Supplementary Material

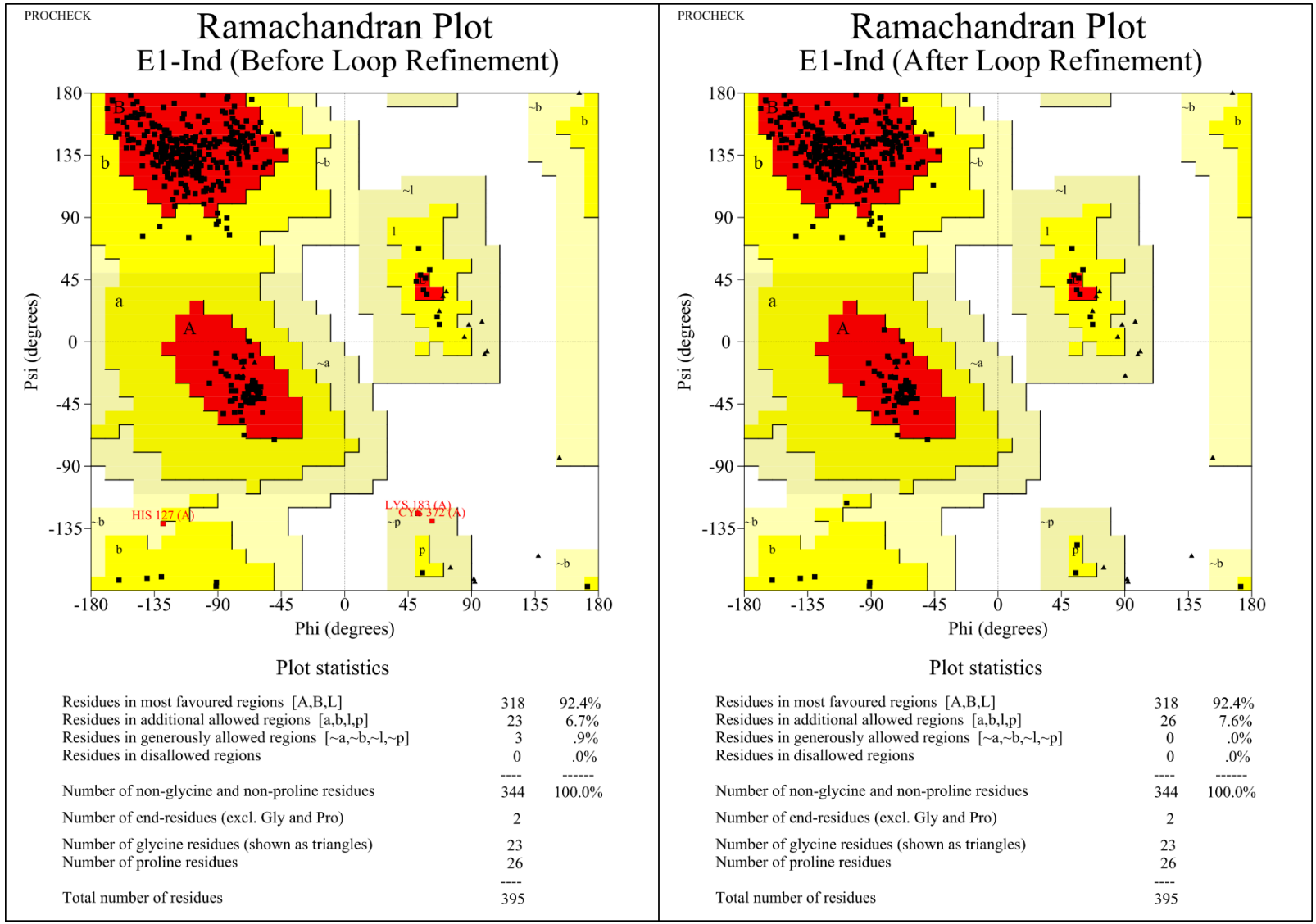

Figure S1. Ramachandran plot of the chain E1-Ind before (left) and after (right) iterative loop refinement.

