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Supplemental Information

Rapid Simulation of Unprocessed DEER Decay Data for Protein Fold Prediction

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Table S1. Benchmark set for evaluation of RosettaDEER.

Protein	Restraints	PDB	Publication
Bax	21	1F16 8	Bleicken (2014) *Mol Cell* (40)
ExoU	11	3TU3	Fischer (2017) *ACS Omega* (11)
CDB3	15	1HYN R/S	Zhou (2005) *Biochemistry* (41)
Rhodopsin	14	1GZM A	Altenbach (2008) *PNAS* (42)
Mhp1	18	2JLN	Kazmier (2014) *PNAS* (14)

Table S2. Benchmark data for simulations of DEER distance distributions (continued on following page). Distances that could not be obtained due to steric hindrance or because residues weren't structurally resolved are marked with * and ‡, respectively.

	Experimental			RosettaDEER		MDDS		MMM (ambient)		MMM (cryogenic)		TagDock		Pronox		MtsslWizard	
Pair	μ	σ	C_β-C_β	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ
Bax (PDB: 1F16 H)																	
101/126	34.0	8.9	32.2	38.9	4.0	37.5	4.1	37.1	3.3	39.1	3.0	32.1	3.2	34.1	3.7	31.0	2.2
101/149	29.1	12.2	19.3	28.4	2.0	29.5	2.6	21.8	4.4	23.4	1.9	28.1	1.4	30.2	2.4	19.9	1.1
101/169	26.2	7.3	26.2	36.1	3.1	34.0	3.8	38.5	3.4	30.6	5.6	32.7	3.1	34.9	4.3	29.1	2.3
126/169	38.8	9.7	34.3	40.3	4.9	38.5	4.3	38.0	3.8	39.3	3.5	30.7	4.4	33.5	3.8	31.7	1.9
16/62	32.7	6.3	22.4	31.9	0.7	25.9	4.2	32.8	1.6	34.6	1.2	33.0	1.7	30.6	3.5	22.5	1.7
55/87	41.8	8.3	19.4	45.3	2.7	41.6	3.8	43.9	3.8	38.9	3.7	40.4	2.6	40.5	1.3	31.3	2.6
55/101	35.9	9.2	37.4	41.6	3.3	38.6	3.2	36.1	4.3	34.1	4.7	39.1	1.5	41.5	1.8	30.2	2.0
55/126	37.8	7.8	32.9	38.6	4.5	39.7	3.2	35.9	4.1	34.9	3.6	35.9	3.5	38.1	2.6	29.2	1.9
55/149	30.7	11.5	31.5	24.0	3.1	19.8	4.5	23.8	4.2	23.1	3.3	23.1	1.6	26.3	1.9	16.9	1.8
62/87	40.6	6.0	14.9	41.9	2.1	37.0	3.9	42.9	2.0	39.5	1.5	38.4	2.1	34.4	1.6	29.0	2.1
62/101	34.1	6.6	31.8	41.9	2.1	29.6	4.1	36.2	1.6	33.6	3.9	35.7	2.0	36.4	2.5	26.6	2.1
62/126	31.5	4.7	26.3	30.0	4.5	31.2	3.5	31.0	3.5	29.2	3.0	28.2	4.1	30.5	3.3	24.9	1.4
62/149	31.5	3.3	25.6	32.8	1.5	25.9	4.4	31.4	1.8	33.7	0.9	31.2	1.1	31.7	3.0	20.8	1.5
62/169	21.4	9.7	20.6	14.4	2.2	15.3	2.9	16.3	1.9	14.9	1.7	13.9	2.4	17.4	3.7	15.8	5.0
72/87	34.9	7.6	25.8	39.7	2.7	41.1	2.4	34.0	4.0	37.7	2.1	32.9	4.4	35.2	3.3	26.0	1.8
72/101	33.0	7.9	30.3	37.7	2.8	35.6	3.5	37.6	2.7	34.9	3.5	31.5	2.5	34.4	3.3	27.2	2.1
72/126	24.7	7.7	27.9	25.0	4.6	25.0	4.0	17.1	4.3	23.8	4.1	18.8	5.9	18.5	4.6	18.8	1.3
72/169	27.4	8.4	21.5	20.4	4.2	16.4	4.5	25.1	4.9	20.3	5.1	14.1	6.3	18.2	4.7	16.6	2.3
72/186	29.7	8.4	15.3	25.6	3.9	25.2	4.2	23.6	4.1	25.1	1.8	24.8	3.5	24.0	3.2	19.1	1.3
87/126	28.9	8.4	20.0	27.2	3.2	29.4	2.9	26.2	3.7	25.9	3.5	20.2	4.1	22.6	3.3	16.4	2.5
87/186	27.0	7.3	25.1	27.2	3.6	30.1	2.6	29.0	3.8	24.6	4.7	28.9	2.1	31.9	3.1	19.6	1.3
Mhp1 (PDB: 2JLN)																	
136/278	27.0	10.2	25.9	32.3	2.4	30.4	3.8	31.4	3.5	31.2	3.8	31.2	3.4	31.2	2.6	25.8	1.8
136/349	33.4	6.0	30.3	36.5	4.4	36.2	3.7	34.3	5.4	32.2	5.6	31.3	5.2	36.6	4.5	30.4	2.0
144/278	33.7	6.8	31.6	39.0	3.5	40.5	3.5	44.6	2.2	38.1	5.4	39.5	2.2	42.4	2.1	32.5	2.1
159/324	24.0	5.6	15.0	15.3	1.6	16.6	3.2	13.8	2.1	13.8	1.3	16.1	2.6	14.9	2.2	17.0	1.6
163/243	45.9	11.8	34.8	42.4	2.7	42.5	3.4	42.3	2.6	39.9	2.2	40.7	2.0	43.6	2.2	35.2	1.7
184/278	27.0	12.3	16.4	25.5	3.0	22.5	3.5	20.7	3.2	22.5	2.7	23.2	2.7	24.5	2.6	24.8	0.9
234/338	42.6	13.6	25.7	25.9	2.2	35.3	4.0	30.3	1.5	29.7	1.2	32.0	3.6	32.9	3.9	n/a*	n/a*
243/338	58.7	14.2	41.8	49.4	2.9	50.9	2.8	49.1	2.9	47.9	1.9	49.3	2.1	32.9	3.9	41.7	2.3
278/349	36.6	7.6	36.9	40.9	4.0	42.8	4.5	46.0	3.3	38.9	6.0	43.7	3.6	45.1	3.4	37.2	2.1
278/362	33.9	2.8	30.5	36.8	4.2	41.6	3.5	42.7	2.6	35.9	4.7	39.7	2.1	40.2	3.0	30.5	1.7
285/362	33.1	12.8	31.0	31.3	3.9	33.8	4.1	31.7	4.8	31.1	4.9	32.1	4.6	31.9	3.7	31.0	1.5
349/362	31.4	10.4	19.1	23.6	4.3	24.4	4.1	25.7	4.3	21.2	5.0	24.3	4.6	24.6	3.9	21.1	1.2
30/163	32.5	9.0	17.5	22.5	3.8	23.7	3.9	23.6	3.7	21.2	3.6	25.5	3.0	22.1	3.1	17.7	1.5
30/243	25.6	6.5	22.9	27.8	4.8	26.2	4.4	26.3	4.7	24.8	4.9	19.8	2.9	28.4	3.5	23.9	1.1
30/338	33.5	4.3	32.3	39.5	3.4	42.3	2.9	40.4	3.5	38.9	3.5	41.3	2.0	41.6	2.6	32.2	1.9
51/278	24.5	7.2	14.3	18.7	3.6	21.4	3.6	21.3	3.8	19.2	3.6	23.4	2.8	21.1	4.1	16.3	2.6
63/285	34.7	4.7	25.9	35.6	3.0	36.8	2.7	35.8	3.0	33.8	3.0	33.4	1.9	36.5	2.3	26.3	1.9
63/362	32.6	7.4	29.4	35.9	3.0	38.9	3.2	39.3	2.5	35.1	2.1	36.5	2.6	38.7	2.5	29.8	1.7
ExoU (PDB: 3TU3)																	
592/636	26.8	4.0	26.2	27.3	4.2	20.3	4.0	25.2	4.3	24.3	3.8	19.3	2.4	26.0	3.8	25.6	1.5
592/649	24.7	6.3	24.8	26.1	4.3	25.7	4.3	25.2	4.7	25.2	4.6	20.3	2.5	26.4	4.7	25.7	1.6
598/680	27.8	4.8	21.5	20.7	3.4	22.5	3.9	25.6	3.2	23.7	3.3	22.4	3.8	26.7	2.7	22.0	1.4
629/645	25.1	7.8	23.5	29.8	4.0	26.0	4.0	27.7	4.3	25.1	5.1	22.4	4.7	26.7	3.7	21.6	1.4
636/645	23.4	6.4	16.2	24.6	3.6	23.4	3.3	23.6	4.2	16.3	3.4	22.0	4.1	20.2	4.1	16.9	1.9
636/649	22.0	5.1	11.9	20.0	3.8	19.2	3.7	19.0	4.3	19.0	2.7	16.3	3.6	14.5	4.3	15.8	4.7
636/657	23.5	7.1	11.9	19.8	3.7	19.9	3.7	17.8	3.9	30.3	1.7	19.0	2.8	15.8	2.6	16.6	2.3
636/672	29.4	4.1	23.9	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡
636/677	27.8	3.8	18.9	28.3	2.7	26.8	3.0	28.0	2.6	23.6	1.9	26.1	1.8	28.7	2.1	18.4	1.3
636/682	25.4	4.7	18.4	25.9	2.8	24.0	2.7	25.3	3.0	20.2	3.2	25.9	2.0	24.9	2.4	19.0	1.1
649/672	26.5	3.5	20.7	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡	_‡

