Supplementary Table 1. Cryo-EM data collection, processing, structural refinement, and validation statistics of wild-type RNase P in the catalytic cycle.

	holoE (consensus) 5 mM Ca^{2+}	holoE (consensus) 5 mM Mg^{2+}	holoES pre-tRNA 5 mM Ca^{2+}	holoES pre-tRNA 1 mM Ca^{2+}	holoES (nc) pre-tRNA	holoES (LB) tilt1 pre-tRNA	holoES (LB) tilt2 pre-tRNA	holoEP mat-tRNA
Magnification	150,000	150,000	150,000	150,000	150,000	150,000	150,000	150,000
Voltage (keV)	200	200	200	200	200	200	200	200
Electron exposure (e^{-}/Å^2)	57	57	57	57	57	57	57	57
Defocus range (μm)	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5
Pixel size (Å)	1.038	1.038	1.038	1.038	1.038	1.038	1.038	1.038
Particle numbers	615,963	998,359	176,972	229,735	113,818	508,154	483,540	50,839
Map resolution (Å)	2.96	2.87	2.82	2.93	2.83	2.70	2.69	3.02
FSC threshold	0.143	0.143	0.143	0.143	0.143	0.143	0.143	0.143
EMDB	EMD-70941	EMD-70942	EMD-70943	EMD-70944	EMD-70946	EMD-70947	EMD-70948	EMD-70945
Model composition								
Non-hydrogen atoms	9,928	9,931	12,017	12,012	11,704	12,230	12,233	11,538
Protein residue	116	116	116	116	116	116	116	116
RNA residue	417	417	515	515	500	525	525	492
Mg^{2+} or Ca^{2+}	19	22	31	26	30	28	31	28
***B* factors (Å^2) mean**								
Protein	99.27	96.53	84.09	103.05	85.92	48.84	68.58	166.39
RNA	227.70	210.10	172.66	217.28	178.64	131.80	137.27	174.08
Mg^{2+} or Ca^{2+}	71.42	74.55	74.41	110.77	97.35	64.02	81.03	91.75
R.m.s. deviations								
Bond lengths (Å)	0.003	0.003	0.003	0.003	0.003	0.003	0.004	0.003
Bond angles (°)	0.672	0.703	0.634	0.661	0.629	0.642	0.645	0.672
Validation								
MolProbity score	2.40	2.13	2.06	2.10	1.78	1.77	1.39	1.64
Clashscore	8.35	8.35	7.53	7.86	7.45	7.51	6.93	7.44
CC_{Mask}	0.84	0.88	0.89	0.84	0.88	0.89	0.88	0.86
CC_{Box}	0.90	0.91	0.91	0.89	0.90	0.91	0.91	0.90
Rotamer outliers (%)	4.04	2.02	7.07	3.03	3.03	2.02	1.01	2.02
Ramachandran outliers (%)	0	0	0	0	0	0	0	0
PDB	9OWR	9OWS	9OWT	9OWU	9OWW	9OWX	9OWY	9OWV

Footnote: Holoenzyme (holoE), holoenzyme-substrate complex (holoES), holoenzyme-product complex (holoEP), correlation coefficient (CC), Fourier shell correlation (FSC), loop-back (LB), non-complementary (nc).

Supplementary Table 2. Cryo-EM data collection, processing, structural refinement, and validation statistics of RNase P tetraloop mutant (TLm) in the catalytic cycle.

	TLm holoE (subclass 1)	TLm holoE (subclass 2)	TLm holoES (consensus)
Magnification	150,000	150,000	150,000
Voltage (kV)	200	200	200
Electron exposure (e^{-}/Å^2)	57	57	57
Defocus range (μm)	-0.5 to -1.5	-0.5 to -1.5	-0.5 to -1.5
Pixel size (Å)	1.038	1.038	1.038
Particle numbers	133,224	157,466	309,996
Map resolution (Å)	3.02	2.96	2.94
FSC threshold	0.143	0.143	0.143
EMDB	EMD-70997	EMD-70998	EMD-71000
Model composition			
Non-hydrogen atoms	9,924	9,925	12,002
Protein residue	116	116	116
RNA residue	417	417	515
Mg^{2+} or Ca^{2+}	24	25	25
***B* factors (Å^2) mean**			
Protein	147.72	106.01	80.63
RNA	213.96	197.81	189.46
Mg^{2+} or Ca^{2+}	75.51	74.12	89.42
R.m.s. deviations			
Bond lengths (Å)	0.003	0.004	0.003
Bond angles (°)	0.673	0.703	0.663
Validation			
MolProbity score	1.84	2.03	1.51
Clashscore	6.80	7.97	6.86
CC_{Mask}	0.82	0.85	0.86
CC_{Box}	0.88	0.88	0.89
Rotamer outliers (%)	4.04	3.03	1.01
Ramachandran outliers (%)	0	0	0
PDB	9OY5	9OY6	9OY7

