Supplementary Information

Statistical Analysis on the Performance of Molecular Mechanics Poisson–Boltzmann Surface Area versus Absolute Binding Free Energy Calculations: Bromodomains as a Case Study

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Table S1

Summary table of the protein-ligand systems considered in the study. †ID refers to the indices used in Aldeghi et al.[1]; ‡ comparison to X-ray not possible due to the lack of a resolved structure.

	Protein	Ligand (ID)†	Lig pose from	Protein PDB	Sim length	No. dock poses	Reference
Test case 1a	BRD4(1)	BI-2356 (1)	X-ray	4OGI	10 ns	n.a.	Ciceri *et al.*[2]
	BRD4(1)	JQ1(+) (2)	X-ray	3MXF	10 ns	n.a.	Filippakopoulos *et al.*[3]
	BRD4(1)	RVX-OH (3)	X-ray	4MR3	10 ns	n.a.	Picaud *et al.*[4]
	BRD4(1)	TG-101348 (4)	X-ray	4OGJ	10 ns	n.a.	Ciceri *et al.*[2]
	BRD4(1)	Isox-2 (5)	X-ray	4J0R	10 ns	n.a.	Hewings *et al.*[5]
	BRD4(1)	BzT-7 (6)	X-ray	3U5L	10 ns	n.a.	Filippakopoulos *et al.*[6]
	BRD4(1)	RVX-208 (7)	X-ray	4MR4	10 ns	n.a.	Picaud *et al.*[4]
	BRD4(1)	Alprazolam (8)	X-ray	3U5J	10 ns	n.a.	Filippakopoulos *et al.*[6]
	BRD4(1)	Isox-1 (9)	X-ray	3SVG	10 ns	n.a.	Hewings *et al.*[7]
	BRD4(1)	Quinaz (10)	X-ray	4HBV	10 ns	n.a.	Fish *et al.*[8]
	BRD4(1)	Isox-cf3 (11)	Model	3SVG	10 ns	n.a.	Vidler *et al.*[9]
Test case 1b	BRD4(1)	BI-2356 (1)	Docking	2OSS	10 ns	3	Ciceri *et al.*[2]
	BRD4(1)	JQ1(+) (2)	Docking	2OSS	10 ns	2	Filippakopoulos *et al.*[3]
	BRD4(1)	RVX-OH (3)	Docking	2OSS	10 ns	5	Picaud *et al.*[4]
	BRD4(1)	TG-101348 (4)	Docking	2OSS	10 ns	1	Ciceri *et al.*[2]
	BRD4(1)	Isox-2 (5)	Docking	2OSS	10 ns	2	Hewings *et al.*[5]
	BRD4(1)	BzT-7 (6)	Docking	2OSS	10 ns	2	Filippakopoulos *et al.*[6]
	BRD4(1)	RVX-208 (7)	Docking	2OSS	10 ns	2	Picaud *et al.*[4]
	BRD4(1)	Alprazolam (8)	Docking	2OSS	10 ns	2	Filippakopoulos *et al.*[6]
	BRD4(1)	Isox-1 (9)	Docking	2OSS	10 ns	3	Hewings *et al.*[7]
	BRD4(1)	Quinaz (10)	Docking	2OSS	10 ns	1	Fish *et al.*[8]
	BRD4(1)	Isox-cf3 (11)	Docking	2OSS	10 ns	2‡	Vidler *et al.*[9]
Test case 2	BRD2(1)	RVX-OH	Docking	4ALG	15 ns	2	Picaud *et al.*[4]
	BRD2(2)	RVX-OH	Docking	4MR5	15 ns	2	Picaud *et al.*[4]
