MOLECULAR MODELING OF BACTERIAL RESISTANCE. THE ROLE OF DYNAMIC BEHAVIOR OF PROTEIN COMPLEXES WITH SUBSTRATES OR INHIBITORS

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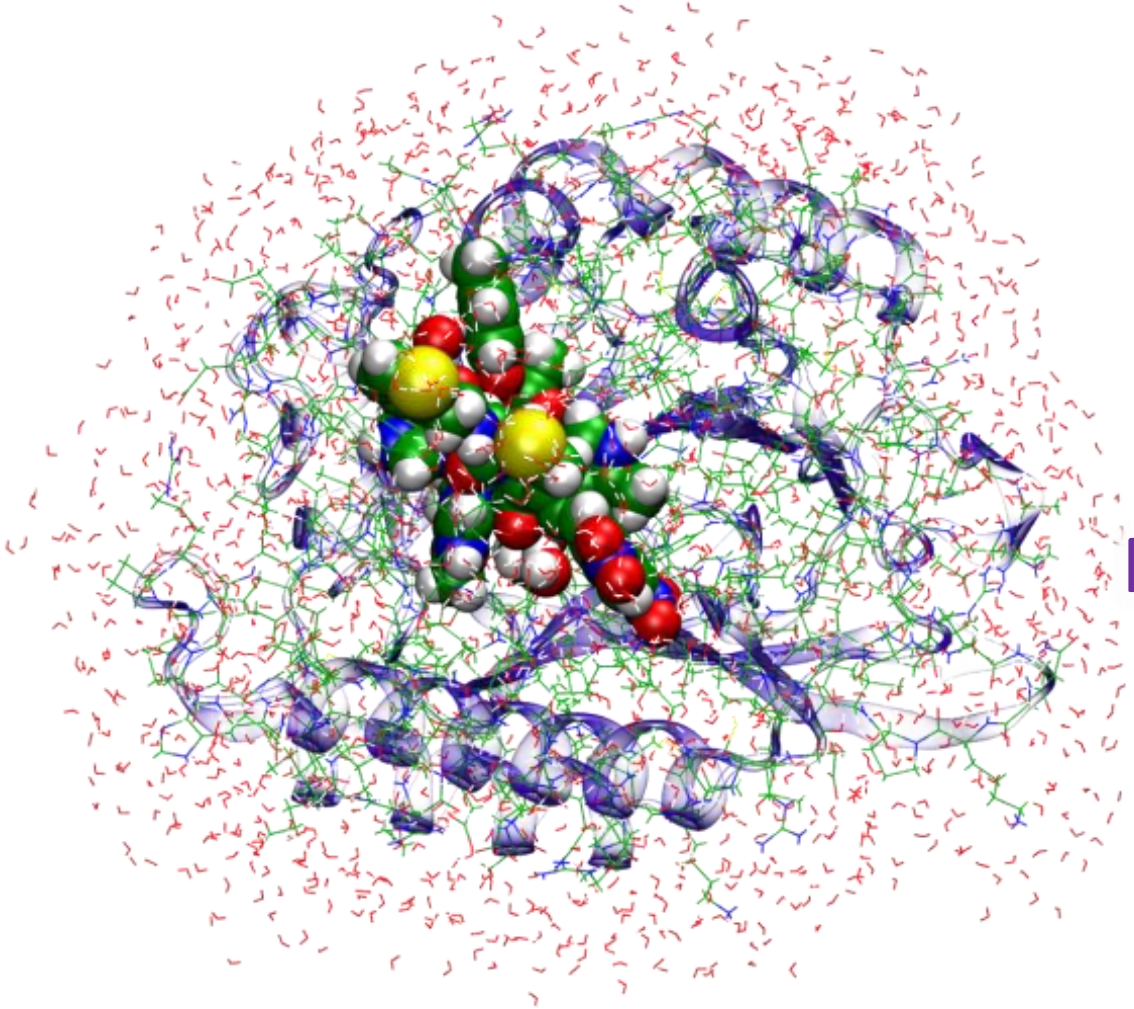
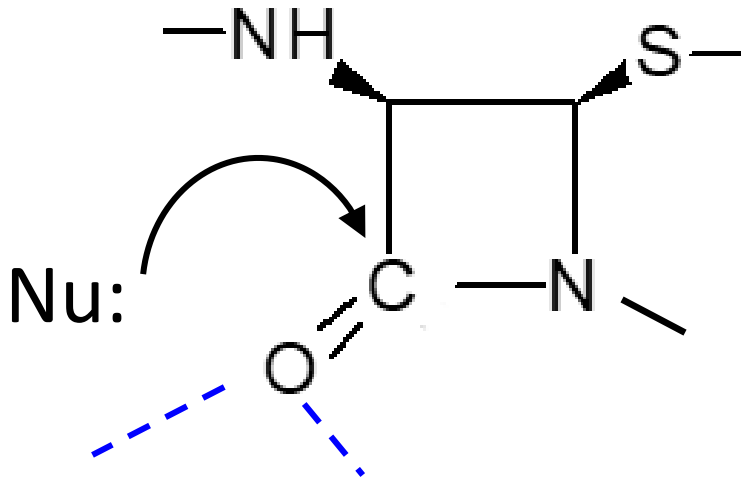
September 16, 2024

Interactions of bacterial enzymes with β-lactams

Enzyme class – EC 3 (hydrolases)





Oxyanion hole

PBP [Penicillin binding proteins

Nu: –OH of Ser

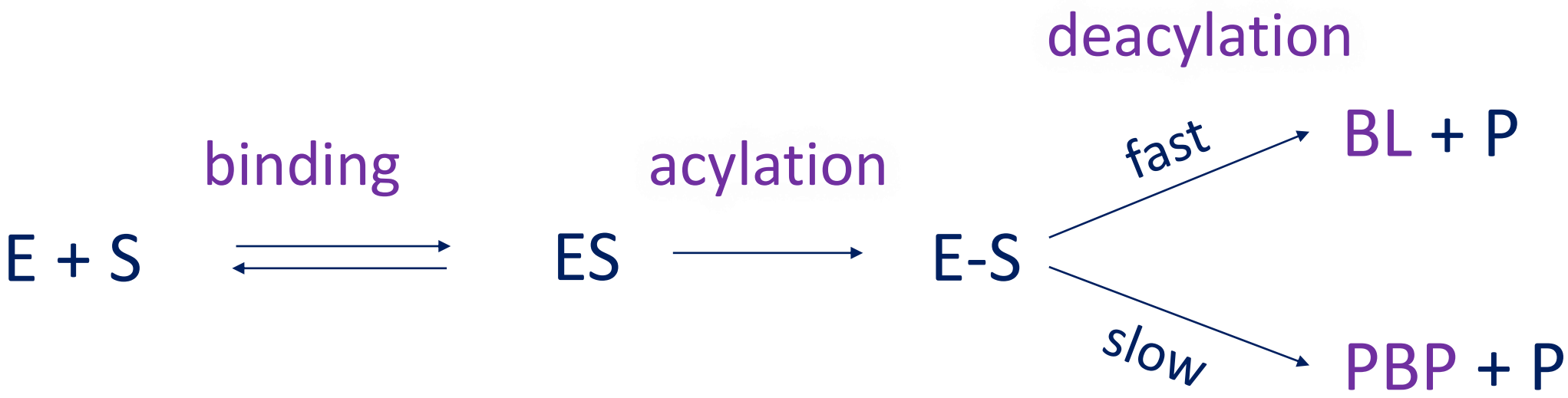
BL [Serine β-lactamases

Nu: –OH of Ser

Metallo-β-lactamases

Nu: OH^-

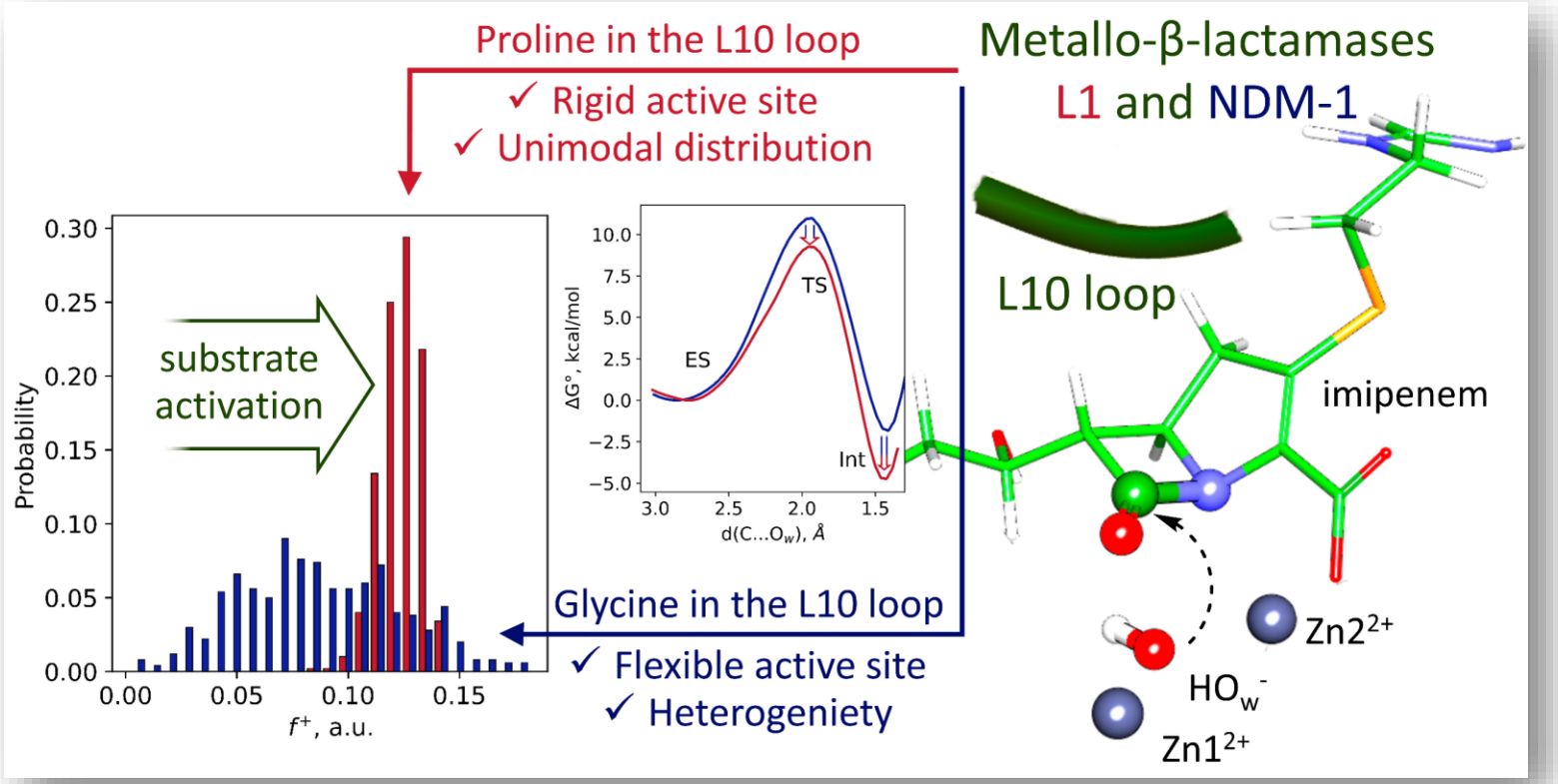
Interactions of bacterial enzymes with β-lactams