	Experimental			RosettaDEER		MDDS		MMM (ambient)		MMM (cryogenic)		TagDock		Pronox		MtsslWizard	
Pair	μ	σ	Cβ-Cβ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ	μ	σ
CDB3 (PDB: 1HYN R/S)																	
105/105	27.9	3.1	31.0	30.4	3.8	21.7	3.6	25.5	3.2	29.6	2.8	32.7	3.1	32.1	1.7	30.9	2.1
116/116	31.2	9.5	33.9	36.4	4.7	31.4	3.4	32.5	4.4	32.2	4.6	30.2	2.7	26.8	2.7	31.9	1.7
142/142	13.6	4.6	14.9	15.7	3.2	14.5	3.9	23.4	2.7	20.4	3.2	11.0	2.1	22.4	4.5	15.7	5.4
199/199	11.1	3.3	23.9	22.5	5.5	22.0	4.0	22.4	4.3	23.7	3.8	24.5	2.5	20.8	3.8	23.5	1.2
208/208	28.5	10.2	37.8	34.1	5.8	34.4	4.5	37.3	4.1	32.6	2.9	44.5	0.9	33.8	2.8	38.3	2.7
277/277	31.8	9.4	29.2	32.5	1.8	30.7	3.9	27.1	3.9	36.2	3.3	22.0	3.1	35.0	0.7	30.1	1.8
290/290	39.2	18.4	44.0	46.5	4.7	37.5	4.0	38.9	4.6	40.3	4.1	36.1	1.1	43.5	5.2	44.2	4.0
312/312	30.6	3.6	27.9	32.6	4.4	36.0	3.8	35.3	3.5	37.4	3.2	31.2	2.7	27.5	1.2	29.0	2.7
340/340	26.2	11.4	33.3	38.4	4.7	40.3	4.5	21.5	1.5	19.8	0.8	26.3	2.2	22.1	1.6	33.9	6.0
342/342	28.7	2.7	24.4	25.8	3.6	21.5	4.7	25.2	3.6	26.4	2.5	20.1	5.1	30.6	2.8	24.1	2.1
343/343	21.6	15.2	24.3	30.6	5.3	30.8	3.4	29.9	3.2	31.9	1.7	25.9	3.8	30.9	2.9	23.9	1.9
344/344	12.6	10.4	20.2	21.1	3.7	25.4	3.7	23.2	1.8	23.8	2.5	30.1	0.9	18.6	2.7	21.9	1.3
345/345	16.9	15.4	25.1	32.1	5.0	31.4	2.1	31.3	3.7	32.4	2.0	23.6	3.5	34.9	2.2	26.7	2.2
84/84	40.5	4.2	33.1	40.7	4.9	43.1	3.1	39.4	4.4	38.5	3.4	42.0	0.9	40.5	2.8	35.7	1.8
96/96	26.7	8.8	30.3	36.6	3.9	37.7	3.7	38.9	3.5	36.9	3.7	33.8	5.0	35.6	4.6	31.4	2.9
Rhodopsin (PDB: 1GZM)																	
63/241	34.4	2.6	26.9	40.6	2.6	39.0	3.4	37.8	4.0	38.8	2.5	36.4	2.2	42.4	3.2	31.0	2.3
63/252	31.0	7.5	25.8	36.3	2.2	35.3	3.0	33.1	3.4	33.9	2.2	31.6	2.2	35.7	2.5	26.0	1.8
63/326	26.3	3.6	17.9	26.3	2.4	24.2	3.5	22.4	2.0	22.7	1.8	24.2	2.6	29.2	2.7	18.5	1.4
74/137	22.3	3.3	14.3	21.1	0.8	19.7	4.0	22.9	2.5	23.1	1.4	19.5	1.5	21.0	0.6	16.6	2.2
74/225	31.4	2.3	21.4	33.7	1.6	30.5	3.8	33.0	2.5	34.1	2.4	31.3	1.9	36.0	0.7	24.1	1.7
74/252	29.2	4.0	18.9	31.0	1.5	30.8	2.5	30.9	1.6	29.4	1.2	29.6	1.0	32.0	1.5	20.9	1.0
74/308	27.8	2.3	19.3	28.6	1.9	29.6	2.5	30.1	1.4	28.9	1.3	26.5	1.8	29.5	1.6	20.4	0.9
137/326	44.0	10.3	32.1	37.7	2.2	37.1	3.4	35.9	1.5	36.0	1.0	34.7	1.0	35.8	4.9	32.9	2.3
151/241	32.5	6.1	29.7	37.0	3.2	38.4	3.0	37.2	3.3	35.5	3.0	33.5	2.5	37.5	3.5	28.0	2.9
151/308	35.9	7.8	29.3	37.3	2.4	39.6	2.8	38.8	2.3	36.7	2.1	35.3	2.8	38.5	2.6	28.8	1.7
151/326	31.8	12.4	31.0	35.1	4.0	36.0	3.9	30.0	3.5	31.1	3.6	34.2	4.2	37.5	3.7	31.2	2.2
225/252	25.0	5.9	16.0	21.8	3.2	24.2	3.1	20.5	3.9	21.1	4.0	24.5	3.2	25.2	3.4	18.1	1.3
225/308	34.0	8.5	11.5	35.2	2.9	36.5	3.0	36.5	2.4	34.0	1.8	32.9	4.1	36.6	2.7	27.7	1.4
252/326	32.7	6.7	23.3	26.1	2.5	25.4	4.3	28.2	3.4	26.0	3.4	21.2	4.7	23.9	4.7	23.9	1.8
Correlation (r-squared)			0.27	0.55	0.06	0.52	0.00	0.53	0.02	0.50	0.00	0.51	0.01	0.42	0.06	0.42	0.04