	BRD3(1)	RVX-OH	Docking	3S91	15 ns	2	Picaud *et al.*[4]
	BRD2(2)	RVX-OH	Docking	3S92	15 ns	2	Picaud *et al.*[4]
	BRD4(1)	RVX-OH	Docking	2OSS	15 ns	2	Picaud *et al.*[4]
	BRD4(2)	RVX-OH	Docking	2OUO	15 ns	2	Picaud *et al.*[4]
	BRDT(1)	RVX-OH	Docking	4KCX	15 ns	2	Picaud *et al.*[4]
	BRD2(1)	RVX-208	Docking	4ALG	15 ns	1	Picaud *et al.*[4]
	BRD2(2)	RVX-208	Docking	4MR5	15 ns	1	Picaud *et al.*[4]
	BRD3(1)	RVX-208	Docking	3S91	15 ns	1	Picaud *et al.*[4]
	BRD2(2)	RVX-208	Docking	3S92	15 ns	1	Picaud *et al.*[4]
	BRD4(1)	RVX-208	Docking	2OSS	15 ns	1	Picaud *et al.*[4]
	BRD4(2)	RVX-208	Docking	2OUO	15 ns	1	Picaud *et al.*[4]
	BRDT(1)	RVX-208	Docking	4KCX	15 ns	1	Picaud *et al.*[4]
Test case 3	CECR2	Bromosporine	Docking	3NXB	15 ns	1	Picaud *et al.*[10]
	FALZ	Bromosporine	Docking	3UV2	15 ns	1	Picaud *et al.*[10]
	PCAF	Bromosporine	Docking	3GG3	15 ns	1	Picaud *et al.*[10]
	BRD2(1)	Bromosporine	Docking	4ALG	15 ns	1	Picaud *et al.*[10]
	BRD2(2)	Bromosporine	Docking	4MR5	15 ns	1	Picaud *et al.*[10]
	BRD3(1)	Bromosporine	Docking	3S91	15 ns	1	Picaud *et al.*[10]
	BRD3(2)	Bromosporine	Docking	3S92	15 ns	1	Picaud *et al.*[10]
	BRD4(1)	Bromosporine	Docking	2OSS	15 ns	1	Picaud *et al.*[10]
	BRD4(2)	Bromosporine	Docking	2OUO	15 ns	1	Picaud *et al.*[10]
	BRDT(1)	Bromosporine	Docking	4KCX	15 ns	1	Picaud *et al.*[10]
	CREBBP	Bromosporine	Docking	4NYX	15 ns	1	Picaud *et al.*[10]
	EP300	Bromosporine	Docking	3I3J	15 ns	1	Picaud *et al.*[10]
	BRD1	Bromosporine	Docking	5AME	15 ns	1	Picaud *et al.*[10]
	BRD9	Bromosporine	Docking	4XY8	15 ns	1	Picaud *et al.*[10]
	BRPF1B	Bromosporine	Docking	4LC2	15 ns	1	Picaud *et al.*[10]
	BAZ2A	Bromosporine	Docking	4QBM	15 ns	1	Picaud *et al.*[10]
	TIF1	Bromosporine	Docking	4YBM	15 ns	1	Picaud *et al.*[10]
	TAF1(1)	Bromosporine	Docking	3UV5	15 ns	1	Picaud *et al.*[10]
	TAF1(2)	Bromosporine	Docking	3UV4	15 ns	1	Picaud *et al.*[10]
	TAF1L(2)	Bromosporine	Docking	3HMH	15 ns	1	Picaud *et al.*[10]
	PB1(5)	Bromosporine	Docking	3MB4	15 ns	1	Picaud *et al.*[10]
	SMARCA4	Bromosporine	Docking	2GRC	15 ns	1	Picaud *et al.*[10]

