Electronic Supplementary Material (ESI) for Physical Chemistry Chemical Physics.

Supplementary information

Table S1. Selected QM(B3LYP/6-31+G(d))/CHARMM bond distances (Å), bond angles (deg), and dihedrals (deg) of the calcium-bound initial model structures. Selected QM(B3LYP/def2-TZVP)/CHARMM bond distances (Å), bond angles (deg), and dihedrals (deg) of the strontium-bound initial model structures. Comparison with values from pseudo-Michaelis complex 4IAC X-ray crystallographic structure (used as model template) and PKAc-Mg_2ATP-SP20 initial model structure (QM(B3LYP/6-31+G(d))/CHARMM level). $Ser_{substrate}$ holds for Ser21 and Ser17 in SP20- and Kemptide-bound ternary complex models, respectively.

	4IAC	$i_{model}S_{Mg}$	$i_{model}S_{Ca}$	$i_{model}S_{Sr}$	$i_{model}K_{Ca}$	$i_{model}K_{Sr}$
O3βATP-PγATP	1.8*	1.70	1.72	1.70	1.74	1.70
PγATP-OγSer$_{substrate}$	3.5	3.76	3.59	3.70	3.68	3.59
OγSer$_{substrate}$-HγSer$_{substrate}$	---	0.98	0.99	0.99	1.00	0.99
Oδ2Asp166-HγSer$_{substrate}$	---	4.23	4.69	4.23	4.24	4.40
O2γATP-HγSer$_{substrate}$	---	3.52	3.47	3.23	3.31	3.39
OγSer$_{substrate}$-Oδ2Asp166	4.4	4.28	4.93	4.24	4.30	4.60
a.$_{O3βATP-PγATP-OγSerSubs.}$	148.2*	141.3	143.2	142.2	141.7	146.6
a.$_{OγSerSubs.-HγSerSubs.-Oδ2Asp166}$	---	86.4	98.0	83.8	86.6	95.3
a.$_{Oδ2Asp166-HγSerSubs.-O2γATP}$	---	60.2	57.1	67.0	64.8	63.9
d.$_{Ser21(N-Cα-Cβ-Oγ)}$	64.0	68.7	61.2	68.7	70.8	69.1
d.$_{Asp166(Cβ-Cγ-Oδ2)-O2γATP}$	160.0	167.2	-179.7	-177.8	178.2	-153.6
M1-O1βATP	2.2	2.08	2.38	2.54	2.38	2.53
M1-O3γATP	2.3	2.16	2.47	2.74	2.47	2.48

M1-Oδ1Asp184	2.4	2.21	2.47	2.63	2.49	2.56
M1-Oδ2Asp184	2.3	2.20	2.58	2.77	2.53	2.68
M1-OH2XWAT1003	2.2	2.07	2.44	2.64	2.43	2.60
M1-OH2XWAT1001	2.2	2.02	2.43	2.63	2.45	2.52
M1-OH2XWAT1029	4.0	3.94	2.51	2.66	2.42	2.52
M1-OH2XWAT1057	6.1	4.97	4.55	2.59	4.20	2.62
M2-O2αATP	2.2	1.99	2.32	2.41	2.32	2.40
M2-O3βATP	3.3*	2.96	2.65	2.89	2.55	2.91
M2-O2γATP	2.2	2.00	2.35	2.42	2.39	2.47
M2-Oδ2Asp184	2.2	2.10	2.37	2.47	2.38	2.46
M2-Oδ1Asn171	2.2	2.01	2.30	2.39	2.30	2.43
M2-OH2XWAT1002	2.3	2.12	2.41	2.53	2.40	2.53

For 4IAC * stands for O3βATP = CβAMPPCP.

Table S2. For the PKAc-Ca_2ATP-SP20 model, selected QM(B3LYP/6-31+G(d))/CHARMM bond distances (Å), bond angles (deg), and dihedrals (deg) in the optimized reactants ($Rd_{Ca/S}$), phosphoryl-transfer transition state ($TSd1P_{Ca/S}$), proton shift intermediate ($Id1_{Ca/S}$), proton shift transition state ($TSd1H_{Ca/S}$), phosphoryl-transfer products ($Pd1_{Ca/S}$), protonated Asp166 side chain-rotation transition state ($TSd2T_{Ca/S}$), and proton-transfer products ($Pd2_{Ca/S}$) for the dissociative mechanism with the O2γATP as the final proton acceptor (O2γATP-dissociative path). Comparison with values from X-ray crystallographic structures characterizing different stages of the phosphoryl-transfer mechanism catalyzed by the PKAc with SP20 or CP20 as substrates and Ca^{2+} ions.