Table S3. List of spin-labeled proteins in the protein data bank.

PDB	Beta carbon			Nitroxide nitrogen			Nitroxide oxygen			Angle	Relative Distance
	x	y	z	x	y	z	x	y	z		
1ZYT (A)	45.34	-7.24	7.94	45.93	-3.81	14.56	46.54	-3.29	15.49	5.23	0.83
2CUU (A)	31.74	9.11	-11.29	37.09	11.32	-15.90	38.33	11.54	-16.04	7.24	0.84
2CUU (A)	31.74	9.11	-11.29	34.84	8.96	-16.89	35.44	8.64	-17.97	3.39	0.78
2IGC (A)	0.63	23.03	97.36	-1.83	18.50	98.24	-2.70	18.07	98.95	11.49	0.81
2NTH (A)	-11.76	35.89	63.20	-6.93	35.75	67.89	-6.25	35.86	68.89	3.06	0.80
2OU8 (A)	0.09	23.50	99.06	-4.09	18.69	97.25	-5.21	18.25	97.24	7.13	0.82
2OU8 (A)	0.09	23.50	99.06	3.34	18.79	103.07	4.14	17.91	102.86	9.08	0.86
2OU9 (A)	20.05	11.78	46.38	18.14	7.36	46.35	18.37	6.51	45.54	15.13	0.84
2Q9D (A)	25.85	29.50	8.70	28.07	33.43	6.03	27.97	34.62	5.86	10.99	0.80
2Q9E (A)	22.82	4.02	77.45	22.72	8.29	80.31	22.16	9.30	79.97	14.55	0.86
2Q9E (B)	16.24	-6.49	118.33	20.24	-6.61	121.49	21.32	-6.13	121.42	13.03	0.83
2Q9E (C)	-13.47	-10.74	116.70	-14.18	-16.97	119.08	-14.98	-17.70	119.55	8.08	0.84
2XGA (A)	27.64	36.01	-12.25	33.29	35.15	-9.09	34.17	35.48	-8.27	6.37	0.80
2XGA (A)	53.10	12.38	32.02	53.37	13.62	25.61	53.22	13.41	24.38	5.53	0.80
2XIU (A)	30.89	11.72	13.12	32.52	18.09	14.29	32.68	19.22	13.87	7.07	0.82
2XIU (A)	31.18	11.32	12.96	12.88	1.28	14.75	12.82	0.14	15.17	3.59	0.96
3G3V (A)	32.01	9.42	-11.26	37.17	11.36	-16.07	38.43	11.38	-16.25	7.83	0.84
3G3V (A)	32.01	9.42	-11.26	34.94	8.76	-16.91	35.47	8.19	-17.91	5.30	0.79
3M8B (A)	-36.14	-15.28	3.58	-34.89	-19.33	7.52	-34.61	-20.63	7.42	12.64	0.82
3M8D (A)	30.56	-24.22	-34.24	33.53	-21.66	-29.05	34.71	-20.81	-28.85	11.98	0.82
3MPN (A)	46.41	34.94	18.10	47.66	35.80	25.63	47.41	35.92	27.00	4.61	0.80
3MPQ (A)	29.69	31.87	-0.63	29.42	28.90	-4.58	29.24	27.53	-4.75	15.33	0.79
3RGM (A)	28.45	-2.69	-21.73	34.19	1.12	-20.61	34.77	2.03	-19.73	9.04	0.82
3RGN (A)	-34.31	44.87	-34.41	-31.48	49.85	-38.26	-31.67	51.20	-38.45	10.25	0.83
3STZ (C)	151.02	114.96	-17.24	156.25	149.05	-22.46	156.30	149.33	-21.26	2.51	1.00
3STZ (C)	155.90	146.71	-29.68	156.25	149.05	-22.46	156.30	149.33	-21.26	1.10	0.81
3V3X (A)	8.06	35.66	44.62	7.17	40.73	39.31	6.94	41.37	38.29	2.62	0.81
3V3X (B)	20.59	27.24	17.69	13.12	27.74	16.14	12.09	27.49	15.56	4.78	0.83
3V3X (B)	20.43	27.14	17.71	14.29	28.00	16.08	13.28	27.42	15.67	8.53	0.82
3V3X (B)	20.826	30.531	-11.28	26.72	40.34	48.40	27.59	40.28	47.56	1.13	1.02
3V3X (C)	5.82	27.48	36.57	9.99	23.00	39.69	10.62	22.08	40.20	2.00	0.80
3V3X (D)	22.506	23.768	6.859	27.85	21.68	8.62	28.91	21.03	8.59	5.65	0.78
3V3X (D)	22.401	24.15	6.799	28.74	23.01	9.39	29.87	22.71	9.77	1.39	0.80
3V3X (D)	28.74	38.19	32.17	31.88	41.91	26.95	32.61	42.58	26.22	2.71	0.81
4EK1 (A)	28.41	-27.82	0.47	29.53	-29.48	7.15	29.96	-30.24	8.29	5.64	0.78
4EK1 (B)	17.67	-27.74	35.80	19.44	-29.86	41.92	19.43	-30.90	42.84	8.46	0.80
5BMG (A)	68.09	8.92	26.62	69.95	14.65	20.03	69.36	15.35	21.21	10.97	1.08
5BMG (B)	70.46	8.41	30.86	68.76	14.18	32.79	68.81	15.31	33.24	4.57	0.79
5BMG (B)	70.46	8.41	30.86	74.05	10.31	26.90	74.00	9.10	27.04	13.52	1.13
5BMG (D)	64.35	-31.08	48.92	66.58	-25.65	51.29	67.33	-24.68	51.37	5.89	0.80
5BMG (E)	61.98	23.34	48.91	61.65	-28.81	44.96	62.20	-29.35	44.01	1.45	0.99
5BMG (F)	59.71	-22.93	53.48	62.05	-28.84	56.84	61.78	-29.62	57.73	7.70	0.84
5BMG (G)	67.80	17.05	26.41	63.62	11.03	27.98	63.33	10.00	27.39	8.47	0.87
5BMH (A)	0.05	-10.04	-13.40	-1.59	-8.86	-18.40	-1.45	-8.90	-19.62	8.29	0.77
5BMH (A)	0.21	-10.02	-13.65	-1.66	-8.67	-18.34	-1.55	-8.70	-19.56	9.38	0.77
5BMI (A)	-12.45	12.28	-1.86	-7.06	15.58	-4.29	-6.04	15.52	-4.96	7.42	0.83
5I26 (A)	94.17	13.89	136.21	97.47	15.70	141.52	97.75	16.31	142.46	4.80	0.81
5I26 (B)	121.29	25.02	121.83	121.69	18.58	121.08	122.34	17.62	121.18	0.55	0.99
5I26 (C)	90.81	62.80	115.49	85.28	65.25	117.86	84.85	65.22	118.94	0.92	1.01
5I26 (D)	75.61	38.89	96.79	78.64	41.52	91.87	78.33	42.28	91.05	0.89	1.00
5I28 (A)	253.06	53.98	41.53	256.95	56.60	46.87	257.07	57.16	47.86	0.50	1.00
5I28 (B)	241.07	51.61	72.62	237.23	53.97	67.15	237.07	54.42	66.10	0.71	1.00
5I28 (C)	254.91	59.75	42.24	248.27	57.34	42.90	247.35	56.89	43.43	0.32	1.01
5I28 (D)	261.00	7.94	25.94	260.59	4.56	22.84	259.61	4.31	22.27	0.25	1.01
5I28 (E)	242.01	18.87	3.05	238.04	15.54	-1.77	237.84	14.60	-2.40	4.49	0.96
5I28 (F)	290.39	18.69	26.81	285.83	18.41	20.66	285.50	18.81	19.63	1.81	1.01
5I28 (G)	223.92	68.30	42.05	222.89	64.06	47.69	222.43	63.01	47.87	2.15	0.98
5I28 (H)	268.83	69.81	68.41	271.91	65.90	65.16	272.94	65.38	65.17	2.10	0.89
5I28 (I)	270.04	41.64	65.86	269.40	45.76	70.37	269.61	46.73	70.97	5.44	0.80
5I28 (J)	222.99	43.38	41.56	219.19	47.62	46.17	219.27	48.46	46.97	6.80	0.85
5JDT (A)	34.90	-28.61	-16.81	32.74	-24.46	-11.81	32.18	-23.58	-11.01	2.52	0.78
5JDT (A)	35.02	-28.35	-16.57	32.87	-24.65	-11.87	32.37	-23.92	-10.88	0.85	0.76

