Supplementary Material

Table S1. Summary of MST3 crystals.

	Mst3_Naformat2	Mst3_Naformat3	Mst3_Li2SO4_PEG	Native	Native_ATP	Native_amppnp1	Native_amppnp2
Space group	C2 / Type I	C2 / Type I	C2 / Type I	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II
Unit cell (Å , °)	99, 59, 62; 94	99, 59, 62; 94	99, 59, 62; 94	56, 95, 61; 93	56, 95, 61; 93	56, 95, 61; 93	56, 95, 62; 93
Resolution (Å)	1.55	1.69	1.77	2.00	1.96	2.20	2.10
R / R_{free} (%)	19.5 / 21.9	34.3	23.3	19.9 / 24.7	20.4 / 25.2	28.7	25.0
Loops	missing 170-175	disordered	missing 170-175	continuous	continuous	disordered	disordered
Phosphorylation	Thr178	unclear	Thr178	Thr178	Thr178	Thr178	Thr178
Ligands	none	none	none	none	ADP	ADP (weak)	ADP

	Mst3_ATP_MgCl2	pMST303_mnadp1	pMST303_mnadp2	pMST303_mnatp1	pMST3_ADP_Mn	pMST3_ADP_Zn	pMST3_ATP_Mn
Space group	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II	$P2_1$ / Type II
Unit cell (Å , °)	55, 95, 62; 93	55, 95, 61; 94	55, 95, 62; 94	56, 95, 62; 93	55, 95, 61; 94	56, 95, 61; 93	55, 95, 61; 94
Resolution (Å)	2.10	1.95	2.05	1.95	1.85	1.95	2.00
R / R_{free} (%)	25.6	22.6	21.7	23.7	25.7	25.6	22.5
Loops	continuous	broken	continuous	continuous	broken	continuous	continuous
Phosphorylation	Thr178	Thr178	Thr178	Thr178	Thr178	Thr178	Thr178
Ligands	adenine (weak)	ADP	ADP / adenine	weak density	ADP / adenine	ADP	adenine

	pMST3_ATP_Zn	Mst3atp	Mst3atp2	Mst3atp3	nMst303_mnATP1	nMst303_mnADP2	MST3_T178E
Space group	$P2_1$ / Type II	$P2_1$ / Type III	$P2_1$ / Type III	$P2_1$ / Type III	$P2_1$ / Type III	$P2_1$ / Type III	$P2_1$ / Type III
Unit cell (Å , °)	55, 95, 62; 94	48, 55, 61; 111	48, 55, 60; 111	48, 56, 60; 111	48, 55, 61; 111	48, 54, 61; 110	47, 55, 61; 112
Resolution (Å)	2.15	1.85	1.83	1.45	1.86	1.50	1.89
R / R_{free} (%)	22.0	22.6	26.1	18.7 / 21.7	27.3	18.6 / 21.0	31.6
Loops	continuous	continuous	disordered G-loop	continuous	continuous	continuous	continuous
Phosphorylation	Thr178	Thr178	Thr178	Thr178	Thr178	Thr178	none
Ligands	ADP	adenine	adenine	adenine	Mn, ADP	Mn, ADP	none

The prefix of p to MST3 indicates that the protein was pretreated with Mg-ATP.

Table S2. Difference between the MST3(1-303) models and relative orientation of the two lobes. The models were superimposed using the LSQ procedures of the program *O* (Jones *et al.*, 1991) with a matching criterion of 3.8 Å between pairs of Cα. The upper-right triangular part contains the root-mean-square deviation (RMSD; Å) calculated for the number of matched Cα atom-pairs. The lower triangle contains rotation angles (°) required to superimpose the small N-terminal lobe after the larger C-terminal lobe is superimposed (see Table S3).

	I-Native	II-Nat-A	II-Nat-B	II-ADP-A	II-ADP-B	III-Adenine	III-MnADP
I-Native		1.17 / 278	1.23 / 280	1.28 / 278	1.30 / 280	0.98 / 270	1.01 / 268
II-Nat-A	9.5		0.85 / 288	0.27 / 290	0.82 / 290	1.06 / 280	0.87 / 279
II-Nat-B	11.1	7.3		0.92 / 290	0.28 / 291	0.88 / 280	0.97 / 279
II-ADP-A	10.7	1.1	7.8		0.84 / 290	1.11 / 280	0.93 / 281
II-ADP-B	11.8	7.1	0.9	7.8		0.93 / 279	0.98 / 279
III-Adenine	10.3	6.7	5.0	7.7	5.3		0.56 / 287
III-MnADP	10.1	3.4	6.9	4.3	7.1	4.3	

Table S3. Difference between the MST3(1-303) models as two individual lobes. The models were superimposed as in Table S2, but by comparing the two lobes (divided at residues 102-103) separately. The upper-right triangle contains the RMSD (Å) calculated for the number of matched Cα atom-pairs in the C-ternimal lobe. The lower triangle contains RMSD for the N-terminal lobe.