Active core geometric features	4XW4	4XW5	$i_{model}S_{Ca}$	$Rd_{Ca/S}$	$TSd1P_{Ca/S}$	$Id1_{Ca/S}$	$TSd1H_{Ca/S}$	$Pd1_{Ca/S}$	$TSd2T_{Ca/S}$	$Pd2_{Ca/S}$	4IAI	4XW6
O3βATP-PγATP	1.74*	1.68	1.72	1.77	2.40	3.04	3.21	3.37	3.35	3.35	4.01	3.33
PγATP-OγSer21	4.70	5.55*	3.59	3.47	2.45	1.94	1.84	1.73	1.69	1.63	1.63	4.44/2.37*
OγSer21-HγSer21	---	---	0.99	0.98	0.99	1.04	1.20	1.56	2.01	2.62	---	---
Oδ2Asp166-HγSer21	---	---	4.69	1.87	1.78	1.57	1.25	1.03	1.00	1.53	---	---
O2γATP-HγSer21	---	---	3.47	3.26	2.86	2.74	2.78	2.72	2.06	1.03	---	---
OγSer21-Oδ2Asp166	4.55	3.36/3.17*	4.93	2.84	2.77	2.60	2.44	2.58	2.91	3.55	5.01	5.80/3.06*
$angle_{O3βATP-PγATP-OγSer21}$	136.8*	161.1	143.2	155.4	164.6	166.1	169.1	175.4	173.6	174.1	155.4	115.1/152.0*
$angle_{OγSer21-HγSer21-Oδ2Asp166}$	---	---	98.0	170.7	176.4	172.0	171.0	169.5	148.8	114.7	---	---
$angle_{Oδ2Asp166-HγSer21-O2γATP}$	---	---	57.1	103.0	107.1	111.5	114.5	112.1	129.7	172.6	---	---
$d._{Ser21(N-Cα-Cβ-Oγ)}$	64.3	-76.6	61.2	-54.9	-56.1	-46.1	-46.1	-52.9	-56.3	-55.7	63.9	56.9/-66.7*
$d._{Asp166(Cβ-Cγ-Oδ2)-O2γATP}$	135.9	159.1	-179.7	144.2	130.5	125.8	124.6	120.3	137.8	170.8	~130	129.5
$d._{phiAsp166}$	-147.2	-142.3	-92.6	-95.4	-90.7	-88.1	-87.2	-94.9	-96.1	-95.7	-150.9	-144.9
$d._{psiAsp166}$	40.5	34.4	88.0	85.6	82.5	80.4	79.2	77.3	78.8	83.9	43.0	36.7
$d._{OγSer21-PγATP-O2γATP-Oδ2Asp166}$	-36.6	-32.2	-37.4	-40.8	-39.2	-37.9	-37.5	-35.3	-39.3	-51.0	25.3*	-3.2/-29.2*
$Oδ1Asp24^{SP20}$-Cε1His87	6.00	5.04	5.7	5.4	5.4	5.4	5.4	5.4	5.4	5.4	5.56	12.33
$Oδ1Asp24^{SP20}$-Nε2Gly84	3.67	3.39	3.0	2.9	2.9	2.9	2.9	2.9	2.9	2.9	5.26	11.36
Oε1Gly84-Nδ1His87	2.74	3.20	2.7	2.70	2.70	2.70	2.70	2.70	2.70	2.70	2.72	2.86
O2pThr197-Nε2His87	2.67	2.59	2.7	2.65	2.65	2.65	2.65	2.65	2.65	2.65	2.72	2.63

OγSer21-Oγ1ATP	5.54*$^{i=2}$ 3.97*$^{i=3}$	5.62*$^{i=2}$ 4.80*$^{i=3}$	4.3$^{i=2}$ 2.6$^{i=3}$	3.20$^{i=2}$ 3.30$^{i=3}$	2.73$^{i=2}$ 2.83$^{i=3}$	2.57$^{=2}$ 2.64$^{i=3}$	2.53$^{=2}$ 2.60$^{i=3}$	2.54$^{i=2}$ 2.58$^{i=3}$	2.49$^{i=2}$ 2.58$^{i=3}$	2.50$^{i=2}$ 2.58$^{i=3}$	2.66 2.55 2.57	5.84$^{i=2}$/3.36$^{i=2}$ 4.22$^{i=3}$/3.01$^{i=3}$
Catalytic core interaction distances	4XW4	4XW5	$i_{model}S_{Ca}$	$Rd_{Ca/S}$	$TSd1P_{Ca/S}$	$Id1_{Ca/S}$	$TSd1H_{Ca/S}$	$Pd1_{Ca/S}$	$TSd2T_{Ca/S}$	$Pd2_{Ca/S}$	4IAI	4XW6
Oγ1Thr201-Oδ1Asp166	2.98	3.47	2.76	2.80	2.74	2.73	2.75	2.89	2.99	2.94	2.88	3.04
Oγ1Thr201-NζLys168	2.78	2.79	2.75	2.74	2.71	2.72	2.73	2.75	2.75	2.73	2.84	2.66
Oγ1Thr201-OγSer21	5.54	3.39	5.21	2.96	3.06	3.04	3.08	3.02	3.07	3.05	5.55	5.99/3.51*
Oδ2Asp166-OγSer21	4.55	3.36/3.17*	4.93	2.84	2.77	2.60	2.44	2.58	2.91	3.55	5.01	5.80/3.06*
Oδ2Asp166-PγATP	6.18	6.02	4.94	4.70	4.11	3.69	3.48	3.43	3.40	3.59	5.09	3.27
Oδ2Asp166-OH2XWAT1003	OH589 2.74	OH595 2.63 OH598 3.02	2.70	2.71	2.74	2.79	2.86	2.92	2.90	2.78	OH1001 2.65 OH1003 2.79	OH714 2.88
Oδ2Asp166-NζLys168	4.35	4.36	3.98	4.31	4.45	4.60	4.62	5.05	4.95	4.88	4.41	3.98
Oδ1Asp166-NζLys168	4.06	3.86	4.32	4.41	4.42	4.43	4.39	4.63	4.67	4.62	4.15	3.77
NζLys168-OγSer21	5.16	3.26	5.15	2.86	3.05	3.27	3.31	3.37	3.37	3.35	5.36	5.94/3.22*
NζLys168-PγATP	5.84	5.97	3.95	3.88	3.64	3.51	3.46	3.31	3.31	3.34	5.33	3.06
NζLys168-O2γATP	5.23	5.22	2.71	2.71	2.72	2.69	2.69	3.11	3.15	3.08	5.03	2.98
NζLys168-O1γATP	5.77	6.14	4.26	4.18	3.84	3.72	3.63	2.81	2.77	2.80	4.76	3.10
Nδ2Asn171-O2γATP	6.08	5.97	4.85	4.64	4.26	4.10	3.92	3.31	3.38	3.70	5.57	3.19
Nδ2Asn171-Oδ2Asp166	2.99	2.85	3.01	3.58	3.84	4.06	4.11	4.23	3.66	3.11	3.00	4.81
NζLys72-O1αATP	3.02	2.75	2.75	2.72	2.71	2.72	2.72	2.73	2.73	2.73	3.84	3.17
NζLys72-O1βATP	3.37	2.91	2.78	2.78	2.75	2.73	2.73	2.73	2.73	2.74	2.82	2.87
NζLys72-Oε2Glu91	2.86	2.67	2.72	2.72	2.75	2.75	2.75	2.75	2.75	2.74	2.71	2.73

