Supporting Information for

Dynamic of non-covalent interactions during the P–O bond cleavage by ribonuclease A

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Electrostatic potential distribution on the solvent accessible surface (SAS) of ribonuclease A was obtained using PDB2PQR и APBS using the free web service (Figure S1).[1] We chose a standard solvent radius 1.4 Å for water molecule for SAS generation. Atomic parameters were generated using AMBER force field implemented in APBS. Enzyme protonation state was assigned to pH = 7.0 using empirical algorithm PROPKA.[2,3] Protein geometry was preliminarily equilibrated from X-ray structure mentioned in the main text by molecular mechanics technique using GROMACS[4,5] v. 2018.2 software program package and ff19SB[6] AMBER force field. PyMOL was used for visualization purpose. Blue areas on the ESP distribution correspond to positively charged protein space in the vicinity of the active site. It stands to reason that negatively charged nucleic acids has a long-range attraction to the active site of the enzyme.

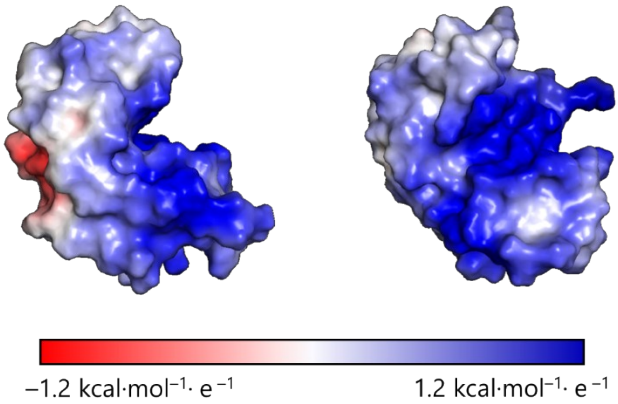

Figure S1. Electrostatic potential distribution on the solvent accessible surface of RNase A.

References

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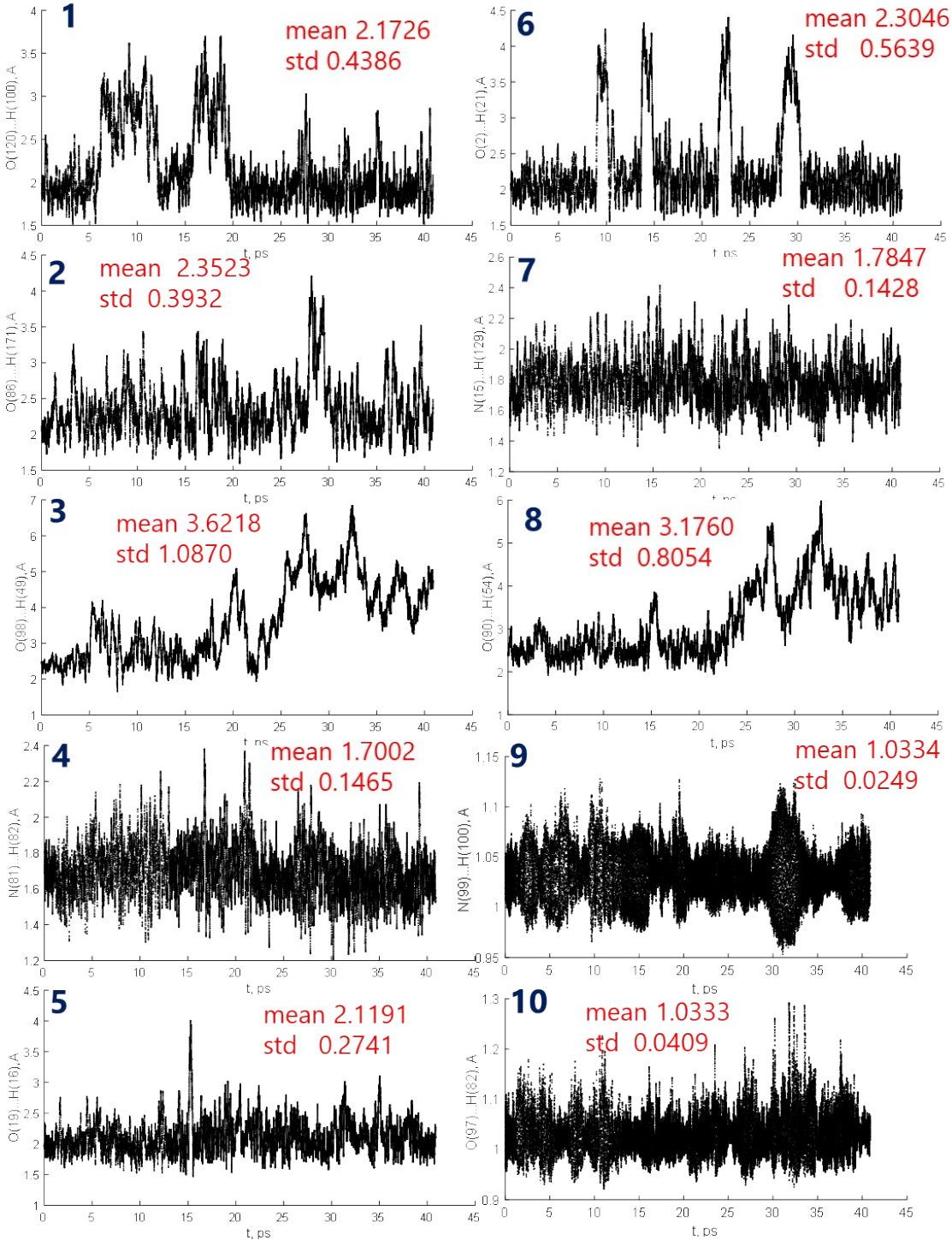
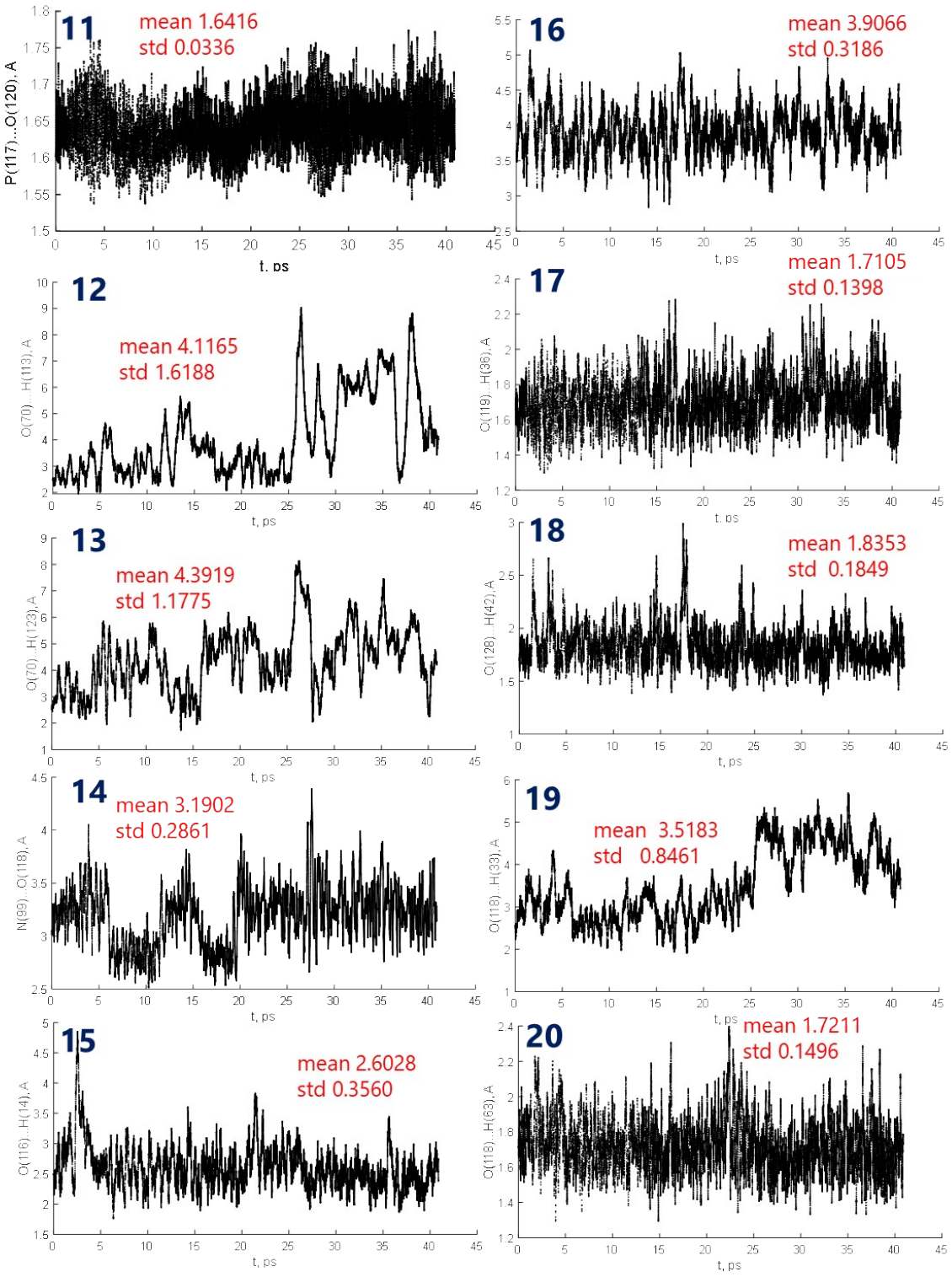
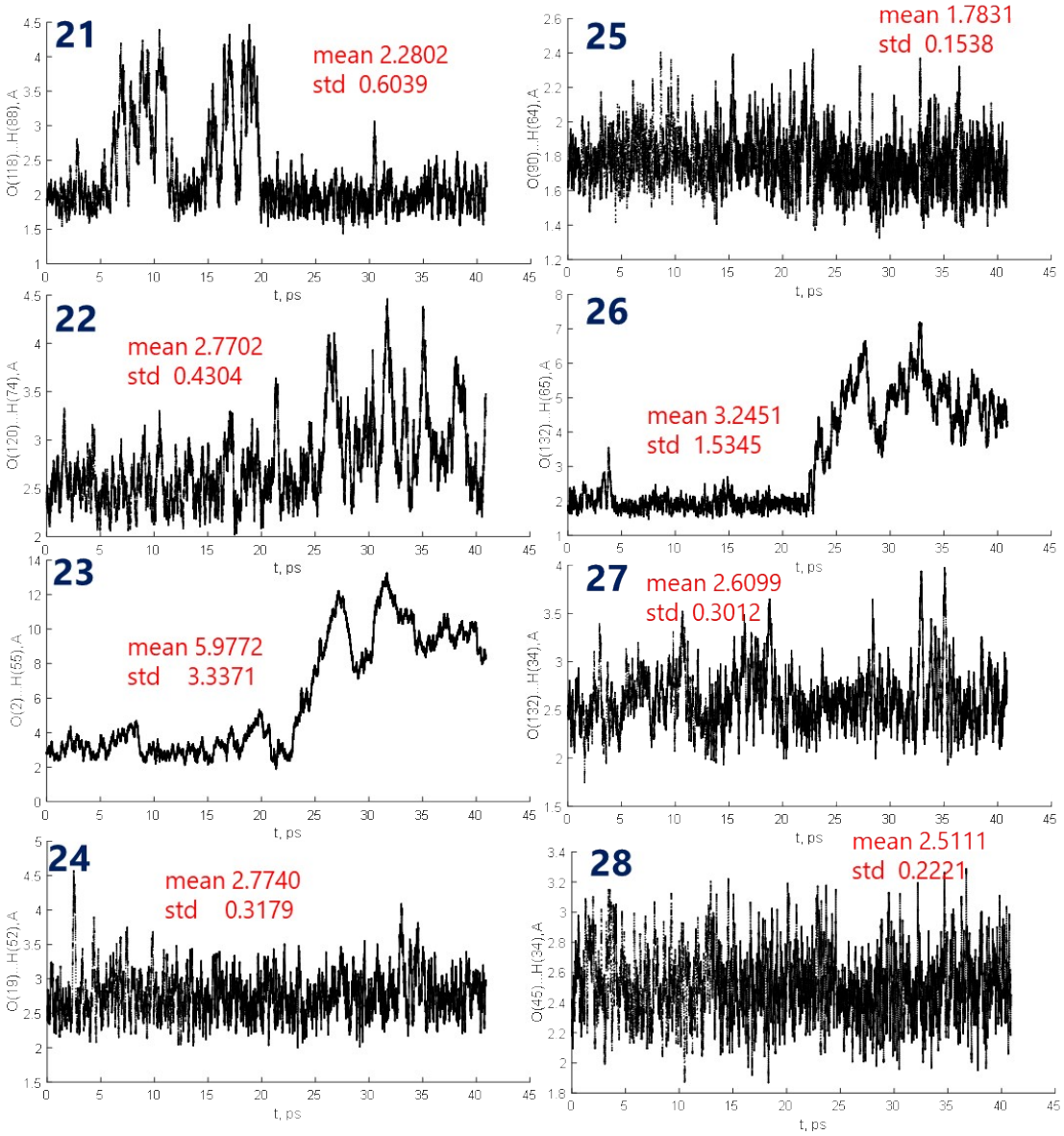
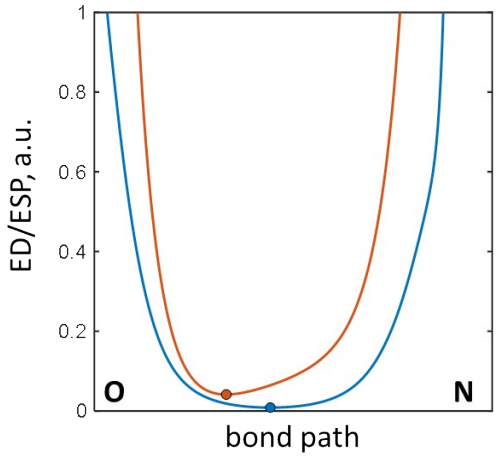
Figure S2. Time evolution of short contacts presented in Table 1 of the main text. Mean values and standard deviations are given in red, all values in Å.