Footnote: Holoenzyme (holoE), holoenzyme-substrate complex (holoES), correlation coefficient (CC), Fourier shell correlation (FSC), Tetraloop mutant (TLm).

Supplementary Table 3. Metal ion binding site validation.

holoE (consensus) in 5 mM Ca^{2+}; PBD ID: 9OWR; EMDB: EMD-70941									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (points)**
Ca1	1.0	**80.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	61.7 (40.4)	O_2	**0.3**	**0.87**	Octahedral	11.0°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca4	1.0	72.9 (43.7)	**O_1N_1**	**0.15**	**0.76**	Octahedral	8.6°	**66%**	Na(2), Mg(2), **Ca(2)**,
Ca5	1.0	40.6 (41.5)	O_3	**0.18**	**0.59**	Octahedral	14.0°	**50%**	**Ca(4)**, Na(4), Mg(4)
Ca6	1.0	79.5 (103.2)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7	1.0	51.5 (82.1)	O_2	**0.4**	**0.94**	Octahedral	**27.4°**	**66%**	Na(3), Mg(3), **Ca(3)**
Ca8	1.0	102.3 (74.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca9 (MeA)	1.0	47.4 (43.4)	O_3	**0.8**	**0.72**	Octahedral	11.0°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca10	1.0	36.4 (28.9)	O_2	**0.2**	**0.83**	Octahedral	20.3°	**66%**	**Ca(3)**, Na(3), Mg(3)
Ca11	1.0	101.1 (76.9)	O_2	**0.5**	**0.86**	Octahedral	**22.0°**	**66%**	Na(3), Mg(3), **Ca(3)**
Ca12	1.0	60.1 (53.7)	**O_2N_2**	**0.4**	**0.41**	**Tetrahedral**	**24.4°**	0	Na(1), Mg(0), **Ca(0)**
Ca13	1.0	**78.7 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca14	1.0	42.9 (39.6)	O_4	**0.7**	**0.6**	Octahedral	8.8°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca15	1.0	98.3 (53.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca16	1.0	**81.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca17	1.0	47.4 (40.5)	O_4	**1.2**	**0.31**	**Trigonal Bipyramidal**	11.3°	20%	Na(5), Mg(2), **Ca(2)**
Ca18	1.0	67.9 (90.0)	**O_3N_1**	**0.7**	**0.57**	Octahedral	18.4°	**33%**	Na(5), Mg(4), **Ca(2)**
Ca19	1.0	89.9 (100.4)	**O_1**	**0.05**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
holoE (consensus) in 5 mM Mg^{2+}; PBD ID: 9OWS; EMDB: EMD-70942									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Mg1	1.0	75.1 (75.6)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg2	1.0	70.4 (83.3)	**N_1**	**0.02**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Mg(0)**, Na(0)
Mg3	1.0	74.5 (64.2)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg4	1.0	51.1 (59.5)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)

