# Supporting information

## The catalytic reaction mechanism of tyrosylprotein sulfotransferase-1

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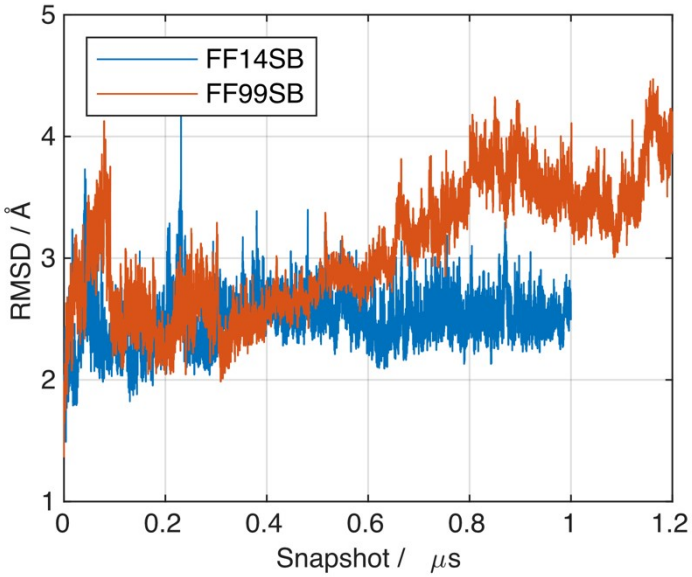


**Fig. S1** Development of the RMSD of protein non-hydrogen atoms towards the initial X-ray structure for the two MD simulations. Two different force fields were used for the description of the enzyme.

| Parameter | Distance / Å |
|---|---|
| a = $d_{OT\text{-}S}$ | 4.28 |
| b = $d_{GH}$ | 2.05 |
| c | 3.71 |
| d | 3.40 |
| e | 2.03 |
| f | 2.73 |
| g | 1.91 |
| h | 1.95 |
| i | 1.77 |
| j | 1.94 |
| k | 2.88 |
| l | 2.32 |
| m | 3.59 |
| n | 3.12 |
| o | 1.95 |
| p | 1.88 |

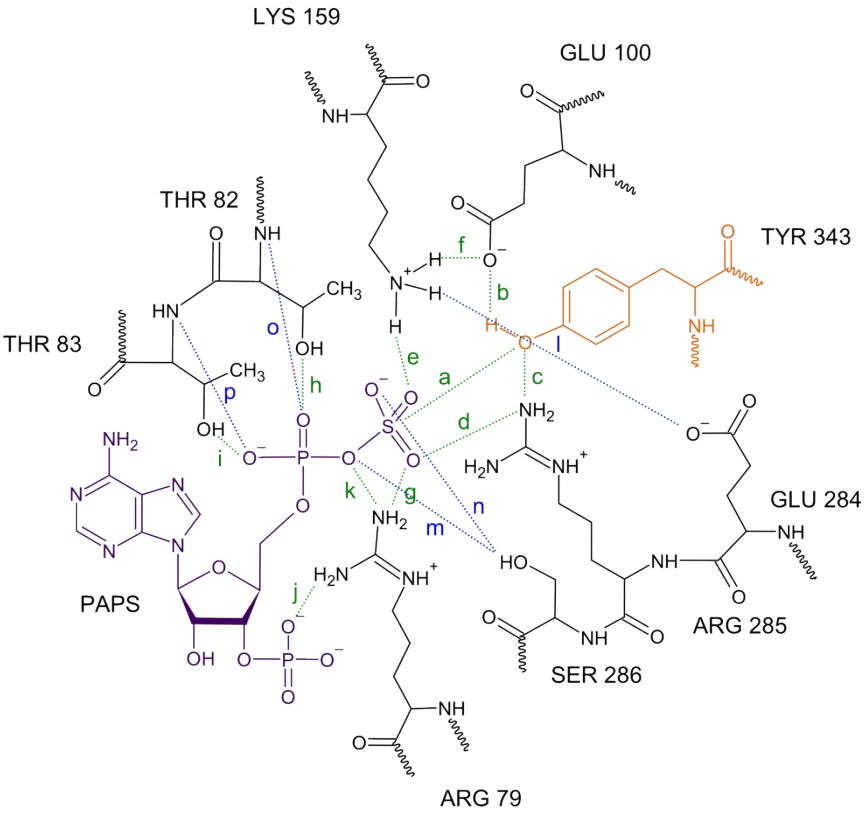


**Fig. S2** Mean values of selected geometrical parameters from MD simulation with FF14SB force field.

**Table S1** Distances (in Angstroms) between selected residues obtained from the metadynamics simulations and potential energy scan. PAP denotes the phosphate part of PAP, the smallest distances between the pairs of residues are provided.

