Conformational and stability analysis of SARS-CoV-2 spike protein variants by molecular simulation

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Supplementary Figures

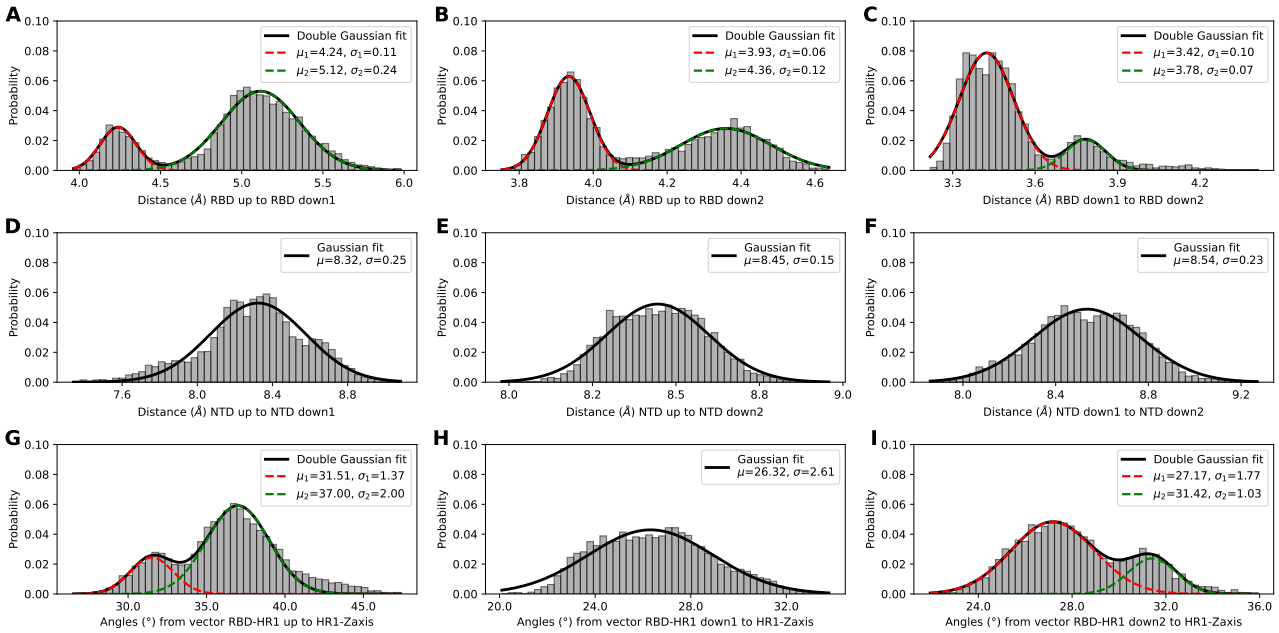


Figure S1: Gaussian distribution of the WT protein for all calculated CVs. The black line indicates a fitting for a single Gaussian distribution while the dotted lines (red or green) for a double distribution.