Table S4. Experimental DEER decay parameters obtained using DeerAnalysis (continued on following page).

Res 1	Res 2	k	λ	Duration	
				Oscillations	μs
Bax (PDB: 1F16 H)					
16	62	-0.01	0.389	4.45	3.00
55	87	-0.01	0.348	2.64	3.70
55	101	0.00	0.342	3.59	3.20
55	126	0.00	0.337	2.99	3.10
55	149	-0.01	0.286	4.85	2.70
62	87	-0.01	0.389	2.71	3.50
62	101	0.00	0.397	3.54	2.70
62	126	-0.27	0.55	7.66	4.62
62	149	-0.01	0.442	4.49	2.70
62	169	-0.01	0.286	12.73	2.40
72	87	-0.01	0.388	3.93	3.20
72	101	-0.01	0.304	4.77	3.30
72	126	0.00	0.237	8.62	2.50
72	169	-0.01	0.382	7.55	3.00
87	126	-0.01	0.234	4.33	2.00
101	126	0.00	0.195	2.26	1.70
101	149	-0.01	0.206	3.18	1.50
101	169	-0.01	0.319	5.23	1.80
126	169	0.00	0.356	2.47	3.00
72	186	-0.01	0.135	3.38	1.70
87	186	-0.01	0.175	4.48	1.70
Mhp1 (PDB: 2JLN)					
30	163	-0.05	0.115	4.47	2.95
30	243	-0.08	0.127	6.32	2.05
30	338	0.00	0.125	1.16	2.14
51	278	-0.05	0.171	6.48	1.74
63	285	-0.05	0.072	3.66	2.94
63	362	-0.04	0.076	3.82	2.54
136	278	-0.04	0.099	6.42	2.44
136	349	-0.04	0.064	4.10	2.94
144	278	-0.03	0.13	3.72	2.74
159	324	-0.12	0.218	7.72	2.05
163	243	-0.02	0.1	1.74	3.24
184	278	-0.07	0.163	6.22	2.34
234	338	-0.03	0.064	1.78	2.65
243	338	-0.03	0.252	0.89	3.45
278	349	-0.05	0.052	3.13	2.94
278	362	-0.04	0.143	3.65	2.74
285	349	-0.02	0.057	3.93	2.74
349	362	-0.04	0.117	4.62	2.74
ExoU (PDB: 3TU3)					
592	636	-0.05	0.127	7.07	2.62
592	649	-0.01	0.081	10.05	2.91
598	680	-0.05	0.096	7.12	2.93
629	645	-0.01	0.088	6.66	2.02
636	645	-0.02	0.117	11.10	2.72
636	649	-0.06	0.123	12.74	2.62
636	657	-0.03	0.115	10.57	2.62
636	672	-0.06	0.149	5.36	2.62
636	677	-0.03	0.18	5.85	2.43
636	682	-0.03	0.164	8.71	2.74
649	672	-0.04	0.2	6.53	2.34

Res 1	Res 2	k	λ	Duration	
				Oscillations	μs
CDB3 (PDB: 1HYN R/S)					
84	84	-0.06	0.185	2.65	1.59
96	96	-0.07	0.177	2.72	1.59
105	105	-0.05	0.122	16.35	0.78
116	116	-0.05	0.108	29.81	0.78
142	142	-0.03	0.071	3.58	1.59
199	199	-0.10	0.146	3.86	2.38
208	208	-0.06	0.062	2.58	2.98
277	277	-0.03	0.19	3.60	1.99
290	290	-0.08	0.195	12.88	4.46
312	312	-0.03	0.078	4.36	1.99
340	340	-0.04	0.243	12.81	2.47
342	342	-0.06	0.163	40.76	1.57
343	343	-0.08	0.17	17.06	1.59
344	344	-0.07	0.162	2.37	3.02
345	345	-0.04	0.228	8.13	2.98
Rhodopsin (PDB: 1GZM A)					
63	241	-0.36	0.326	3.26	2.56
63	252	-0.05	0.233	3.42	1.97
63	326	-0.09	0.101	4.17	1.46
74	137	-0.16	0.218	8.29	1.77
74	225	-0.13	0.234	2.96	1.77
74	252	-0.10	0.214	3.70	1.77
74	308	-0.14	0.413	4.77	1.98
137	326	0.02	0.25	1.55	2.54
151	241	-0.18	0.085	3.84	2.54
151	308	-0.03	0.133	2.21	1.97
151	326	-0.05	0.146	4.01	2.47
225	252	-0.35	0.277	3.90	1.17
225	308	-0.25	0.292	3.28	2.47
252	326	-0.04	0.282	2.92	1.97