Figure S1

Distributions of Spearman correlation values for the protocol W0 and for ABFE calculations, obtained by bootstrap and based on the uncertainties of the experimental affinity measurements and computational predictions. The probability densities on the bottom row show the distribution of r_s for ABFE subtracted from the distribution of r_s for MMPBSA. The fraction of the area above or below zero is reported on the plots, the median is shown as a dashed line, and a difference value of zero is marked with a vertical gray dotted line.

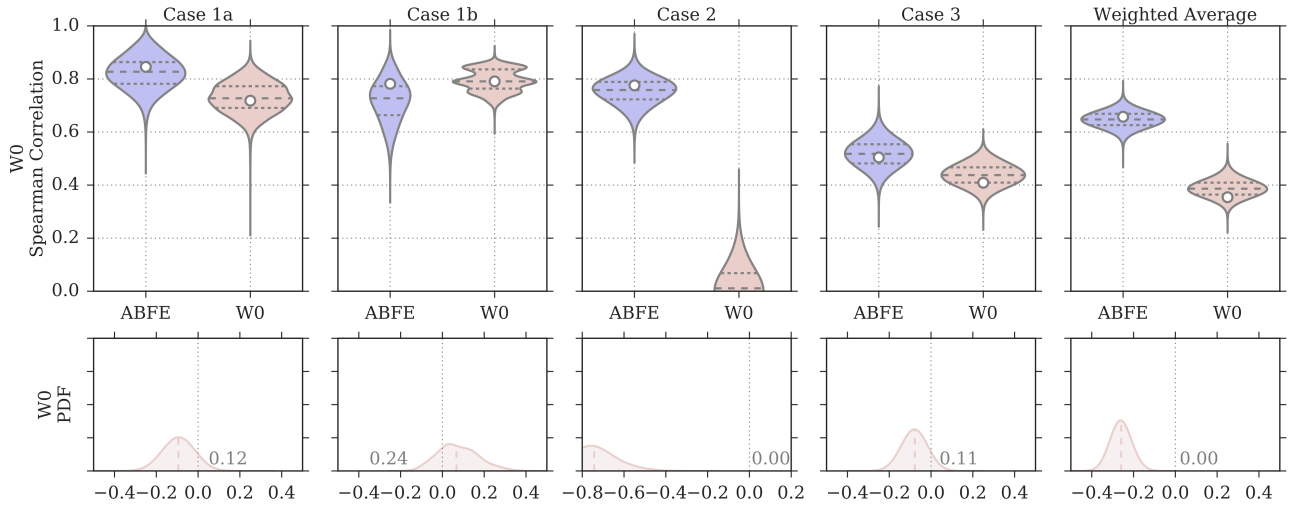

Figure S2

Distributions of Spearman correlation values for the protocol W0e and for ABFE calculations, obtained by bootstrap and based on the uncertainties of the experimental affinity measurements and computational predictions. The probability densities on the bottom row show the distribution of r_s for ABFE subtracted from the distribution of r_s for MMPBSA. The fraction of the area above or below zero is reported on the plots, the median is shown as a dashed line, and a difference value of zero is marked with a vertical gray dotted line.