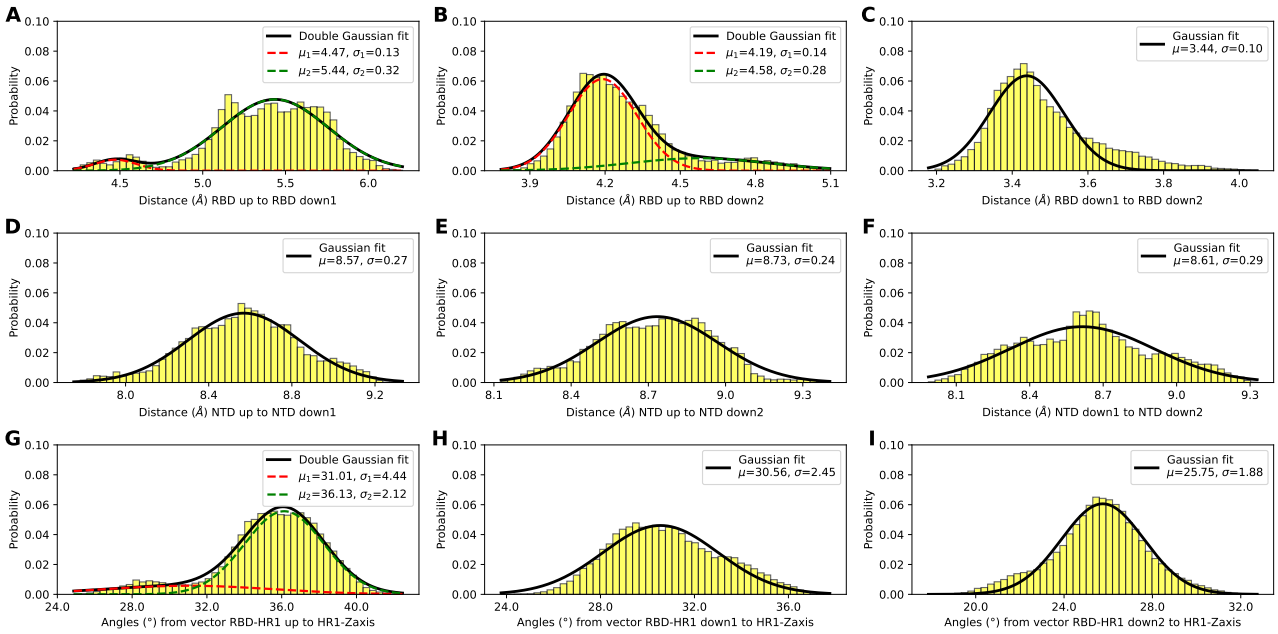


Figure S2: Gaussian distribution of the Delta variant for all calculated CVs. The black line indicates a fitting for a single Gaussian distribution while the dotted lines (red or green) for a double distribution.

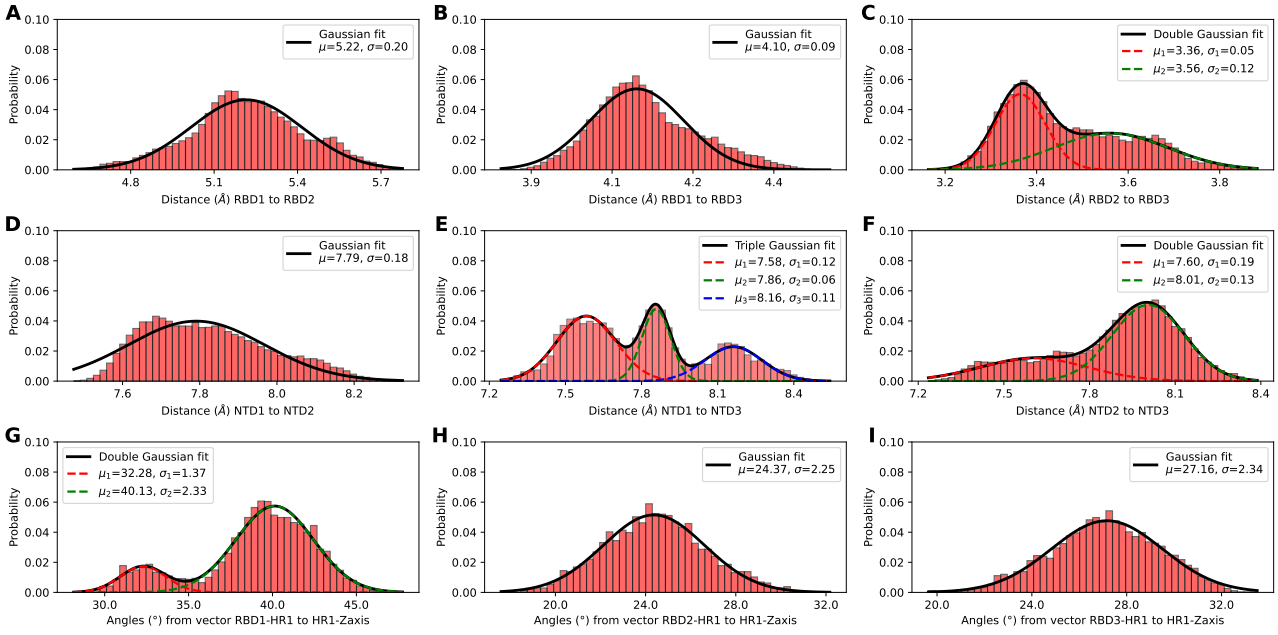


Figure S3: Gaussian distribution of the BA.1 variant for all calculated CVs. The black line indicates a fitting for a single Gaussian distribution while the dotted lines (red, green, or blue) for a double or triple distribution.

