

**Exploring the Substrate-Assisted Acetylation Mechanism by UDP-linked Sugar
N-Acetyltransferase from QM/MM Calculations: the Role of Residue Asn84 and
the Effects of Starting Geometries**

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Figure S1 The constructed solvation model for the MD simulation.

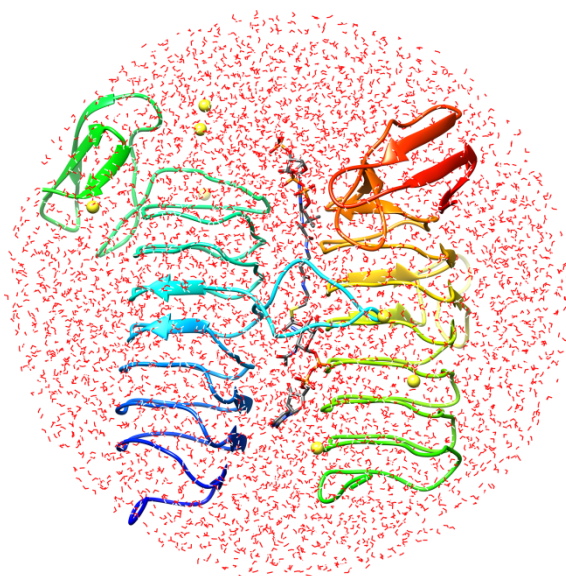


Figure S2 Time dependencies of the root-mean-square deviation (RMSD) for the backbone atoms of WlbB.

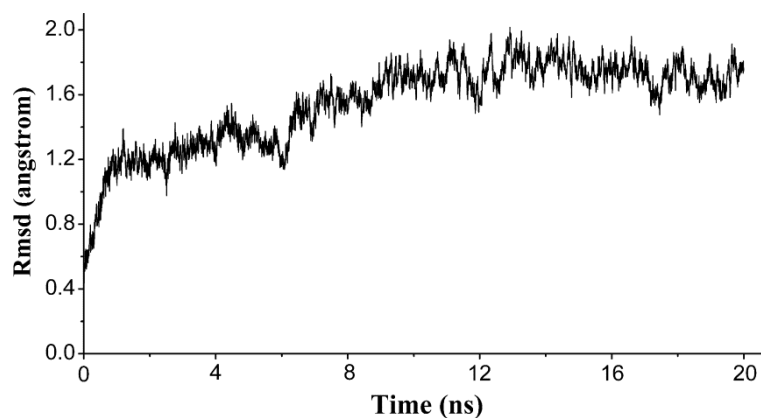


Figure S3 Optimized structures of reactant (ES), transition states (TS1 and TS2), intermediate (EI) and product (EP) for model A, in which Asn84 was left out to the QM part and mutated into Ala84.

