1	400.00	400.00	1.00
2	371.13	739.18	1.99
3	345.54	1039.92	3.01
4	322.82	1306.85	4.05
5	303.70	1531.53	5.04
6	286.71	1731.16	6.04
7.5	264.20	1995.61	7.55
9	245.14	2219.64	9.05

(b)

Approximate [Peptide]/[TD]	AP180 peptide 1 expt. [TD] uM	AP180 peptide 1 expt. [Peptide] uM	AP180 peptide 1 expt. Exact [Peptide]/[TD]
0	500.00	0.00	0.00
1	469.26	494.37	1.05
2	443.27	912.21	2.06
3	420.09	1284.90	3.06
4	399.43	1617.12	4.05
5	381.04	1912.91	5.02
6	363.49	2195.08	6.04
7.5	340.04	2572.17	7.56
9	320.17	2891.72	9.03

(c)

Approximate [Peptide]/[TD]	AP180 peptide 2 expt. [TD] uM	AP180 peptide 2 expt. [Peptide] uM	AP180 peptide 2 expt. Exact [Peptide]/[TD]
0	500.00	0.00	0.00
1	453.07	459.87	1.01
2	414.58	837.14	2.02
3	383.11	1145.54	2.99
4	355.20	1419.04	4.00
5	331.52	1651.11	4.98
6	310.45	1857.56	5.98
7.5	283.15	2125.13	7.51
9	260.80	2344.20	8.99

(d)

Approximate [Peptide]/[TD]	AP2 mutant peptide expt. [TD] uM	AP2 mutant peptide expt. [Peptide] uM	AP2 mutant peptide expt. Exact [Peptide]/[TD]
0	500.00	0.00	0.00
1	463.58	470.60	1.02
2	432.67	869.89	2.01
3	405.18	1225.11	3.02
4	380.70	1541.32	4.05
5	358.91	1822.86	5.08
6	340.66	2058.64	6.04
7.5	315.85	2379.27	7.53
9	293.81	2663.97	9.07

Figure S1. Peptide and TD concentrations used for each step of the four NMR titrations performed in the study. AP2 peptide (panel a), AP180 peptide 1 (panel b), AP180 peptide 2 (panel c), and AP2 mutant peptide (panel d).

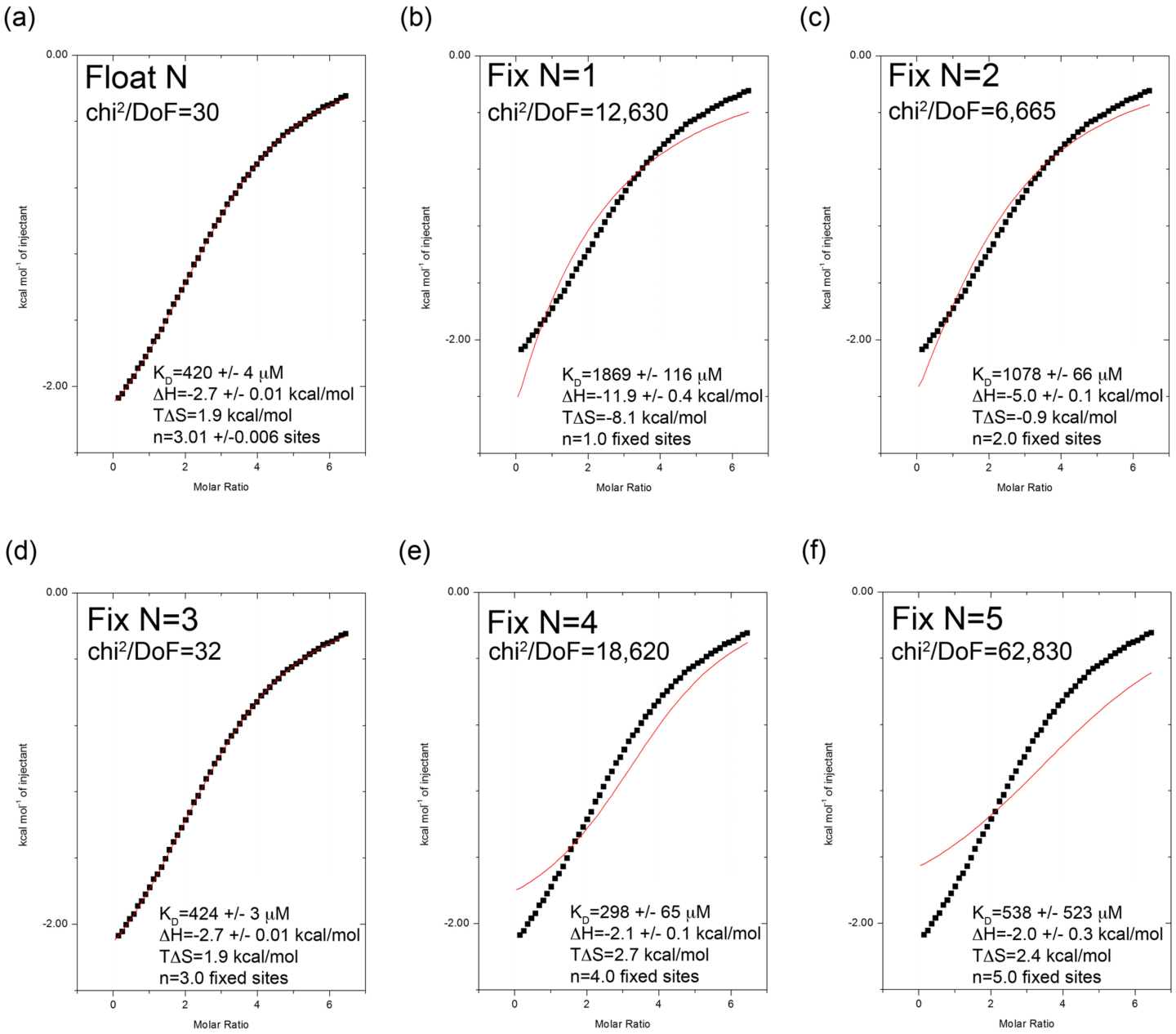

Figure S2. Analysis of ITC data for the titration of AP2 peptide into clathrin TD. The thermogram displayed in Fig. 1a was integrated and fit with Microcal modified Origin 7 software to an N independent binding sites model with either N floated (panel a), or fixed to 1 (panel b), 2 (panel c), 3 (panel d), 4 (panel e) or 5 (panel f).