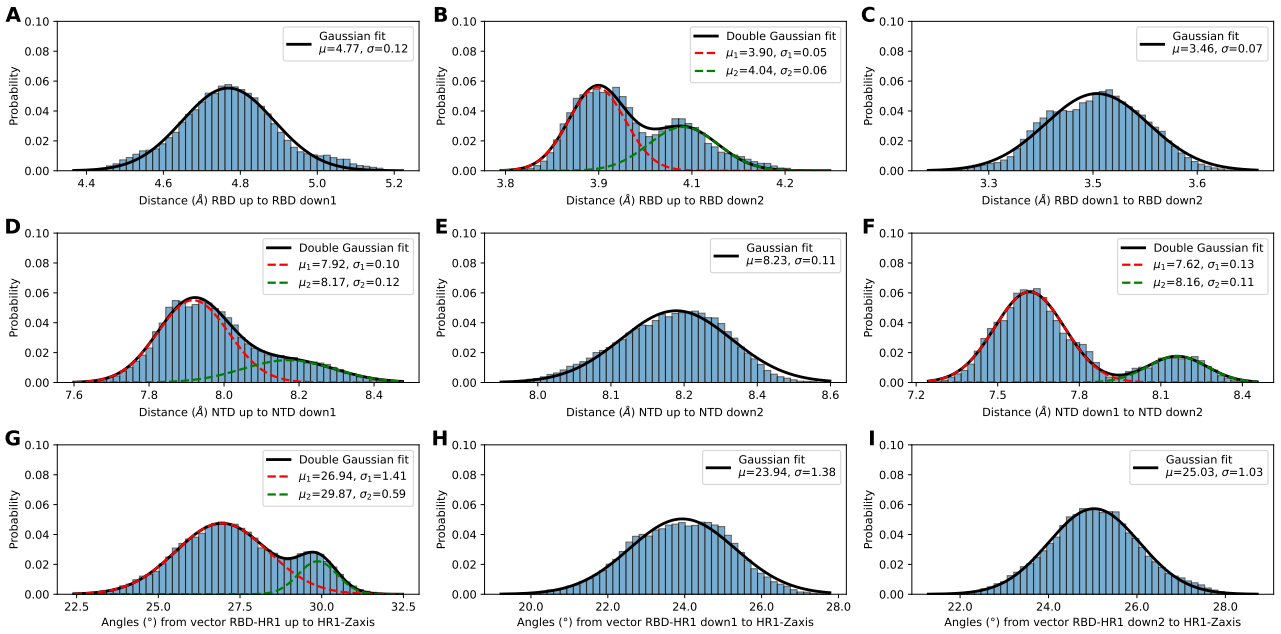


Figure S4: Gaussian distribution of the XBB.1.5 variant for all calculated CVs. The black line indicates a fitting for a single Gaussian distribution while the dotted lines (red or green) for a double distribution.

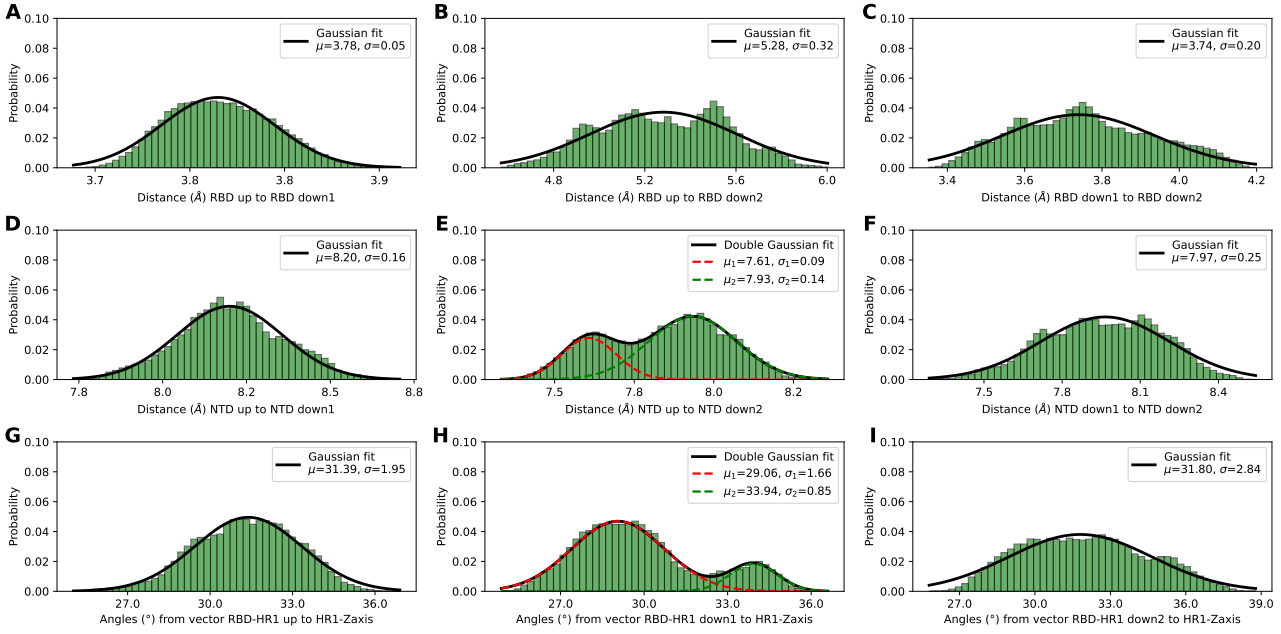


Figure S5: Gaussian distribution of the JN.1 variant for all calculated CVs. The black line indicates a fitting for a single Gaussian distribution while the dotted lines (red or green) for a double distribution.