| Distance / Å | CPMD Metadynamics | | | Optimized geometries | | |
|---|---|---|---|---|---|---|
| | MC | TS | P | MC | TS | P |
| $d_{OT-S}$ / Å | 3.6 | 2.5 | 1.9 | 3.5 | 2.2 | 1.9 |
| $d_{OP-S}$ / Å | 1.7 | 2.4 | 2.8 | 1.7 | 2.4 | 2.9 |
| $d_{T-H}$ / Å | 1.0 | 1.4 | 1.6 | 1.3 | 1.5 | 1.7 |
| $d_{G-H}$ / Å | 1.7 | 1.2 | 1.0 | 1.1 | 1.0 | 1.0 |
| PAP – ARG 79 | 2.56 | 2.49 | 1.88 | 2.42 | 2.24 | 1.42 |
| PAP – SER 286 | 3.07 | 3.49 | 4.09 | 1.72 | 1.50 | 1.59 |
| PAP – THR 82 | 2.86 | 1.83 | 1.99 | 2.07 | 1.81 | 1.85 |
| PAP – THR 83 | 1.71 | 1.46 | 1.53 | 2.12 | 1.80 | 1.86 |
| PAP – GLY 81 | 2.20 | 2.48 | 2.19 | 2.50 | 2.65 | 2.58 |
| PAP – SER 80 | 2.18 | 3.04 | 2.89 | 2.36 | 2.60 | 2.49 |
| Sulfate – SER 286 | 1.93 | 4.33 | 4.91 | 3.49 | 3.74 | 4.19 |
| Sulfate – THR 83 | 2.77 | 2.85 | 3.39 | 3.01 | 3.28 | 3.58 |
| Sulfate – THR 82 | 2.94 | 3.52 | 3.66 | 2.99 | 3.25 | 3.41 |
| Sulfate – LYS 159 | 2.49 | 2.60 | 2.69 | 4.57 | 4.44 | 4.60 |
| Sulfate – ARG 79 | 2.25 | 2.36 | 2.70 | 1.61 | 1.83 | 2.66 |
| Sulfate – ARG 285 | 2.11 | 2.28 | 1.95 | 3.48 | 2.70 | 2.61 |

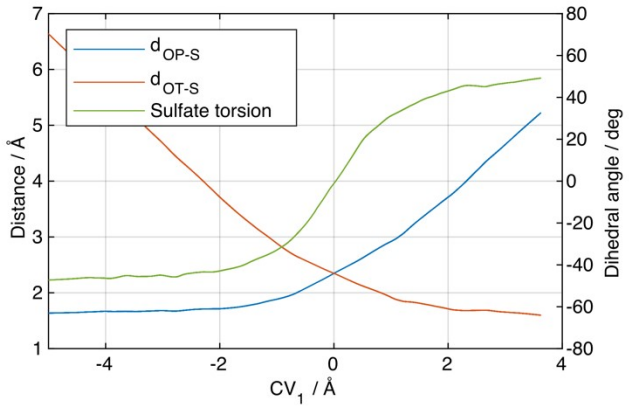


**Fig. S3** Dependence of distances $d_{OP\text{-}S}$ and $d_{OT\text{-}S}$, and the shape of the transferred sulfate (torsion of 0° indicates planarity of the group), on the collective variable $CV_1$. The data from the metadynamics simulation were smoothed using a moving average filter.