	I-Native	II-Nat-A	II-Nat-B	II-ADP-A	II-ADP-B	III-Adenine	III-MnADP
I-Native		0.401 /190	0.364 /190	0.402 /190	0.391 /190	0.338 /190	0.333 /190
II-Nat-A	0.902 / 91		0.287 /196	0.141 /196	0.273 /196	0.392 /196	0.433 /196
II-Nat-B	0.652 / 94	0.780 / 93		0.294 /196	0.161 /196	0.353 /196	0.379 /196
II-ADP-A	0.917 / 92	0.321 / 94	0.728 / 94		0.258 /196	0.398 /196	0.439 /196
II-ADP-B	0.861 / 94	0.686 / 94	0.372 / 95	0.544 / 94		0.369 /196	0.396 /196
III-Adenine	0.999 / 85	1.285 / 84	0.991 / 85	1.207 / 84	1.087 / 85		0.233 /197
III-MnADP	1.108 / 85	1.305 / 85	1.057 / 85	1.199 / 85	1.058 / 85	0.567 / 90	

Table S4. Difference between MST3(1-303) and some related models found in the PDB. The models were superimposed using the LSQ procedures of *O* as in Tables S2 and S3, with a matching criterion of 3.8 Å. The table contains the RMSD (Å) calculated for the number of matched Cα atom-pairs.

	I-Native	II-Nat-A	II-Nat-B	II-ADP-A	II-ADP-B	III-Adenine	III-MnADP
3ckw	0.97 / 253	0.74 / 252	0.56 / 254	0.82 / 252	0.65 / 254	0.64 / 254	0.68 / 253
3ckx	0.73 / 267	0.96 / 274	1.02 / 275	1.05 / 275	1.07 / 275	0.73 / 276	0.74 / 276
3com-A	1.44 / 251	1.03 / 261	1.04 / 261	1.01 / 261	1.02 / 261	1.31 / 260	1.19 / 262
3com-B	1.55 / 243	1.25 / 261	1.16 / 267	1.22 / 261	1.14 / 268	1.45 / 264	1.46 / 266
2j7t	1.49 / 234	1.73 / 230	1.65 / 240	1.77 / 227	1.68 / 238	1.45 / 229	1.56 / 229
2j51	1.31 / 238	1.64 / 240	1.71 / 235	1.71 / 237	1.76 / 237	1.51 / 236	1.56 / 238
2q0n	1.74 / 261	1.72 / 240	1.72 / 242	1.54 / 233	1.61 / 228	1.63 / 256	1.68 / 254