11
mean 1.6416
std 0.0336
P(117)...O(120), A
t, ps
16
mean 3.9066
std 0.3186
t, ps
12
mean 4.1165
std 1.6188
O(70)...H(113), A
t, ps
17
mean 1.7105
std 0.1398
O(119)...H(36), A
t, ps
13
mean 4.3919
std 1.1775
O(70)...H(123), A
t, ps
18
mean 1.8353
std 0.1849
O(128)...H(42), A
t, ps
14
mean 3.1902
std 0.2861
N(99)...O(118), A
t, ps
19
mean 3.5183
std 0.8461
O(118)...H(33), A
t, ps
15
mean 2.6028
std 0.3560
O(116)...H(14), A
t, ps
20
mean 1.7211
std 0.1496
O(118)...H(63), A
t, ps

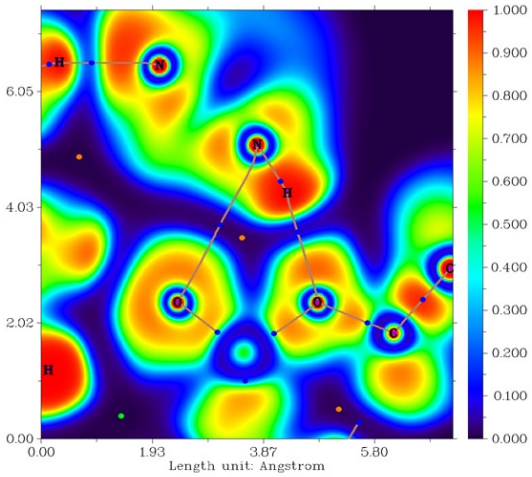

21
mean 2.2802
std 0.6039
O(116)...H(88), A
t, ps
25
mean 1.7831
std 0.1538
O(90)...H(64), A
t, ps
22
mean 2.7702
std 0.4304
O(120)...H(74), A
t, ps
26
mean 3.2451
std 1.5345
O(132)...H(65), A
t, ps
23
mean 5.9772
std 3.3371
O(2)...H(55), A
t, ps
27
mean 2.6099
std 0.3012
O(132)...H(34), A
t, ps
24
mean 2.7740
std 0.3179
O(19)...H(52), A
t, ps
28
mean 2.5111
std 0.2221
O(45)...H(34), A
t, ps

For "short contact **21** P=O^{118}···N^{99} the phylicity of the interacting partners was checked. The latter was done using the criterion proposed in [10.1107/S2052520618018280; 10.3390/molecules27154848]. The main idea of this criterion is that the relative location of the positions of the electron density (ED) and electrostatic potential (ESP) minima along the bond path determines the nucleophile and electrophile of the interacting partners. In particular, the minimum of ESP along the bond path is located closer to the atom that donates electrons, whereas the minimum of ED is located closer to the atom that delivers its electrophilic site for non-covalent bonding. Regarding P=O^{118}···N^{99} short contact, ED and ESP distribution along the bond path looks as follows:



The ESP curve (red) minimum is located closer to O atom, while ED curve (blue) – to N atom. Therefore, oxygen atom in O···N interaction is a nucleophile, while nitrogen – electrophile. Which make a reason to conclude that O···N interaction is a pnictogen bond.