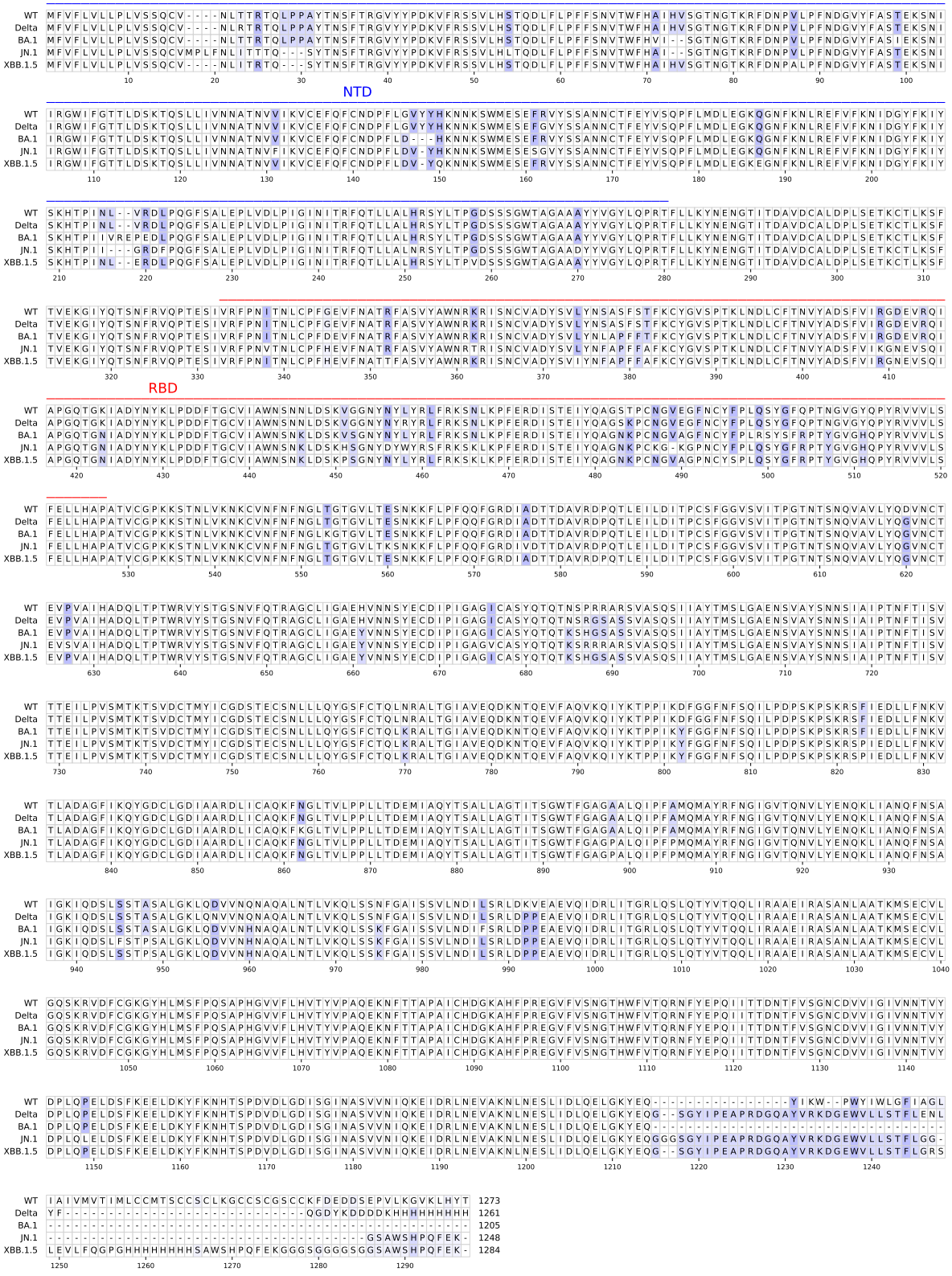


Figure S6: Alignment of the primary sequences of the WT S protein and its 4 variants. The WT sequence (PDB ID: 7CWL), as well as the sequences from Delta (PDB ID 7W92), BA.1 (PDB ID: 7XO5), JN.1 (PDB ID: 8Y5J), and XBB.1.5 (PDB ID: 8VKM) variants, were obtained in fasta format from the Protein Data Bank. Regions with at least one mutation are colored by identity (See pyMSAviz documentation). Note that insertions and deletions in the sequences of the variants modify the reading frame alignment.

Supplementary Tables

Table S1: Values of each CV (calculated between RBDs), σ, and χ^2 averages of the different states obtained by the probability distribution.

Collective Variables	Variants	States	μ	σ	χ^2
CV_1 (nm)	WT	1	4.24	0.11	0.04
		2	5.12	0.24	
	Delta	1	4.47	0.13	0.07
		2	5.44	0.32	
	BA.1	1	5.22	0.20	0.04
	XBB.1.5	1	4.77	0.12	0.09
	JN.1	1	3.78	0.05	0.03
CV_2 (nm)	WT	1	3.93	0.06	0.03
		2	4.36	0.12	
	Delta	1	4.19	0.14	0.03
		2	4.58	0.28	
	BA.1	1	4.10	0.09	0.23
	XBB.1.5	1	3.90	0.05	0.09
		2	4.04	0.06	
	JN.1	1	5.28	0.32	0.07

Table S2: Values of each CV (calculated between NTDs), σ, and χ^2 averages of the different states obtained by the probability distribution.