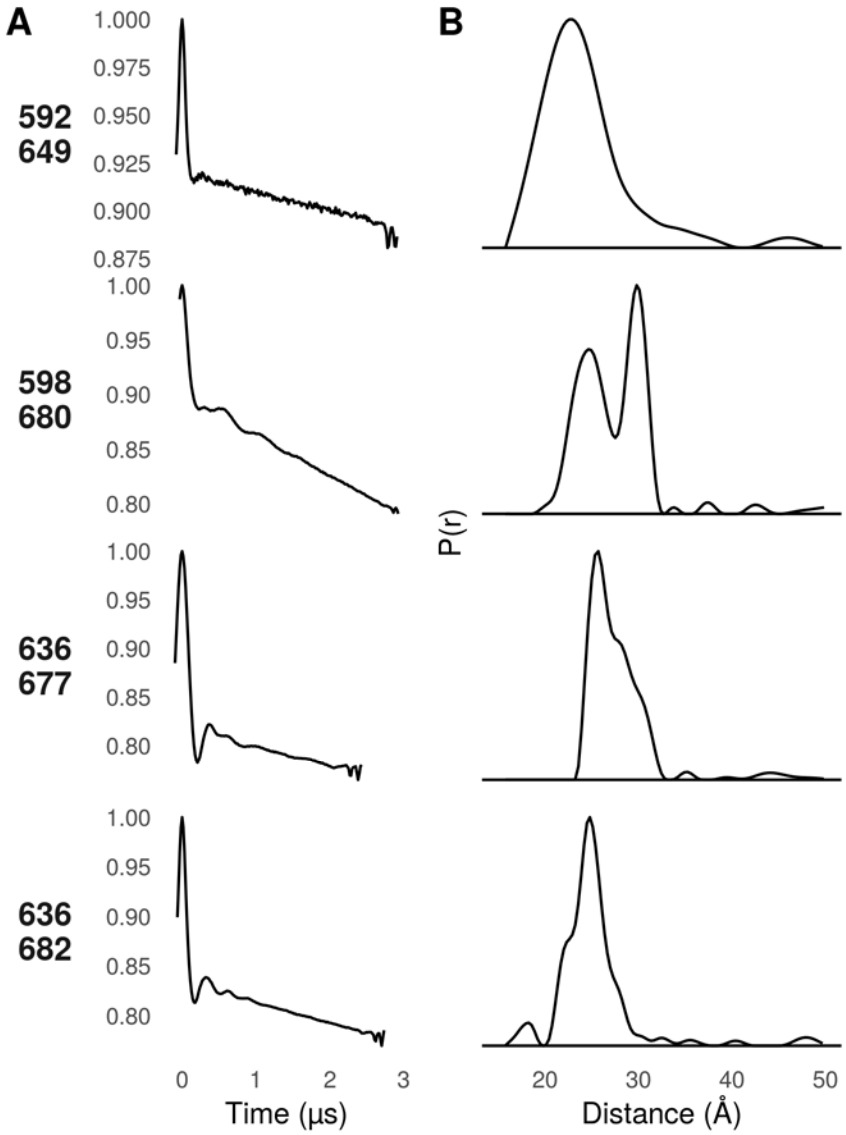


Figure S1 . Data gathered in the ExoU C-terminus for this study.

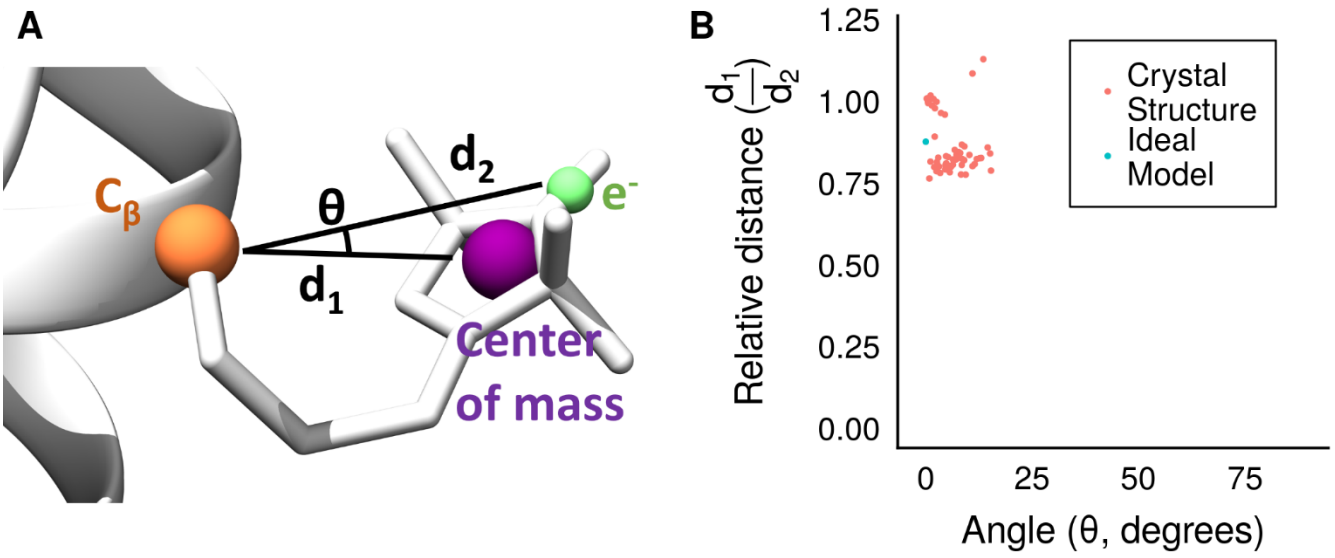


Figure S2. Nitroxide centers of mass fall along the C_β-electron vector. A) Depiction of spin label from PDB: 2Q9D showing the nitroxide center of mass (purple) and the nitroxide bond midpoint (green). **B).** Angle and relative distance of nitroxide center of mass along the C_β-electron vector.