To independently confirm this conclusion, we analyzed the distribution of the electron localization function in the plane of the NH-group of His119 and O118 phosphate moiety:



It can be seen that the bond path (shown in brown) for O···N interaction passes through the electron-depleted region of the electron shell of the N atom and the electron-rich region around the O atom.

Figure S3. Electronic energy along moderedundant scan.

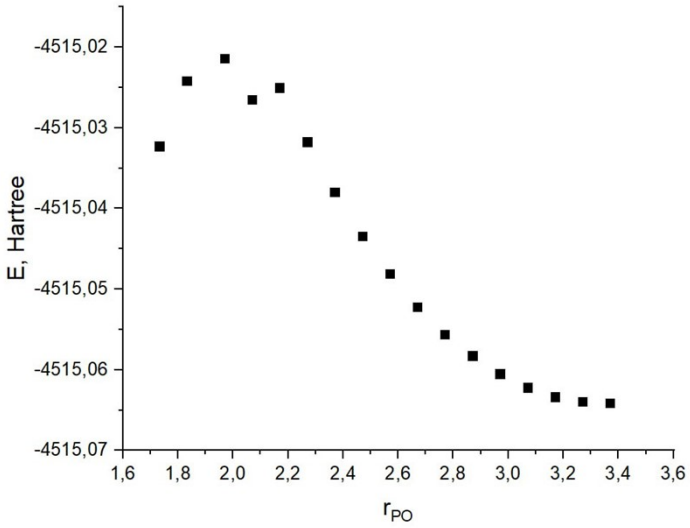
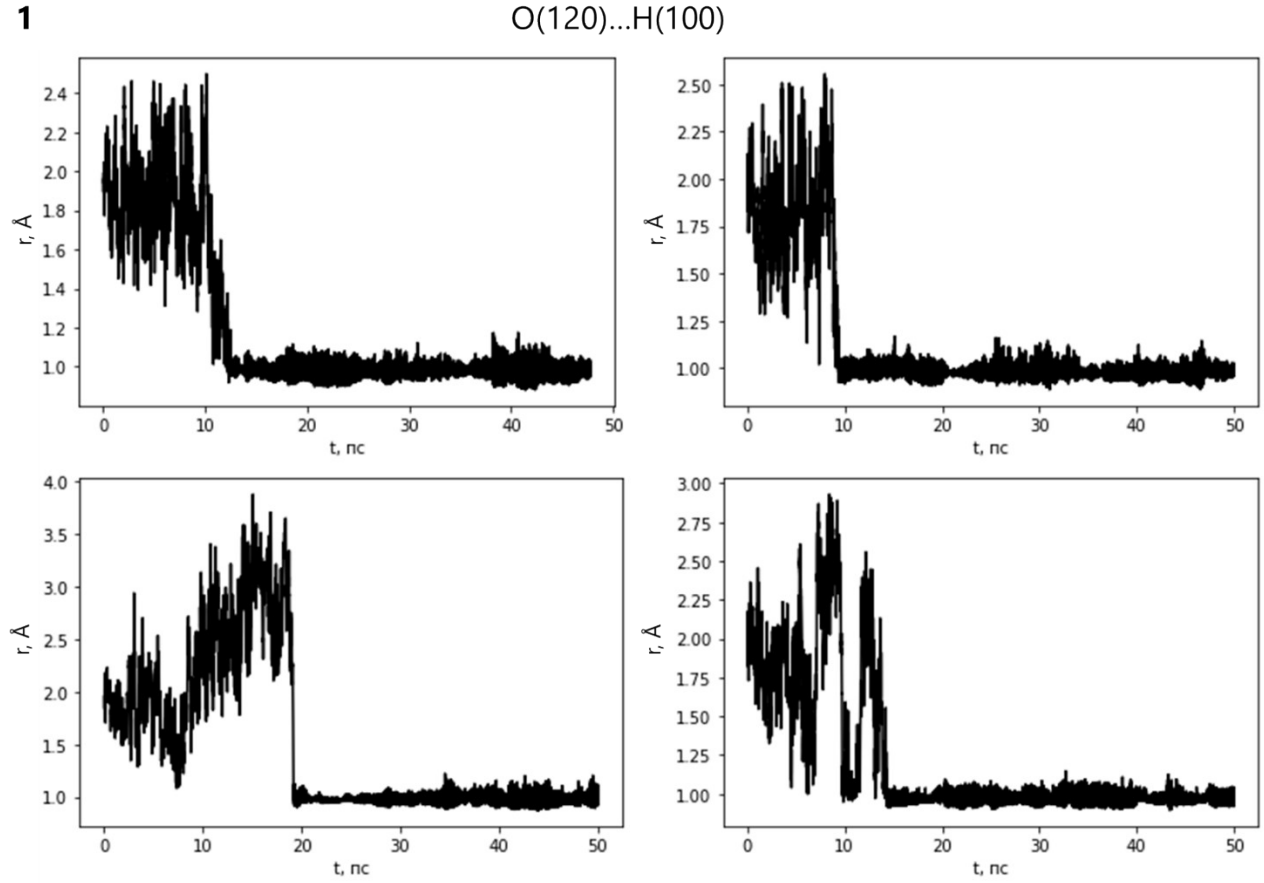
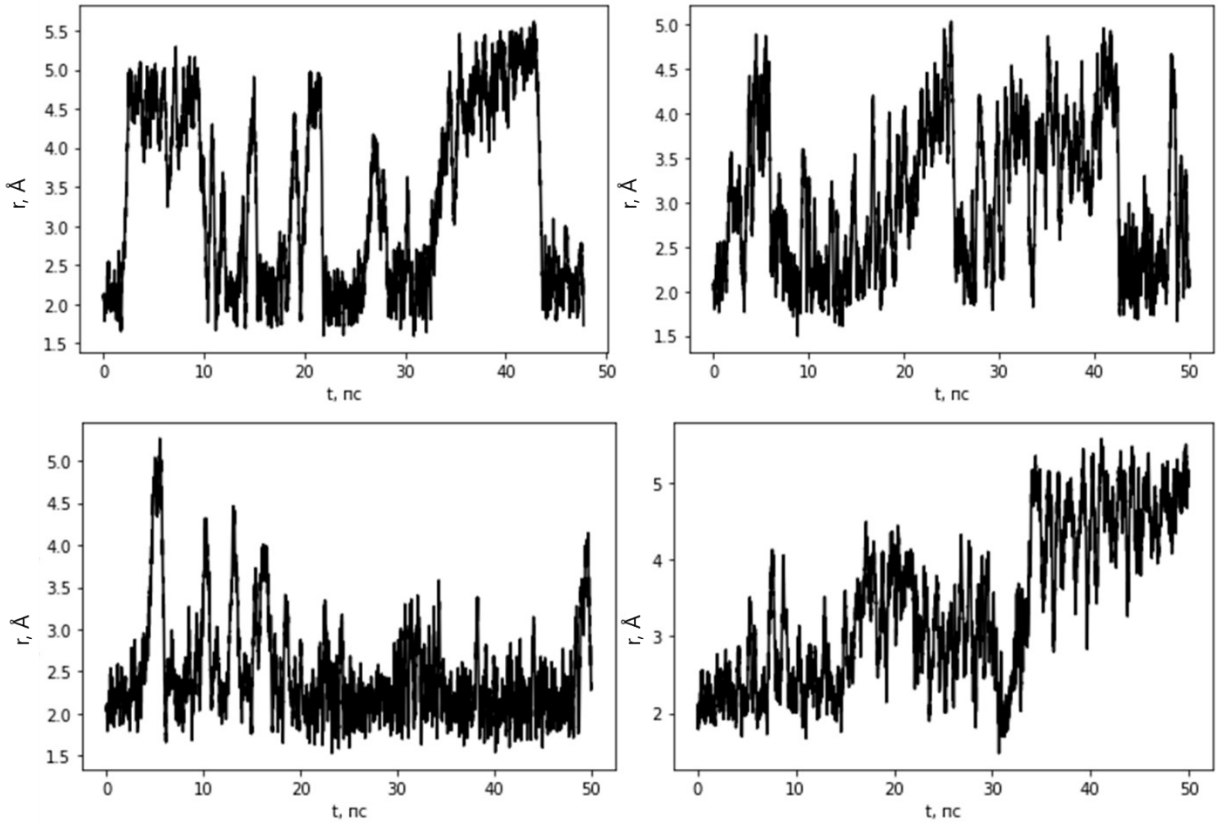