Mg5	1.0	83.0 (77.4)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg6	1.0	83.9 (79.1)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg7	1.0	66.8 (64.6)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Mg(0)**, Na(0)
Mg8	1.0	50.6 (59.6)	O_2	**0.7**	**0.71**	Octahedral	0.9°	**66%**	Na(5), **Mg(3)**
Mg9	1.0	**78.1 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg10	1.0	53.7 (55.4)	O_3	**0.9**	**0.31**	Octahedral	11.9°	**50%**	Na(6), **Mg(4)**
Mg11	1.0	72.1 (92.3)	O_1N_1	**0.5**	**0.72**	Octahedral	0.5°	**66%**	Na(4), **Mg(3)**
Mg12	1.0	64.5 (62.4)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg13	1.0	**69.9 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg14	1.0	**84.1 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg15 (MeA)	1.0	58.0 (61.3)	O_3	**0.5**	**0.56**	**Tetrahedral**	9.7°	25%	Na(5), **Mg(2)**
Mg17	1.0	87.4 (84.9)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg18	1.0	86.2 (97.6)		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg19	1.0	**71.4 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Mg(0)**, Na(0)
Mg20	1.0	75.8 (76.9)	O_2	**0.8**	**0.7**	Octahedral	0.8°	**66%**	Na(5), **Mg(3)**
Mg21	1.0	98.2 (84.3)	**N_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Mg(0)**, Na(0
Mg22	1.0	108.7 (117.0)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(1), **Mg(0)**
holoES (pre-tRNA) in 5 mM Ca^{2+}; PBD ID: 9OWT; EMDB: EMD-70943									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**70.8 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**96.1 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**72.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	70.9 (52.6)	**O_1**	**0.09**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5 (MeB)	1.0	48.6 (45.5)	O_3	**1.0**	**0.66**	Octahedral	10.7°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca6	1.0	69.4 (38.1)	**O_1**	**0.08**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7 (MeC)	1.0	76.2 (57.7)	O_2	**0.4**	**0.81**	Octahedral	1.6°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca8	1.0	70.1 (46.9)	O_2	**0.3**	**0.78**	Octahedral	13.1°	**66%**	Na(3), Mg(3), **Ca(3)**

Ca9	1.0	73.6 (43.6)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	72.9 (49.0)	O_2	**0.11**	**0.79**	Octahedral	9.7°	**66%**	**Ca(3)**, Na(3), Mg(3)
Ca11	1.0	83.1 (54.6)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	91.0 (62.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca13	1.0	54.7 (51.4)	O_3	**1.2**	**0.43**	Octahedral	19.1°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca14	1.0	86.8 (72.0)	O_2	**0.6**	**0.76**	Octahedral	9.2°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca15	1.0	57.2 (39.7)	O_2	**0.3**	**0.81**	Octahedral	16.3°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca16	1.0	78.7 (74.2)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca17	1.0	36.4 (36.0)	O_4	**1.1**	**0.46**	**Tetrahedral**	**34.6°**	0	Na(5), Mg(2), **Ca(2)**
Ca18	1.0	83.5 (67.0)	O_2	**0.6**	**0.75**	Octahedral	7.9°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca19	1.0	57.0 (50.6)	O_3	**0.7**	**0.69**	Octahedral	14.7°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca20	1.0	**81.7 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca21	1.0	65.8 (63.6)	O_2	**0.5**	**0.66**	**Tetrahedral**	7.1°	**50%**	Na(2), Mg(1), **Ca(1)**
Ca22	1.0	87.3 (60.5)	**N_1**	**0.12**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca23	1.0	97.5 (92.7)	O_2	**0.6**	**0.61**	**Tetrahedral**	4.0°	**50%**	Na(3), Mg(1), **Ca(1)**
Ca24	1.0	80.2 (74.5)	**O_2N_1**	**0.3**	**0.76**	**Trigonal Bipyramidal**	14.3°	**40%**	Na(1), Mg(0), **Ca(0)**
Ca25	1.0	81.3 (72.5)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26	1.0	70.2 (57.7)	**O_1**	**0.15**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca27	1.0	66.6 (64.4)	**O_3N_1**	**0.4**	**0.7**	Octahedral	**29.1°**	**33%**	Na(2), Mg(2), **Ca(2)**
Ca28	1.0	**84.7 (45.1)**	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca29	1.0	**101.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca30 (MeA)	1.0	51.8 (39.9)	O_5	1.7	**0.24**	Octahedral	19.8°	16%	**Ca(6)**, Na(6), Mg(4)
Ca31	1.0	88.9 (60.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
holoES (pre-tRNA) in 1 mM Ca^{2+}; PBD ID: 9OWU; EMDB: EMD-70944									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**102.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	121.0 (95.3)	**N_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)