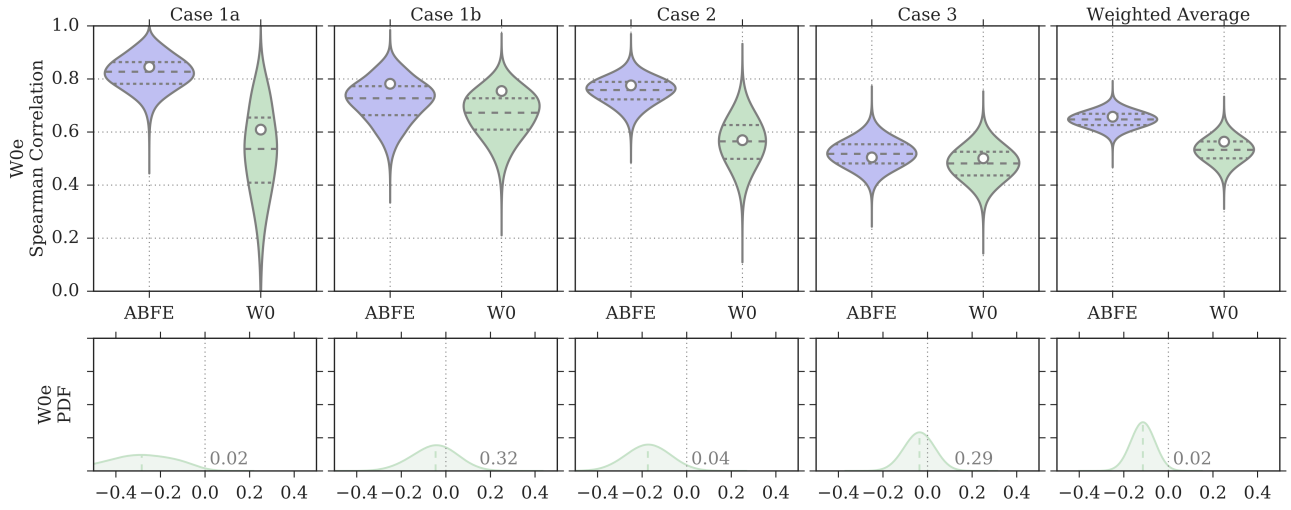

Figure S3

Distributions of Spearman correlation values for the protocols W10, W20, W30, W40, and W5, and for ABFE calculations, obtained by bootstrap and based on the uncertainties of the experimental affinity measurements and computational predictions. The probability densities on the bottom row show the distribution of r_s for ABFE subtracted from the distribution of r_s for MMPBSA. The fraction of the area above or below zero is reported on the plots, the median is shown as a dashed line, and a difference value of zero is marked with a vertical gray dotted line.