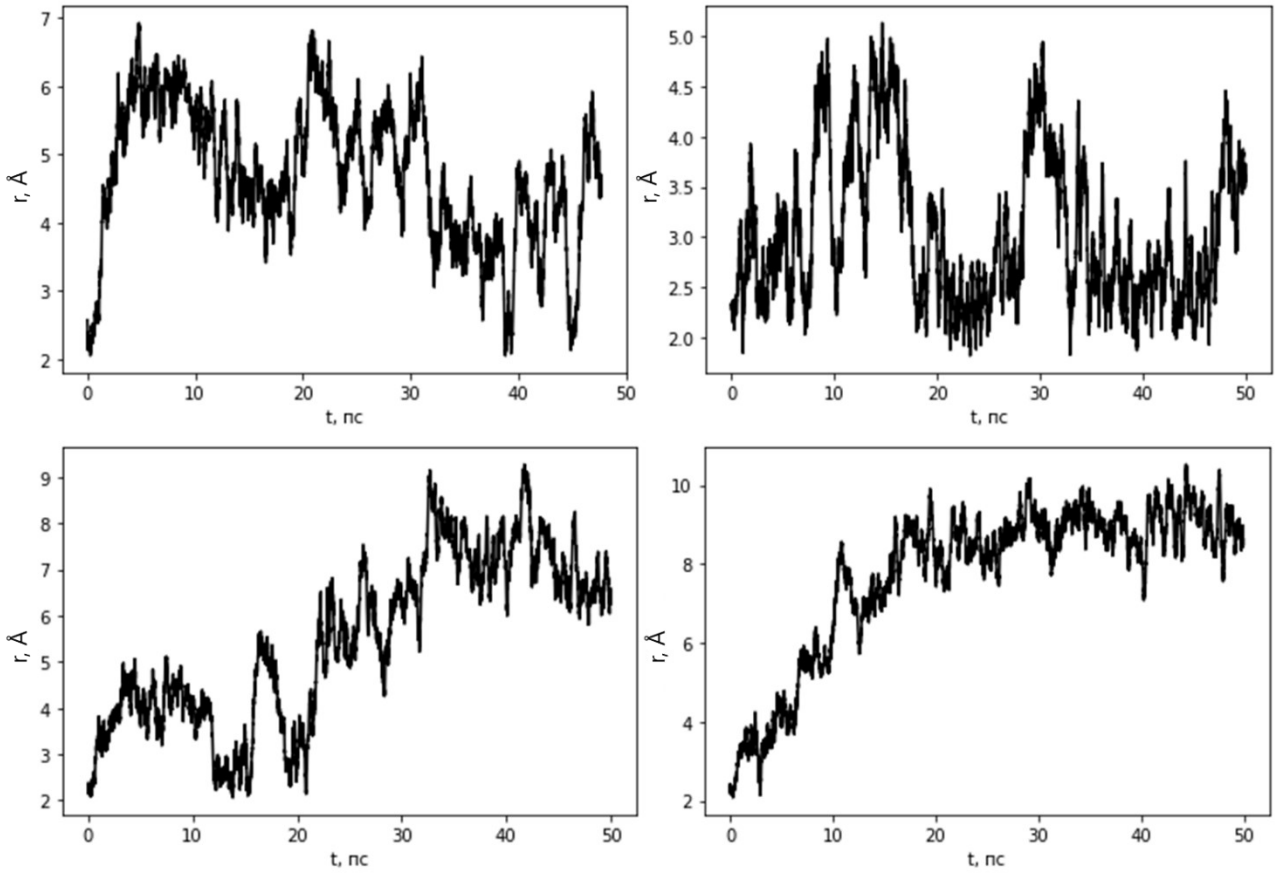
Figure S4. Time evolution of short contacts simulations presented in Table 1 of the main text in metadynamics simulations for successful trajectories.



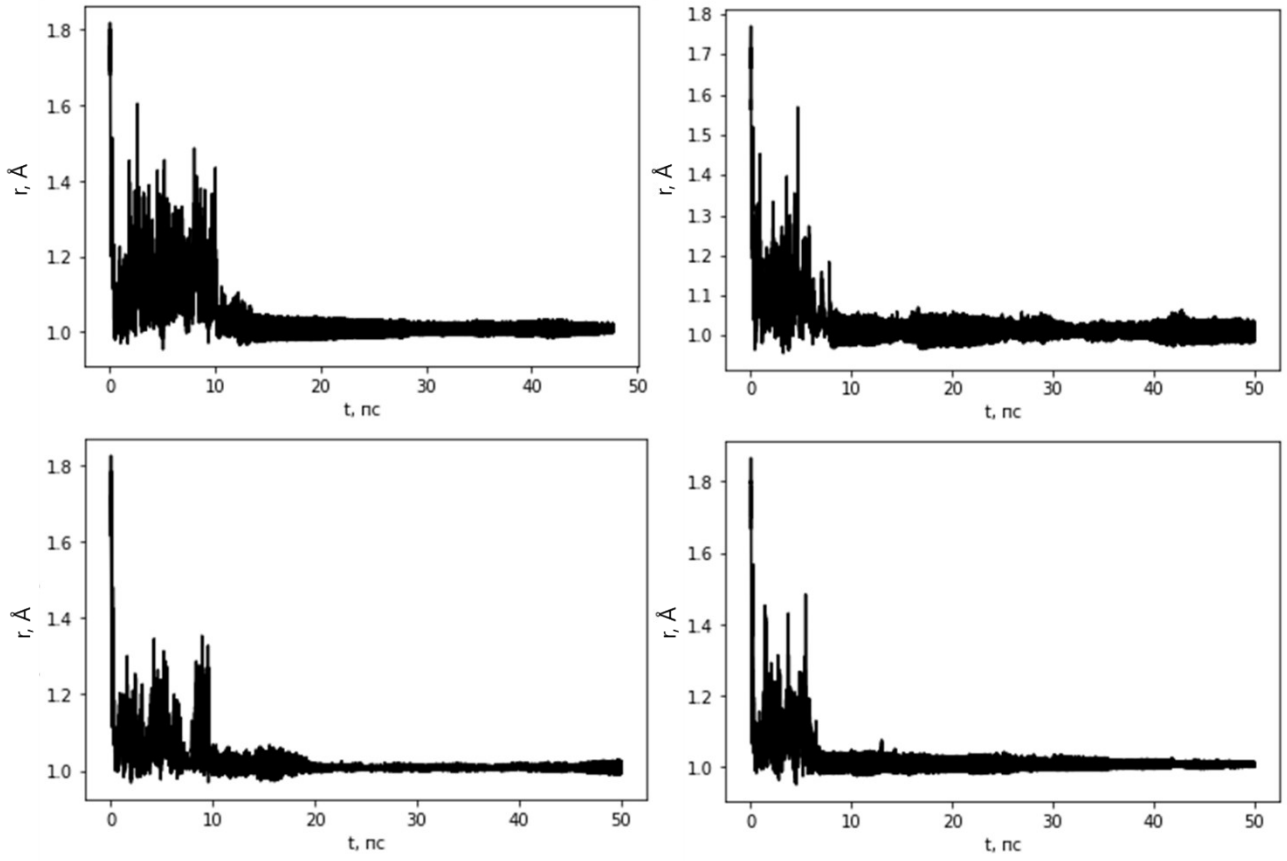
2 O(86)...H(171)