OγSer53-O1γATP	6.01	2.57	5.63	5.64	5.63	5.67	5.78	6.55	6.62	6.72	2.39	2.97
NSer53-O2βATP	4.54	4.34	4.24	4.47	4.17	4.48	4.46	4.45	4.47	4.49	4.13	3.26
NPhe54-O2βATP	5.45	4.94	5.38	5.85	5.39	5.79	5.77	5.77	5.79	5.83	4.58	3.49
NGly55-O2βATP	5.59	5.09	5.57	5.83	5.96	5.68	5.66	5.68	5.71	5.77	3.53	5.11
OγSer53-OSer21	6.45	7.25	4.8	4.70	4.64	4.70	4.72	4.67	4.61	4.53	4.46	4.80
Ca^{2+} ions coordination sphere	4XW4	4XW5	$i_{model}S_{Ca}$	$Rd_{Ca/S}$	$TSd1P_{Ca/S}$	$Id1_{Ca/S}$	$TSd1H_{Ca/S}$	$Pd1_{Ca/S}$	$TSd2T_{Ca/S}$	$Pd2_{Ca/S}$	4IAI	4XW6
Ca1-O1βATP	2.42	2.26	2.38	2.41	2.38	2.36	2.35	2.36	2.36	2.36	2.26	2.37
Ca1-O3γATP	2.63	2.37	2.47	2.40	2.41	2.41	2.40	2.44	2.46	2.53	2.29*	2.13
Ca1-Oδ1Asp184	2.59	2.37	2.47	2.47	2.46	2.47	2.46	2.52	2.54	2.52	2.41	2.73
Ca1-Oδ2Asp184	2.54	2.42	2.58	2.60	2.61	2.59	2.59	2.53	2.53	2.55	2.45	2.25
Ca1-OH2XWAT1003	OH589 2.42	OH598 2.69	2.44	2.46	2.48	2.50	2.53	2.54	2.53	2.48	OH1003 2.41	OH714 2.19
Ca1-OH2XWAT1001	OH592 2.42	OH802 2.39	2.43	2.44	2.45	2.47	2.48	2.48	2.47	2.43	OH702 2.43	OH713 2.34
Ca1-OH2XWAT1029	OH711 2.61 OH813 2.69	OH594 2.64	2.51	2.53	2.50	2.50	2.50	2.51	2.52	2.51	OH1004 2.47	
Ca2-O2αATP	2.34	2.32	2.32	2.31	2.31	2.33	2.34	2.36	2.37	2.36	2.27	1.92
Ca2-O3βATP	4.21*	4.23	2.65	2.52	2.40	2.32	2.31	2.31	2.30	2.30	2.25	2.07
Ca2-O2γATP	2.68 O3γATP 2.78	2.42	2.35	2.33	2.34	2.38	2.39	2.45	2.49	2.63	OH701 2.57	2.47
Ca2-Oδ2Asp184	2.29	2.40	2.37	2.40	2.41	2.42	2.43	2.40	2.39	2.38	2.39	2.29
Ca2-Oδ1Asn171	2.40	2.38	2.30	2.29	2.32	2.35	2.36	2.34	2.35	2.35	2.43	2.20
Ca2-OH2XWAT1002	OH587 2.46	OH596 2.57	2.41	2.42	2.44	2.46	2.46	2.47	2.47	2.45	OH1002 2.42	2.35

		OH590	OH595					OH1001
		2.66	2.63					2.46

For 4IAI * means that is not possible to establish the chemical correspondence of the O*i*γATP atoms in relation to the 4IAC nomenclature

Table S3. For the PKAc-Sr_2ATP-SP20 model, selected QM(B3LYP/def2-TZVP)/CHARMM bond distances (Å), bond angles (deg), and dihedrals (deg) in the optimized reactants ($Rd_{Sr/S}$), phosphoryl-transfer transition state ($TSd1P_{Sr/S}$), phosphoryl-transfer products ($Pd1_{Sr/S}$), protonated Asp166 side chain-rotation transition state ($TSd2T_{Sr/S}$), and proton-transfer products ($Pd2_{Sr/S}$) for the dissociative mechanism with the O2γATP as the final proton acceptor (O2γATP-dissociative path). Comparison with values from X-ray crystallographic structures characterizing the product complex stage of the phosphoryl-transfer mechanism catalyzed by the PKAc with SP20 as substrate and with Sr^{2+} ions.