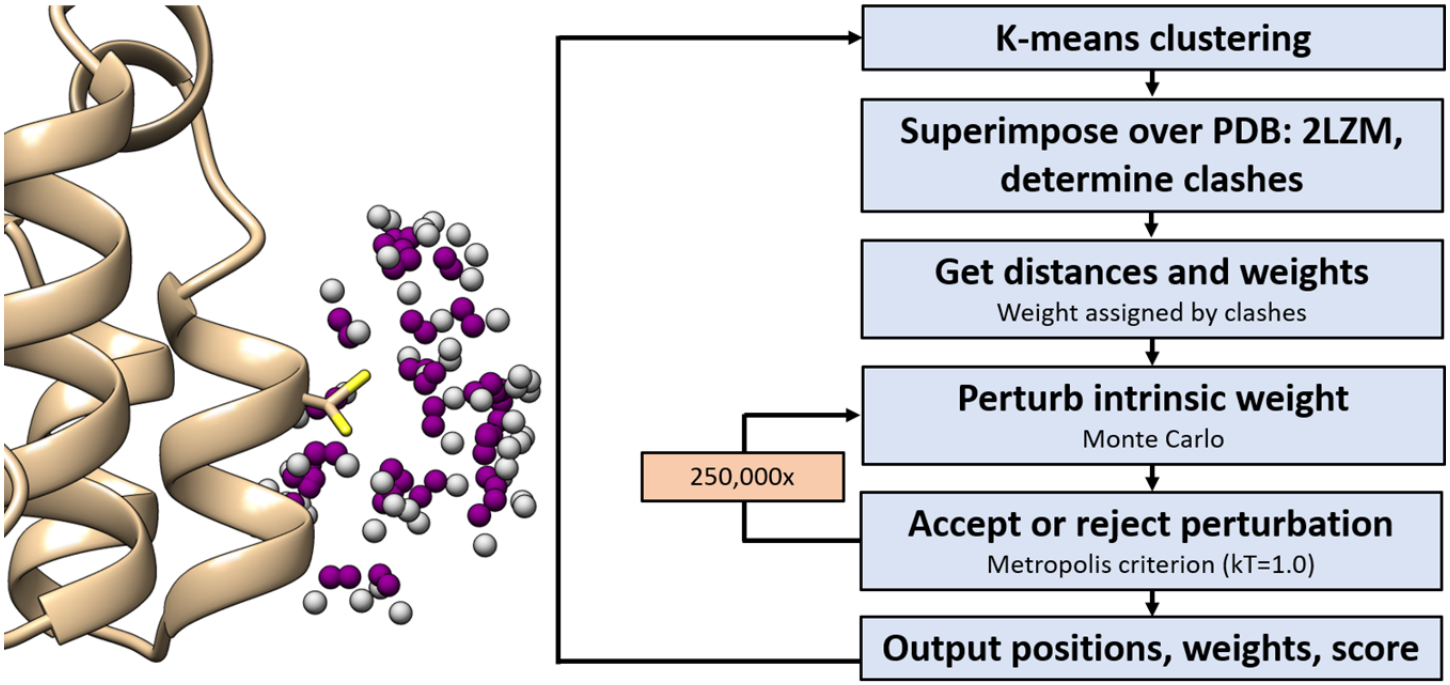
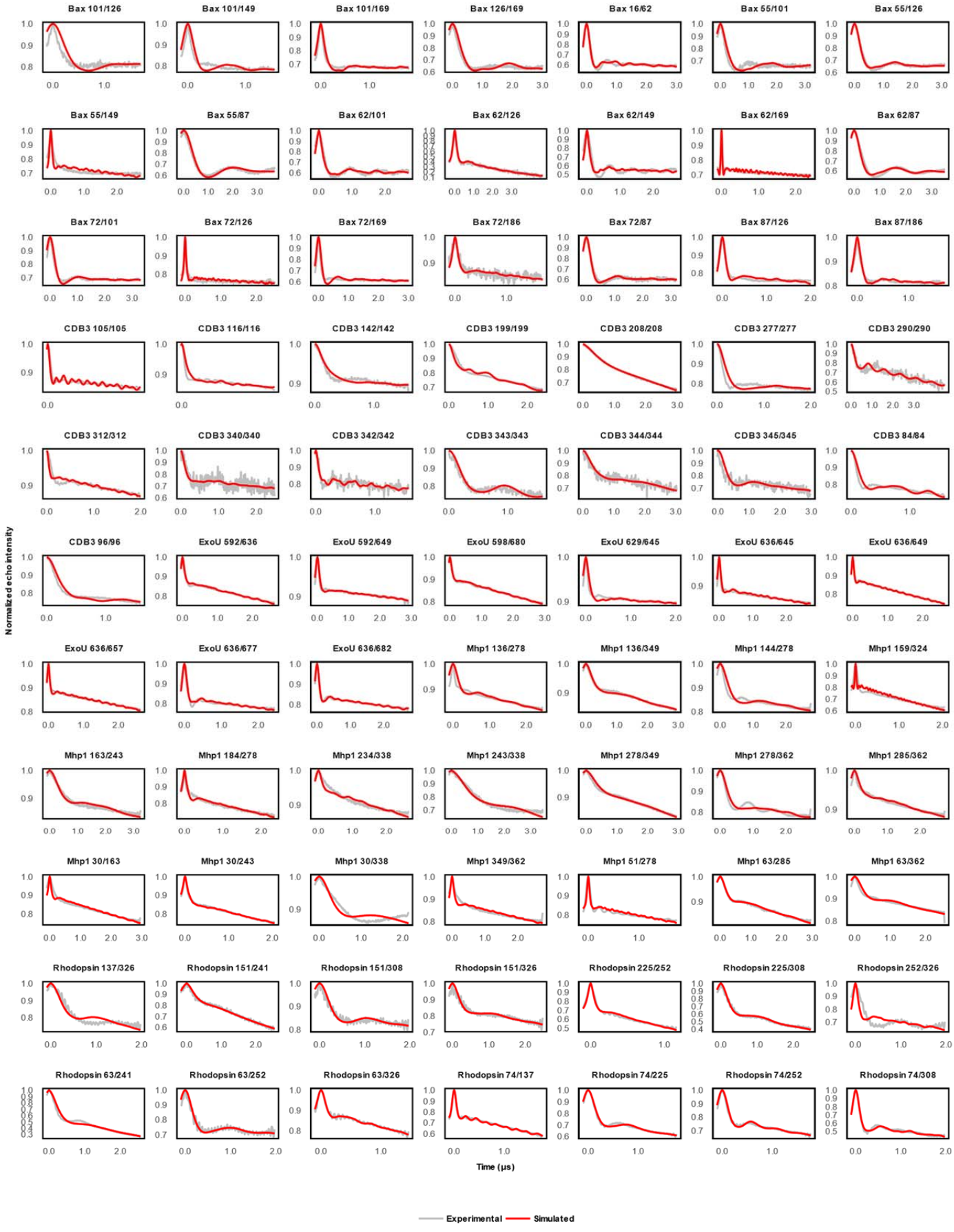