3 O(98)...H(49)



4 N(81)…H(82)

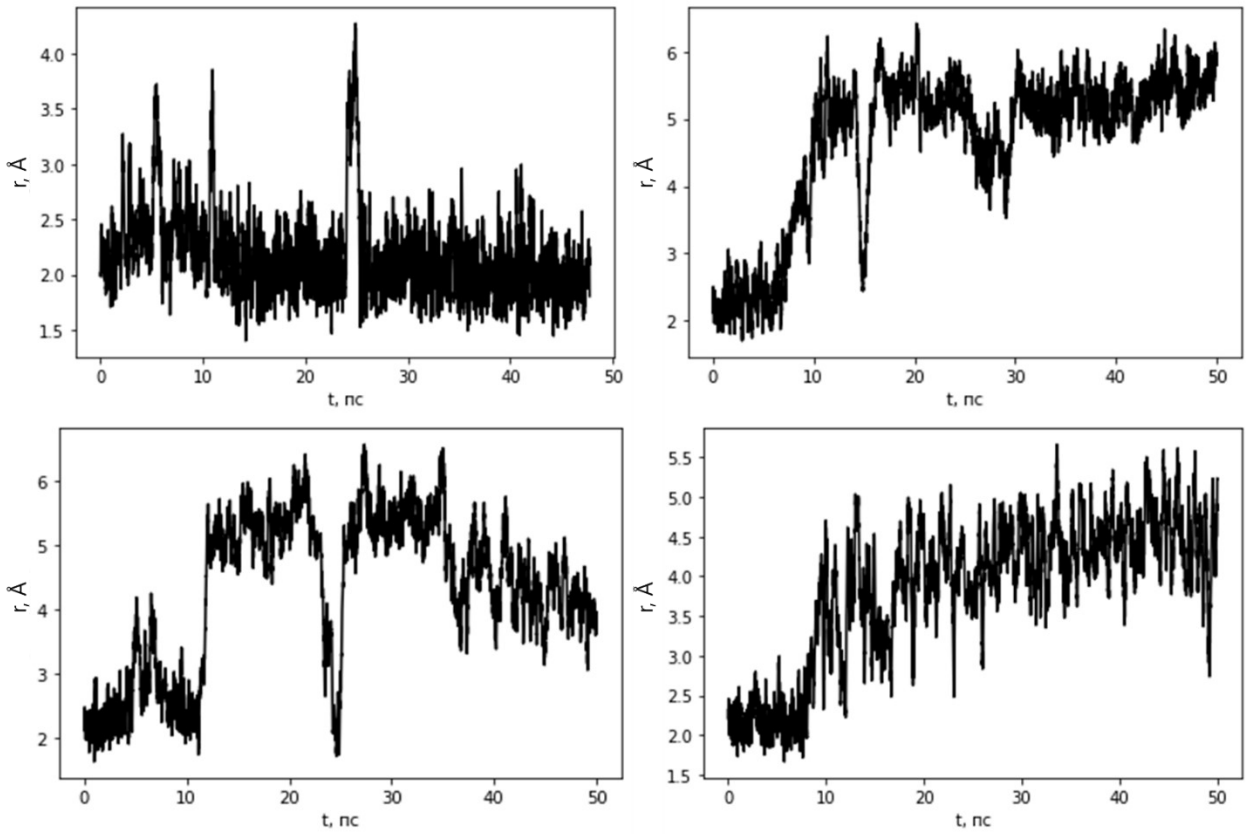

r, Å
t, nc
1.8
1.6
1.4
1.2
1.0
0
10
20
30
40
50

r, Å
t, nc
1.8
1.7
1.6
1.5
1.4
1.3
1.2
1.1
1.0
0
10
20
30
40
50

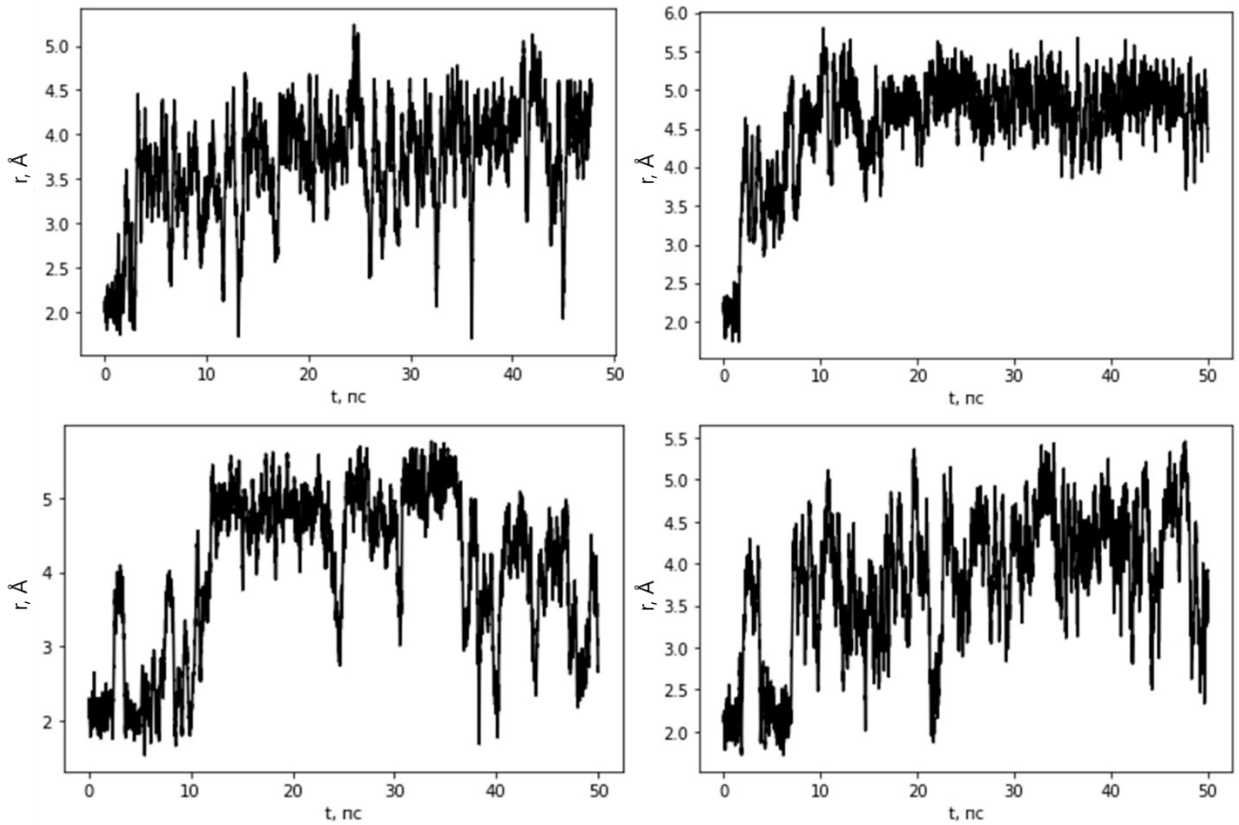
r, Å
t, nc
1.8
1.6
1.4
1.2
1.0
0
10
20
30
40
50

r, Å
t, nc
1.8
1.6
1.4
1.2
1.0
0
10
20
30
40
50

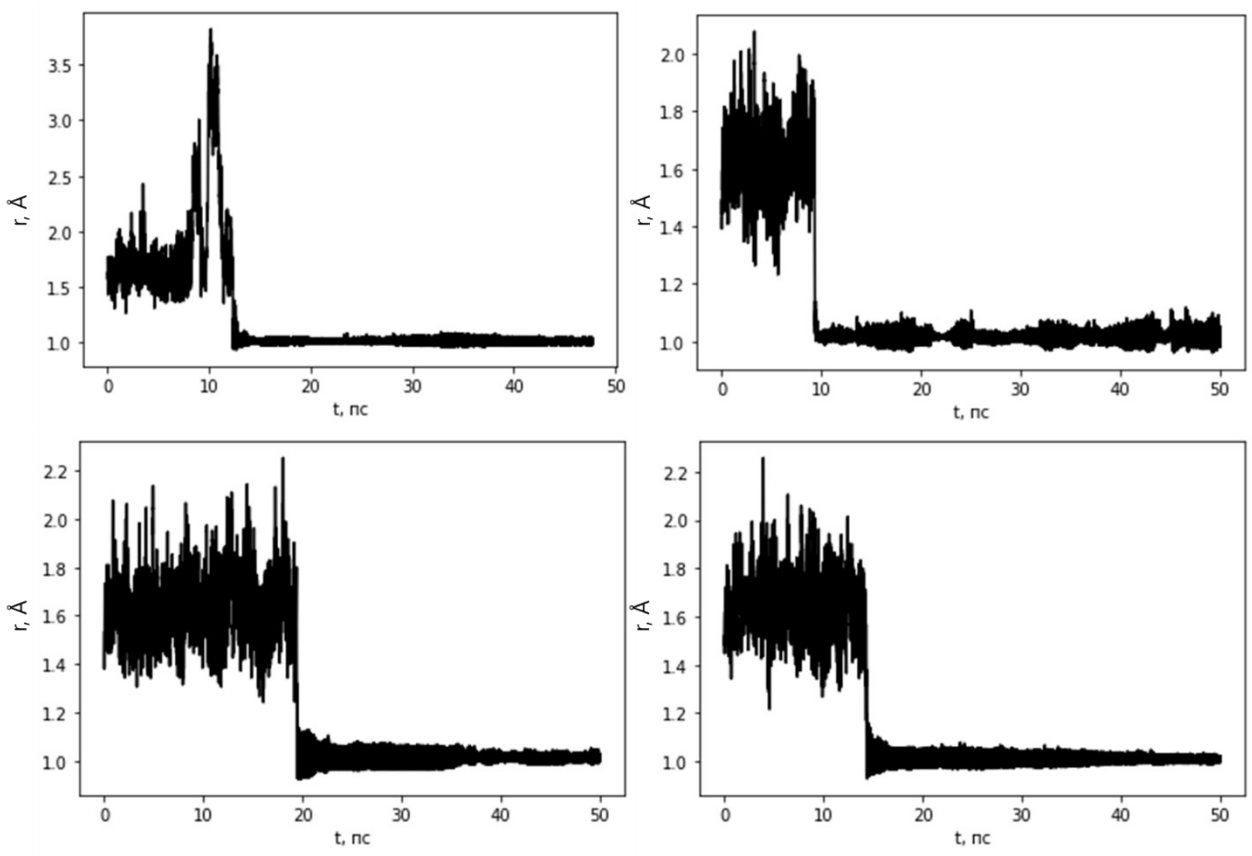
5 O(19)…H(12)