Active core geometric features	$i_{model}S_{Sr}$	$Rd_{Sr/S}$	$TSd1P_{Sr/S}$	$Pd1_{Sr/S}$	$TSd2T_{Sr/S}$	$Pd2_{Sr/S}$	4IAK	4IAY
O3βATP-PγATP	1.70	1.77	2.60	4.05	4.21	4.34	4.72	4.49
PγATP-OγSer21	3.70	3.59	2.73	1.72	1.67	1.61	1.62	1.63
OγSer21-HγSer21	0.99	0.98	0.99	1.67	2.03	2.64	---	---
Oδ2Asp166-HγSer21	4.23	1.77	1.72	1.01	0.99	1.41	---	---
O2γATP-HγSer21	3.23	3.27	2.98	2.89	2.11	1.05	---	---
OγSer21-Oδ2Asp166	4.24	2.74	2.71	2.66	2.93	3.47	5.21	5.09
$angle_{O3\beta ATP\text{-}P\gamma ATP\text{-}O\gamma Ser21}$	142.2	154.1	158.5	161.5	164.3	152.9	159.0	159.2
$angle_{O\gamma Ser21\text{-}H\gamma Ser21\text{-}O\delta 2Asp166}$	83.8	170.2	178.4	166.6	149.7	114.5	---	---
$angle_{O\delta 2Asp166\text{-}H\gamma Ser21\text{-}O2\gamma ATP}$	67.0	105.7	110.2	124.6	131.4	174.3	---	---
$d._{Ser21(N\text{-}C\alpha\text{-}C\beta\text{-}O\gamma)}$	68.7	-53.3	-42.2	-39.4	-49.9	-51.3	72.1	72.1
$d._{Asp166(C\beta\text{-}C\gamma\text{-}O\delta 2)\text{-}O2\gamma ATP}$	-177.8	160.5	140.0	130.8	133.9	155.9	-163.1	-132.5
$d._{phiAsp166}$	-121.6	-119.6	-119.7	-119.5	-125.8	-128.1	-151.4	-142.8
$d._{psiAsp166}$	75.6	80.5	76.6	70.4	65.4	66.1	45.2	41.2
$d._{O\gamma Ser21\text{-}P\gamma ATP\text{-}O2\gamma ATP\text{-}O\delta 2Asp166}$	-27.8	-39.3	-36.5	-42.6	-39.2	-56.3	-85.7	31.4
$O\delta 1Asp24^{SP20}$-Cε1His87	5.5	5.5	5.5	5.5	5.5	5.5	10.23	10.99

Oδ1Asp24^{SP20}-Nε2Gly84	3.0	2.9	2.9	3.0	2.9	2.9	7.14	9.32
Oε1Gly84-Nδ1His87	2.8	2.8	2.8	2.8	2.8	2.8	2.76	2.78
O2pThr197-Nε2His87	2.7	2.6	2.6	2.6	2.6	2.6	2.69	2.84
OγSer21-O/γATP	4.1$^{i=2}$ 2.7$^{i=3}$	3.15$^{i=2}$ 3.49$^{i=3}$	2.83$^{i=2}$ 3.01$^{i=3}$	2.46$^{i=2}$ 2.59$^{i=3}$	2.46$^{i=2}$ 2.54$^{i=3}$	2.47$^{i=2}$ 2.54$^{i=3}$	2.58*$^{i=1}$ 2.58*$^{i=2}$ 2.71*$^{i=3}$	2.57*$^{i=1}$ 2.66*$^{i=2}$ 2.66*$^{i=3}$
Catalytic core interaction distances	$i_{model}S_{Sr}$	$Rd_{Sr/S}$	$TSd1P_{Sr/S}$	$Pd1_{Sr/S}$	$TSd2T_{Sr/S}$	$Pd2_{Sr/S}$	4IAK	4IAY
Oγ1Thr201-Oδ1Asp166	2.68	2.79	2.73	2.77	2.93	2.93	2.93	2.88
Oγ1Thr201-NζLys168	2.74	2.80	2.74	2.79	2.79	2.77	2.74	2.84
Oγ1Thr201-OγSer21	4.86	2.96	3.18	3.31	3.28	3.28	5.41	5.55
Oδ2Asp166-OγSer21	4.24	2.74	2.71	2.66	2.93	3.47	5.09	5.01
Oδ2Asp166-PγATP	5.17	4.87	4.33	3.53	3.43	3.42	5.19	5.09
Oδ2Asp166-OH2XWAT1003	2.72	2.70	2.74	3.04	2.94	2.74	OH1002 2.80 OH1003 2.67	OH1001 2.65 OH1003 2.79
Oδ2Asp166-NζLys168	3.99	4.18	4.30	4.53	4.81	4.82	4.06	4.41
Oδ1Asp166-NζLys168	4.38	4.27	4.17	4.06	4.32	4.36	4.04	4.15
NζLys168-OγSer21	4.90	2.80	2.96	3.21	3.25	3.22	5.28	5.36
NζLys168-PγATP	3.99	3.99	3.76	3.51	3.35	3.40	5.21	5.33
NζLys168-O2γATP	2.72	2.79	2.81	2.70	3.17	3.15	4.89*	5.03*
NζLys168-O1γATP	4.28	4.22	4.01	3.86	2.92	2.98	4.56*	4.76*
Nδ2Asn171-O2γATP	5.08	4.88	4.54	4.20	3.51	3.52	5.79*	5.57*
Nδ2Asn171-Oδ2Asp166	2.95	3.26	3.66	4.11	3.83	3.39	3.13	3.00