Figure S3. Optimization of RosettaDEER measurement coordinates. Left: Each of the rotamers in Rosetta's MTSSL rotamer library was converted into two coordinates: one representing the nitroxide ring center of mass (purple), which was used to evaluate clashes; and one representing the nitroxide bond midpoint (silver), from which distances were measured. Shown over PDB 2CUU residue 131. **Right:** Optimization scheme for reducing the number of measurement coordinates using experimental T4 Lysozyme distance data. One thousand replicates were performed for each of *N* clusters, with *N* ranging from 3 to 53 coordinates.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Bax 101/149
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Bax 16/62
Bax 55/101
Bax 55/126
Bax 55/149
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Bax 62/126
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CDB3 116/116
CDB3 142/142
CDB3 199/199
CDB3 208/208
CDB3 277/277
CDB3 290/290
CDB3 312/312
CDB3 340/340
CDB3 342/342
CDB3 343/343
CDB3 344/344
CDB3 345/345
CDB3 84/84
CDB3 96/96
ExoU 592/636
ExoU 592/649
ExoU 598/680
ExoU 629/645
ExoU 636/645
ExoU 636/649
ExoU 636/657
ExoU 636/677
ExoU 636/682
Mhp1 136/278
Mhp1 136/349
Mhp1 144/278
Mhp1 159/324
Mhp1 163/243
Mhp1 184/278
Mhp1 234/338
Mhp1 243/338
Mhp1 278/349
Mhp1 278/362
Mhp1 285/362
Mhp1 30/163
Mhp1 30/243
Mhp1 30/338
Mhp1 349/362
Mhp1 51/278
Mhp1 63/285
Mhp1 63/362
Rhodopsin 137/326
Rhodopsin 151/241
Rhodopsin 151/308
Rhodopsin 151/326
Rhodopsin 225/252
Rhodopsin 225/308
Rhodopsin 252/326
Rhodopsin 63/241
Rhodopsin 63/252
Rhodopsin 63/326
Rhodopsin 74/137
Rhodopsin 74/225
Rhodopsin 74/252
Rhodopsin 74/308
Normalized echo intensity
Time (µs)
Experimental
Simulated

Figure S4 (on previous page). All simulated and experimental DEER decay data used in this study between experimentally resolved residues. RosettaDEER could generally, but not always, simulate DEER traces from native-like models that are comparable to the experimental data.

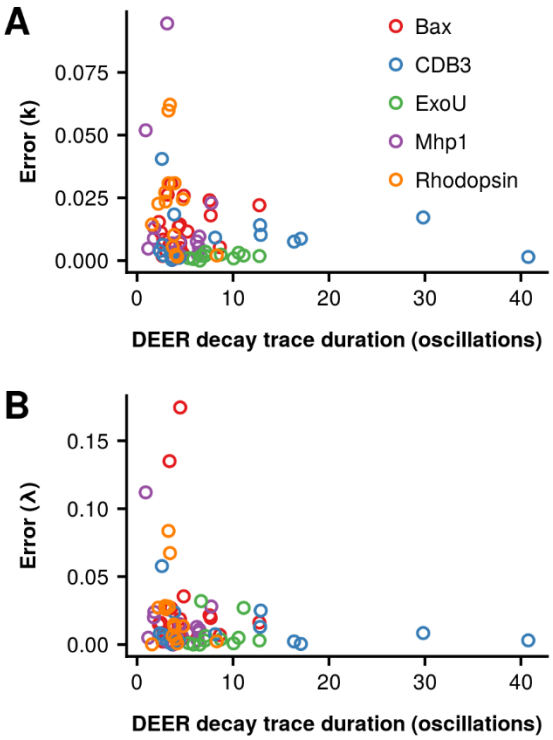

Figure S5. Deviation between experimental and simulated background decay (*k*) and modulation depths (*λ*).

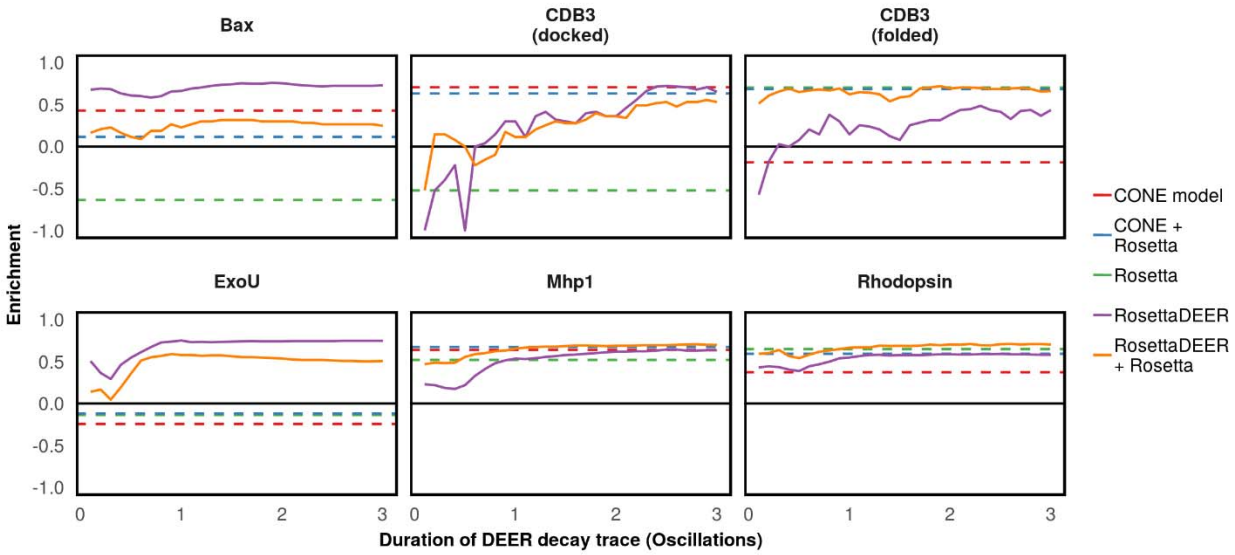

Figure S6. Enrichment of misfolded and misdocked decoys as a function of DEER decay trace duration. Enrichment was quantified as the logarithm of the percentage of native-like models (top 10% by RMSD100SSE) that were also in the top 10% by score. An enrichment of 1 indicates that the set of models constituting the top 10% by RosettaDEER score was identical to the set of models constituting the top 10% by Rosetta score.