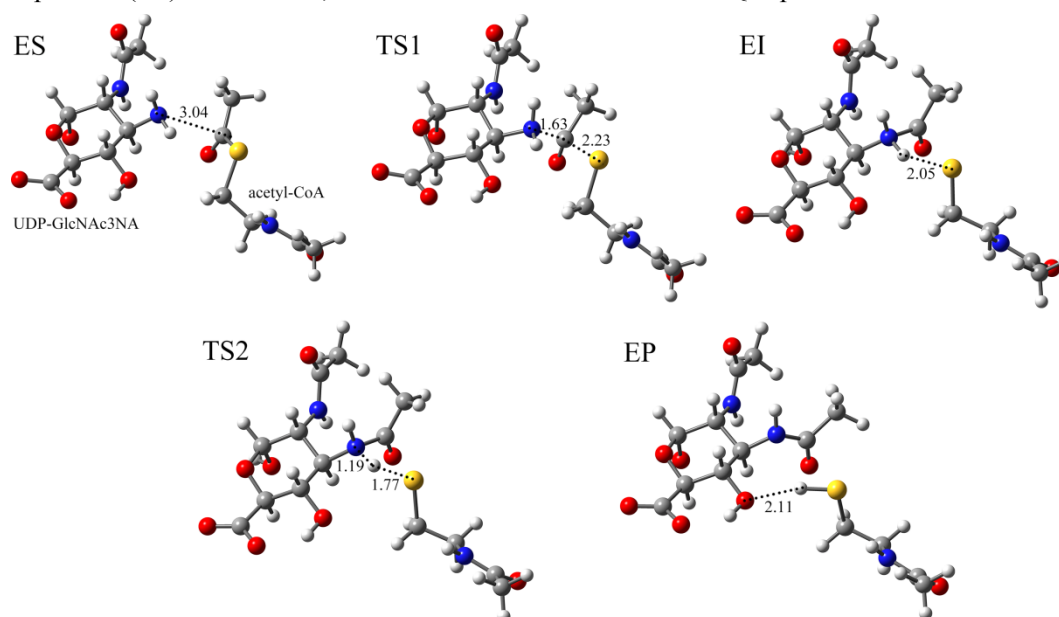


Figure S4 Optimized structures of reactant (ES), transition states (TS1 and TS2), intermediate (EI) and product (EP) for model B, in which Asn84 was left out to the QM part.

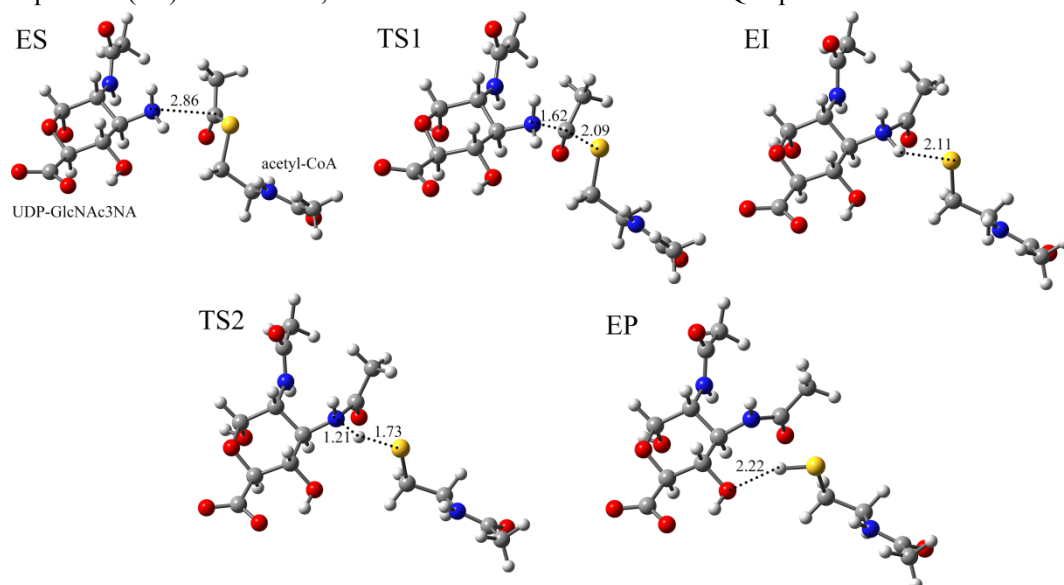


Figure S5 Optimized structures of reactant (ES), transition states (TS1 and TS2), intermediate (EI) and product (EP) for model C, in which the backbone NH group of Arg94, Gln59 and three solvent water molecules were added to the QM part.