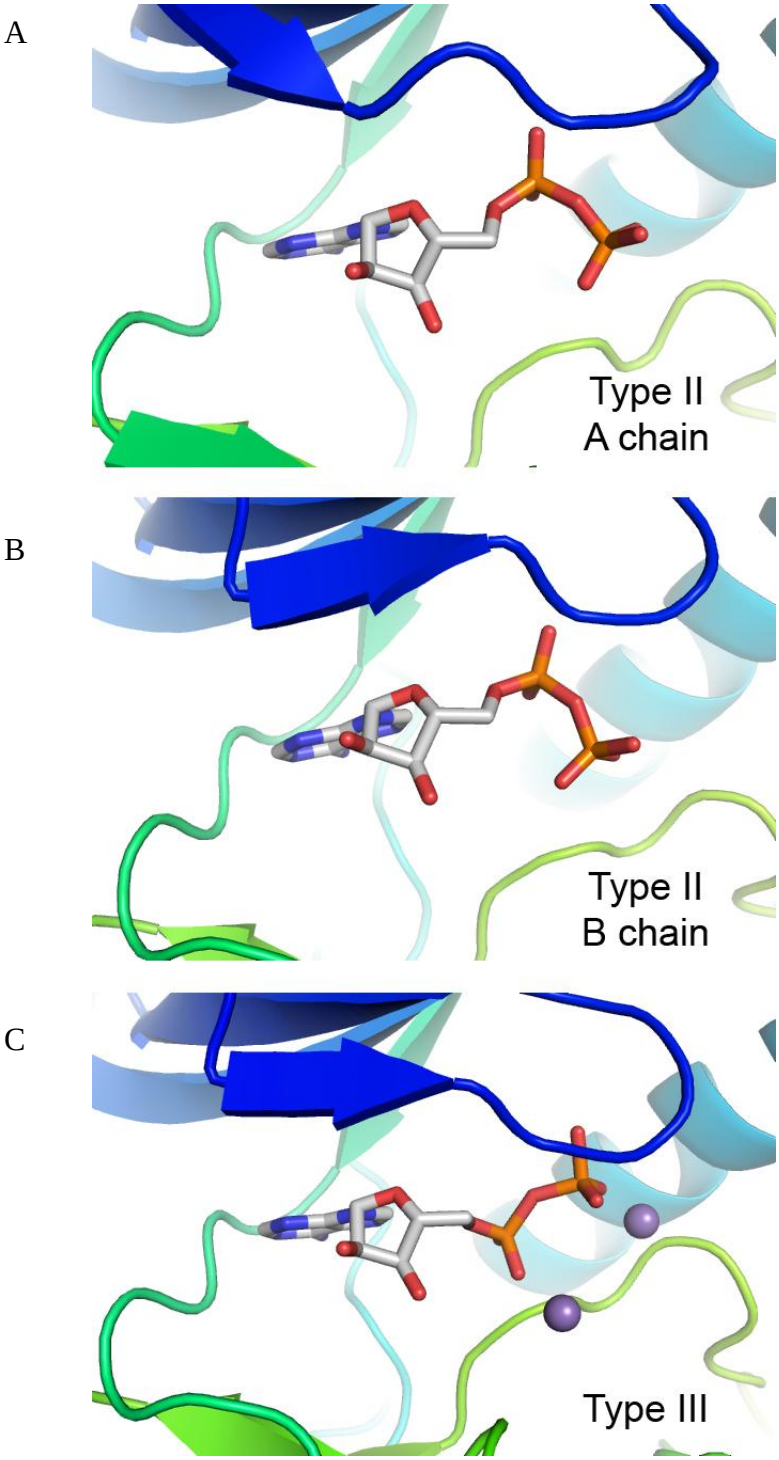

Figure S1. Comparison of the conformations of bound ADP in different crystals. (A) and (B) are for the two independent MST3(1-303) molecules in the type II crystal of ADP complex. (C) is for the type III Mn-ADP complex. The Mn ions are shown as purple spheres.

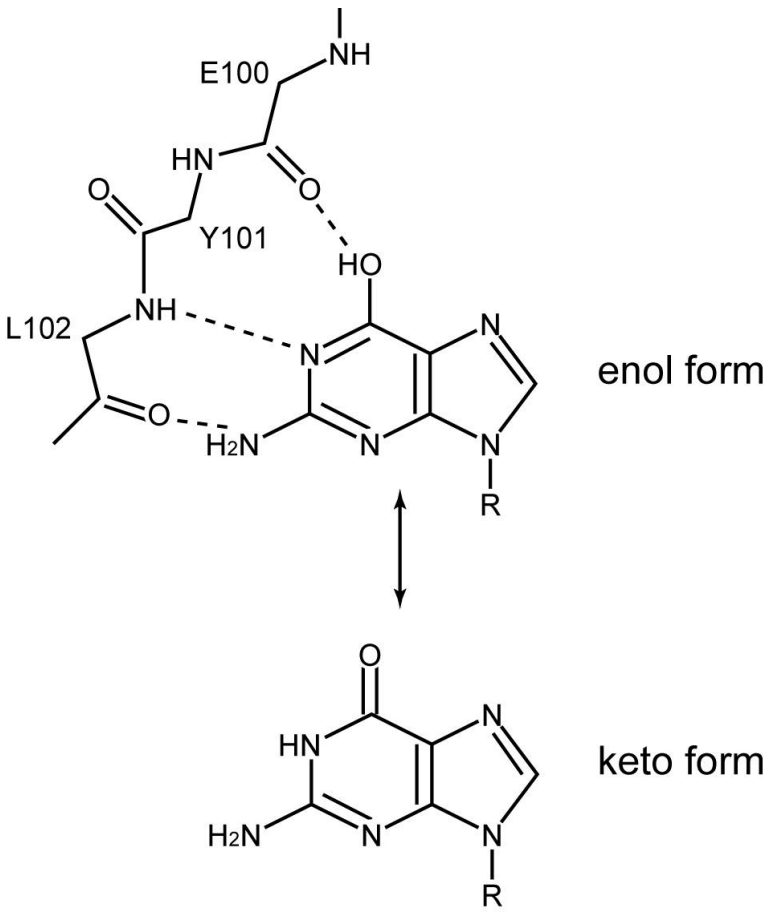

Figure S2. Tautomeric conversion of GTP. In order to form proper hydrogen bonds with the backbone atoms in the hinge region, the guanine base must undergo proton shift from N1 to O6, a less common formation than those observed in the conventional G-C base pairs of DNA.