Ca3	1.0	**112.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	118.0 (89.5)	**O_1**	**0.05**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5 (MeB)	1.0	113.0 (97.2)	O_3	**1.0**	**0.65**	Octahedral	9.6°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca6	1.0	100.5 (81.8)	**O_1**	**0.07**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7 (MeC)	1.0	130.4 (108.5)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca8	1.0	97.6 (83.9)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca9	1.0	105.8 (75.3)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	95.3 (82.6)	O_4	**0.2**	**0.59**	Octahedral	14.5°	**33%**	Na(4), Mg(4), **Ca(4)**
Ca11	1.0	111.9 (116.1)	**O_1**	**0.07**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	125.9 (116.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca13	1.0	112.2 (110.2)	O_3	**1.0**	**0.48**	Octahedral	**23.2°**	**50%**	Na(6), Mg(4), **Ca(4)**
Ca14	1.0	115.1 (107.3)	O_2	**0.5**	**0.8**	Octahedral	13.3°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca15	1.0	102.5 (78.2)	O_3	**0.19**	**0.63**	Octahedral	**24.2°**	**50%**	**Ca(4)**, Na(4), Mg(4)
Ca16	1.0	63.2 (71.2)	O_4	1.3	**0.37**	Octahedral	14.8°	**33%**	Na(6), **Ca(5)**, Mg(4)
Ca17	1.0	118.2 (119.0)	O_2	**0.5**	**0.83**	Octahedral	18.2°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca18	1.0	87.5 (85.1)	O_2	**0.6**	**0.78**	Octahedral	12.5°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca19	1.0	**123.4 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca20	1.0	140.2 (142.1)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca21	1.0	130.9 (106.6)	**N_1**	**0.4**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca22	1.0	101.7 (119.8)	**O_2N_1**	**0.15**	**0.61**	**Trigonal Bipyramidal**	16.0°	**40%**	Na(1), Mg(0), **Ca(0)**
Ca23	1.0	124.2 (105.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca24	1.0	104.5 (93.2)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca25	1.0	**123.6 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26 (MeA)	1.0	99.2 (93.0)	O_5	**1.2**	**0.25**	**Tetrahedral**	8.2°	25%	Na(5), Mg(2), **Ca(2)**
holoES (nc-pre-tRNA) in 5 mM Ca^{2+}; PBD ID: 9OWW; EMDB: EMD-70946									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	85.4 (71.1)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)

Ca2	1.0	106.7 (89.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**99.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	94.3 (76.1)	**O_1**	**0.11**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5 (MeB)	1.0	71.6 (69.4)	O_3	**0.9**	**0.65**	Octahedral	11.4°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca6	1.0	96.5 (60.0)	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7 (MeC)	1.0	99.4 (76.2)	O_2	**0.3**	**0.7**	Octahedral	6.5°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca8	1.0	110.6 (72.6)	O_2	**0.3**	**0.8**	Octahedral	14.6°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca9	1.0	109.6 (68.4)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	86.1 (70.0)	O_3	**0.13**	**0.7**	Octahedral	16.3°	**50%**	**Ca(4)**, Na(4), Mg(4)
Ca11	1.0	97.8 (95.4)	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	107.9 (73.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca13	1.0	77.2 (78.6)	O_3	**1.2**	**0.4**	Octahedral	15.2°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca14	1.0	113.4 (98.7)	O_2	**0.6**	**0.8**	Octahedral	16.2°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca15	1.0	89.5 (62.0)	O_2	**0.2**	**0.82**	Octahedral	19.4°	**66%**	**Ca(3)**, Na(3), Mg(3)
Ca16	1.0	56.0 (60.9)	O_4	**1.2**	**0.44**	Octahedral	21.5°	**33%**	Na(6), Mg(4), **Ca(4)**
Ca17	1.0	105.6 (94.2)	O_2	**0.6**	**0.78**	Octahedral	13.4°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca18	1.0	79.7 (78.4)	O_4	**0.8**	**0.49**	Octahedral	10.3°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca19	1.0	**100.6 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca20	1.0	91.3 (90.7)	O_2	**0.4**	**0.71**	Octahedral	9.2°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca21	1.0	117.9 (94.9)	**N_1**	**0.06**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca22	1.0	125.0 (117.1)	O_3	**0.17**	**0.31**	**Tetrahedral**	**29.2°**	25%	Na(3), Mg(2), **Ca(2)**
Ca23	1.0	95.9 (111.3)	**O_2N_1**	**0.7**	**0.5**	**Trigonal Bipyramidal**	11.5°	**40%**	Na(4), Mg(2), **Ca(0)**
Ca24	1.0	104.3 (95.8)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca25	1.0	94.8 (78.8)	**O_1**	**0.08**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26	1.0	**119.5 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca27	1.0	97.6 (78.3)	**O_1**	**0.18**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca28	1.0	**120.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)