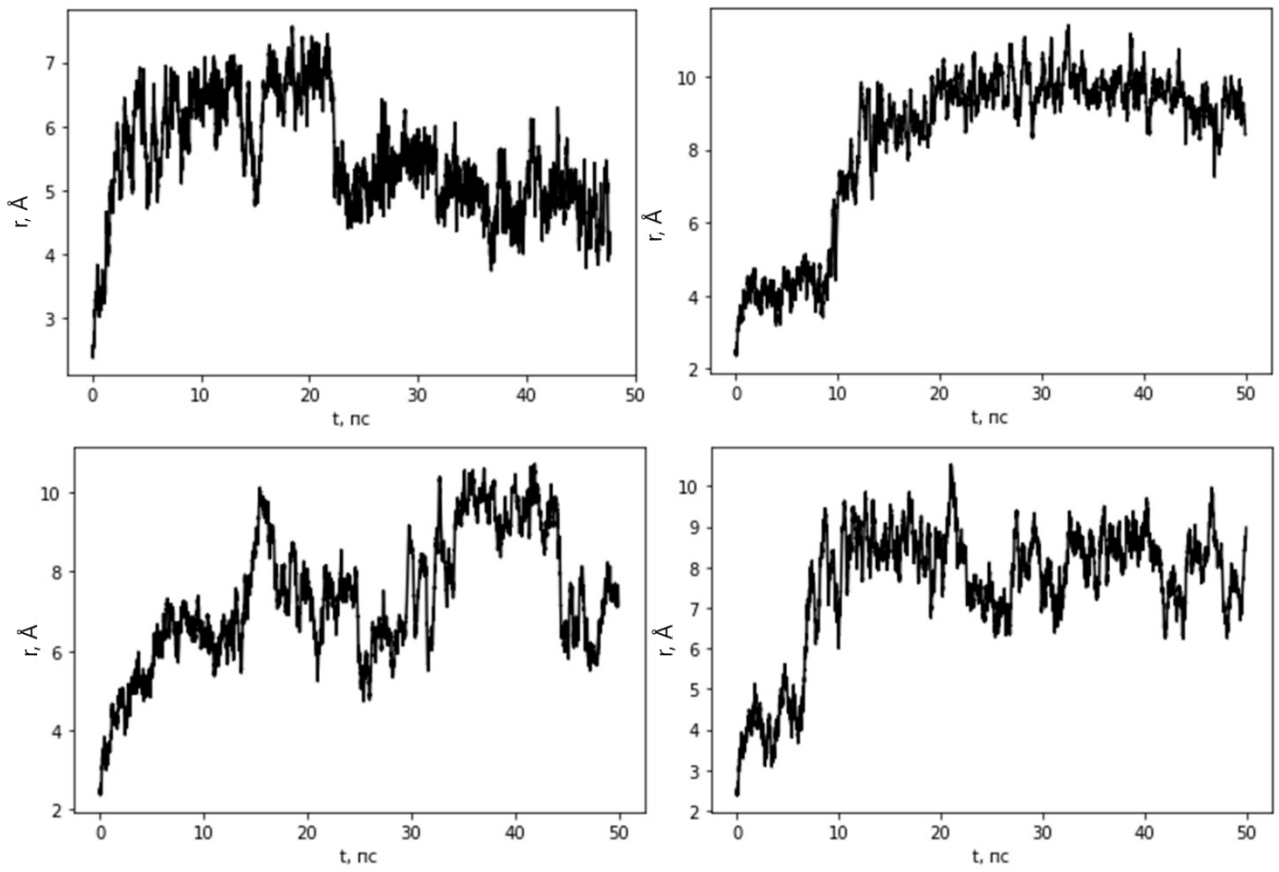
6 O(2)...H(21)



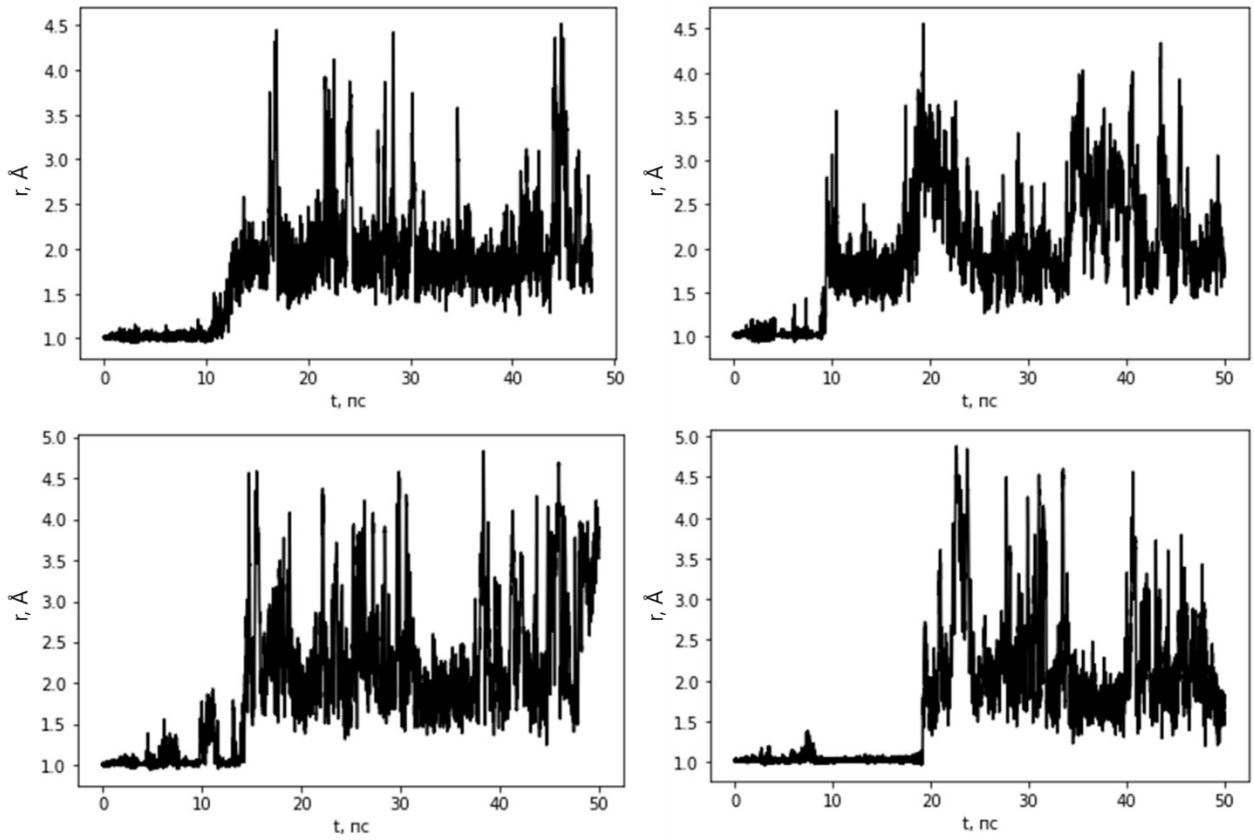
7 N(15)…H(129)