Collective Variables	Variants	States	μ	σ	χ^2
CV_3 (nm)	WT	1	3.42	0.10	0.14
		2	3.78	0.07	
	Delta	1	3.44	0.10	0.01
	BA.1	1	3.36	0.05	0.01
		2	3.56	0.12	
	XBB.1.5	1	3.46	0.07	0.02
	JN.1	1	3.74	0.20	0.05
CV_4 (nm)	WT	1	8.32	0.25	0.14
	Delta	1	8.57	0.27	0.04
	BA.1	1	7.79	0.18	0.09
	XBB.1.5	1	7.92	0.10	<0.01
		2	8.17	0.12	
	JN.1	1	8.20	0.16	0.02
CV_5 (nm)	WT	1	8.45	0.15	0.04
	Delta	1	8.73	0.24	0.04
	BA.1	1	7.58	0.12	0.01
		2	7.86	0.06	
		3	8.16	0.11	
	XBB.1.5	1	8.23	0.11	0.03
	JN.1	1	7.61	0.09	<0.01
		2	7.93	0.14	

Table S3: Values of each CV (calculated with angles of RBDs), σ, and χ^2 averages of the different states obtained by the probability distribution.

Collective Variables	Variants	States	μ	σ	χ^2
CV_7 (°)	WT	1	31.51	1.37	0.04
		2	37.00	2.00	
	Delta	1	31.01	4.44	0.01
		2	36.13	2.12	
	BA.1	1	32.28	1.37	0.02
		2	40.13	2.33	
	XBB.1.5	1	26.94	1.41	<0.01
		2	29.84	0.59	
	JN.1	1	31.39	1.95	0.02
CV_8 (°)	WT	1	26.32	2.61	0.05
	Delta	1	30.56	2.45	0.04
	BA.1	1	24.37	2.25	0.02
	XBB.1.5	1	23.94	1.38	0.17
	JN.1	1	29.06	1.66	0.01
		2	33.94	0.85	
CV_9 (°)	WT	1	27.17	1.77	0.25
		2	31.42	1.03	
	Delta	1	25.75	1.88	0.12
	BA.1	1	27.16	2.34	0.02
	XBB.1.5	1	25.03	1.03	<0.01
	JN.1	1	31.80	2.84	0.05

Table S4: Missing residues range remodeled into the chains of the S protein variants.

Delta			BA.1		
A	B	C	A	B	C
70-76	70-76	70-76	71-79	71-79	71-79
156-157	156-157	156-157	146-156	146-156	146-157
248-254	248-254	248-254	177-186	177-186	177-186
624-631	622-640	622-629	212-214	212-214	212-214
677-688	677-688	677-688	621-639	621-640	621-640
828-853	828-853	828-853	677-690	677-689	677-659
			829-853	829-854	829-584
XBB.1.5			JN.1		
A	B	C	A	B	C
72-77	72-77	72-77	70-80	70-80	66-68
145-152	145-152	145-152	140-143	139-143	70-80
179-186	179-186	179-186	145-152	145-153	140-143
250-255	250-255	250-255	173-187	173-187	145-152
621-640	621-640	621-640	244-163	244-263	173-187
676-690	676-690	676-690	677-688	517-529	244-263
828-847	828-847	828-847	828-654	677-688	677-688
				828-854	828-854

Table S5: High frequency NCs (freq > 60%) between the RBDs in the WT protein.

CV_1		CV_2		CV_3	
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=6)	State 2 (NCs=4)	State 1 (NCs=2)	State 2 (NCs=8)
—	—	Specific NCs F374–F486 F377–F486 Nonspecific NCs S375–F486 T376–F486 S383–F456 T385–K458	Nonspecific NCs S375–F486 F377–Y489 T385–K458 T385–F456	Nonspecific NCs T415–P384 K417–Y369	Specific NCs F456–A372 Nonspecific NCs K417–A372 F456–Y369 F456–N370 R457–Y369 K458–Y369 K458–T385 K458–N388

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S6: High frequency NCs (freq > 60%) between the RBDs in the Delta variant.

CV_1		CV_2		CV_3
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=9)	State 2 (NCs=10)	State 1 (NCs=5)
—	—	Specific NCs Y369–N487 N370–S477 A372–F486 F374–F486 F377–F486 P384–F456 P384–Y489 T385–Y473 Nonspecific NC S383–F456	Specific NCs Y369–N487 N370–S477 F374–F486 F377–F486 Nonspecific NCs Y369–G476 N370–K478* S371–K478* A372–K478* P384–Y489 T385–A475	Specific NCs T415–T385 Nonspecific NCs D405–S375 R408–T376 T415–P384 K417–N370

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S7: High frequency NCs (freq > 60%) between the RBDs in the BA.1 variant.