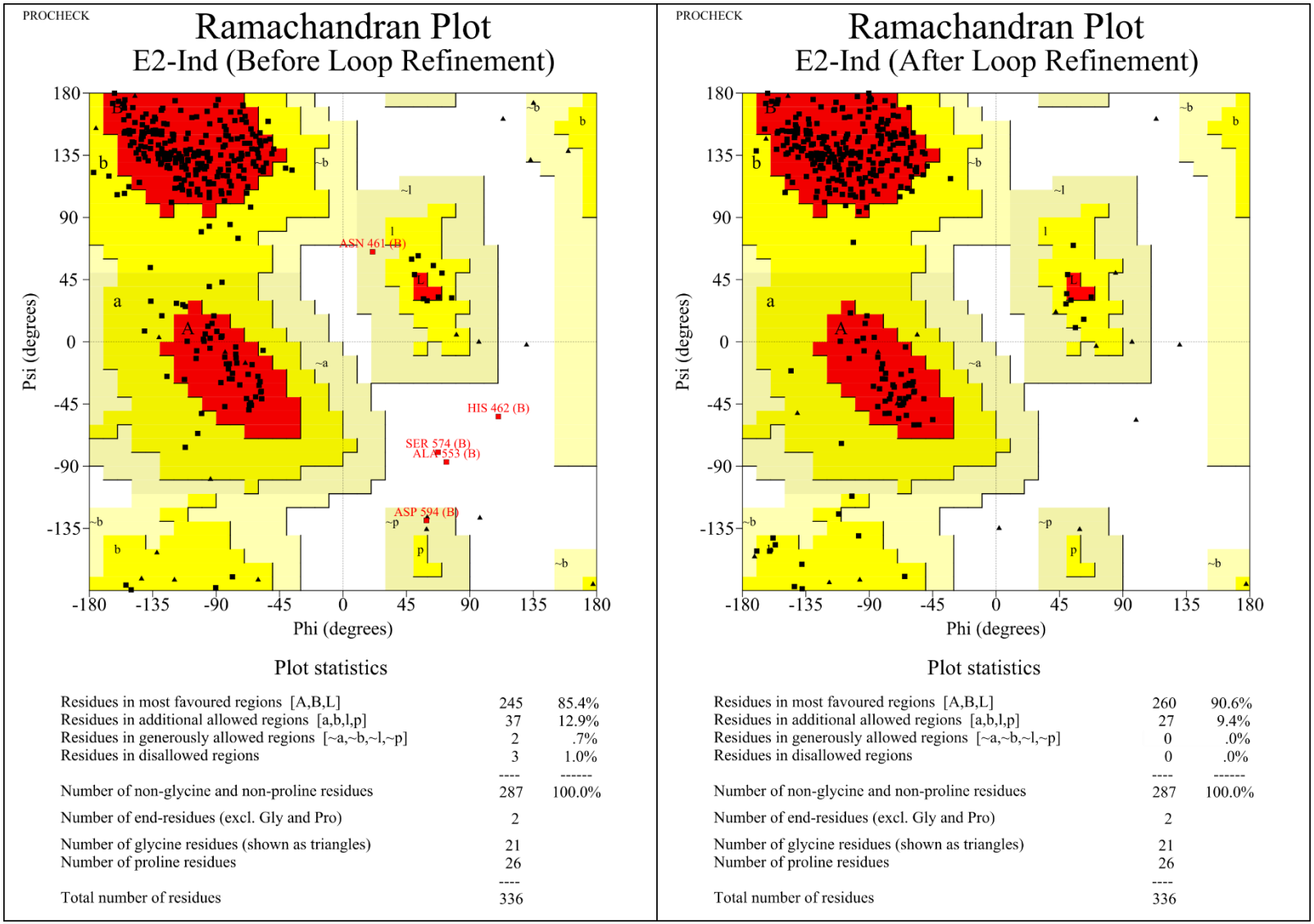

Figure S2. Ramachandran plot of the chain E2-Ind before (left) and after (right) iterative loop refinement.