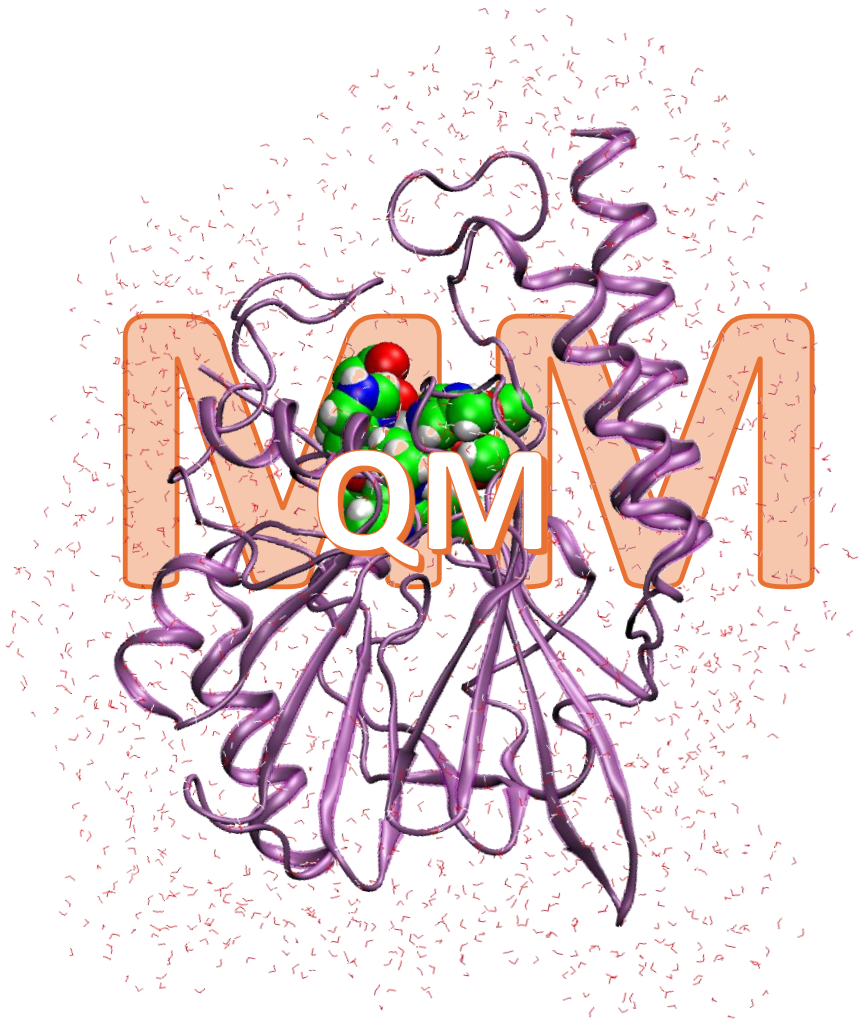
- ✓ Serine β-lactamases were evolutionary originated from PBPs
- ✓ Covalent inhibitors can be found for serine β-lactamases

Antimicrobial Agents and Chemotherapy (2018) DOI: 10.1128/AAC.01076-18

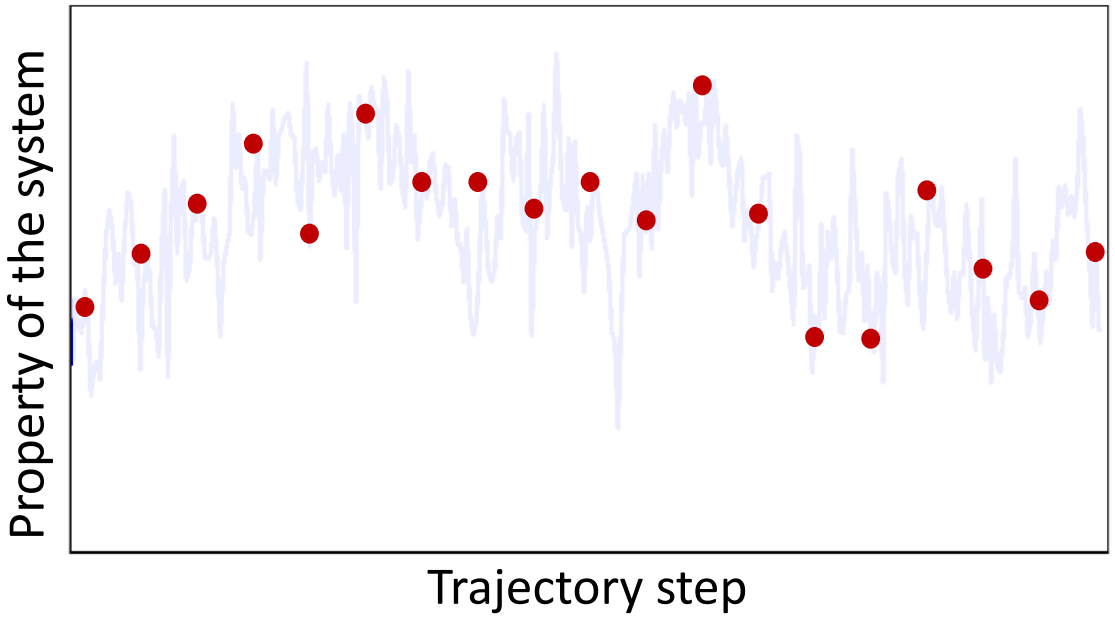
Catalytic efficiency and dynamic behavior of enzyme-substrate complexes: metallo-β-lactamases NDM-1 and L1