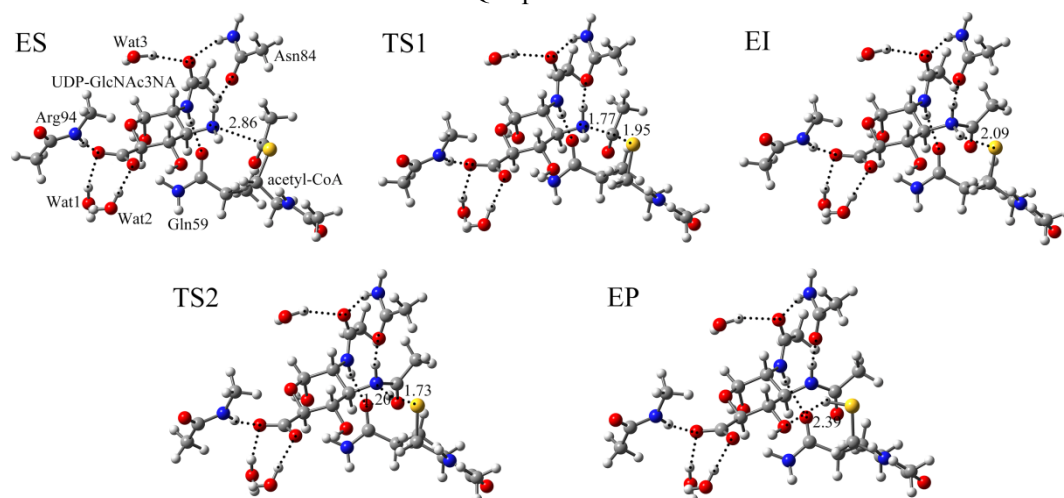


Figure S6 The superposition of the active sites of 6 optimized geometries after 5, 8, 11, 14, 17 and 20 ns MD simulations.

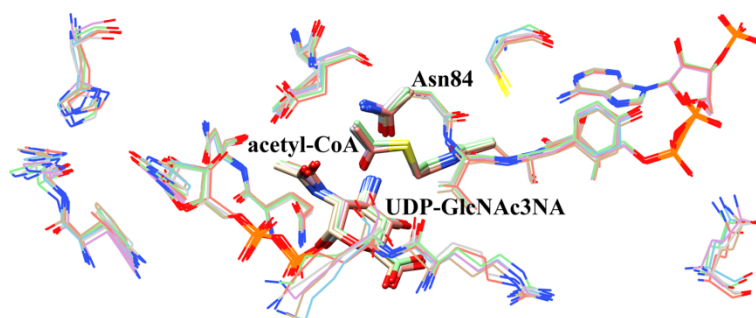


Table S1 Key distances (Å) in the optimized structures of all species along the reaction pathways for the six models from NAT-1 to NAT-6.

model	structure	N3-C	C-S	S-H2	N3-H2	O-H1
NAT-1	ES	2.96	1.80	2.68	1.00	2.18
	TS1	1.62	2.14	2.50	1.00	1.93
	EI	1.49	3.18	2.17	1.05	1.84
	TS2	1.45	3.18	1.73	1.23	1.88
	EP	1.35	3.28	1.32	2.40	2.05
NAT-2	ES	2.91	1.80	2.60	1.00	2.16
	TS1	1.64	2.08	2.41	1.00	1.88
	EI	1.49	3.10	2.08	1.06	1.87
	TS2	1.46	3.11	1.73	1.20	1.90
	EP	1.36	3.21	1.32	2.31	2.05
NAT-3	ES	2.94	1.81	2.69	1.00	2.17
	TS1	1.81	1.96	2.52	1.00	1.98
	EI	1.49	3.11	2.14	1.05	1.88
	TS2	1.45	3.12	1.76	1.19	1.91
	EP	1.36	3.26	1.32	2.35	2.09
NAT-4	ES	2.93	1.80	2.64	1.00	2.17
	TS1	1.69	2.02	2.47	1.00	1.92
	EI	1.49	3.15	2.15	1.05	1.86
	TS2	1.45	3.16	1.75	1.19	1.90
	EP	1.35	3.29	1.32	2.45	2.08
NAT-5	ES	2.90	1.80	2.68	1.00	2.07
	TS1	1.73	1.97	2.49	1.00	1.89
	EI	1.49	3.15	2.13	1.05	1.84
	TS2	1.45	3.19	1.73	1.21	1.88
	EP	1.35	3.25	1.32	2.49	1.99
NAT-6	ES	2.95	1.80	2.66	1.00	2.15
	TS1	1.64	2.07	2.46	1.00	1.89
	EI	1.49	3.11	2.16	1.05	1.87
	TS2	1.46	3.10	1.78	1.19	1.90
	EP	1.36	3.25	1.32	2.36	2.07