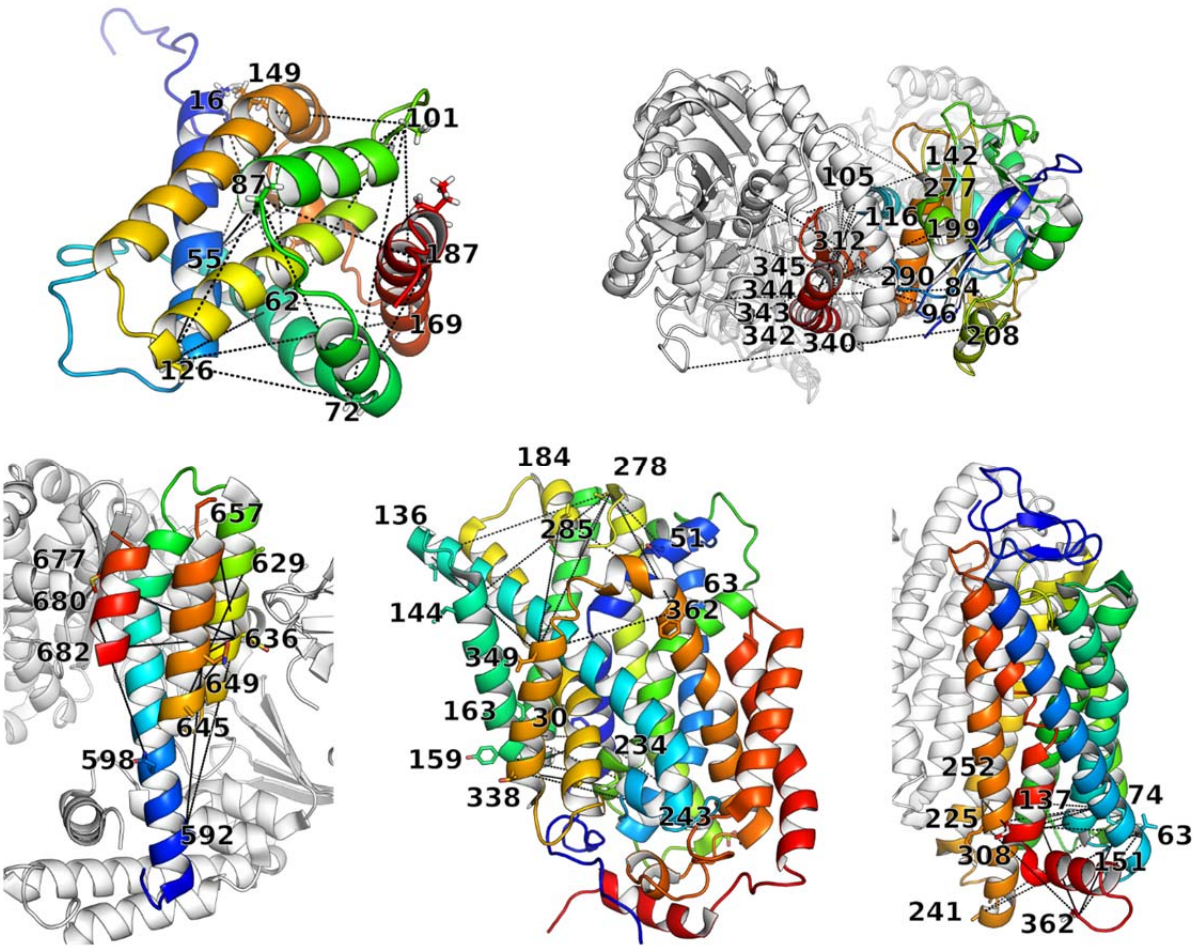

Figure S7. Placement of experimental DEER restraints on protein structures used in this study. Clockwise from top left: Bax (PDB: 1F16 model 8), CDB3 (PDB: 1HYN chains R/S), Rhodopsin (1GZM chain A), Mhp1 (PDB: 2JLN), and ExoU (PDB: 3TU3, C-terminus only).

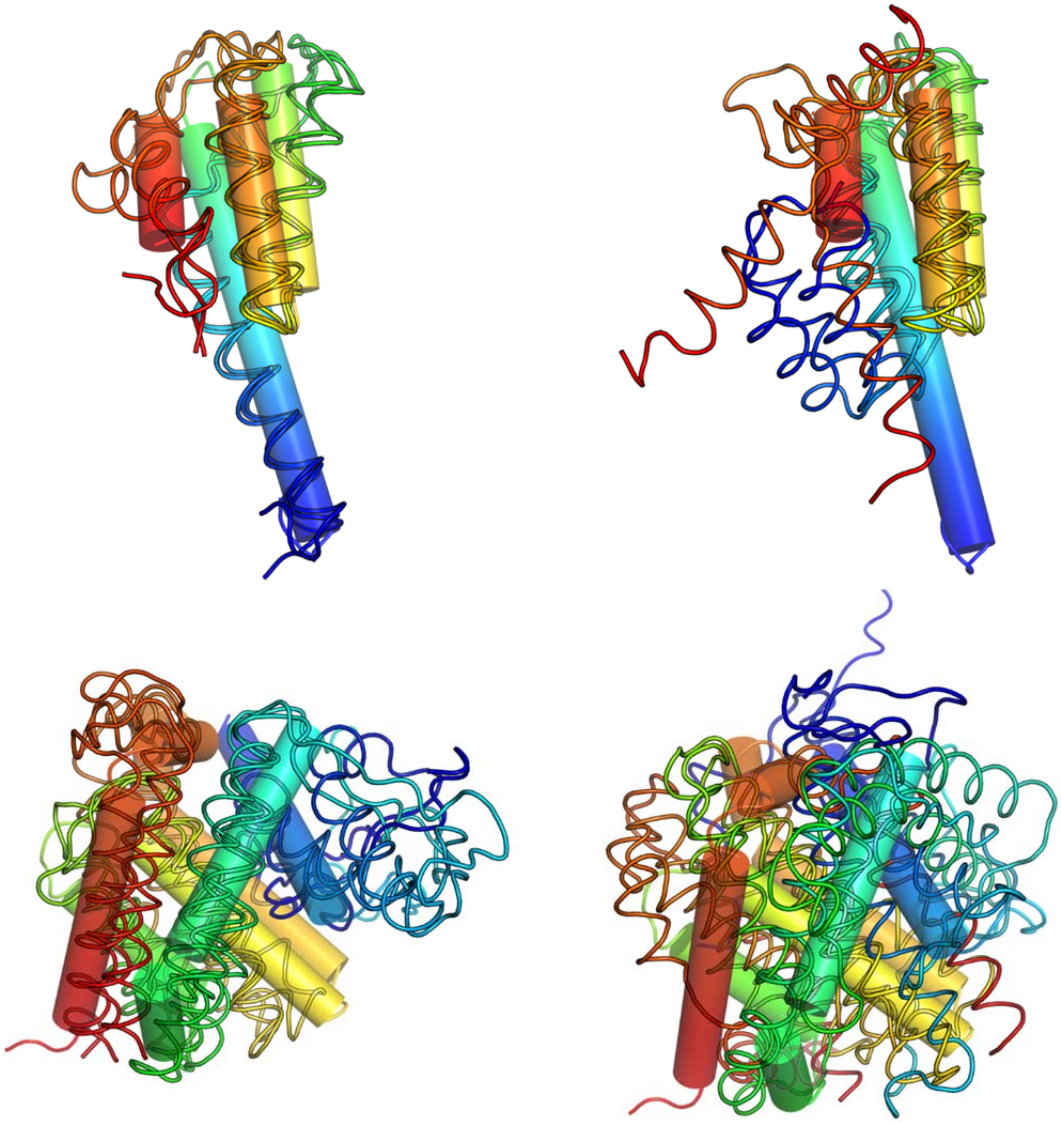

Figure S8. Effect of DEER restraints on structure prediction of Bax and ExoU. Top 3 best-scoring models of ExoU (top) and Bax (bottom) folded either with (left) or without (right) experimental DEER restraints. The native models are shown as cylinders for comparison.