Supporting Material

Role of THz Fluctuations in the Allosteric Properties of the PDZ Domains

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Table S1. Cbeta of PDZ3

progressive #	residue #
1	306
2	307
3	308
4	309
5	310
6	311
7	312
8	313
9	314
10	315
11	316
12	317
13	318
14	320
15	321
16	323
17	325
18	326
19	327
20	328
21	331
22	332
23	334
24	336
25	337
26	338
27	339
28	340
29	341
30	342
31	343
32	346
33	347
34	348
35	349
36	350
37	352
38	353
39	354
40	355
41	357
42	358
43	359
44	360
45	361
46	362
47	363
48	365
49	366
50	367
51	368
52	369
53	370

54	371
55	372
56	373
57	374
58	375
59	376
60	377
61	378
62	379
63	380
64	381
65	382
66	384
67	385
68	386
69	387
70	388
71	389
72	390
73	391
74	392
75	393
76	394
77	395
78	396
79	397
80	398
81	399
82	400
83	401
84	402
85	403
86	404
87	405
88	406
89	407
90	408
91	409
92	411
93	412
94	413
95	414
96	415

Table S2. C of CH_3 in PDZ3

progressive #	#resid
1	307
2	307
3	314
4	314
5	315
6	315
7	316
8	316
9	321
10	323
11	323
12	327
13	327
14	328
15	328
16	336
17	336
18	338
19	338
20	341
21	341
22	342
23	342
24	343
25	347
26	349
27	349
28	353
29	353
30	359
31	359
32	360
33	360
34	362
35	362
36	365
37	365
38	367
39	367
40	370
41	375
42	376
43	377
44	377
45	378
46	379
47	379
48	382
49	385
50	386
51	386
52	387
53	388

54	388
55	389
56	389
57	390
58	402
59	406
60	406
61	412
62	412
63	413
64	413
65	414

Table S3. Cbeta of PDZ2

progressive #	residue #
1	1
2	2
3	3
4	5
5	6
6	7
7	8
8	9
9	10
10	11
11	12
12	13
13	14
14	15
15	16
16	17
17	18
18	20
19	21
20	22
21	23
22	26
23	27
24	28
25	29
26	30
27	31
28	32
29	35
30	36
31	37
32	38
33	39
34	40
35	41
36	42
37	43
38	45
39	46
40	47
41	48
42	49
43	51
44	52
45	53
46	54
47	56
48	57
49	58
50	59
51	60
52	61
53	62

54	64
55	65
56	66
57	67
58	69
59	70
60	71
61	72
62	73
63	74
64	75
65	76
66	77
67	78
68	79
69	80
70	81
71	83
72	84
73	85
74	86
75	87
76	88
77	89
78	90
79	91
80	93
81	94

Table S4. C of CH_3 in PDZ2

progressive #	residue #
1	6
2	6
3	9
4	9
5	11
6	11
7	12
8	18
9	18
10	20
11	20
12	22
13	22
14	23
15	26
16	26
17	28
18	30
19	30
20	35
21	35
22	37
23	37
24	39
25	40
26	40
27	41
28	41
29	45
30	46
31	52
32	52
33	58
34	58
35	59
36	59
37	60
38	61
39	61
40	64
41	64
42	66
43	66
44	69
45	70
46	74
47	75
48	75
49	77
50	78
51	78
52	81

53	84
54	84
55	85
56	85
57	87
58	87
59	88
60	88
61	89
62	89

The vibrational density of states of the responding subsets.

We have focussed on the THz vibrational dynamics of the response nuclei defined by means of the RMSF analyses in the THz regime, as described in detail in the main text.

THz-frequency vibrational modes determine protein flexibility and the presence of additional intermolecular interactions between the protein and the ligand can lead to changes in the spectrum of the vibrational density of states (VDOS) (Turton, D. A. et al., *Nature Comm*. **2013**, *5*, 3999.)

The VDOS is defined as the Fourier transform of the autocorrelation function of the atomic velocities and here it has been computed considering the set of trajectories b).