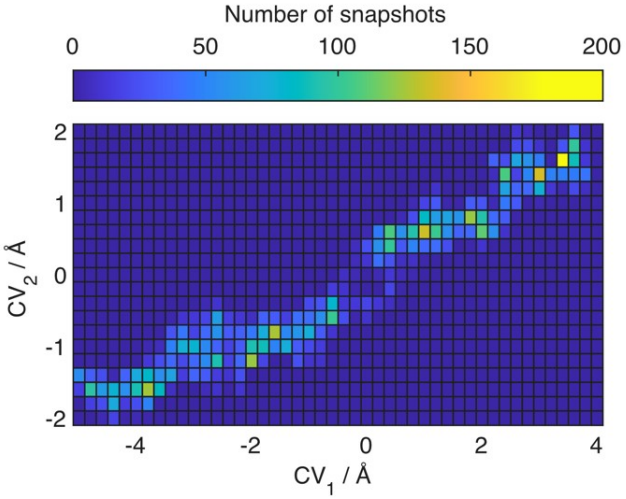


**Fig. S4** Probability of the different structures with respect to the collective variables $CV_1$ and $CV_2$ within the CPMD simulation. The local minima of free energy are implied by the maxima of probability in the graph. A snapshot was recorded for every 10 MD steps.

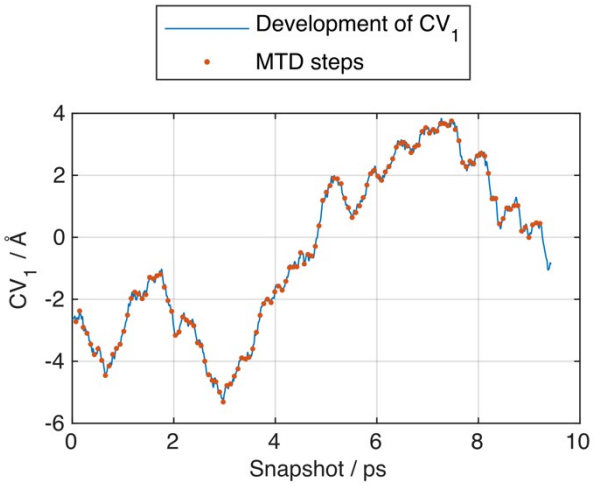


**Fig. S5** Development of the value of $CV_1$ within the metadynamics (MTD) simulation (blue line) and positions of added metadynamics potentials (orange dots). After adding the last shown metadynamics potential, the walker crossed the reaction barrier to the side of the MC.

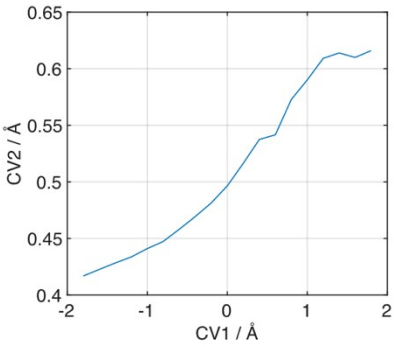


**Fig. S6** The development of the value of CV2 within the potential energy scan along CV1.

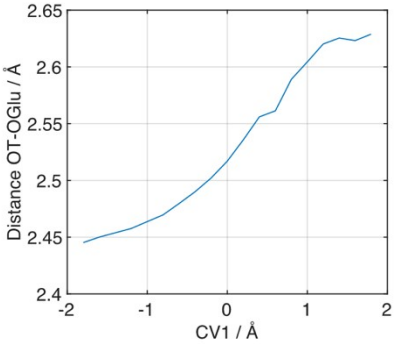


**Fig. S7** The development of the distance between the hydroxyl oxygen of TYR 343 (acceptor) and the proximal oxygen of the carboxyl group of GLU 100 (base) within the potential energy scan along CV1.

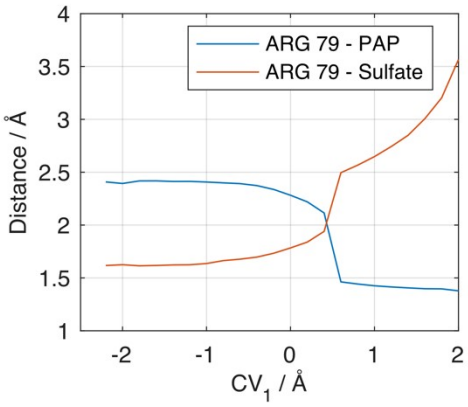


**Fig. S8** Distance between ARG 79 and transferred sulfate residue within the potential energy scan.

The schemes were created with the use of Marvin.[68] Trajectories were processed with the use of cpptraj (Amber Tools),[69] and the visualization created with VMD.[53]