CV_1	CV_2	CV_3	
State 1 (NCs=0)	State 1 (NCs=7)	State 1 (NCs=3)	State 2 (NCs=1)
—	Specific NC F377–F486 D428–H505* Nonspecific NCs Y369–F486 S383–L455 S383–F456 P384–Y489 T385–F456	Specific NCs Q414–T385 T415–T385 Nonspecific NCs H505*–A372	Specific NC T415–T385

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S8: High frequency NCs (freq > 60%) between the RBDs in the XBB.1.5 variant.

CV_1	CV_2		CV_3
State 1 (NCs=0)	State 1 (NCs=24)	State 2 (NCs=14)	State 1 (NCs=4)
—	Specific NCs P373*–P486* Y380–Q493 G381–L455 V382–L455 P384–F456 T385–Y473 D427–H505* D428–H505* Nonspecific NCs Y369–A475 P373*–N487 F374–N487 F374–Y489 F377–Y489 C379–L455 C379–Q493 G381–N417* G381–Y453 S383–L455 P384–Y473 T385–F456 T385–R457 G413–R498* Q414–R498* D427–Y501*	Specific NCs F374–P486 F377–P486 P384–L455 T385–Y489 T385–Y473 Nonspecific NCs S383–L455 S383–F456 P384–Y489 T385–F456 G413–R498* D427–Y501* D427–G502	Specific NCs T415–T385 Nonspecific NCs N405*–F374 N405*–F375* H505*–F375*

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S9: High frequency NCs (freq > 60%) between the RBDs in the JN.1 variant.

CV_1	CV_2	CV_3
State 1 (NCs=0)	State 1 (NCs=24)	State 1 (NCs=0)
	Specific NCs	
	G416–P520	
	P485*–F374*	
	Y421–T522	
	T499–T415	
	S455*–S383	
	Y500*–Q414	
	F456–V382	
	Y500*–T415	
	Nonspecific NCs	
	N405*–D427	
	N405*–D428	
—	T415–A519	—
	N417*–G381	
	Y421–A521	
	Y421–P520	
	Y421–V382	
	S455*–V382	
	F456–S383	
	R457–V382	
	R457–T522	
	K458–T385	
	K458–T522	
	A475–T385	
	Y488–F377	
	H504*–G413	

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S10: High frequency NCs (freq > 60%) between an NTD and RBD in the WT protein.

CV_4	CV_5	CV_6
State 1 (NCs=4)	State 1 (NCs=6)	State 1 (NCs=1)
	Nonspecific NCs	
Nonspecific NCs	R357–F168	
F168–R357	R357–P230	Nonspecific NC
T167–R357	Y396–P230	P230–N394
Y200–P521	H519–V42	
P230–T523	A520–K41	
	P521–K41	

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S11: High frequency NCs (freq > 60%) between an NTD and RBD in the Delta variant.

CV_4	CV_5	CV_6
State 1 (NCs=0)	State 1 (NCs=3)	State 1 (NCs=3)
—	Nonspecific NCs R357–P230 A520–K41 P521–K41	Nonspecific NCs R357–P230 A520–K41 P521–K41

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S12: High frequency NCs (freq > 60%) between an NTD and RBD in the BA.1 variant.

CV_4	CV_5			CV_6	
State 1 (NCs=2)	State 1 (NCs=21)	State 2 (NCs=26)	State 3 (NCs=4)	State 1 (NCs=7)	State 2 (NCs=6)
Specific NC P521–P230 Nonspecific NC S359–E167	Specific NCs R357–E167 F464–G232 F464–I233 Nonspecific NCs R355–Q116 R355–T165 R357–F166 R357–Y168 N394–P230 Y396–P230 Y396–I231 Y396–G232 P463–N234 F464–N234 E465–N234 R466–T115 R466–Q116 R466–I233 R466–N234 D467–T115 I468–K114 H519–V45	Specific NCs R357–E167 F464–G197 F464–G232 F464–I233 Nonspecific NCs R355–Q116 R355–F166 R355–G232 K356–F166 R357–F166 R357–T165 R357–Y168 N394–D228 N394–P230 Y396–I231 Y396–G232 Y396–P230 P426–D196 F464–N234 E465–N234 R466–Q116 R466–I233 R466–T115 R466–N234 D467–T115 I468–K114 H519–V45	Nonspecific NCs K44–A520 K44–P521 V45–H519 P230–R357	Specific NCs G197–F464 G197–P463 Nonspecific NCs K44–A520 K44–P521 V45–H519 Q116–I468 P230–R357	Specific NC G197–P463 Nonspecific NCs K44–A520 K44–P521 V45–H519 Q116–I468 P230–R357

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S13: High frequency NCs (freq > 60%) between an NTD and RBD in the XBB.1.5 variant.