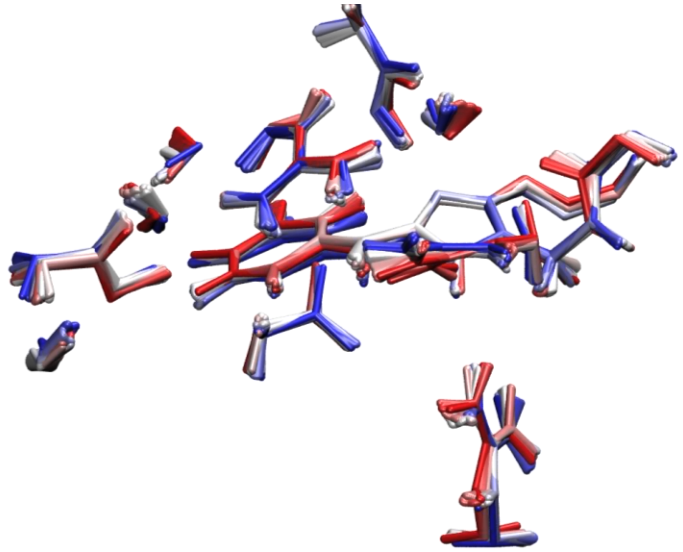


From local minima to trajectories

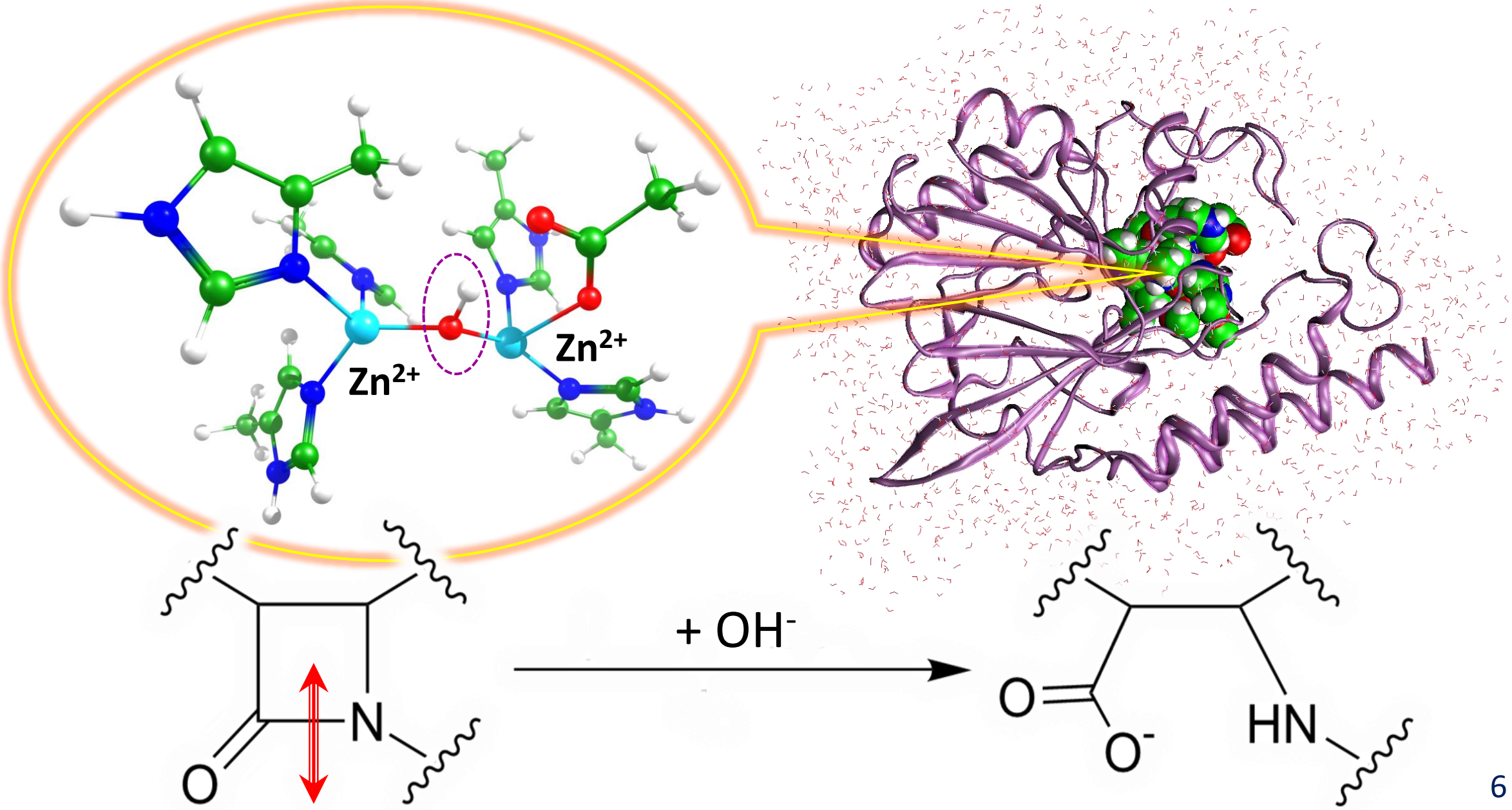


MD

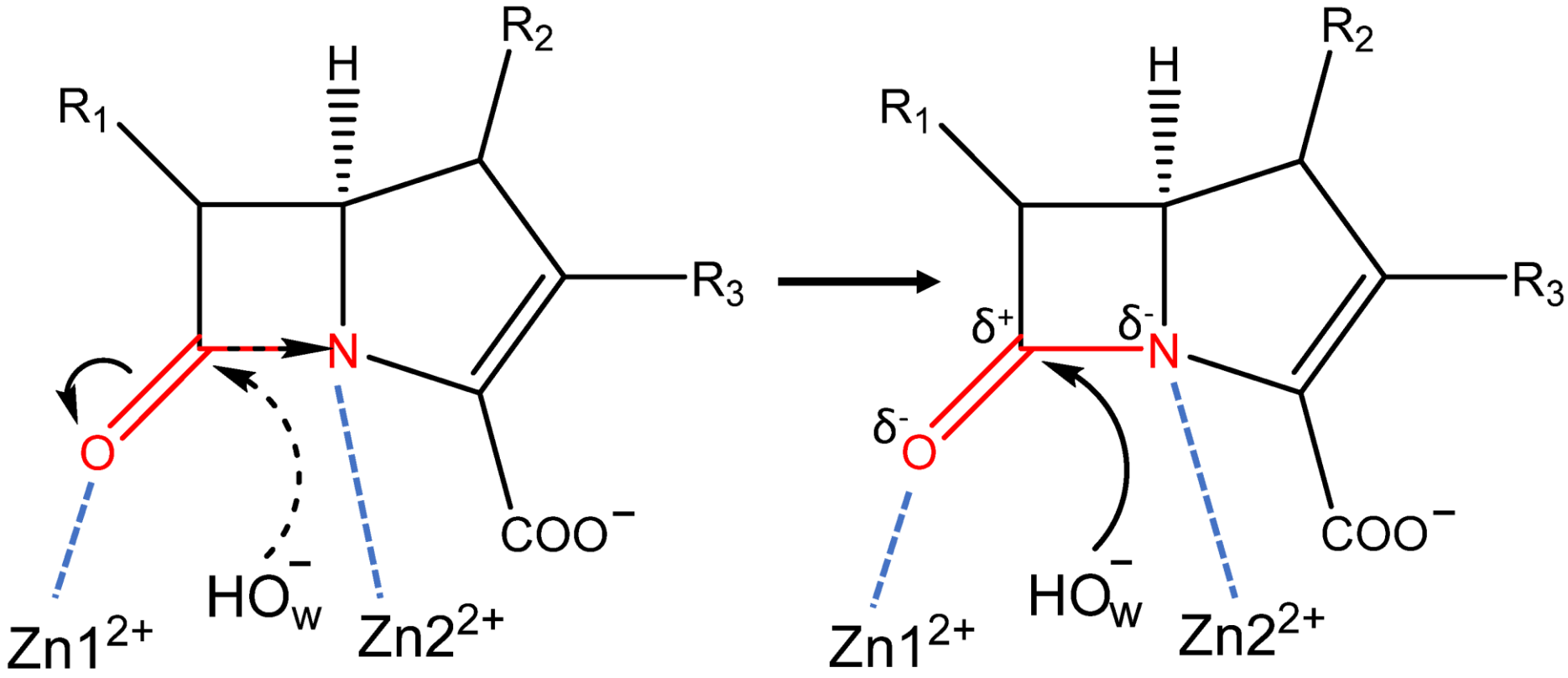




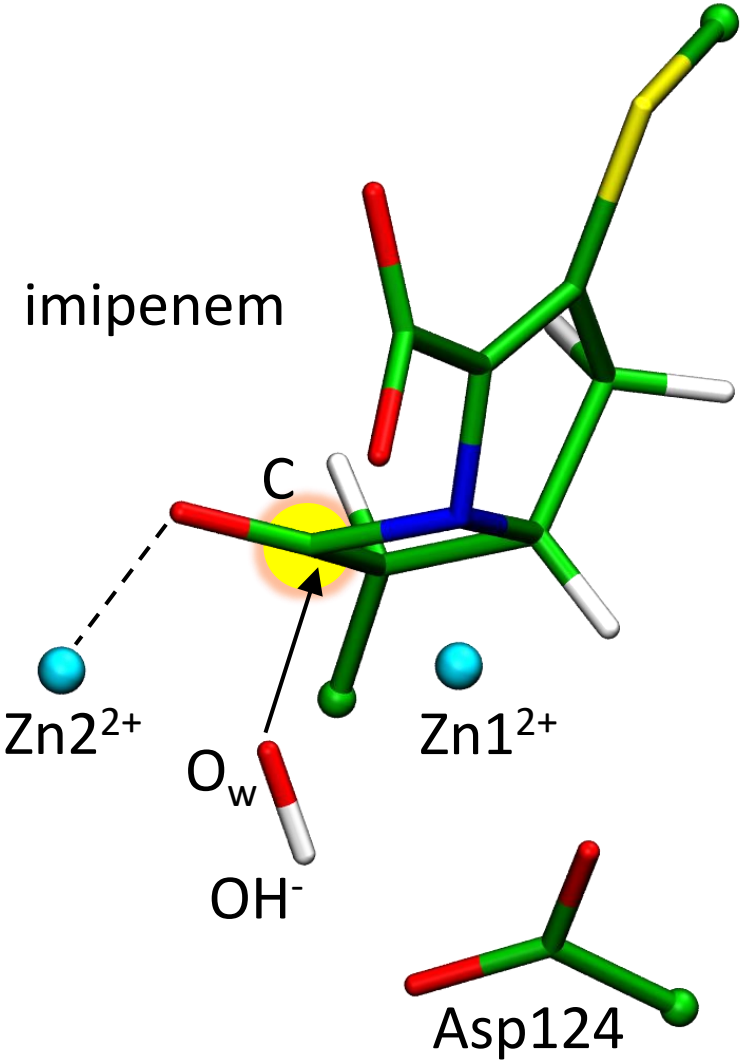
Hydrolysis mechanism in the active site of metallo-β-lactamases