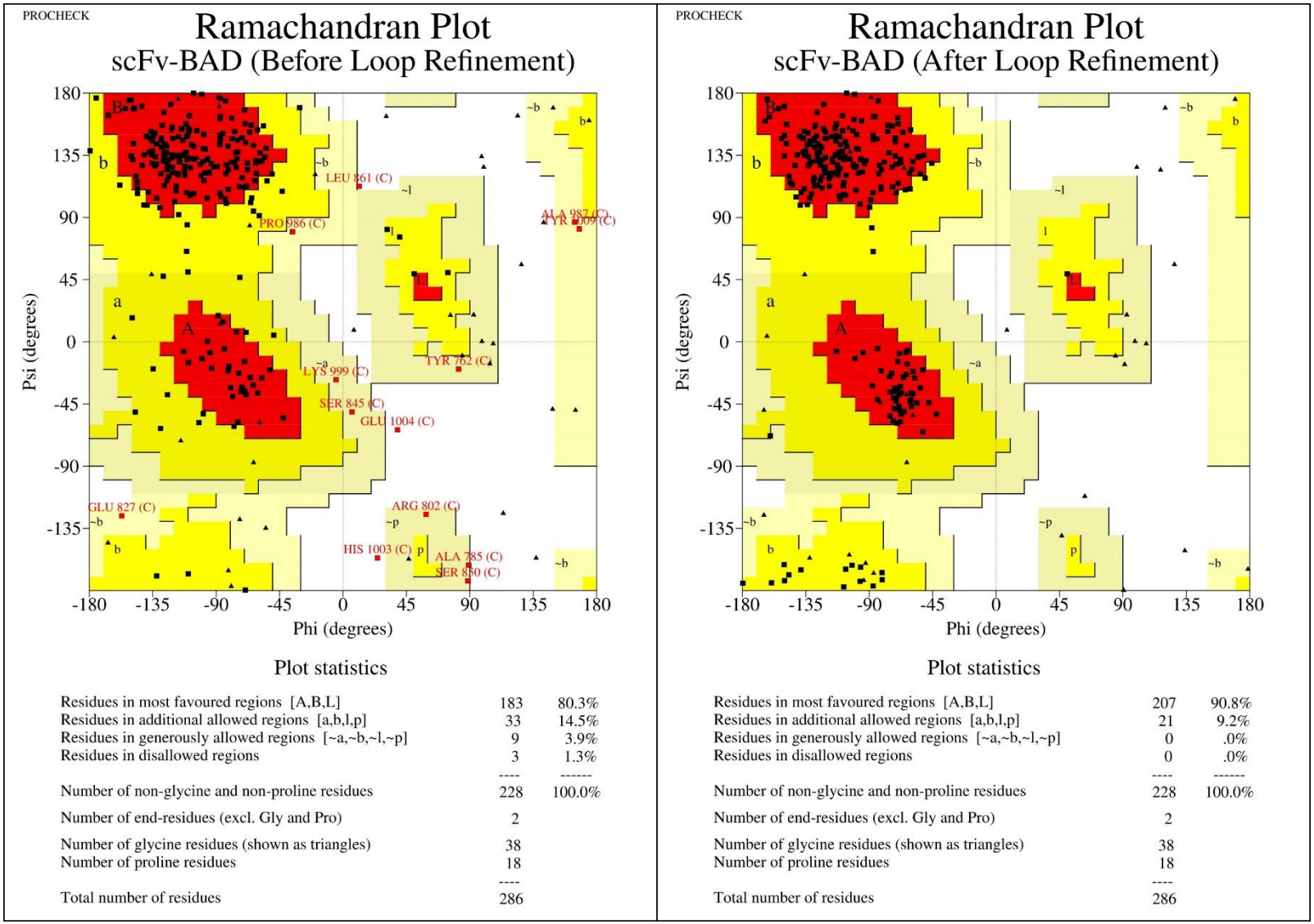

Figure S3. Ramachandran plot of the chain anti-E1E2-scFv-BAD before (left) and after (right) iterative loop refinement.