Ca29	1.0	104.7 (96.5)	**O_4N_1**	**0.9**	**0.34**	Octahedral	**27.4°**	16%	Na(3), Mg(2), **Ca(2)**
Ca30 (MeA)	1.0	62.3 (61.4)	O_5	1.7	**0.24**	Octahedral	18.9°	16%	**Ca(6)**, Na(6), Mg(4)
holoES (LB-pre-tRNA-tilt1) in 5 mM Ca^{2+}; PBD ID: 9OWX; EMDB: EMD-70947									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**60.9 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**79.8 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**60.8 (23.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	65.5 (44.9)	**O_1**	**0.08**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5 (MeB)	1.0	41.4 (35.4)	O_3	**1.0**	**0.65**	Octahedral	12.3°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca6	1.0	51.4 (31.6)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7 (MeC)	1.0	59.6 (42.5)	O_2	**0.5**	**0.72**	Octahedral	3.0°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca8	1.0	**75.1 (30.4)**	O_2	**0.2**	**0.81**	Octahedral	17.2°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca9	1.0	**70.5 (31.1)**	**O_1**	**0.06**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	64.6 (38.4)	**O_1**	**0.05**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca11	1.0	68.5 (64.7)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	**77.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca13	1.0	48.3 (43.2)	O_3	**1.1**	**0.41**	Octahedral	15.7°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca14	1.0	72.0 (57.8)	O_2	**0.5**	**0.75**	Octahedral	2.2°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca15	1.0	51.0 (33.1)	O_2	**0.3**	**0.81**	Octahedral	17.8°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca16	1.0	29.0 (28.3)	O_4	1.4	**0.34**	Octahedral	9.7°	**33%**	Na(6), **Ca(5)**, Mg(4)
Ca17	1.0	74.4 (52.7)	O_2	**0.5**	**0.79**	Octahedral	10.8°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca18	1.0	47.5 (41.7)	O_4	**0.8**	**0.55**	Octahedral	13.4°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca19	1.0	**85.3 (33.2)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca20	1.0	65.7 (55.3)	O_2	**0.5**	**0.61**	**Tetrahedral**	1.0°	**50%**	Na(2), Mg(1), **Ca(1)**
Ca21	1.0	76.5 (56.5)	**O_1**	**0.1**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca22	1.0	64.9 (65.3)	**O_2N_1**	**0.5**	**0.6**	**Trigonal Bipyramidal**	10.0°	**40%**	Na(3), Mg(2), **Ca(0)**
Ca23	1.0	78.8 (54.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)

Ca24	1.0	65.9 (38.9)	**O_1**	**0.2**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca25	1.0	**92.8 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26	1.0	56.2 (54.6)	**O_3N_1**	**0.6**	0.16	Octahedral	**29.1°**	**33%**	Na(4), Mg(4), **Ca(2)**
Ca27	1.0	61.3 (60.5)	O_3	**0.8**	**0.6**	Octahedral	20.8°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca28 (MeA)	1.0	48.0 (31.9)	O_5	1.4	**0.24**	Octahedral	**21.9°**	16%	Na(6), **Ca(5)**, Mg(4)
holoES (LB-pre-tRNA-tilt2) in 5 mM Ca^{2+}; PBD ID: 9OWY; EMDB: EMD-70948									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	78.0 (53.1)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**86.1 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**80.1 (43.1)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	74.6 (61.3)	**O_1**	**0.07**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5 (MeB)	1.0	56.1 (55.5)	O_3	**1.1**	**0.64**	Octahedral	12.2°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca6	1.0	78.6 (51.7)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7 (MeC)	1.0	76.4 (62.6)	O_2	**0.5**	**0.69**	Octahedral	5.7°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca8	1.0	**85.3 (43.2)**	O_2	**0.2**	**0.87**	Octahedral	**22.2°**	**66%**	**Ca(3)**, Na(3), Mg(3)
Ca9	1.0	82.3 (49.9)	**O_1**	**0.07**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	88.6 (70.2)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca11	1.0	**83.8 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	58.6 (61.0)	O_3	**1.1**	**0.39**	Octahedral	16.5°	**50%**	Na(6), Mg(4), **Ca(4)**
Ca13	1.0	87.7 (72.8)	O_2	**0.3**	**0.82**	Octahedral	18.7°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca14	1.0	69.8 (51.7)	O_2	**0.3**	**0.84**	Octahedral	18.1°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca15	1.0	45.3 (48.6)	O_4	1.3	**0.42**	Octahedral	14.6°	**33%**	Na(6), **Ca(5)**, Mg(4)
Ca16	1.0	85.5 (69.6)	O_2	**0.5**	**0.76**	Octahedral	7.6°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca17	1.0	61.5 (64.0)	O_4	**0.8**	**0.53**	Octahedral	13.3°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca18	1.0	92.8 (59.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca19	1.0	76.3 (72.2)	O_2	**0.4**	**0.73**	**Tetrahedral**	6.1°	**50%**	Na(2), Mg(1), **Ca(1)**
Ca20	1.0	99.4 (64.9)	**N_1**	**0.14**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)