Prior to investigating the vibrational features of the two response nuclei, we may wonder whether and how the features we find with RMSF analysis reflect on the vibrational properties of the investigated systems, i.e. of the whole proteins. This can be assessed quantitatively by calculating the vibrational density of states (VDOS) of the molecule. As shown in Fig. S1 for the PDZ3, the VDOS is characterized by the above described peak, commonly observed in proteins. Our results show that there is no appreciable difference between the VDOS of the bound and the unbound system.

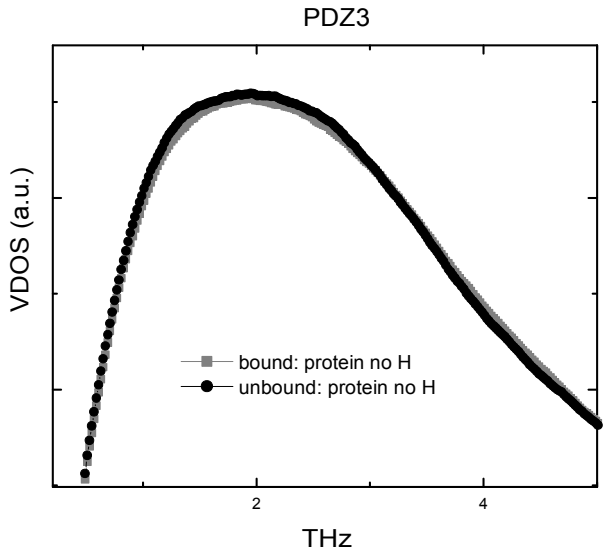

Fig. S1: VDOS of the PDZ3 in the bound and the unbound state, taking into account the heavy atoms of the protein.

This is in agreement with a recent study that suggested that the contribution of specific modes involved in substrate binding is even more important than the net difference between the bound and unbound state VDOS (Niessen, K. A. et al., Biophysical Journal 112, 933–942, March

14, 2017).

This is why we decided to quantitatively estimate the local and delocalized density of vibrational modes involving the atoms belonging to the two response nuclei revealed by the RMSF analysis. To verify its alteration due to the binding event, we have calculated the VDOS of the response nuclei in the bound and unbound state.

As shown in Fig. S2, we find that the presence of the ligand causes a blue-shift of the vibrational modes of the response nucleus (first response nucleus). This finding, together with the results of the RMSF analysis, points to a stiffening and a reduction in flexibility of the binding site at THz frequencies, due to strengthened interactions between the latter and the ligand. Vice versa, the second responding set shows an opposite behaviour, i.e. its vibrational frequencies are red-shifted in the presence of the ligand (cf. Fig. S3). Thus, the vibrational features of the two identified responding sets significantly depend on the presence of the ligand.