Carbapenem activation in the active site of metallo-β-lactamases

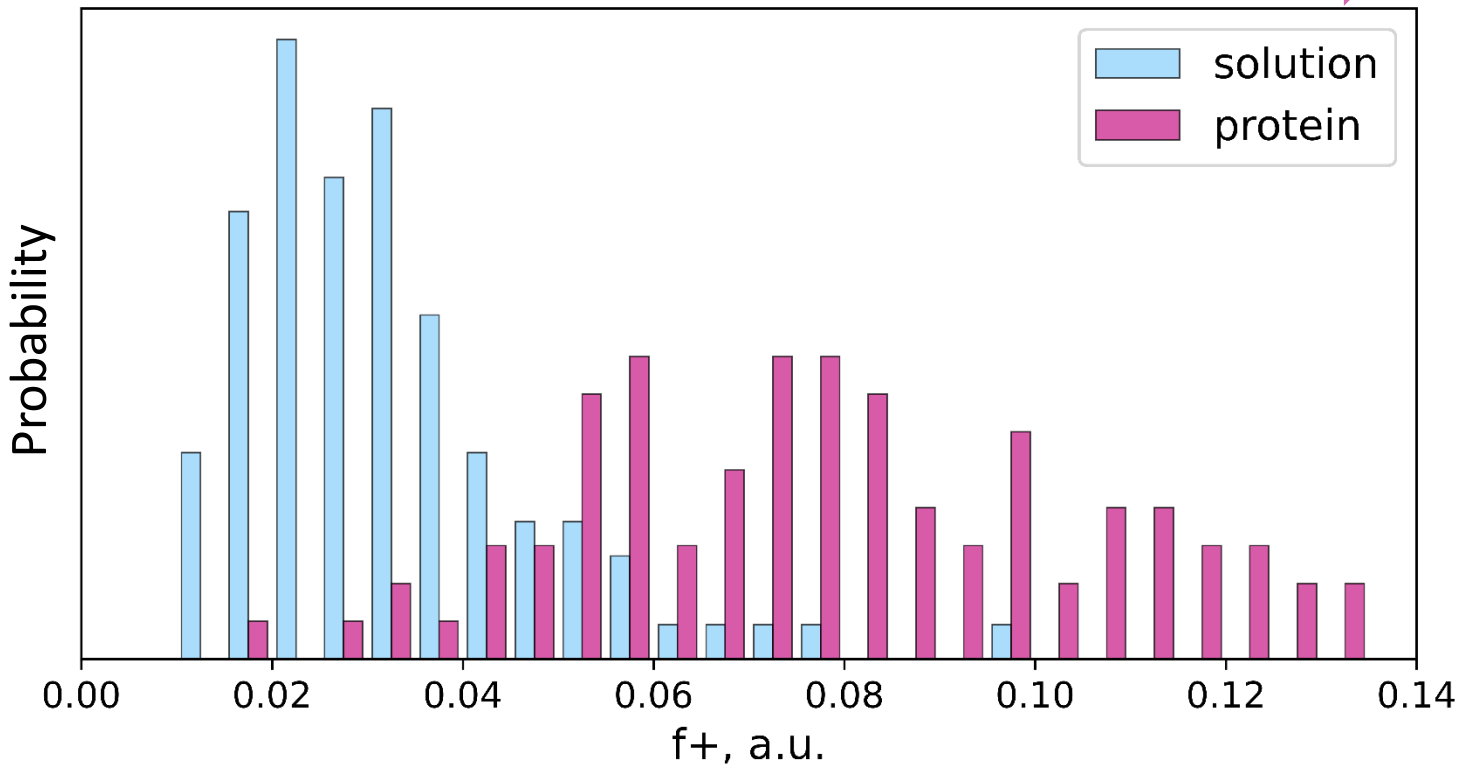


Substrate activation in the enzyme active site



QM(PBE0-D3/6-31G**)/MM MD

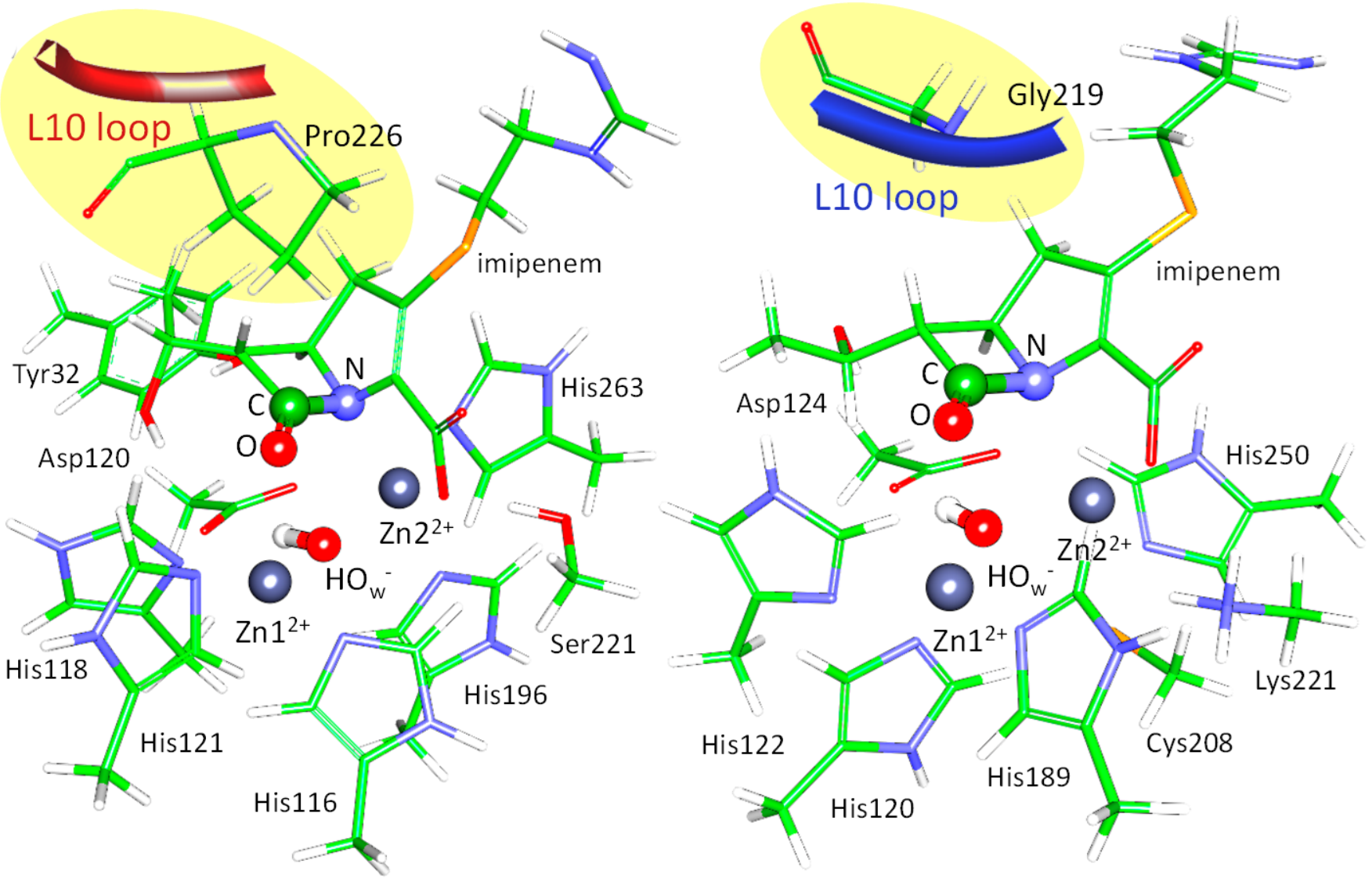
An enzyme activates a substrate



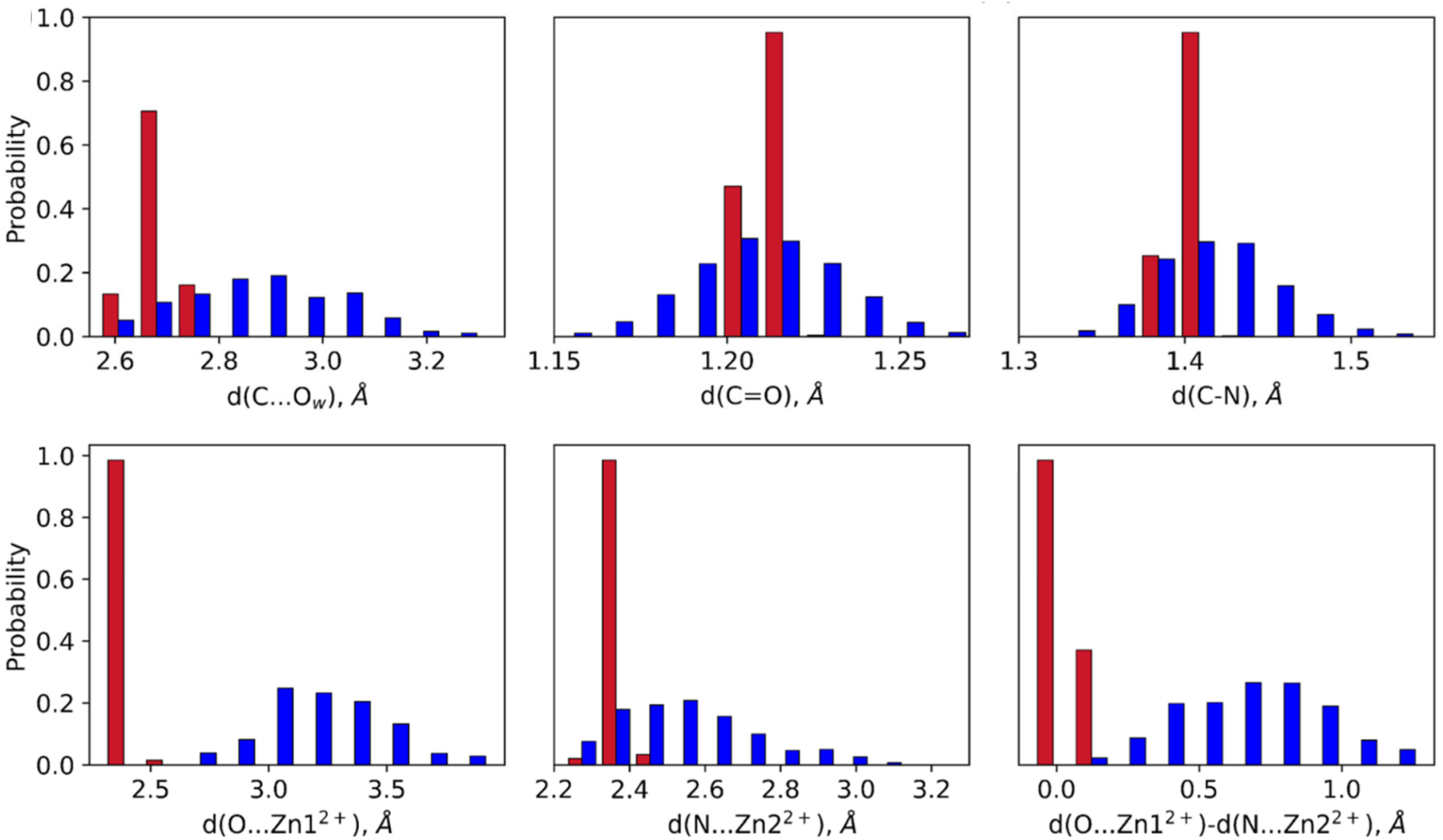
Fukui atomic index

$$f^{+}(C) = q_{N+1}(C) - q_{N}(C)$$

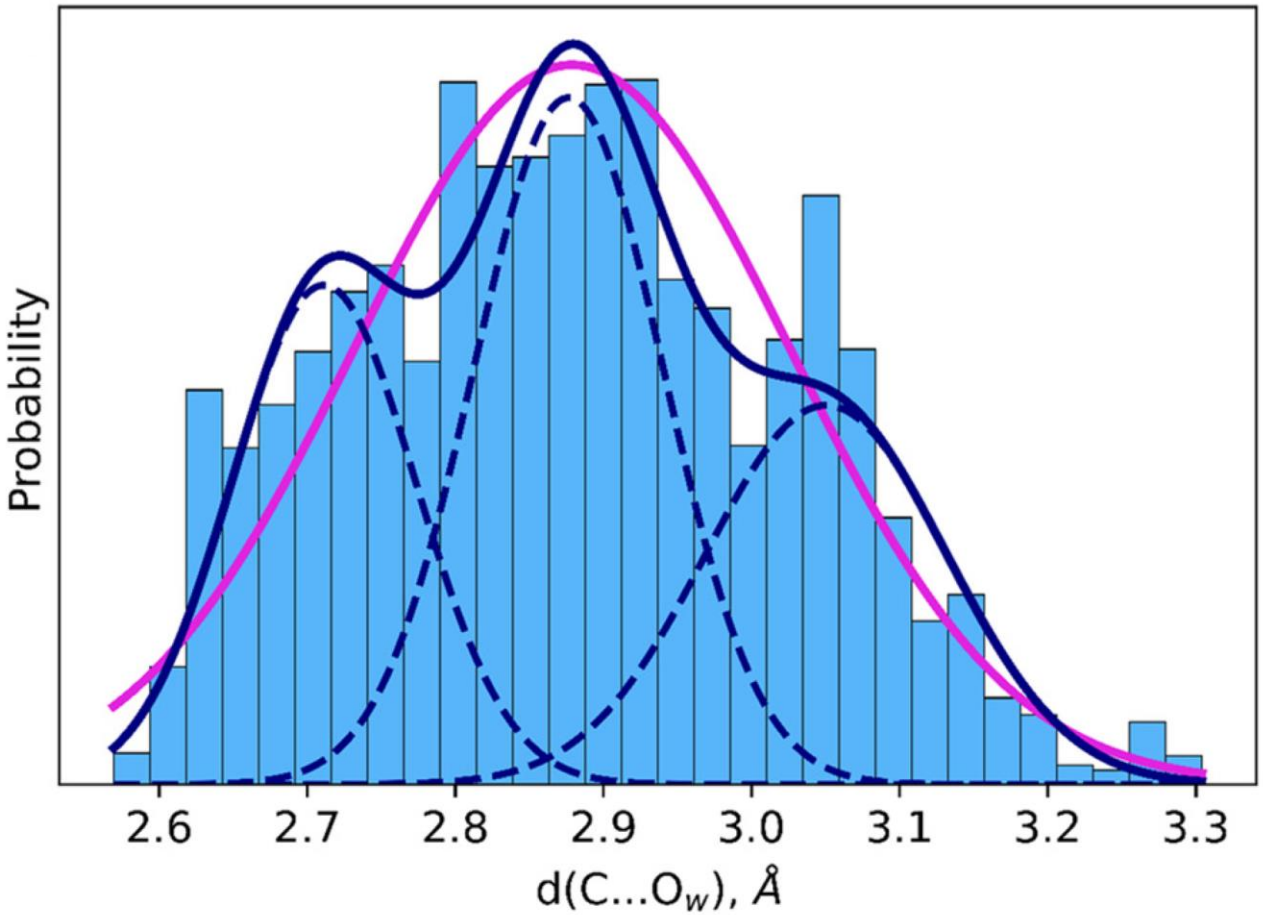
Active sites of metallo-β-lactamases NDM-1 and L1