Ca21	1.0	101.9 (98.6)	O_2	**0.6**	**0.55**	**Tetrahedral**	4.3°	**50%**	Na(3), Mg(1), **Ca(1)**
Ca22	1.0	79.3 (83.9)	**O_2N_1**	**0.6**	**0.61**	**Trigonal Bipyramidal**	10.5°	**40%**	Na(2), Mg(0), **Ca(0)**
Ca23	1.0	**87.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca24	1.0	76.0 (59.3)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca25	1.0	**107.8 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26	1.0	103.6 (90.7)	O_2	**0.7**	**0.29**	**Linear**	**33.8°**	0	Na(2), Mg(1), **Ca(1)**
Ca27	1.0	95.5 (91.3)	O_2	**0.4**	**0.72**	**Tetrahedral**	0.7°	**50%**	Na(2), Mg(1), **Ca(1)**
Ca28	1.0	76.3 (70.7)	**O_3N_1**	**0.5**	**0.37**	Octahedral	**30.2°**	**33%**	Na(2), Mg(2), **Ca(2)**
Ca29	1.0	77.9 (77.1)	O_3	**0.8**	**0.6**	Octahedral	**21.6°**	**50%**	Na(5), Mg(4), **Ca(4)**
Ca30	1.0	**93.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca31 (MeA)	1.0	66.6 (52.7)	O_5	1.4	0.23	Octahedral	20.7°	16%	Na(6), **Ca(5)**, Mg(4)
holoEP (mat-tRNA) in 5 mM Ca^{2+}; PBD ID: 9OWV; EMDB: EMD-70945									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**80.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**117.5 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	87.7 (76.3)	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4 (MeA)	1.0	66.4 (67.4)	O_5	1.5	0.17	**Tetrahedral**	11.6°	25%	Na(5), **Ca(3)**, Mg(2)
Ca5	1.0	93.7 (70.5)	O_2	**0.3**	**0.94**	Octahedral	17.2°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca6	1.0	82.1 (79.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7	1.0	89.3 (82.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca8	1.0	67.6 (71.9)	O_3	**1.0**	**0.31**	Octahedral	**21.8°**	**50%**	Na(6), Mg(4), **Ca(4)**
Ca9	1.0	96.7 (91.1)	O_2	**0.7**	**0.73**	Octahedral	3.1°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca10	1.0	55.7 (58.7)	O_2	**0.3**	**0.82**	Octahedral	20.3°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca11	1.0	109.7 (90.3)	**O_1**	**0.2**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	88.9 (95.7)	O_3	**0.6**	**0.71**	Octahedral	20.0°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca13	1.0	59.1 (63.2)	O_4	1.4	**0.34**	Octahedral	14.3°	**33%**	Na(6), **Ca(5)**, Mg(4)
Ca14	1.0	90.8 (88.8)	O_2	**0.7**	**0.78**	Octahedral	11.6°	**66%**	Na(4), Mg(3), **Ca(3)**