NζLys72-O1αATP	2.73	2.73	2.72	2.71	2.70	2.70	2.77	3.84
NζLys72-O1βATP	2.83 XW1029 2.83	2.81 XW1029 2.83	2.72 XW1029 2.92	3.23 XW1029 2.81	3.45 XW1029 2.80	3.62 XW1029 2.79	2.85 XW1003 2.97	2.82
NζLys72-Oε2Glu91	2.71	2.73	2.75	2.72	2.70	2.69	2.94	2.71
OγSer53-O1γATP	5.44	5.39	5.21	5.18	6.05	6.20	2.34	2.39
NSer53-O2βATP	4.21	4.14	4.16	4.27	4.15	4.09	3.75	4.13
NPhe54-O2βATP	5.33	5.40	5.39	5.43	5.43	5.41	4.85	4.58
NGly55-O2βATP	5.74	5.84	5.82	5.82	5.88	5.89	5.67	3.53
OγSer53-OSer21	4.55	4.66	4.63	4.64	4.52	4.45	5.32	4.46
Sr^{2+} ions coordination sphere	$i_{model}S_{Sr}$	$Rd_{Sr/S}$	$TSd1P_{Sr/S}$	$Pd1_{Sr/S}$	$TSd2T_{Sr/S}$	$Pd2_{Sr/S}$	4IAK	4IAY
Sr1-O1βATP	2.54	2.53	2.50	2.54	2.50	2.52	2.33	2.27
Sr1-O3γATP	2.74	2.57	2.63	2.49	2.59	2.62	2.44*	2.44*
Sr1-Oδ1Asp184	2.63	2.64	2.59	2.60	2.66	2.61	2.64	2.61
Sr1-Oδ2Asp184	2.77	2.79	2.79	2.78	2.72	2.73	2.64	2.55
Sr1-OH2XWAT1003	2.64	2.68	2.70	2.88	2.93	2.84	OH1001 2.52	OH1002 2.88
Sr1-OH2XWAT1001	2.63	2.56	2.57	2.62	2.61	2.58	OH1003 2.49	OH1006 2.86
Sr1-OH2XWAT1029	2.66	2.68	2.62	2.64	2.68	2.67	OH701 2.49	OH701 2.79
Sr1-OH2XWAT1057	2.59	2.69	2.68	2.65	2.66	2.68	OH702 2.56	OH1001 2.87
Sr2-O2αATP	2.41	2.42	2.40	2.47	2.48	2.46	2.35	2.37
Sr2-O3βATP	2.89	2.83	2.57	2.56	2.60	2.61	2.59	2.42
Sr2-O2γATP	2.42	2.39	2.46	2.51	2.63	2.87	3.90*	3.82*

Sr2-Oδ2Asp184	2.47	2.49	2.52	2.63	2.61	2.59	2.46	2.50
Sr2-Oδ1Asn171	2.39	2.40	2.46	2.55	2.52	2.54	2.62	2.70
Sr2-OH2XWAT1002	2.53	2.52	2.53	2.56	2.59	2.56	2.61	OH703 2.88
Sr2-O1βATP	3.48	3.71	3.60	2.75	2.68	2.65	3.22	3.70
Sr2-	O1γATP 4.53not directed	O1γATP 4.48not directed	O1γATP 4.50not directed	O1γATP 4.37not directed	O1γATP 3.57not directed	O1γATP 3.13	OH703 2.66	OH702 3.06
Sr2-							OH1004 2.47	OH1003 2.77

For 4IAK and 4IAY * means that is not possible to establish the chemical correspondence of the O*i*γATP atoms in relation to the 4IAC nomenclature

Table S4. For the PKAc-Ca_2ATP-Kemptide model, selected QM(B3LYP/6-31+G(d))/CHARMM bond distances (Å), bond angles (deg), and dihedrals (deg) in the optimized reactants ($Rd_{Ca/K}$), phosphoryl-transfer transition state ($TSd1P_{Ca/K}$), proton shift intermediate ($Id1_{Ca/K}$), proton shift transition state ($TSd1H_{Ca/K}$), phosphoryl-transfer products ($Pd1_{Ca/K}$), protonated Asp166 side chain-rotation transition state ($TSd2T_{Ca/K}$), and proton-transfer products ($Pd2_{Ca/K}$) for the dissociative mechanism with the O2γATP as the final proton acceptor (O2γATP-dissociative path).

Active core geometric features	$i_{model}K_{Ca}$	$Rd_{Ca/K}$	$TSd1P_{Ca/K}$	$Id1_{Ca/K}$	$TSd1H_{Ca/K}$	$Pd1_{Ca/K}$	$TSd2T_{Ca/K}$	$Pd2_{Ca/K}$
O3βATP-PγATP	1.74	1.79	2.62	3.26	3.39	3.56	3.65	3.66
PγATP-OγSer21	3.68	3.69	2.53	1.97	1.89	1.76	1.71	1.65
OγSer21-HγSer21	1.00	0.99	1.00	1.05	1.18	1.70	2.05	2.50
Oδ2Asp166-HγSer21	4.24	1.80	1.74	1.52	1.28	1.02	1.00	1.57
O2γATP- HγSer21	3.31	3.27	2.83	2.68	2.75	2.92	2.00	1.03
OγSer21-Oδ2Asp166	4.30	2.79	2.74	2.56	2.46	2.71	2.91	3.28
$angle_{O3βATP\text{-}PγATP\text{-}OγSer21}$	141.7	148.7	157.4	161.2	161.7	161.2	171.6	170.2
$angle_{OγSer21\text{-}HγSer21\text{-}Oδ2Asp166}$	86.6	174.3	173.5	170.3	171.1	167.0	142.1	105.0