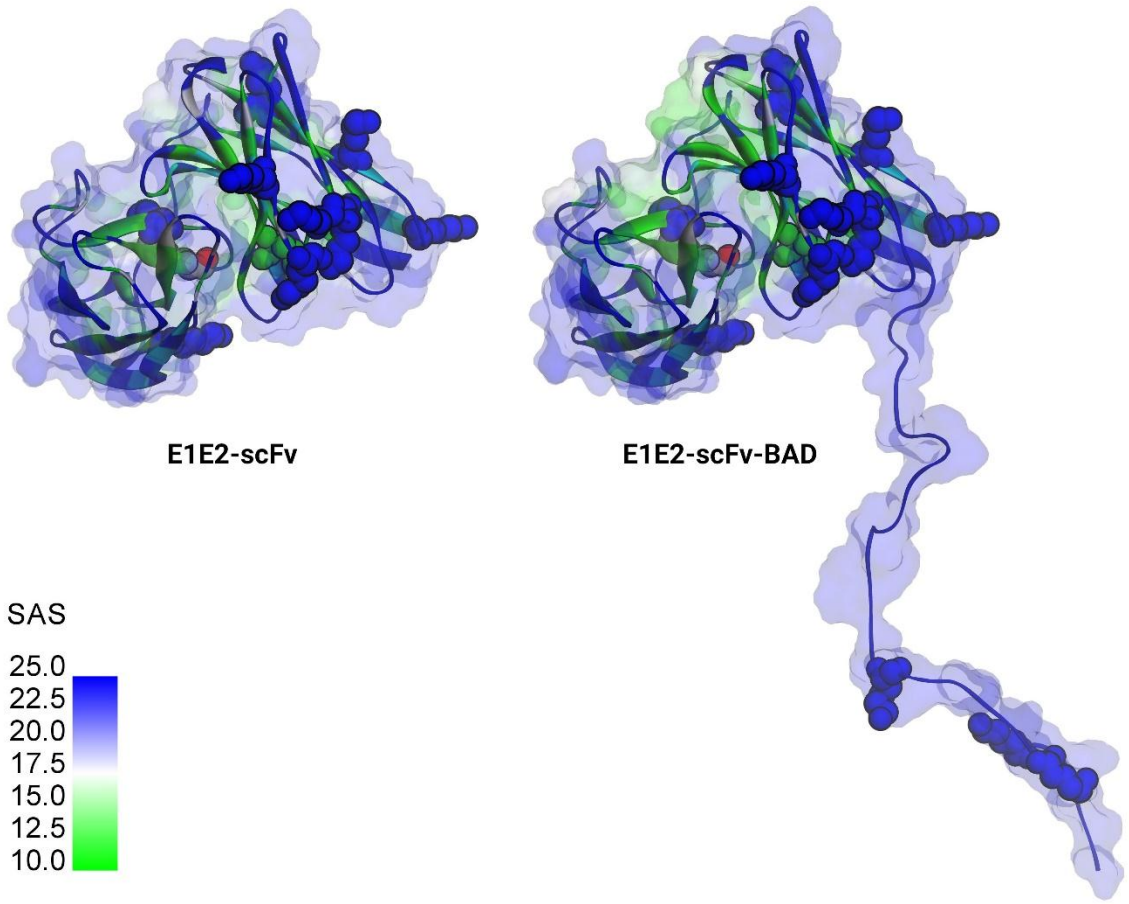

Figure S4. The distribution of lysine residues in the structures of anti-E2E2-scFv and anti-E1E2-scFv-BAD. The lysine residues are represented by space-filling atoms. The exposed lysine residues are in blue color, whereas the buried ones are in green color.

Table S1. Non-bonded interactions formed by scFv-BAD and E2, from E1E2-African and E1E2-Ind, before and after MD simulations. The simulations include CGMD and all-atom MD. The interactions were analyzed from a snapshot at 9 ns of each all-atom MD trajectory.

Interaction Category	Interaction Type	Number of interaction(s)			
		Before MD simulations		After MD simulations	
		E1E2-African	E1E2-Ind	E1E2-African	E1E2-Ind
H-Bond;Electrostatic	Salt Bridge;Attractive Charge	3	2	3	6
Electrostatic	Attractive Charge	1	2	3	2
Electrostatic	π-Cation			1	1
Electrostatic	π-Anion		1		1
H-Bond	Conventional H-Bond	6	9	15	17
H-Bond	Carbon H-Bond	5	1	10	9
H-Bond	π-Donor H-Bond			2	1
Hydrophobic	π- π T				2
Hydrophobic	π-Sigma			1	
Hydrophobic	Amide-Pi Stacked			1	
Hydrophobic	Alkyl	1	1	2	
Hydrophobic	π-Alkyl	3	5	2	4

Table S2. Hydrogen bond (H-bond) interactions between E1E2-African and scFv-BAD and their conservation levels. H-bond analysis was calculated from 10-ns all-atom MD trajectory.