Ca15	1.0	69.6 (71.7)	O_4	**0.8**	**0.48**	Octahedral	9.6°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca16	1.0	**85.7 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca17	1.0	85.8 (93.6)	**O_1**	**0.17**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca18	1.0	101.3 (91.6)	**O_1N_1**	**0.16**	**0.61**	**Tetrahedral**	3.5°	**50%**	Na(1), Mg(0), **Ca(0)**
Ca19	1.0	113.2 (107.1)	O_2	**0.3**	**0.8**	**Linear**	12.4°	0	Na(1), Mg(1), **Ca(1)**
Ca20	1.0	90.5 (97.7)	**O_2N_1**	**0.3**	**0.82**	**Trigonal Bipyramidal**	11.5°	**40%**	Na(1), Mg(0), **Ca(0)**
Ca22	1.0	88.4 (78.6)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca23	1.0	118.1 (109.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca24	1.0	103.7 (98.1)	**O_1**	**0.03**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca25	1.0	96.3 (82.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca26	1.0	**102.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca27	1.0	121.3 (85.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca28	1.0	92.2 (67.8)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
TLm holoE (subclass 1) in 5 mM Ca^{2+}; PBD ID: 9OY5; EMDB: EMD-70997									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**63.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**74.5 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**77.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	89.4 (50.8)	**O_1**	**0.05**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5	1.0	57.4 (58.1)	O_2	**0.3**	**0.96**	Octahedral	19.7°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca6	1.0	67.5 (49.8)	**O_1**	**0.02**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7	1.0	79.9 (78.6)	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca8	1.0	79.3 (61.5)	O_2	**0.4**	**0.85**	Octahedral	12.7°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca9	1.0	84.1 (75.8)	O_2	**0.6**	**0.72**	Octahedral	1.9°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca10	1.0	46.7 (38.3)	O_2	**0.5**	**0.79**	Octahedral	14.4°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca11	1.0	74.8 (68.3)	**O_1**	**0.12**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca12	1.0	25.4 (33.0)	O_4	1.6	**0.37**	Octahedral	6.0°	**33%**	Na(6), **Ca(5)**, Mg(4)

Ca13	1.0	99.9 (109.2)	O_2	**0.3**	**0.93**	Octahedral	4.0°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca14	1.0	46.9 (47.7)	O_3	**0.7**	**0.58**	Octahedral	13.7°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca15	1.0	**83.6 (39.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca16	1.0	91.4 (98.0)	O_2	**0.2**	**0.89**	Octahedral	7.6°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca17	1.0	95.3 (80.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca18	1.0	79.8 (58.7)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca19 (MeA)	1.0	48.3 (46.5)	O_3	**1.0**	**0.6**	Octahedral	13.7°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca20	1.0	86.9 (66.3)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca21	1.0	132.5 (175.0)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca22	1.0	79.4 (59.8)	**O_1**	**0.09**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca23	1.0	70.9 (58.6)	**O_1**	**0.02**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca24	1.0	77.9 (87.9)	O_3	**0.6**	**0.58**	Octahedral	**24.2°**	**50%**	Na(5), Mg(4), **Ca(4)**
TLm holoE (subclass 2) in 5 mM Ca^{2+}; PBD ID: 9OY6; EMDB: EMD-70998									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**68.7 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	76.5 (59.1)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**74.6 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	81.2 (48.8)	**O_1**	**0.06**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5	1.0	74.7 (57.0)	O_2	**0.3**	**0.76**	Octahedral	6.8°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca6	1.0	67.7 (47.6)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca7	1.0	67.9 (68.8)	**O_1**	**0.06**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca8	1.0	104.2 (109.0)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca9	1.0	81.8 (59.4)	O_2	**0.4**	**0.95**	Octahedral	20.6°	**66%**	Na(3), Mg(3), **Ca(3)**,
Ca10	1.0	84.5 (67.9)	O_2	**0.5**	**0.82**	Octahedral	13.9°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca11	1.0	39.7 (33.6)	O_2	**0.5**	**0.8**	Octahedral	15.9°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca12	1.0	76.5 (66.7)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca13	1.0	28.0 (32.4)	O_4	1.4	**0.4**	Octahedral	10.2°	**33%**	Na(6), **Ca(5)**, Mg(4)