$angle_{Oδ2Asp166\text{-}HγSer21\text{-}O2γATP}$	64.8	108.7	112.8	115.4	117.6	126.4	139.1	175.4
$d._{Ser21(N\text{-}Cα\text{-}Cβ\text{-}Oγ)}$	70.8	-48.1	-33.1	-33.8	-34.7	-33.7	-36.2	-36.5
$d._{Asp166(Cβ\text{-}Cγ\text{-}Oδ2)\text{-}O2γATP}$	178.2	152.9	141.1	132.6	131.1	136.9	154.6	171.8
$d._{phiAsp166}$	-128.1	-124.7	-122.9	-120.7	-119.4	-120.1	-123.4	-121.8
$d._{psiAsp166}$	80.8	76.0	73.2	71.7	71.4	71.1	73.0	76.0
$d._{OγSer21\text{-}PγATP\text{-}O2γATP\text{-}Oδ2Asp166}$	-31.1	-41.7	-40.2	-38.7	-38.1	-43.2	-43.0	-50.3
OγSer21-O/γATP	$4.1^{i=2}$	$3.18^{i=2}$	$2.71^{i=2}$	$2.52^{i=2}$	$2.50^{i=2}$	$2.45^{i=2}$	$2.44^{i=2}$	$2.46^{i=2}$
	$2.7^{i=3}$	$3.65^{i=3}$	$2.96^{i=3}$	$2.70^{i=3}$	$2.65^{i=3}$	$2.60^{i=3}$	$2.62^{i=3}$	$2.63^{i=3}$
Catalytic core interaction distances	$i_{model}K_{Ca}$	$Rd_{Ca/K}$	$TSd1P_{Ca/K}$	$Id1_{Ca/K}$	$TSd1H_{Ca/K}$	$Pd1_{Ca/K}$	$TSd2T_{Ca/K}$	$Pd2_{Ca/K}$
Oγ1Thr201-Oδ1Asp166	2.74	3.00	2.84	2.81	2.81	2.91	3.17	3.23
Oγ1Thr201-NζLys168	2.82	2.88	2.78	2.81	2.82	2.84	2.82	2.79
Oγ1Thr201-OγSer21	5.21	2.93	3.24	3.33	3.35	3.39	3.27	3.24
Oδ2Asp166-OγSer21	4.30	2.79	2.74	2.56	2.46	2.71	2.91	3.28
Oδ2Asp166-PγATP	5.05	4.80	4.14	3.68	3.54	3.60	3.41	3.48
Oδ2Asp166-OH2XWAT1003	2.69	2.74	2.78	2.84	2.88	2.97	2.95	2.77
Oδ2Asp166-NζLys168	3.96	4.37	4.38	4.44	4.42	4.56	4.82	4.87
Oδ1Asp166-NζLys168	4.45	4.46	4.36	4.34	4.30	4.32	4.53	4.43
NζLys168-OγSer21	5.00	2.74	2.94	3.13	3.15	3.11	3.15	3.17
NζLys168-PγATP	4.11	4.16	3.81	3.61	3.56	3.52	3.36	3.41
NζLys168-O2γATP	2.82	2.87	2.83	2.79	2.77	2.74	3.16	3.21
NζLys168-O1γATP	4.52	4.62	4.15	2.87	3.82	3.87	2.94	2.93
Nδ2Asn171-O2γATP	4.79	4.77	4.34	4.09	4.03	4.08	3.47	3.66
Nδ2Asn171-Oδ2Asp166	3.01	3.26	3.42	3.62	3.67	3.64	3.19	2.98

NζLys72-O1αATP	2.74	2.72	2.71	2.72	2.72	2.72	2.72	2.72
NζLys72-O1βATP	2.76	2.76	2.73	2.74	2.74	2.75	2.76	2.76
NζLys72-Oε2Glu91	2.70	2.69	2.71	2.72	2.72	2.73	2.73	2.72
OγSer53-O1γATP	5.49	5.17	5.35	5.47	5.49	5.44	6.25	6.39
NSer53-O2βATP	4.21	4.29	4.28	4.28	4.27	4.28	4.24	4.23
NPhe54-O2βATP	5.46	5.46	5.47	5.45	5.44	5.44	5.44	5.47
NGly55-O2βATP	5.77	5.70	5.70	5.65	5.64	5.62	5.64	5.70
OγSer53-OSer21	4.75	4.96	4.94	4.97	4.98	4.95	5.07	5.05
Ca^{2+} ions coordination sphere	$i_{model}K_{Ca}$	$Rd_{Ca/K}$	$TSd1P_{Ca/K}$	$Id1_{Ca/K}$	$TSd1H_{Ca/K}$	$Pd1_{Ca/K}$	$TSd2T_{Ca/K}$	$Pd2_{Ca/K}$
Ca1-O1βATP	2.38	2.37	2.33	2.32	2.32	2.32	2.32	2.32
Ca1-O3γATP	2.47	2.41	2.47	2.47	2.45	2.43	2.56	2.66
Ca1-Oδ1Asp184	2.49	2.45	2.44	2.45	2.46	2.47	2.55	2.52
Ca1-Oδ2Asp184	2.53	2.60	2.58	2.58	2.58	2.61	2.53	2.54
Ca1-OH2XWAT1003	2.43	2.45	2.47	2.48	2.50	2.51	2.49	2.46
Ca1-OH2XWAT1001	2.45	2.40	2.41	2.42	2.42	2.41	2.41	2.40
Ca1-OH2XWAT1029	2.42	2.63	2.57	2.59	2.59	2.60	2.61	2.59
Ca2-O2αATP	2.32	2.31	2.30	2.31	2.32	2.34	2.36	2.35
Ca2-O3βATP	2.55	2.57	2.39	2.35	2.34	2.34	2.35	2.35
Ca2-O2γATP	2.39	2.36	2.38	2.39	2.38	2.37	2.41	2.55
Ca2-Oδ2Asp184	2.38	2.42	2.44	2.46	2.46	2.48	2.43	2.40
Ca2-Oδ1Asn171	2.30	2.31	2.34	2.36	2.37	2.39	2.36	2.36
Ca2-OH2XWAT1002	2.40	2.40	2.41	2.42	2.43	2.43	2.42	2.40