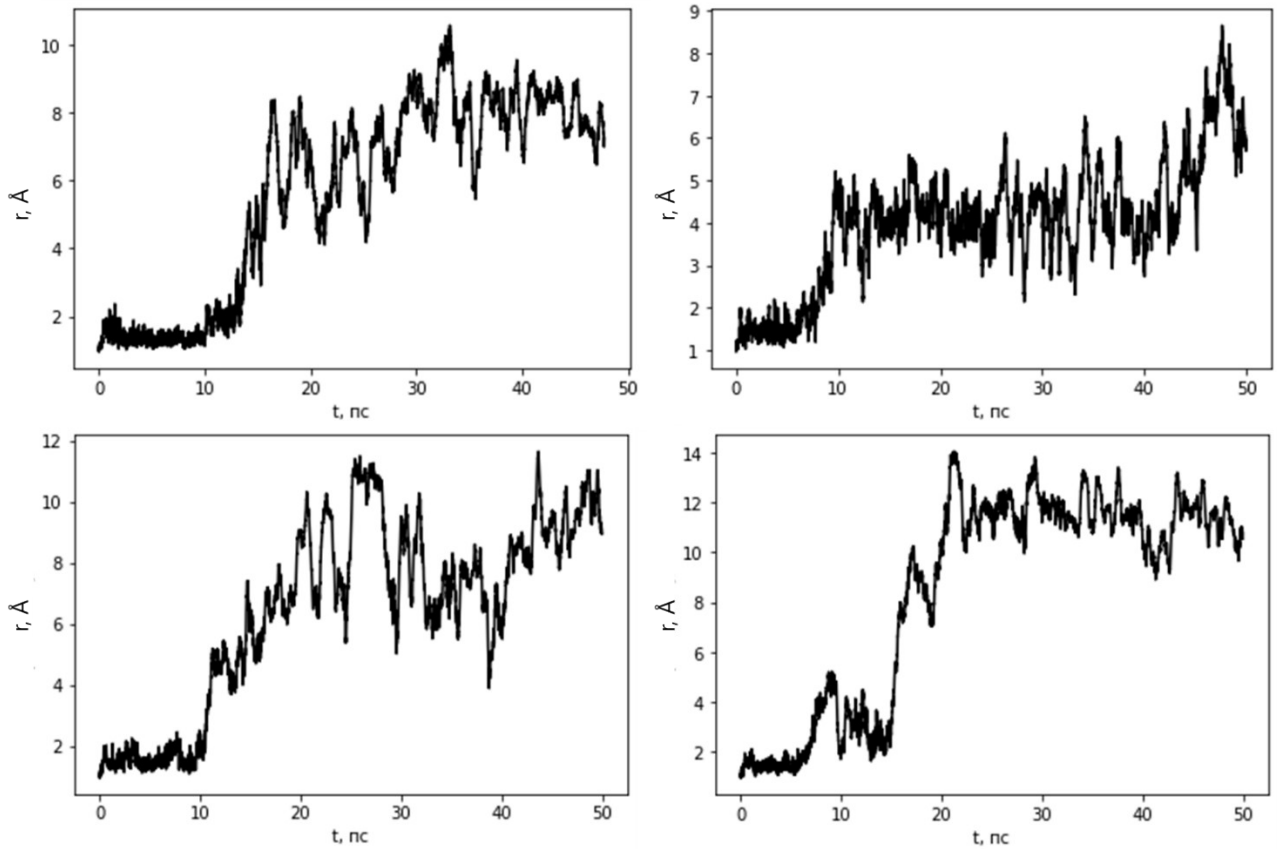
8 O(90)…H(54)



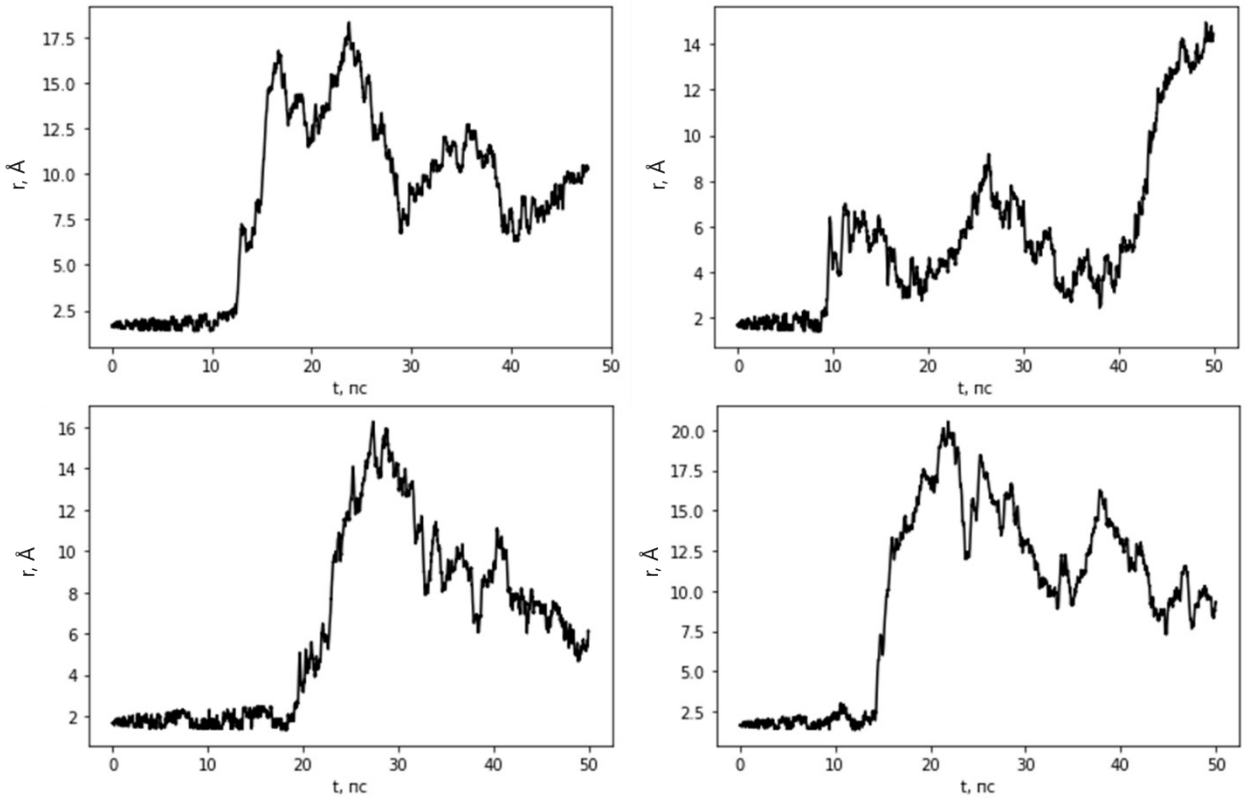
9 N(99)...H(100)



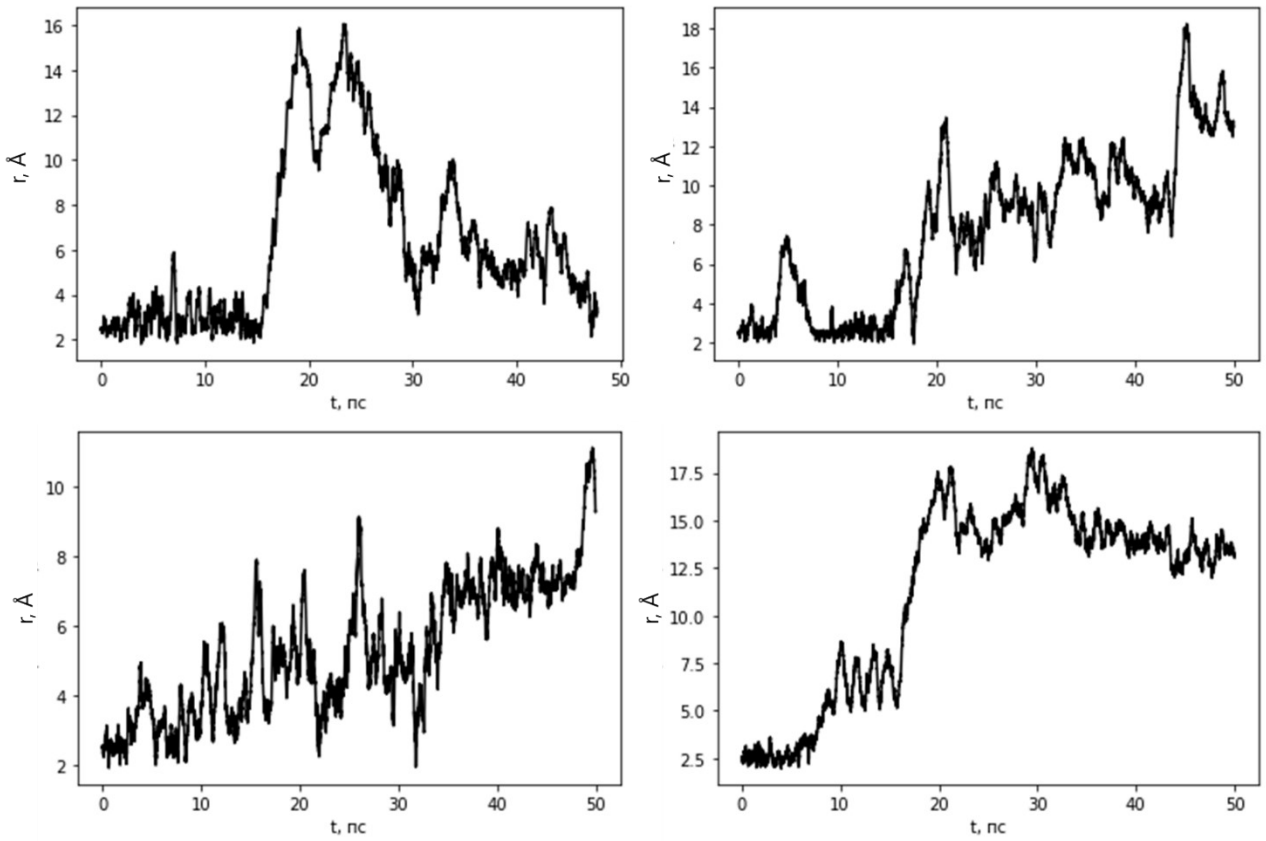
10 O(97)…H(82)