No.	#Acceptor			DonorH	H-Bond Conservation (%)
1	scFv-BAD	ASN_95@OD1	E2	LYS_10@HZ3	0.8
2	scFv-BAD	ASN_95@OD1	E2	LYS_10@HZ1	0.8
3	scFv-BAD	ASN_95@O	E2	LYS_10@HZ1	0.2
4	scFv-BAD	ASN_95@O	E2	LYS_10@HZ2	0.1
5	scFv-BAD	ASN_95@OD1	E2	LYS_10@HZ2	0.1
6	scFv-BAD	ASN_95@O	E2	LYS_10@HZ3	0.1
7	scFv-BAD	GLY_152@O	E2	ASN_193@HD22	5.2
8	scFv-BAD	ASP_226@OD2	E2	GLN_195@HE22	17.9
9	scFv-BAD	ASP_226@OD1	E2	GLN_195@HE22	12.7
10	scFv-BAD	ASP_231@OD1	E2	GLN_195@HE21	46.6
11	scFv-BAD	ASP_231@OD2	E2	GLN_195@HE21	19.8
12	scFv-BAD	GLU_58@O	E2	THR_196@HG1	3.7
13	scFv-BAD	GLU_58@OE2	E2	LYS_233@HZ2	32.1
14	scFv-BAD	GLU_58@OE2	E2	LYS_233@HZ1	23.9
15	scFv-BAD	GLU_58@OE2	E2	LYS_233@HZ3	23.6
16	scFv-BAD	GLU_58@OE1	E2	LYS_233@HZ3	10.2
17	scFv-BAD	GLU_58@OE1	E2	LYS_233@HZ2	7.1
18	scFv-BAD	GLU_58@OE1	E2	LYS_233@HZ1	7.1
19	scFv-BAD	ASP_226@OD2	E2	LYS_233@HZ3	0.3
20	scFv-BAD	ASP_226@OD2	E2	LYS_233@HZ2	0.1
21	scFv-BAD	ASP_231@OD2	E2	LYS_233@HZ2	20.8
22	scFv-BAD	ASP_231@OD2	E2	LYS_233@HZ1	19.3
23	scFv-BAD	ASP_231@OD2	E2	LYS_233@HZ3	14.6
24	scFv-BAD	ASP_226@OD1	E2	LYS_234@HZ1	16
25	scFv-BAD	ASP_226@OD2	E2	LYS_234@HZ3	14.5
26	scFv-BAD	ASP_226@OD2	E2	LYS_234@HZ2	14.3
27	scFv-BAD	ASP_226@OD1	E2	LYS_234@HZ3	14
28	scFv-BAD	ASP_226@OD1	E2	LYS_234@HZ2	12.6
29	scFv-BAD	ASP_226@OD2	E2	LYS_234@HZ1	10.2
30	scFv-BAD	GLY_227@O	E2	LYS_234@HZ3	0.2
31	scFv-BAD	ASN_33@O	E2	TRP_235@HE1	4.5
32	scFv-BAD	GLY_32@O	E2	LYS_254@H	1.3
33	scFv-BAD	TYR_31@OH	E2	HIE_256@H	0.1
34	E2	ASN_7@OD1	scFv-BAD	TYR_31@HH	0.2
35	E2	HIE_256@ND1	scFv-BAD	TYR_31@HH	4.6
36	E2	HIE_256@O	scFv-BAD	TYR_31@HH	0.2
37	E2	LYS_254@O	scFv-BAD	GLY_32@H	1
38	E2	TRP_235@NE1	scFv-BAD	ASN_33@H	0.1
39	E2	HIE_232@O	scFv-BAD	TYR_52@HH	29.2
40	E2	HIE_232@ND1	scFv-BAD	ASN_56@HD22	0.5
41	E2	HIE_232@ND1	scFv-BAD	ASN_56@HD21	0.5
42	E2	THR_58@O	scFv-BAD	TYR_159@HH	2

43	E2	ASP_60@OD1	scFv-BAD	TYR_159@HH	79.4
44	E2	ASP_60@OD2	scFv-BAD	TYR_159@HH	4.6
45	E2	GLY_194@O	scFv-BAD	ARG_224@HH22	85.4
46	E2	GLY_194@O	scFv-BAD	ARG_224@HH12	34.8
47	E2	GLN_195@OE1	scFv-BAD	ARG_224@HH12	0.4
48	E2	GLN_195@NE2	scFv-BAD	ARG_224@HH12	0.1
49	E2	ASP_60@OD1	scFv-BAD	TYR_228@HH	76.2
50	E2	ASP_60@OD2	scFv-BAD	TYR_228@HH	18.1

Table S3. Hydrogen bond (H-bond) interactions between E1E2-Ind and scFv-BAD and their conservation levels. H-bond analysis was calculated from 10-ns all-atom MD trajectory.