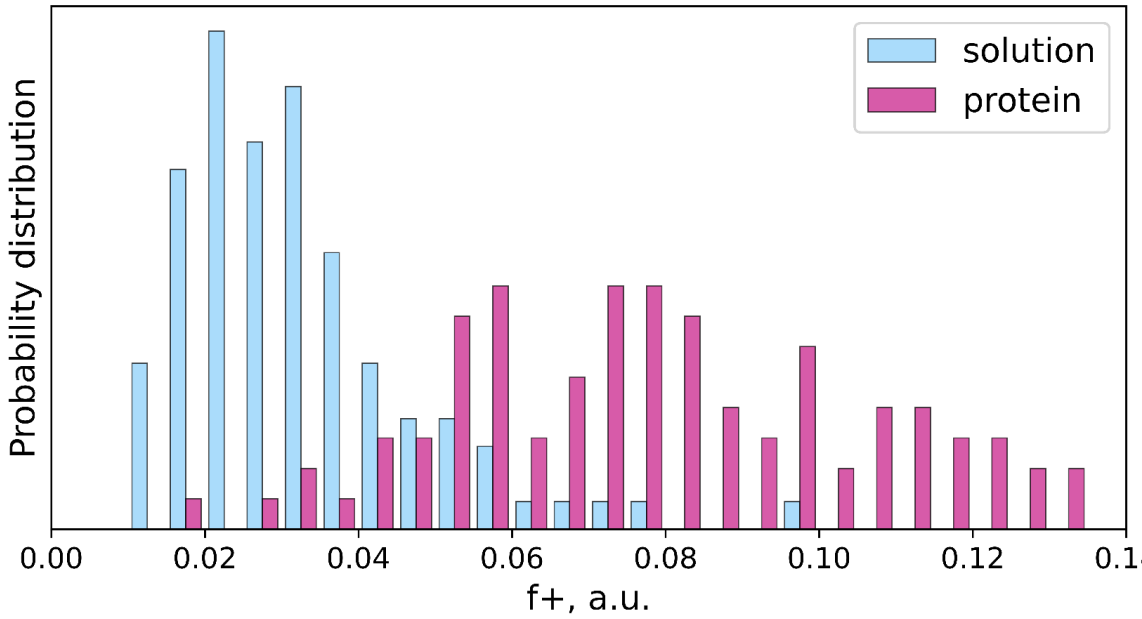


Dynamics of ES complexes of metallo-β-lactamases NDM-1 and L1

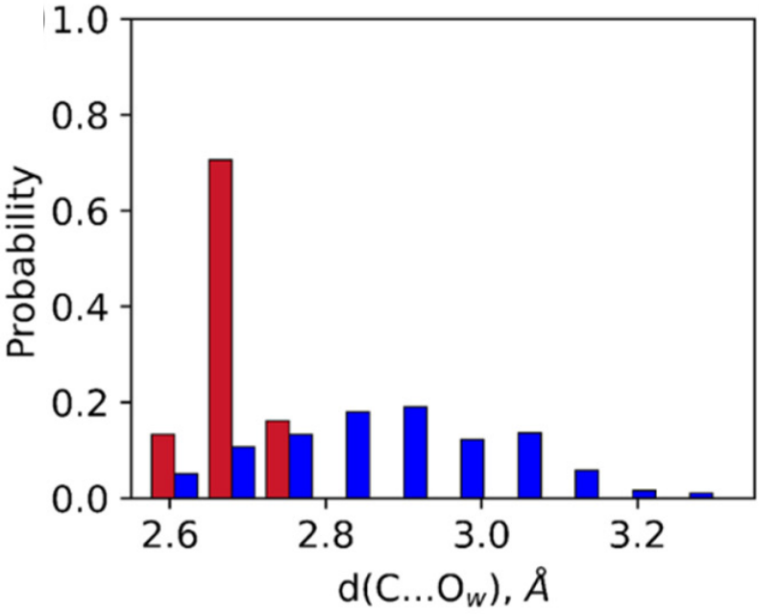


Dynamics of ES complexes of metallo-β-lactamases NDM-1 and L1

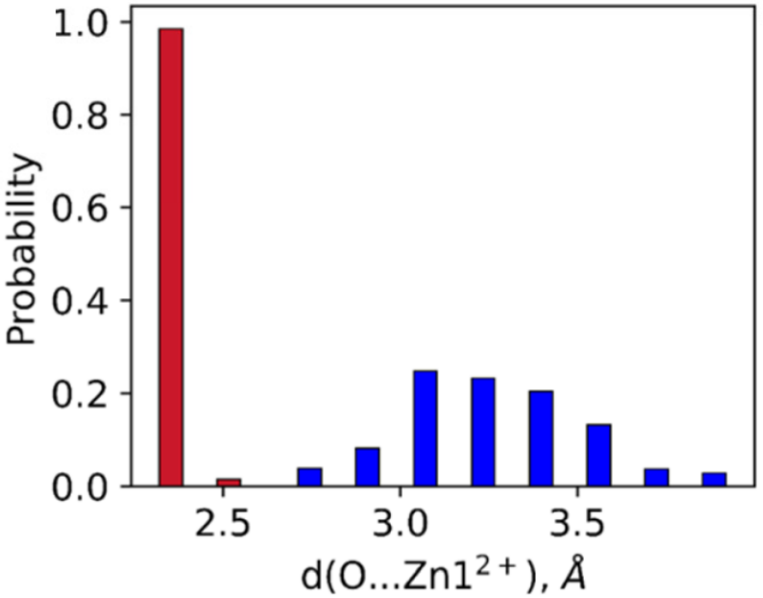


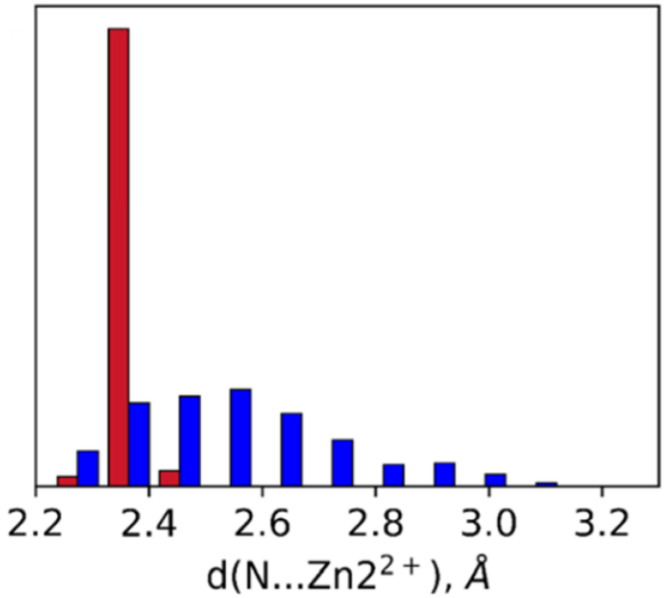


Dynamics of ES complexes of metallo-β-lactamases NDM-1 and L1

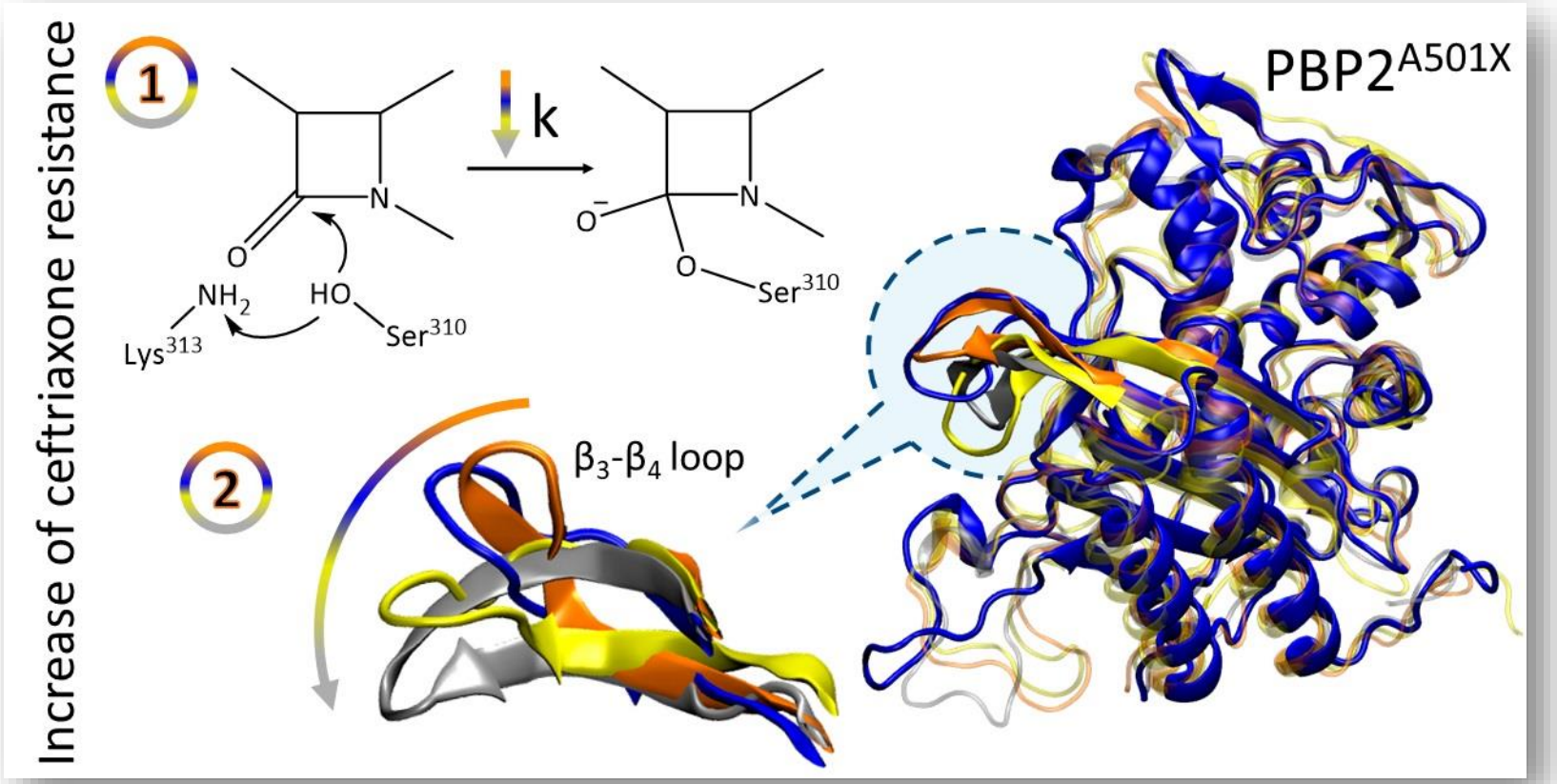


	k_{cat}, s^{-1}
L1 – imipenem	384
NDM-1 - imipenem	64

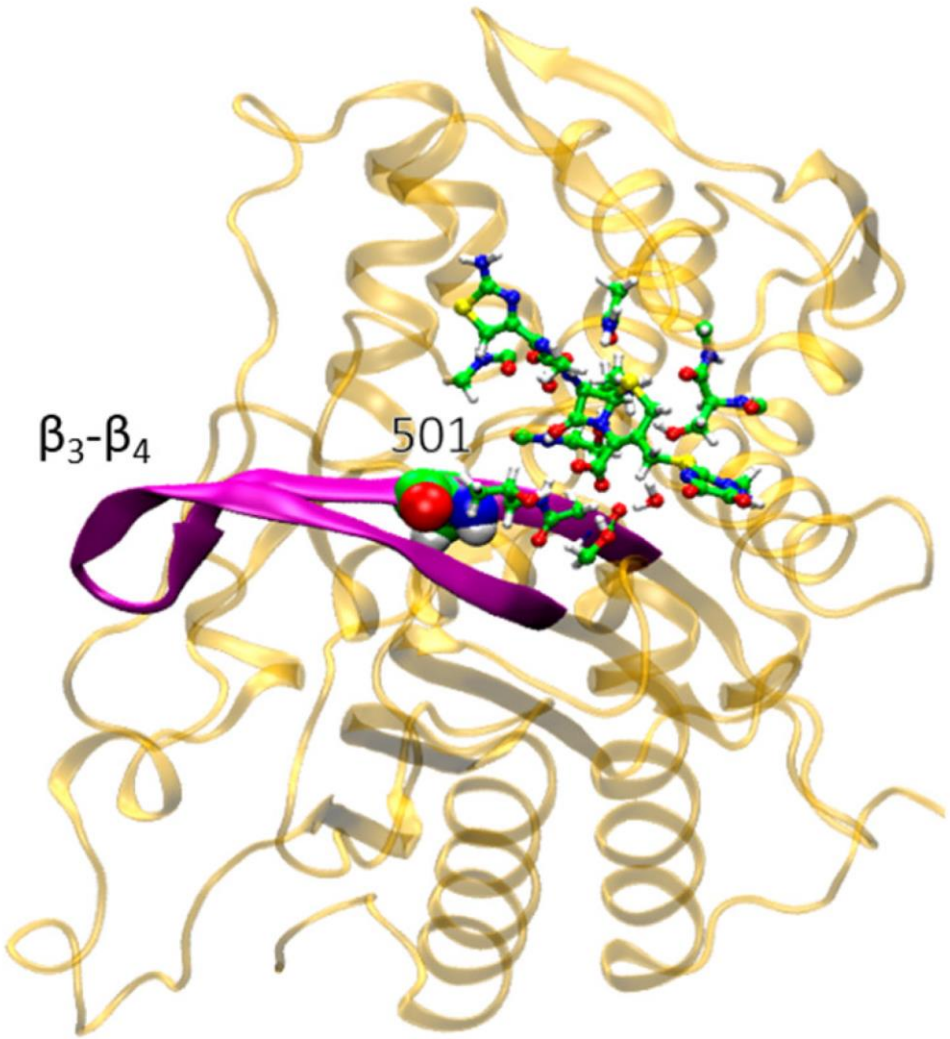