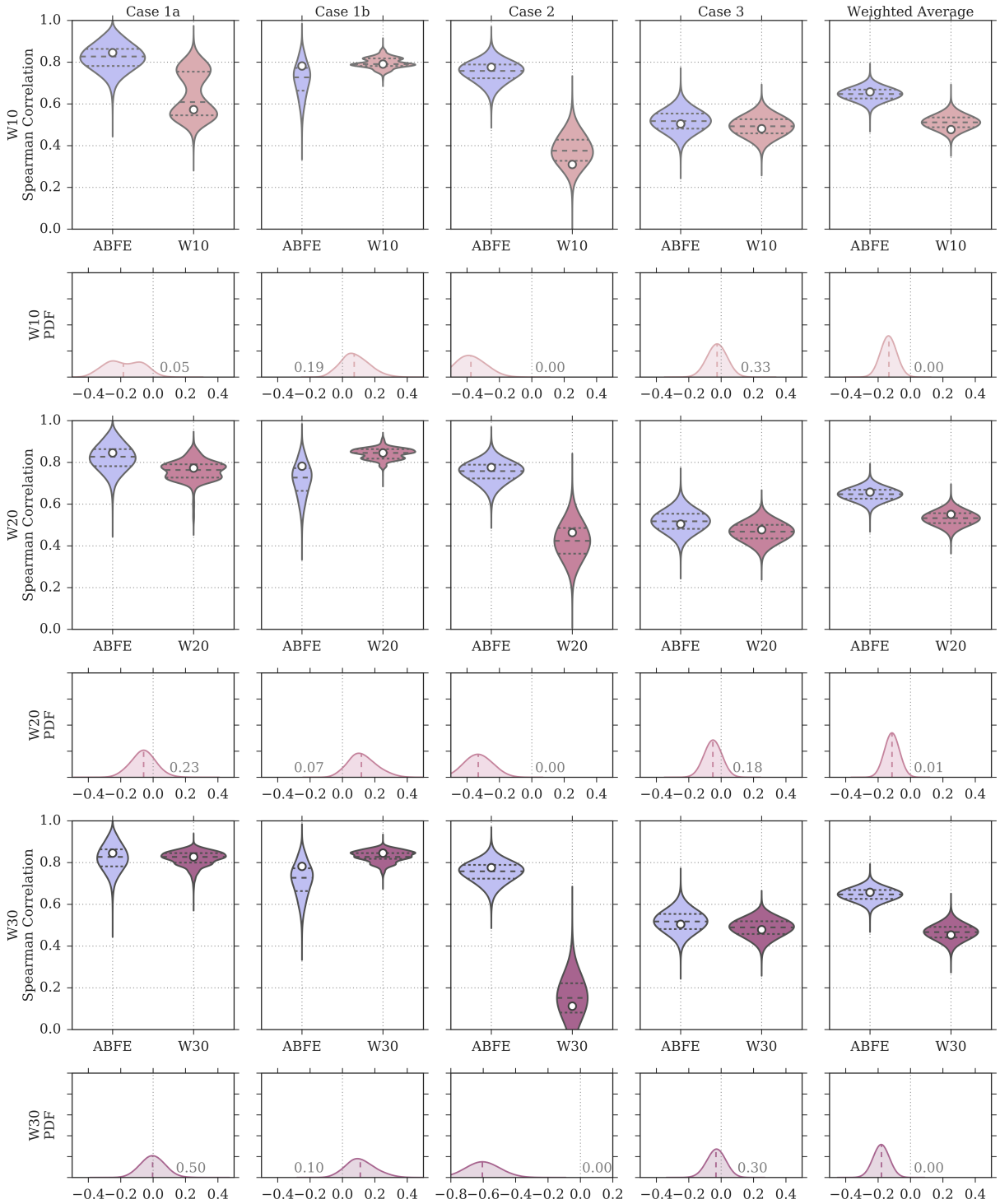

Figure S3 (continued)