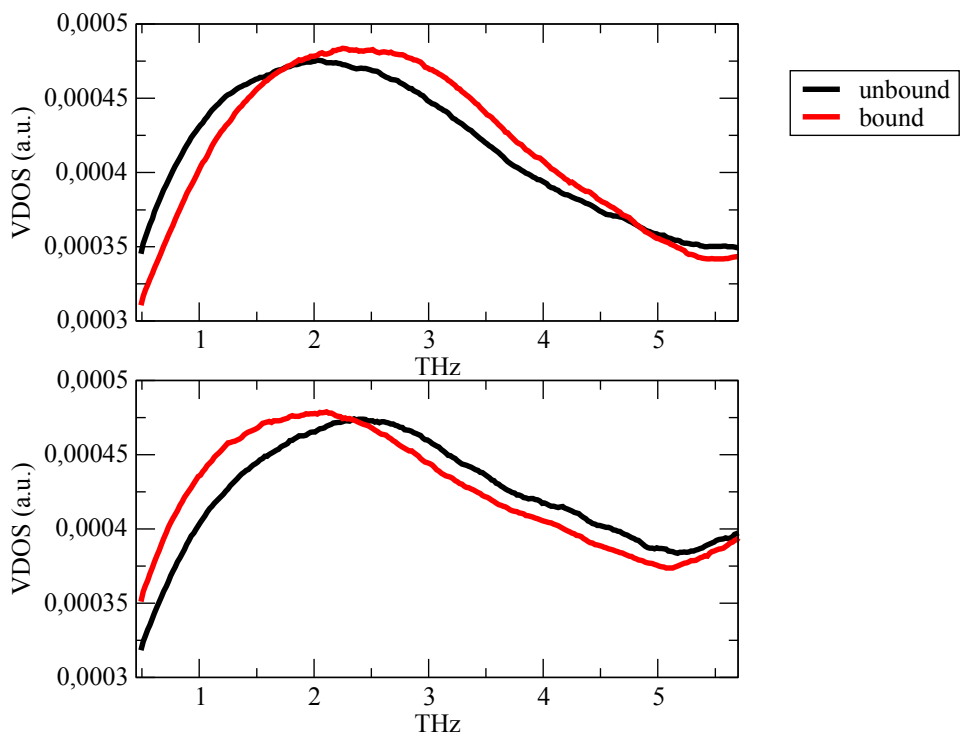

Figure S2. PDZ3 system. Vibrational Density of States (VDoS) of the first (top) and the second responding subsets (bottom).

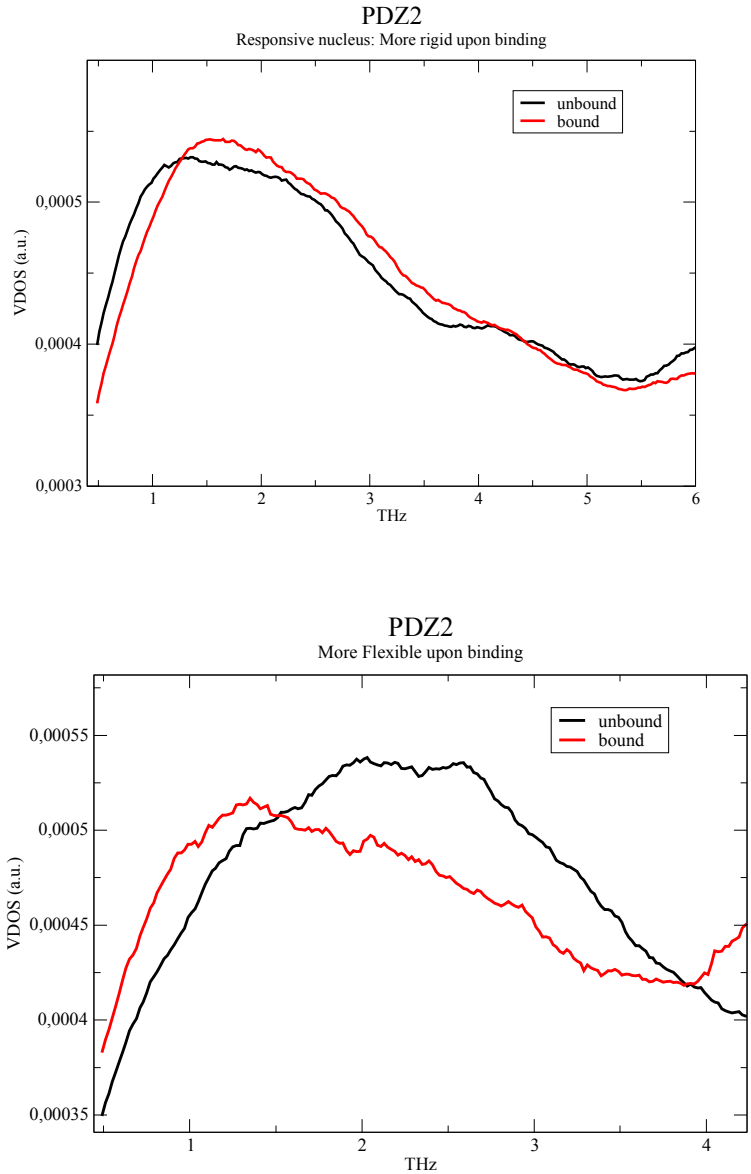

Figure S3. PDZ2 system. Vibrational Density of States (VDoS) of the first (top) and the second responding subsets (bottom).