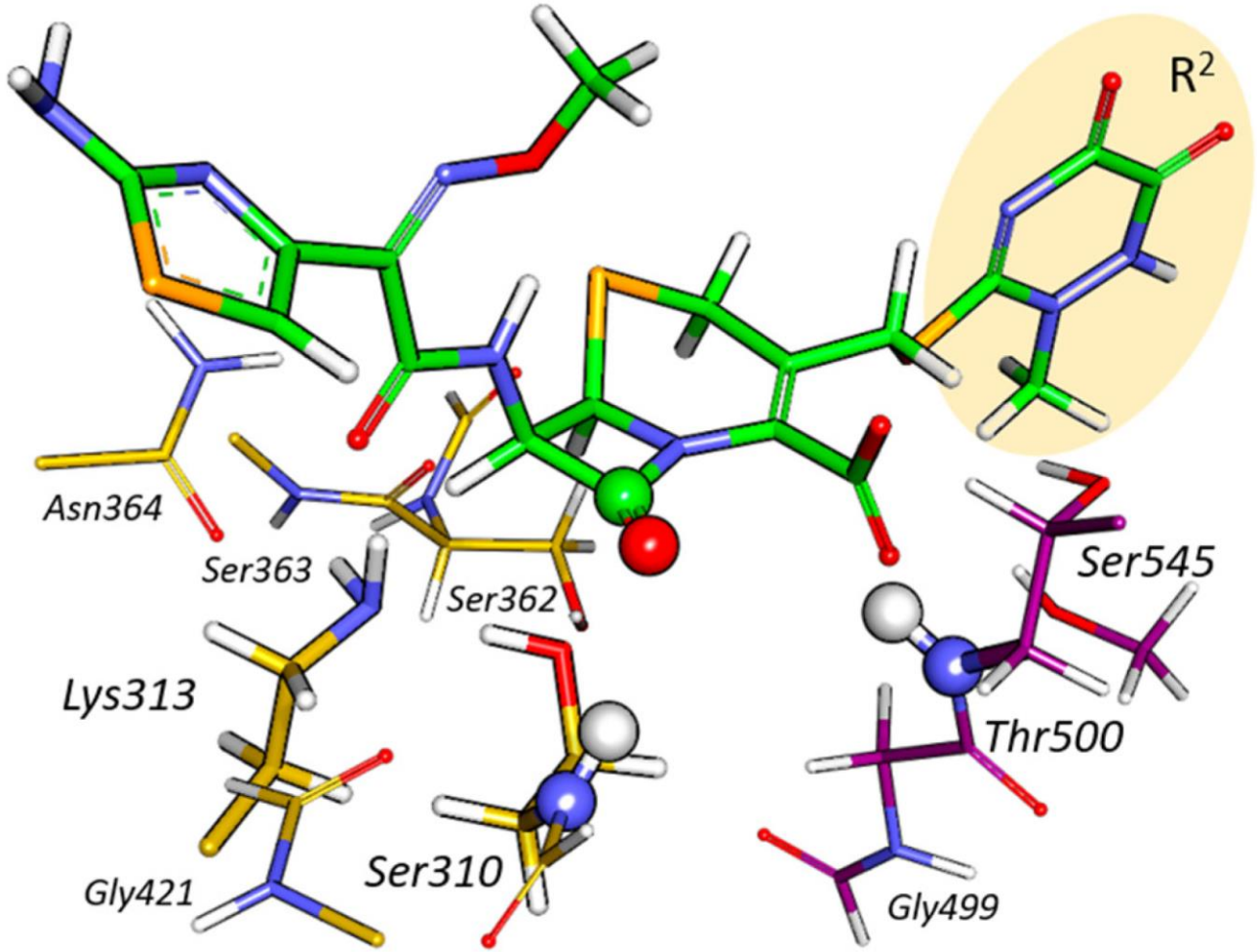


Dynamic behavior of penicillin binding protein PBP2 and its A501X variants

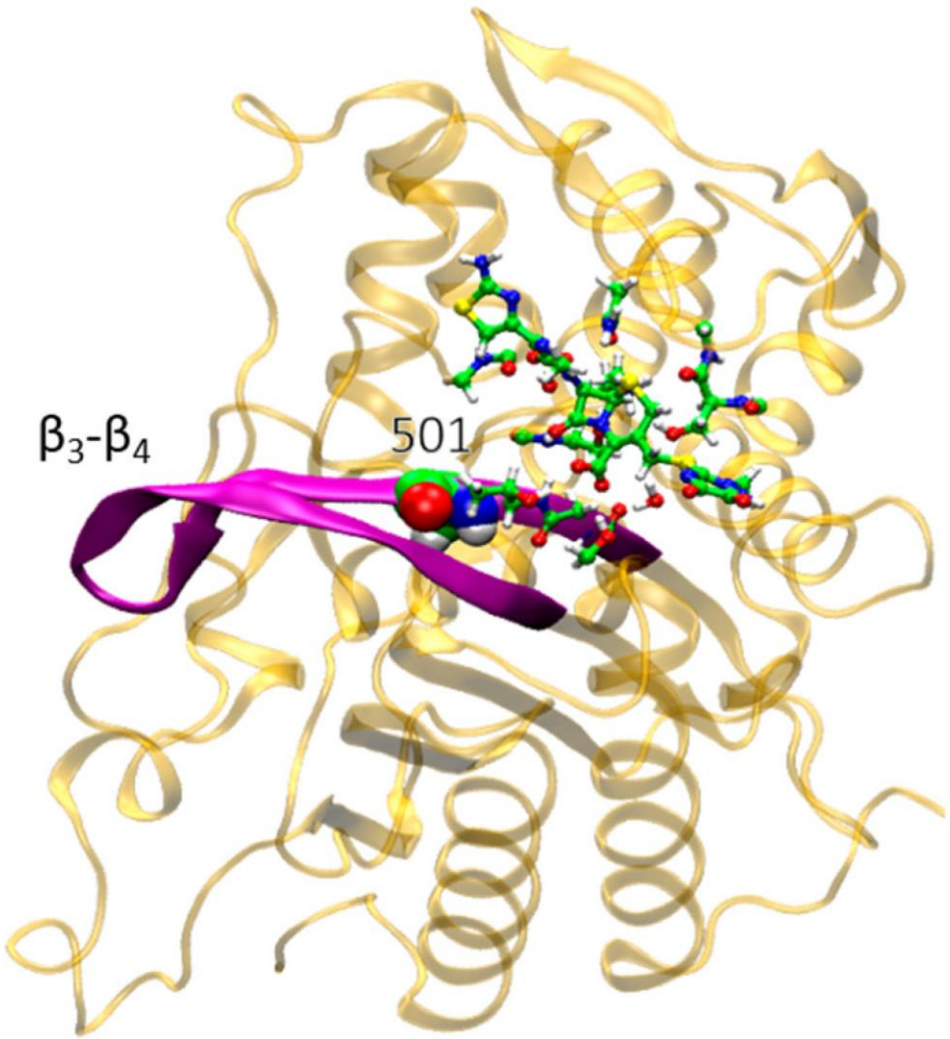


Structure of the PBP2





Penicillin binding protein PBP2



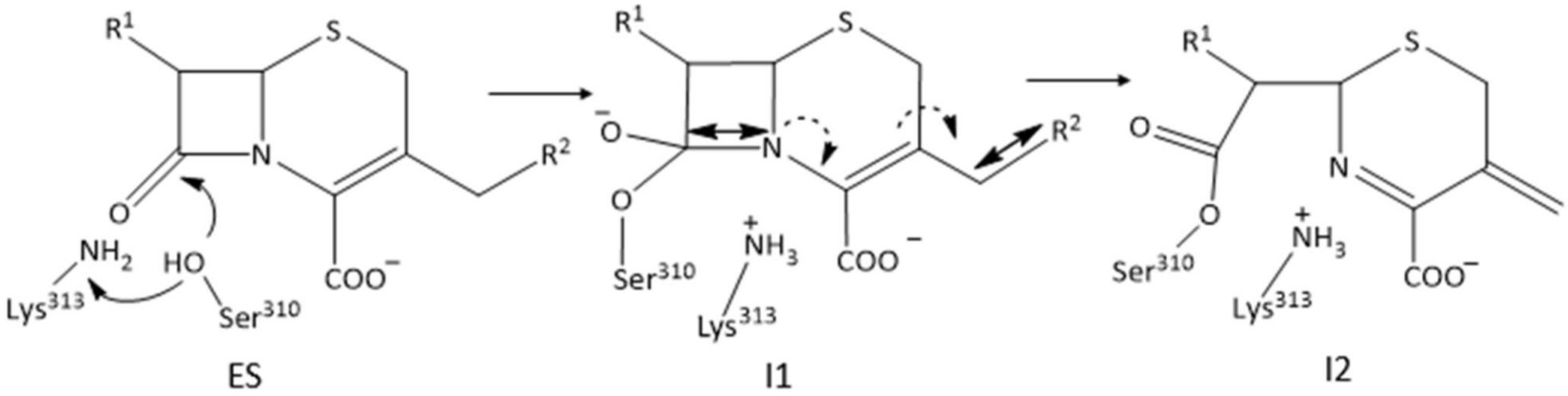
PBP2 with A501X variants demonstrate different catalytic activity *

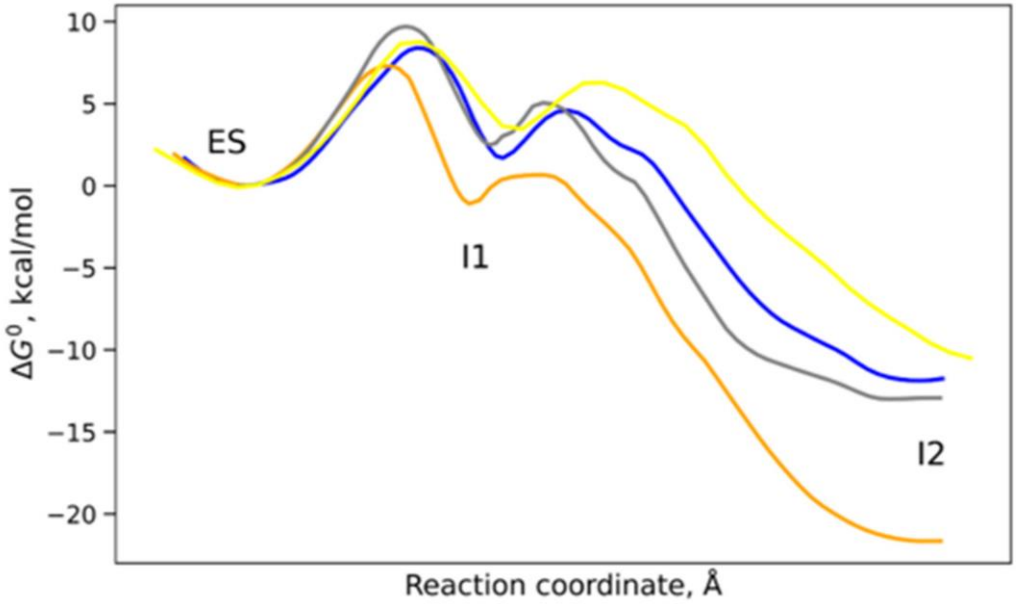
PBP2 variant	Relative k_2/K_s
A501V	1.8
A501	1
A501R	0.5
A501P	0.02

These variations can by due to changes of k_2 and/or K_s values.