CV_4		CV_5	CV_6	
State 1 (NCs=10)	State 2 (NCs=12)	State 1 (NCs=9)	State 1 (NCs=8)	State 2 (NCs=13)
Specific NCs A520–G232 P521–G199 P521–G232 P521–P230 Nonspecific NCs R357–C166 N360–E169 N360–F168 P521–Y200 T523–P230 A522–Y200	Specific NCs N360–Y170 A520–G232 P521–G199 P521–G232 P521–P230 Nonspecific NCs R357–C166 N360–F168 N360–E169 P521–Y200 A522–Y200 T523–F168 T523–P230	Specific NCs N394–Y200 F464–G232 Nonspecific NCs R357–F168 R357–P230 Y396–P230 K462–N234 F464–N234 E465–N234 P521–K42	Specific NCs Y200–N394 Y200–Y396 Nonspecific NCs K42–P521 T167–R357 F168–R357 P230–R357 P230–Y396 N234–E465	Specific NCs G199–F464 Y200–N394 Y200–Y396 Nonspecific NCs K42–P521 F168–R357 D198–F464 G199–Y396 P230–R357 P230–Y396 G232–R355 G232–Y396 N234–E465 N234–F464

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S14: High frequency NCs (freq > 60%) between an NTD and RBD in the JN.1 variant.

CV_4	CV_5		CV_6
State 1 (NCs=0)	State 1 (NCs=14)	State 2 (NCs=13)	State 1 (NCs=10)
—	Specific NC G200–F464 Nonspecific NCs K45–P520 Q117–R466 F169–R357 D199–P463 D199–F464 P230–R357 P230–N394 P230–Y396 I231–R355 G232–R355 G232–R466 N234–D467 N234–E465	Specific NC G200–F464 Nonspecific NCs K45–P520 F169–R357 D199–P463 D199–F464 P230–R357 P230–Y396 I231–R355 G232–R355 G232–R466 I233–R466 N234–E465 N234–R466	Specific NC Y396–Y201 Nonspecific NCs R355–G232 R357–P230 V395–Y201 Y396–G200 Y396–P230 Y396–G232 E465–N234 I468–T116 H518–K45

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S15: High frequency NCs (freq > 60%) for the CVs calculated with angles in the WT protein.

NCs between two RBDs				
CV_7		CV_8	CV_9	
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=7)	State 1 (NCs=0)	State 2 (NCs=6)
—	—	Specific NCs F374–F486 F377–F486 Nonspecific NCs S375–F486 T376–F486 S383–F456 T385–F456 T385–K458	—	Nonspecific NCs T415–F377 T415–P384 K417–Y369 F456–Y369 F456–N370 K458–N388
NCs between an RBD and an NTD				
CV_7		CV_8	CV_9	
State 1 (NCs=11)	State 2 (NCs=7)	State 1 (NCs=6)	State 1 (NCs=3)	State 2 (NCs=5)
Nonspecific NCs T167–R355 T167–K356 T167–R357 F168–R357 F168–S359 D228–N360 L229–N360 P230–S359 P230–T523 I231–R357 G232–R357	Specific NCs P230–P521 P230–A520 G232–L518 Nonspecific NCs C166–R357 T167–R357 Y200–P521 P230–T523	Nonspecific NCs R357–F168 R357–P230 Y396–P230 H519–V42 A520–K41 P521–K41	Nonspecific NCs Y200–A520 D228–P521 P230–T523	Nonspecific NCs P230–N394 P230–Y396 G232–Y396 N234–E465 N234–R466

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S16: High frequency NCs (freq > 60%) for the CVs calculated with angles in the Delta variant.

NCs between two RBDs			
CV_7		CV_8	CV_9
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=6)	State 1 (NCs=4)
—	—	Specific NCs Y369–N487 N370–S477 F374–F486 F377–F486 P384–F456 Nonspecific NCs P384–Y489	Specific NCs T415–T385 Nonspecific NCs R408–T376 T415–P384 K417–N370
NCs between an RBD and an NTD			
CV_7		CV_8	CV_9
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=3)	State 1 (NCs=1)
—	—	Nonspecific NCs R357–P230 A520–K41 P521–K41	Nonspecific NCs P230–R357

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S17: High frequency NCs (freq > 60%) for the CVs calculated with angles in the BA.1 variant.