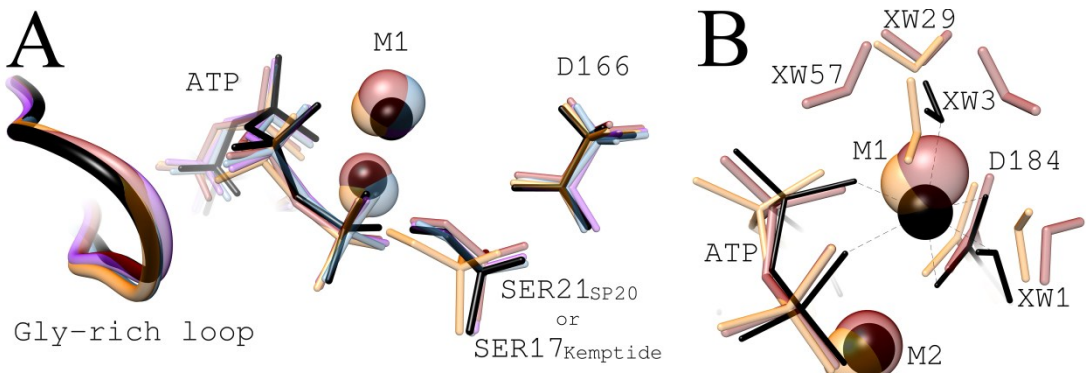

Figure S1. (A) Superposition of the active sites in the initial model structures of PKAc-Ca_2ATP-SP20 ($i_{model}S_{Ca}$, colored in orange), PKAc-Sr_2ATP-SP20 ($i_{model}S_{Sr}$, colored in dark red), PKAc-Ca_2ATP-Kemptide ($i_{model}K_{Ca}$, colored in purple), PKAc-Sr_2ATP-Kemptide ($i_{model}K_{Sr}$, colored in blue), and PKAc-Mg_2ATP-SP20 ($i_{model}S_{Mg}$ shown for comparison, colored in black). **(B)** Superposition of the coordination sphere for the M1 site in the initial model structures of PKAc-Ca_2ATP-SP20 (colored in orange), PKAc-Sr_2ATP-SP20 (colored in dark red), and PKAc-Mg_2ATP-SP20 (colored in black).