* J. Tomberg et al., Biochemistry (2017)

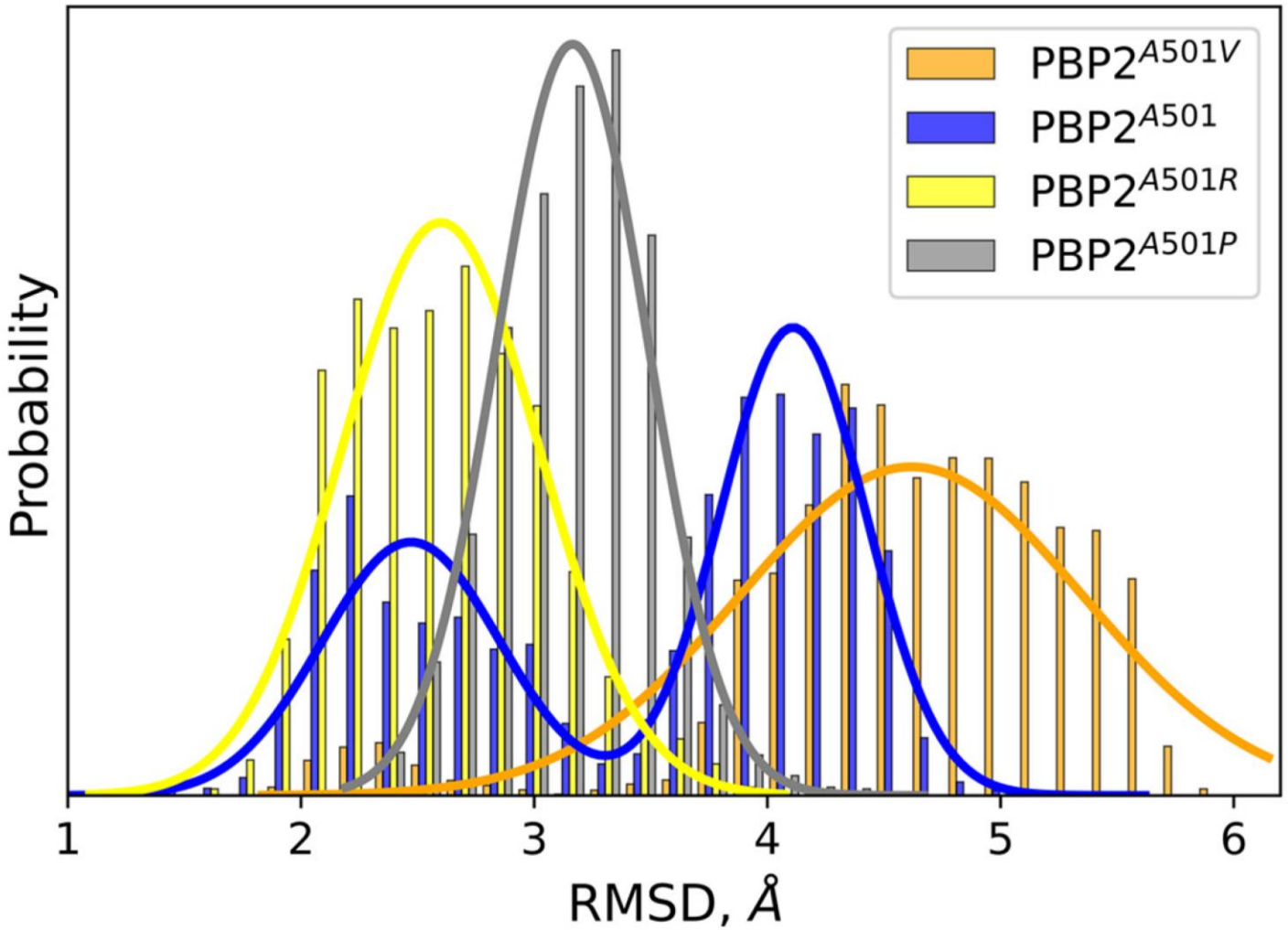
Acylation mechanism in the active site of the PBP2





QM(PBE0-D3/6-31G**)/MM MD

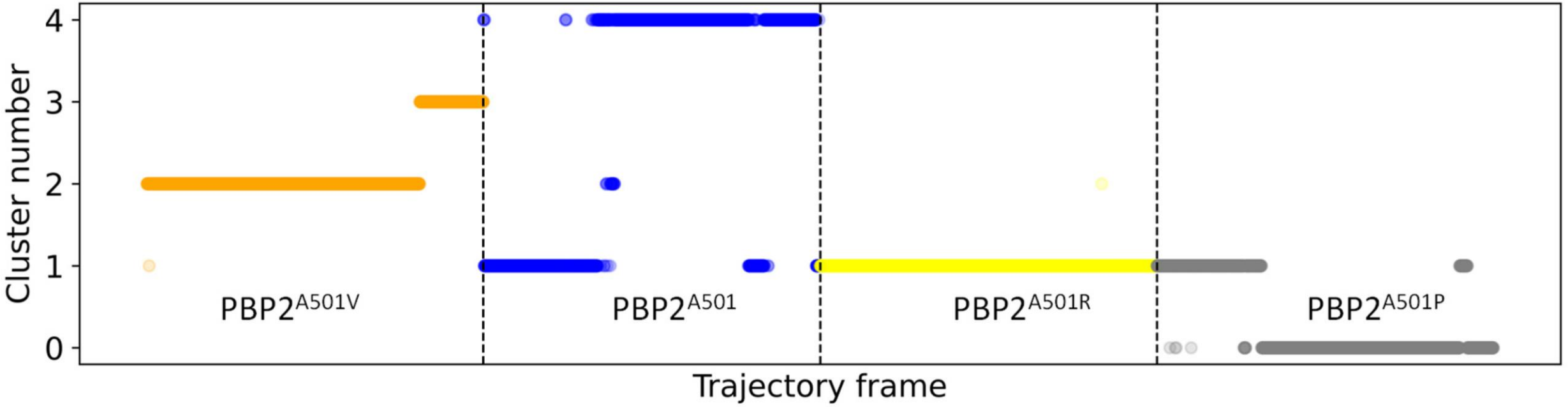
Dynamic behavior of the β_3-β_4 loop

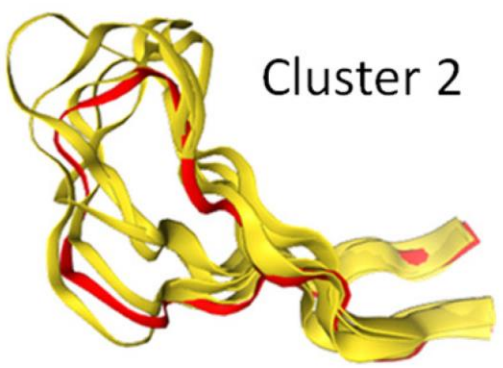


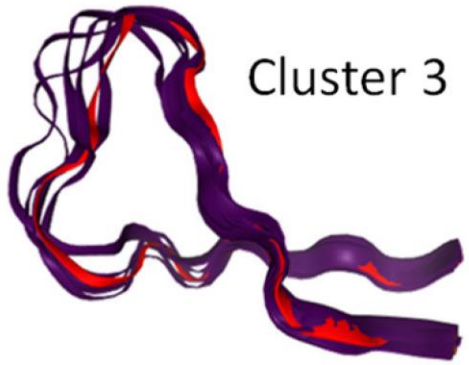
PBP2 variant	RMSD of β_3-β_4 loop mean ± st.dev., Å
$PBP2^{A501V}$	4.61 ± 0.54
$PBP2^{A501}$	4.11 ± 0.10 (w = 0.6) 2.47 ± 0.15 (w = 0.4)
$PBP2^{A501R}$	2.60 ± 0.18
$PBP2^{A501P}$	3.16 ± 0.10