No.	**#Acceptor**		**DonorH**		**H-Bond Conservation (%)**
1	E2	VAL_8@O	scFv-BAD	TYR_8@H	1
2	E2	THR_58@OG1	scFv-BAD	ASN_58@HD21	11
3	E2	THR_58@O	scFv-BAD	TYR_58@HD22	1
4	E2	THR_58@O	scFv-BAD	ASN_58@HD21	1
5	E2	ASP_59@O	scFv-BAD	TYR_59@HH	65
6	E2	ASP_59@OD1	scFv-BAD	ASN_59@HD21	14
7	E2	ASP_59@OD2	scFv-BAD	TYR_59@HH	11
8	E2	ASP_59@OD2	scFv-BAD	ASN_59@HD22	4
9	E2	ASP_59@OD1	scFv-BAD	ASN_59@HD22	2
10	E2	ASP_59@OD2	scFv-BAD	LYS_59@HZ3	2
11	E2	ASP_59@OD2	scFv-BAD	LYS_59@HH	2
12	E2	ASP_59@OD2	scFv-BAD	ASN_59@HZ1	1
13	E2	ASP_59@OD1	scFv-BAD	TYR_59@H	1
14	E2	ALA_162@O	scFv-BAD	TYR_162@HD21	48
15	E2	ASN_193@OD1	scFv-BAD	TYR_193@	5
16	E2	ASN_193@OD1	scFv-BAD	GLY_193@H	1
17	E2	SER_194@OG	scFv-BAD	GLY_194@HH	1
18	E2	GLN_195@OE1	scFv-BAD	GLY_195@H	2
19	E2	GLN_195@OE1	scFv-BAD	SER_195@H	1
20	E2	GLN_195@OE1	scFv-BAD	SER_195@HG	1
21	E2	THR_196@O	scFv-BAD	SER_196@H	12
22	E2	HIE_232@O	scFv-BAD	ASN_232@HD22	72
23	E2	HIE_232@ND1	scFv-BAD	LEU_232@HG	4
24	E2	HIE_232@ND1	scFv-BAD	ASN_232@HD21	1
25	E2	LYS_233@O	scFv-BAD	TYR_233@HD22	31
26	E2	LYS_233@O	scFv-BAD	TYR_233@HH	2
27	E2	LYS_234@O	scFv-BAD	ASN_234@H	9
28	E2	LYS_254@O	scFv-BAD	ASN_254@HD21	20
29	E2	HID_256@NE2	scFv-BAD	ASN_256@HD22	1
30	E2	HID_256@NE2	scFv-BAD	ASN_256@HH	1
31	scFv-BAD	ASN_33@OD1	E2	HID_33@HH	6
32	scFv-BAD	ALA_54@O	E2	ARG_54@H	1