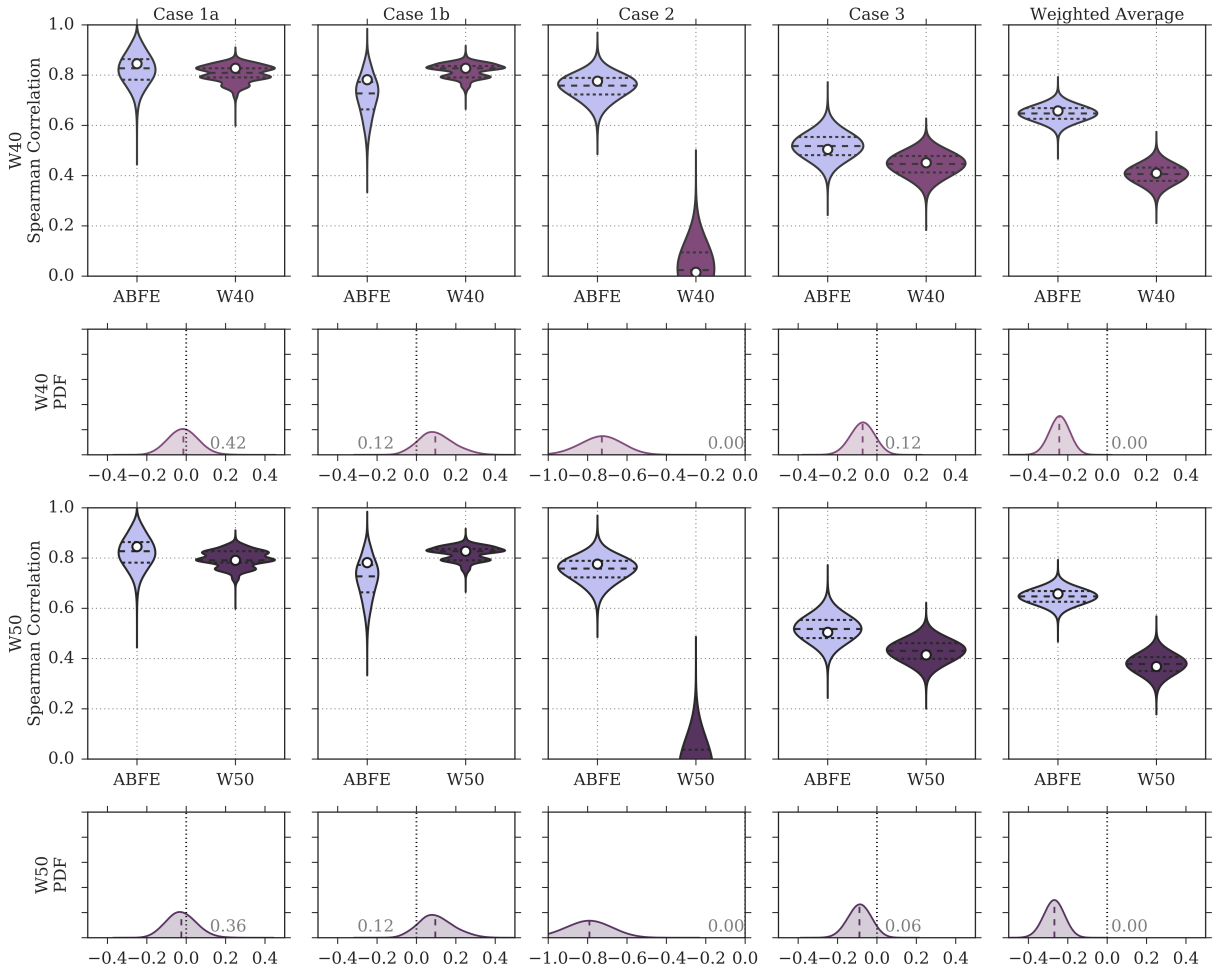

Table S1 Overview of the results obtained with ABFE and MMPBSA calculations, in terms of Pearson and Spearman correlation to experimental binding free energies, for the protocols W10e to W50e. In square brackets are the 95% confidence intervals of the statistics.

Pearson Correlation

Test Case N°	W10e	W20e	W30e	W40e	W50e
1a	0.31 [-0.09, 0.66]	0.62 [0.39, 0.75]	0.80 [0.60, 0.89]	0.87 [0.61, 0.93]	0.90 [0.74, 0.94]
1b	0.51 [0.35, 0.62]	0.85 [0.75, 0.90]	0.88 [0.80, 0.93]	0.92 [0.86, 0.95]	0.91 [0.86, 0.94]
2	0.64 [0.48, 0.74]	0.62 [0.30, 0.73]	0.58 [0.28, 0.72]	0.38 [0.06, 0.57]	0.34 [0.01, 0.55]
3	0.35 [0.24, 0.43]	0.24 [0.11, 0.34]	0.29 [0.16, 0.40]	0.22 [0.06, 0.36]	0.22 [0.05, 0.35]
Weighted Average	0.45 [0.28, 0.57]	0.47 [0.27, 0.57]	0.51 [0.32, 0.61]	0.43 [0.22, 0.56]	0.41 [0.22, 0.55]

Spearman Correlation

Test Case N°	W10e	W20e	W30e	W40e	W50e
1a	0.09 [-0.17, 0.49]	0.54 [0.19, 0.61]	0.61 [0.35, 0.88]	0.88 [0.45, 0.94]	0.90 [0.58, 0.95]
1b	0.35 [0.19, 0.56]	0.71 [0.60, 0.85]	0.83 [0.69, 0.91]	0.85 [0.77, 0.95]	0.88 [0.76, 0.95]
2	0.44 [0.26, 0.64]	0.50 [0.22, 0.71]	0.48 [0.13, 0.68]	0.29 [-0.05, 0.49]	0.31 [-0.08, 0.47]
3	0.35 [0.23, 0.49]	0.37 [0.21, 0.49]	0.42 [0.21, 0.53]	0.31 [0.11, 0.46]	0.27 [0.11, 0.46]
Weighted Average	0.34 [0.19, 0.54]	0.47 [0.26, 0.61]	0.51 [0.26, 0.66]	0.43 [0.18, 0.58]	0.43 [0.18, 0.58]

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