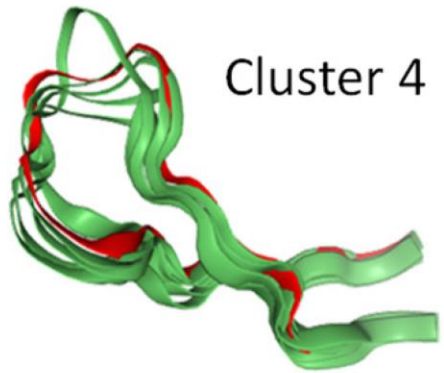
MD: 500ns, CHARMM + TIP3P

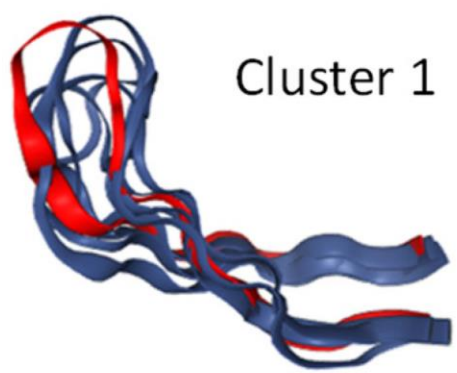
Clustering of β3–β4 loop states

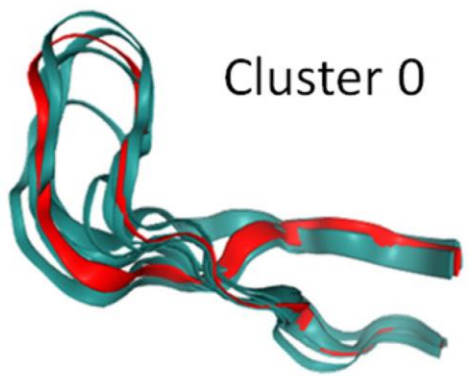






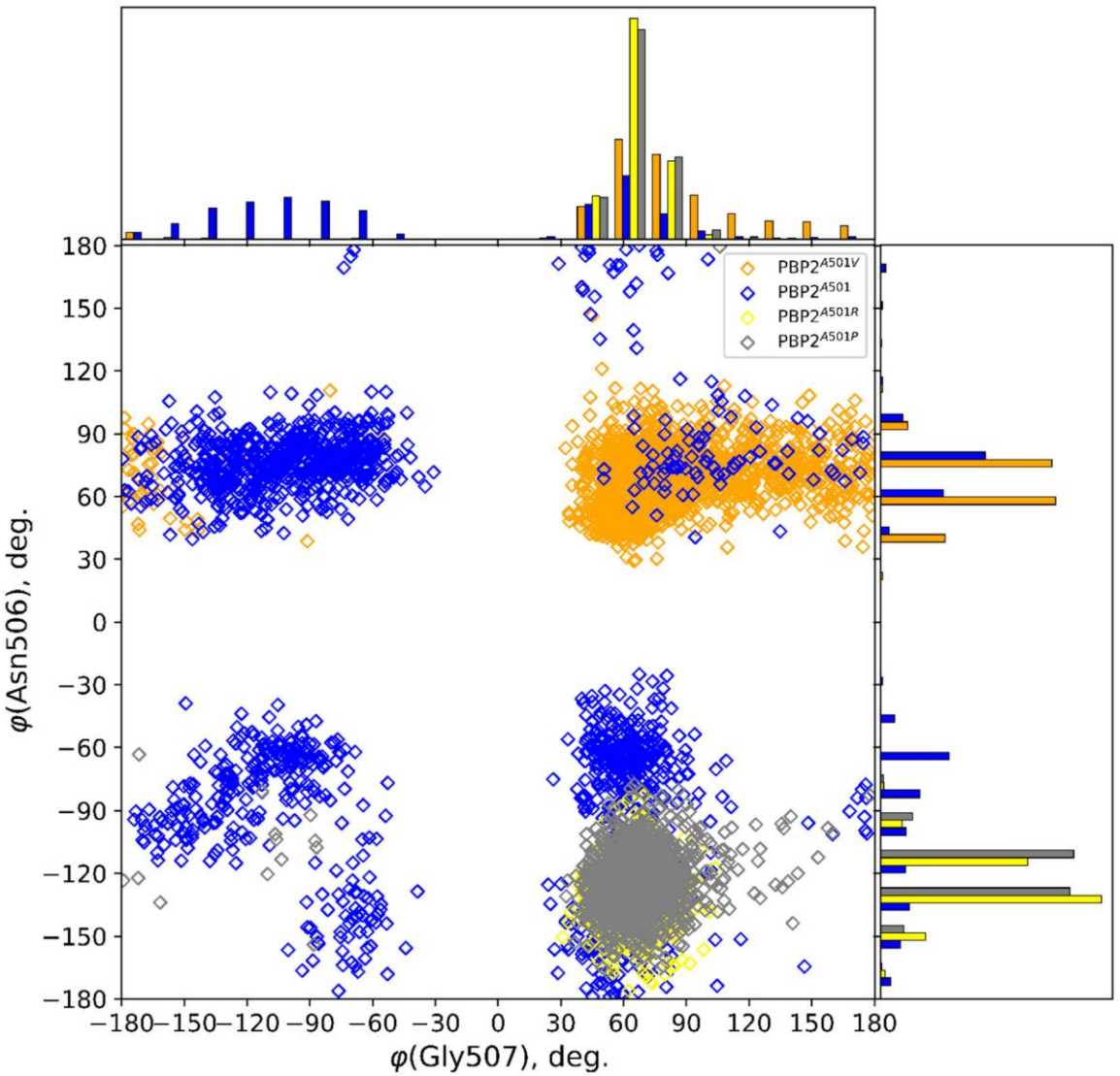


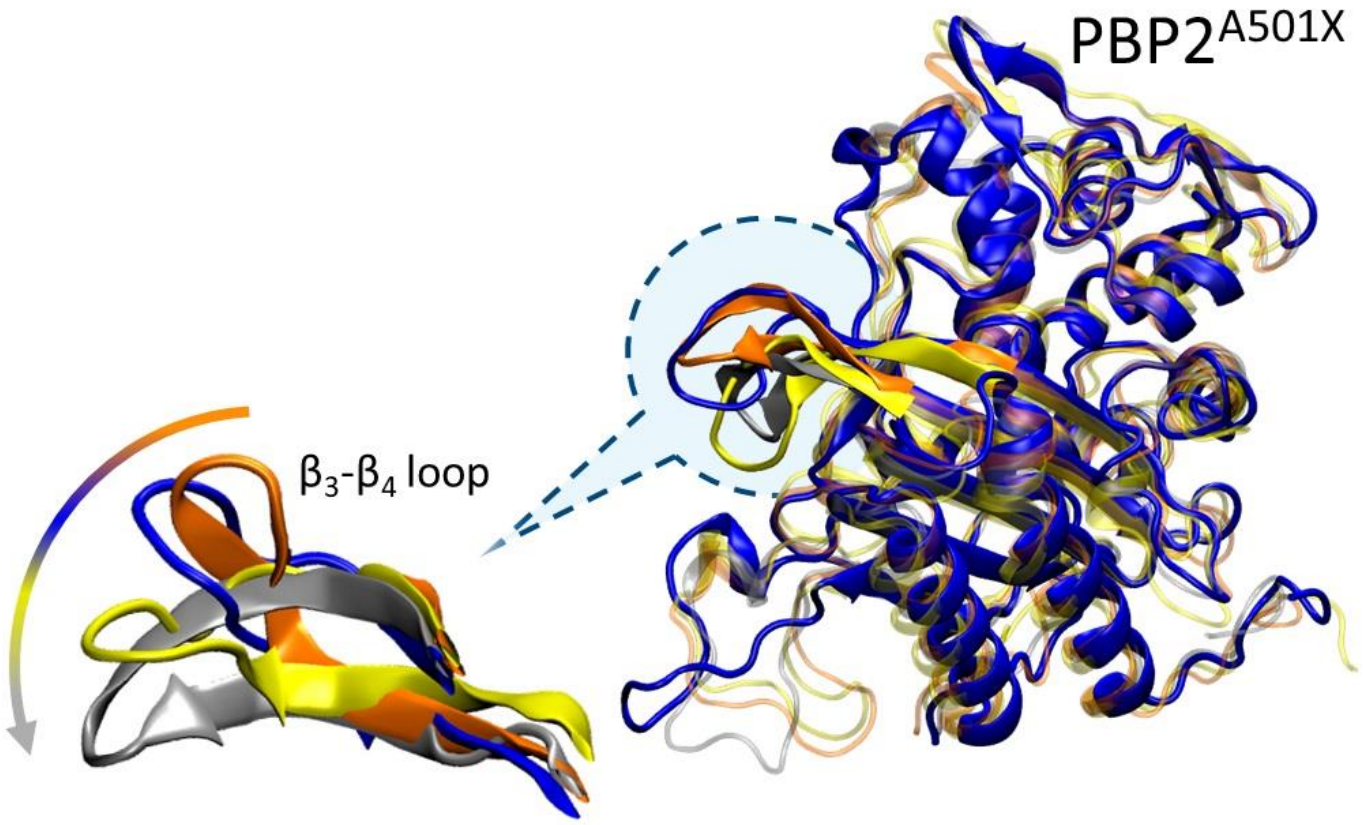




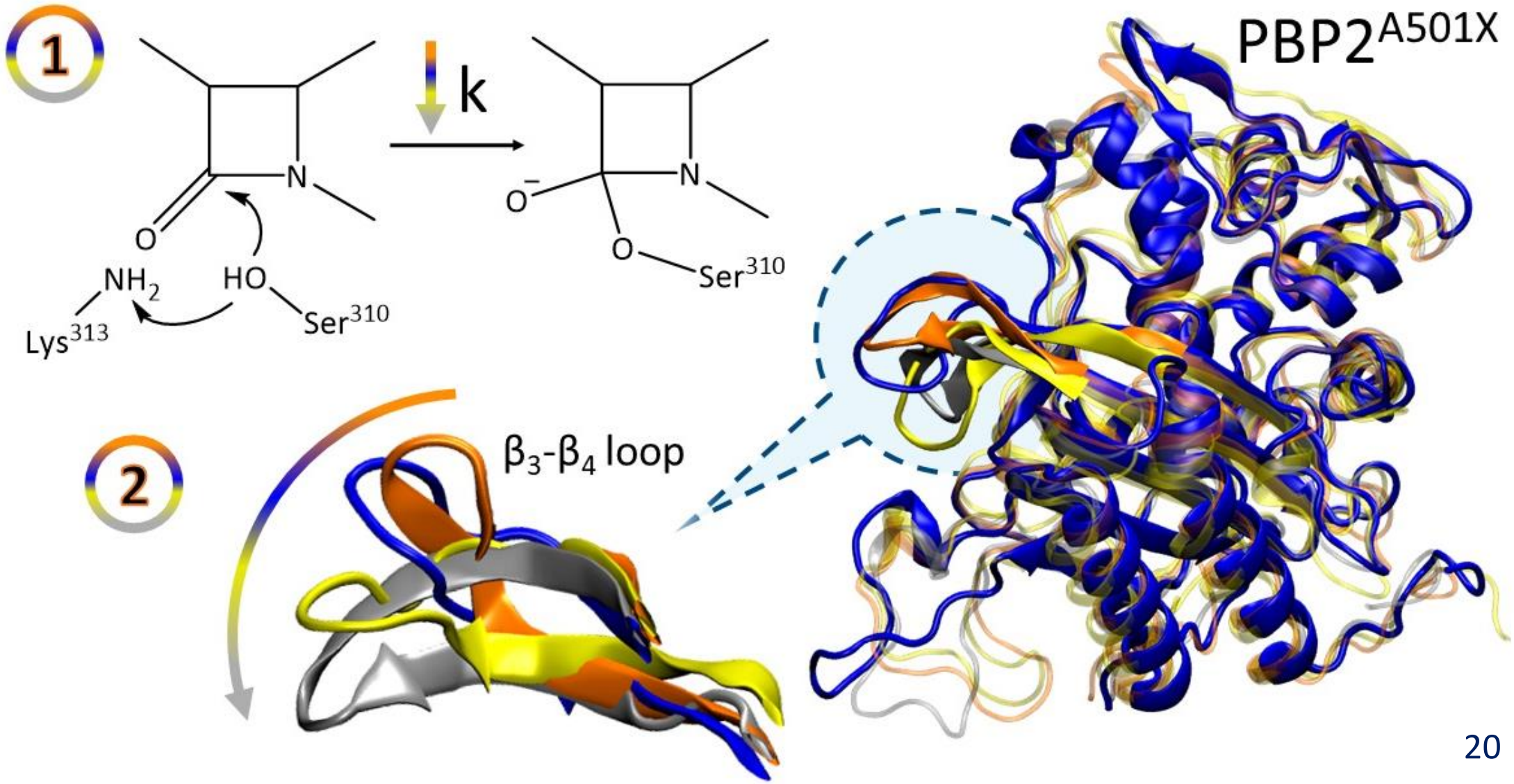
- 500 ns MD trajectories (NAMD software)
- Clustering over backbone dihedrals of the β3–β4 loop, 14 principal components (EnGens service)

Distribution of φ(Gly507) and φ(Asn506) over MD trajectories





Increase of ceftriaxone resistance is due to both changes of the acylation rate constant and binding constant



Any questions?

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