NCs between two RBDs			
CV_7		CV_8	CV_9
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=9)	State 1 (NCs=1)
—	—	Specific NCs F377–F486 P384–L455 T385–Y489 D428–H505* Nonspecific NCs Y369–F486 S383–L455 S383–F456 P384–Y489 T385–F456	Specific NC T415–T385
NCs between an RBD and an NTD			
CV_7		CV_8	CV_9
State 1 (NCs=21)	State 2 (NCs=1)	State 1 (NCs=21)	State 1 (NCs=4)
Specific NCs L335–V169 P337–V169 N354–N163 R357–E167 N360–Y168 R466–E133 P521–P230 Nonspecific NCs L335–S170 L335–Q171 P337–E167 R355–T165 K356–T165 R357–T165 R357–F166 S359–F166 S359–E167 N360–P230 R466–N162 R466–N163 I468–N163 T523–P230	Specific NC P521–P230	Specific NCs R357–E167 F464–G232 F464–I233 Nonspecific NCs R355–Q116 R355–T165 R357–F166 R357–Y168 N394–P230 Y396–P230 Y396–I231 Y396–G232 P426–D196 F464–N234 E465–N234 R466–T115 R466–Q116 R466–I233 R466–N234 D467–T115 I468–K114 H519–V45	Nonspecific NCs K44–A520 K44–P521 V45–H519 P230–R357

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S18: High frequency NCs (freq > 60%) for the CVs calculated with angles in the XBB.1.5 variant.

NCs between two RBDs			
CV_7		CV_8	CV_9
State 1 (NCs=0)	State 2 (NCs=0)	State 1 (NCs=17)	State 1 (NCs=3)
—	—	Specific NCs P373*–P486* F374–P486* P384–F456 P384–L455 T385–Y473 D427–H505* D428–H505* Nonspecific NCs F377–Y489 C379–L455 C379–Q493 G381–Y453 S383–L455 T385–F456 G413–R498* G413–Y501* Q414–R498* D427–Y501*	Specific NC T415–T385 Nonspecific NCs N405*–F374 N405*–F375*
NCs between an RBD and an NTD			
CV_7		CV_8	CV_9
State 1 (NCs=11)	State 2 (NCs=12)	State 1 (NCs=9)	State 1 (NCs=9)
Specific NCs N360–Y170 A520–G232 P521–G199 P521–P230 P521–G232 Nonspecific NCs R357–C166 N360–F168 N360–E169 P521–Y200 A522–Y200 T523–P230	Specific NCs S359–T167 A520–G232 P521–G199 P521–P230 P521–G232 Nonspecific NCs R357–C166 N360–F168 N360–E169 P521–Y200 A522–Y200 T523–P230 T523–F168	Specific NC N394–Y200 Nonspecific NCs R357–F168 R357–P230 Y396–P230 Y396–G232 K462–N234 F464–N234 E465–N234 P521–K42	Specific NC Y200–N394 Y200–Y396 Nonspecific NCs F168–R357 K42–P521 P230–R357 P230–Y396 G232–R355 G232–Y396 N234–E465

Note: – ionic interactions, – polar interactions, – nonpolar interactions, – nonspecific interactions. * Mutated amino acids.

Table S19: High frequency NCs (freq > 60%) for the CVs calculated with angles in the JN.1 variant.

NCs between two RBDs			
CV_7	CV_8		CV_9
State 1 (NCs=0)	State 1 (NCs=22)	State 2 (NCs=36)	State 1 (NCs=0)
—	Specific NCs G416—P520 Y421—T522 S455—S383 F456—V382 T499—T415 Y500*—Q414 Y500*—T415 H504*—D427 Nonspecific NCs N405*—D428 T415—A519 N417*—G381 Y421—V382 Y421—P520 Y421—A521 S455*—V382 F456—S383 R457—V382 R457—T522 K458—T522 Y488—F377 R497*—T415 H504*—G413	Specific NCs G416—P520 Y421—T522 S455—S383 F456—V382 F456—P384 Y473—T385 P485*—F374 N486—T385 Y488—T385 T499—T415 Y500*—Q414 Y500*—T415 Nonspecific NCs N405*—D427 N405*—D428 T415—A519 T415—P520 N417*—G381 Y421—V382 Y421—P520 S455*—V382 F456—S383 F456—T385 R457—V382 R457—T522 K458—T385 K458—T522 A475—T385 A475—K386 K483*—Y369 K483*—N370 G484—Y369 P485*—Y369 N486—K386 Y488—F377 H504*—G413 H504*—Q414	—
NCs between an RBD and an NTD			
CV_7	CV_8		CV_9
State 1 (NCs=0)	State 1 (NCs=15)	State 2 (NCs=2)	State 1 (NCs=10)
—	Specific NCs G200—P463 G200—F464 Nonspecific NCs K45—P520 F169—R357 D199—P463 D199—F464 P230—R357 P230—Y396 I231—R355 G232—R355 G232—R466 I233—R466 N234—E465 N234—R466 N234—D467	Nonspecific NCs K45—P520 P230—R357	Specific NCs Y396—Y201 Nonspecific NCs R355—G232 R357—P230 Y396—G200 Y396—P230 Y396—G232 E465—N234 I468—K115 I468—T116 H518—K45

Note: — ionic interactions, — polar interactions, — nonpolar interactions, — nonspecific interactions. * Mutated amino acids.