Ca14	1.0	95.1 (96.2)	O_2	**0.4**	**0.76**	Octahedral	2.0°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca15	1.0	39.9 (41.9)	O_4	**0.8**	**0.55**	Octahedral	13.3°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca16	1.0	62.7 (41.5)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca17	1.0	104.5 (113.1)	O_2	**0.7**	**0.67**	Octahedral	6.3°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca18	1.0	**90.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca19	1.0	66.4 (58.4)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca20 (MeA)	1.0	45.9 (36.1)	O_3	**0.9**	**0.65**	Octahedral	10.4°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca21	1.0	82.5 (46.2)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca22	1.0	**108.3 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca23	1.0	76.8 (54.4)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca24	1.0	77.2 (52.1)	**O_1**	**0.04**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca25	1.0	77.4 (81.5)	O_3	**0.7**	**0.29**	Octahedral	16.5°	**50%**	Na(5), Mg(4), **Ca(4)**
TLm holoES (pre-tRNA) in 5 mM Ca^{2+}; PBD ID: 9OY7; EMDB: EMD-71000									
Metal ID	**Occupancy**	**Bfactor(env.)[1]**	**Atomic contacts**	**Valence[2]**	**nVECSUM[3]**	**Geometry[1,4]**	**gRMSD(°)[1]**	**Vacancy[1]**	**Ranking (Points)**
Ca1	1.0	**86.2 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca2	1.0	**86.1 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca3	1.0	**86.7 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca4	1.0	99.3 (78.8)	**O_1**	**0.09**	**1.0**	**Poorly Coordinated**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca5	1.0	70.9 (70.9)	O_3	**0.9**	**0.67**	Octahedral	12.9°	**50%**	Na(5), Mg(4), **Ca(4)**
Ca6	1.0	100.1 (82.8)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca7	1.0	81.6 (66.4)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca8	1.0	94.6 (95.1)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca9	1.0	98.3 (79.0)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca10	1.0	84.1 (74.2)	O_4	**1.2**	**0.25**	**Linear**	12.9°	0	Na(4), Mg(2), **Ca(2)**
Ca11	1.0	99.3 (90.9)	O_2	**0.5**	**0.81**	Octahedral	17.3°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca12	1.0	70.6 (63.2)	O_3	**0.3**	**0.7**	Octahedral	20.5°	**50%**	Na(4), Mg(4), **Ca(4)**
Ca13	1.0	107.5 (85.9)	**O_1**	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**

Ca14	1.0	52.1 (61.0)	O_4	**1.2**	**0.37**	Octahedral	13.9°	**33%**	Na(6), Mg(4), **Ca(4)**
Ca15	1.0	90.4 (87.5)	O_2	**0.6**	**0.76**	Octahedral	8.0°	**66%**	Na(4), Mg(3), **Ca(3)**
Ca16	1.0	78.6 (75.3)	O_4	**0.8**	**0.45**	Octahedral	12.7°	**66%**	Na(5), Mg(4), **Ca(4)**
Ca17	1.0	**102.0 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca18	1.0	101.8 (103.7)	O_2	**0.5**	**0.59**	**Tetrahedral**	2.3°	**50%**	Na(2), Mg(1), **Ca(1)**
Ca19	1.0	97.1 (119.2)	O_2N_1	**0.3**	**0.68**	**Trigonal Bipyramidal**	13.7°	**40%**	Na(1), Mg(0), **Ca(0)**
Ca20	1.0	90.7 (81.7)	O_1	**0.3**	**1.0**	**Poorly Coordinated**	N/A	N/A	Na(0), Mg(0), **Ca(0)**
Ca21	1.0	**105.6 (0.0)**		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca22	1.0	92.8 (76.9)	O_2	**0.4**	**0.83**	Octahedral	2.9°	**66%**	Na(3), Mg(3), **Ca(3)**
Ca23	1.0	83.8 (62.8)		N/A	N/A	**Free**	N/A	N/A	**Ca(0)**, Na(0), Mg(0)
Ca24	1.0	85.4 (95.2)	O_3	**0.8**	**0.56**	Octahedral	**21.6°**	**50%**	Na(5), Mg(4), **Ca(4)**
Ca25	1.0	90.0 (67.6)	O_5	1.5	**0.24**	**Trigonal Bipyramidal**	17.5°	0	Na(5), **Ca(3)**, Mg(2)

Supplementary Table 4. Kinetic rate constants of pre-tRNA hydrolysis by RNase P holoenzyme derived from quantitative mass analysis.

	k_{on} (μM^{-1} min^{-1})	k_{cat} (min^{-1})	[holoE] (μM)	[pre-tRNA] (μM)	Percentage of singular value
Experimental 1	3.55 ± 0.20	1.05 ± 0.059	0.2*	2*	87 %
Experimental 2	2.17 ± 0.26	0.95 ± 0.11	0.2*	2*	92 %

Footnote: *fixed constant during the nonlinear regression fit.