33	scFv-BAD	GLU_58@OE1	E2	LYS_58@HZ3	14
34	scFv-BAD	GLU_58@OE1	E2	LYS_58@HZ1	11
35	scFv-BAD	GLU_58@OE1	E2	LYS_58@HZ3	11
36	scFv-BAD	GLU_58@OE2	E2	GLN_58@HE21	10
37	scFv-BAD	GLU_58@OE2	E2	LYS_58@HZ2	9
38	scFv-BAD	GLU_58@OE1	E2	GLN_58@HE22	7
39	scFv-BAD	GLU_58@OE2	E2	GLN_58@HE22	6
40	scFv-BAD	GLU_58@OE2	E2	LYS_58@HZ1	6
41	scFv-BAD	GLU_58@OE1	E2	GLN_58@HE22	3
42	scFv-BAD	GLU_58@OE2	E2	LYS_58@HZ2	3
43	scFv-BAD	SER_59@O	E2	THR_59@H	7
44	scFv-BAD	SER_59@O	E2	GLN_59@HE21	3
45	scFv-BAD	SER_59@O	E2	GLN_59@HE21	3
46	scFv-BAD	SER_59@OG	E2	HIE_59@HZ2	2
47	scFv-BAD	SER_59@OG	E2	THR_59@H	1
48	scFv-BAD	ASN_95@O	E2	LYS_95@HZ2	25
49	scFv-BAD	ASN_95@O	E2	LYS_95@HZ1	24
50	scFv-BAD	ASN_95@O	E2	LYS_95@HZ1	14
51	scFv-BAD	ASN_95@OD1	E2	LYS_95@HZ3	13
52	scFv-BAD	ASN_95@OD1	E2	LYS_95@HZ2	9
53	scFv-BAD	ASN_95@OD1	E2	LYS_95@HZ1	6
54	scFv-BAD	GLY_123@O	E2	SER_123@HG	29
55	scFv-BAD	GLY_123@O	E2	LEU_123@HG1	3
56	scFv-BAD	GLY_123@O	E2	THR_123@HE2	1
57	scFv-BAD	SER_124@O	E2	SER_124@HG	4
58	scFv-BAD	GLY_127@O	E2	ASN_127@HD21	2
59	scFv-BAD	SER_181@O	E2	THR_181@HD21	8
60	scFv-BAD	ASN_183@OD1	E2	LYS_183@HZ2	5
61	scFv-BAD	ASN_183@OD1	E2	LYS_183@HZ3	4
62	scFv-BAD	ASN_183@OD1	E2	LYS_183@HG1	2
63	scFv-BAD	ASP_226@OD2	E2	LYS_226@HZ3	14
64	scFv-BAD	ASP_226@OD2	E2	LYS_226@HZ1	12
65	scFv-BAD	ASP_226@OD2	E2	LYS_226@HZ3	8
66	scFv-BAD	ASP_231@OD1	E2	LYS_231@HZ2	17
67	scFv-BAD	ASP_231@OD1	E2	LYS_231@HZ1	16
68	scFv-BAD	ASP_231@OD1	E2	LYS_231@HZ1	8
69	scFv-BAD	ASP_231@OD1	E2	GLN_231@HE21	3
70	scFv-BAD	ASP_231@OD2	E2	LYS_231@HZ3	2
71	scFv-BAD	ASP_231@OD2	E2	LYS_231@HZ2	2
72	scFv-BAD	ASP_231@OD2	E2	SER_231@HE21	1
73	scFv-BAD	ASP_231@OD2	E2	GLN_231@H	1
74	scFv-BAD	ASP_231@OD2	E2	LYS_231@HH12	1
75	scFv-BAD	TYR_232@OH	E2	ASN_232@HG	8

Table S4. MMGBSA binding energy values and their components for the E1E2-African and the E1E2-Ind binding to scFv-BAD.

	E1E2-African:scFv-BAD		**E1E2-Ind:scFv-BAD**	
Energy Component*	**Average**	**Std. Dev.**	**Average**	**Std. Dev.**
E_{vdW}	-136.9617	8.7632	-170.1476	9.7280
$E_{electrostatic}$	-487.3570	47.3147	-561.0648	74.9945
E_{GB}	601.3991	44.7639	708.3794	69.5596
E_{surf}	-19.9217	0.7053	-24.0793	1.5356
ΔG_{gas}	-624.3186	48.2403	-731.2125	79.3188
ΔG_{solv}	581.4773	44.4412	684.3001	68.7659
ΔG_{total}	-42.8413	7.2594	-46.9123	14.9607

* Molecular mechanic energy terms and 1-4 interaction scaling factors were canceled out due to MMGBSA calculation from single trajectory. All energy components are in kcal/mol.

Tabel S5. The sensitivity assay of biotinylated scFv against NS1 DENV and BSA by ELISA method

[scFv-BAD] µM	Absorbance 490 nm		
	E2 CHIKV	NS1 DENV	BSA
0.2	0.151	0.136	0.063
0.3	0.151	0.153	0.072
0.6	0.184	0.152	0.084
1.3	0.190	0.157	0.091
2.6	0.217	0.161	0.107
5.3	0.296	0.171	0.111