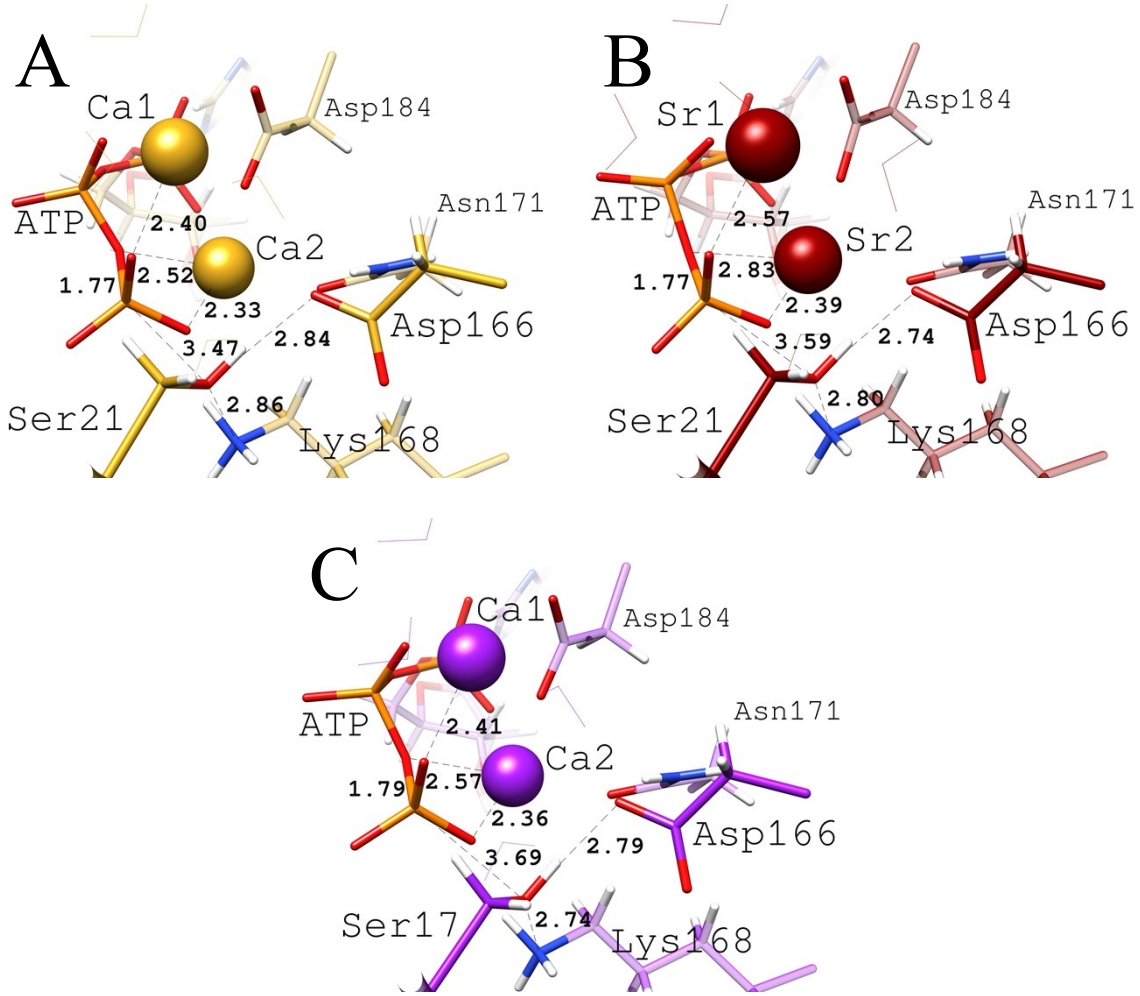

Figure S2. Active site structures at: **(A)** QM(B3LYP/6-31+G(d))/CHARMM optimized $Rd_{Ca/S}$; **(B)** QM(B3LYP/def2-TZVP)/CHARMM optimized $Rd_{Sr/S}$; and **(C)** QM(B3LYP/6-31+G(d))/CHARMM optimized $Rd_{Ca/K}$. Selected distances are given in Å.

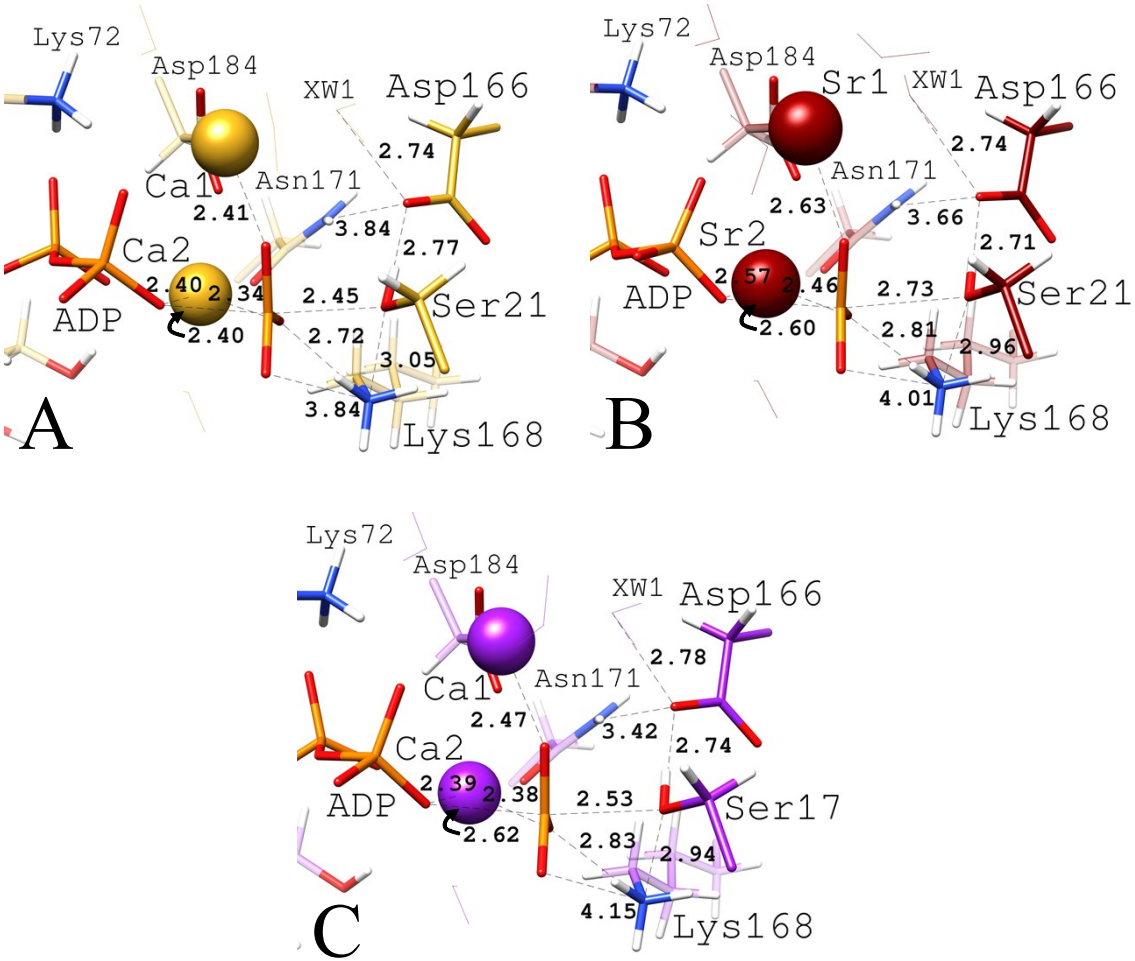

Figure S3. Active site structure at: **(A)** QM(B3LYP/6-31+G(d))/CHARMM optimized TSd1P$_{Ca/S}$; **(B)** QM(B3LYP/def2-TZVP)/CHARMM optimized TSd1P$_{Sr/S}$; and **(C)** QM(B3LYP/6-31+G(d))/CHARMM optimized TSd1P$_{Ca/K}$. Selected distances are given in Å.