11 P(117)…O(120)

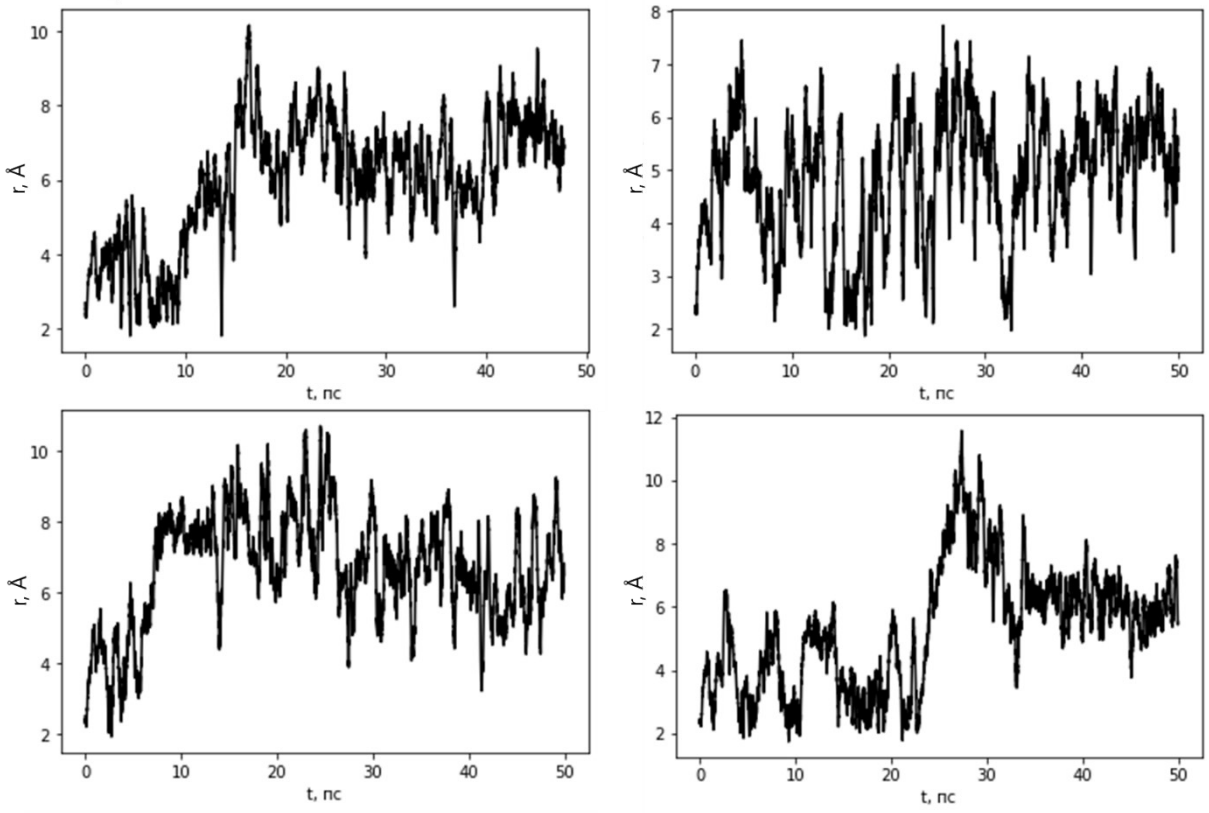


12 O(70)…H(113)

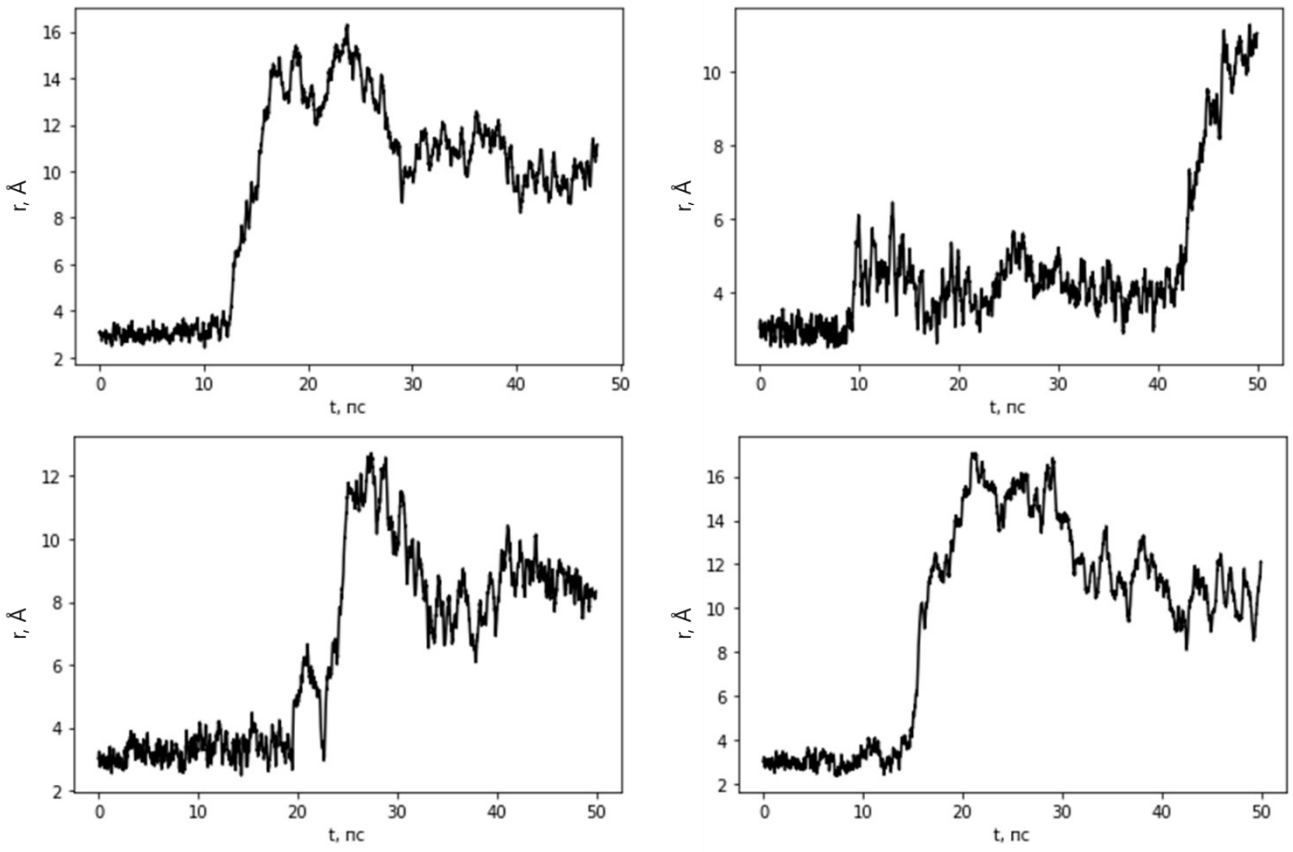


13

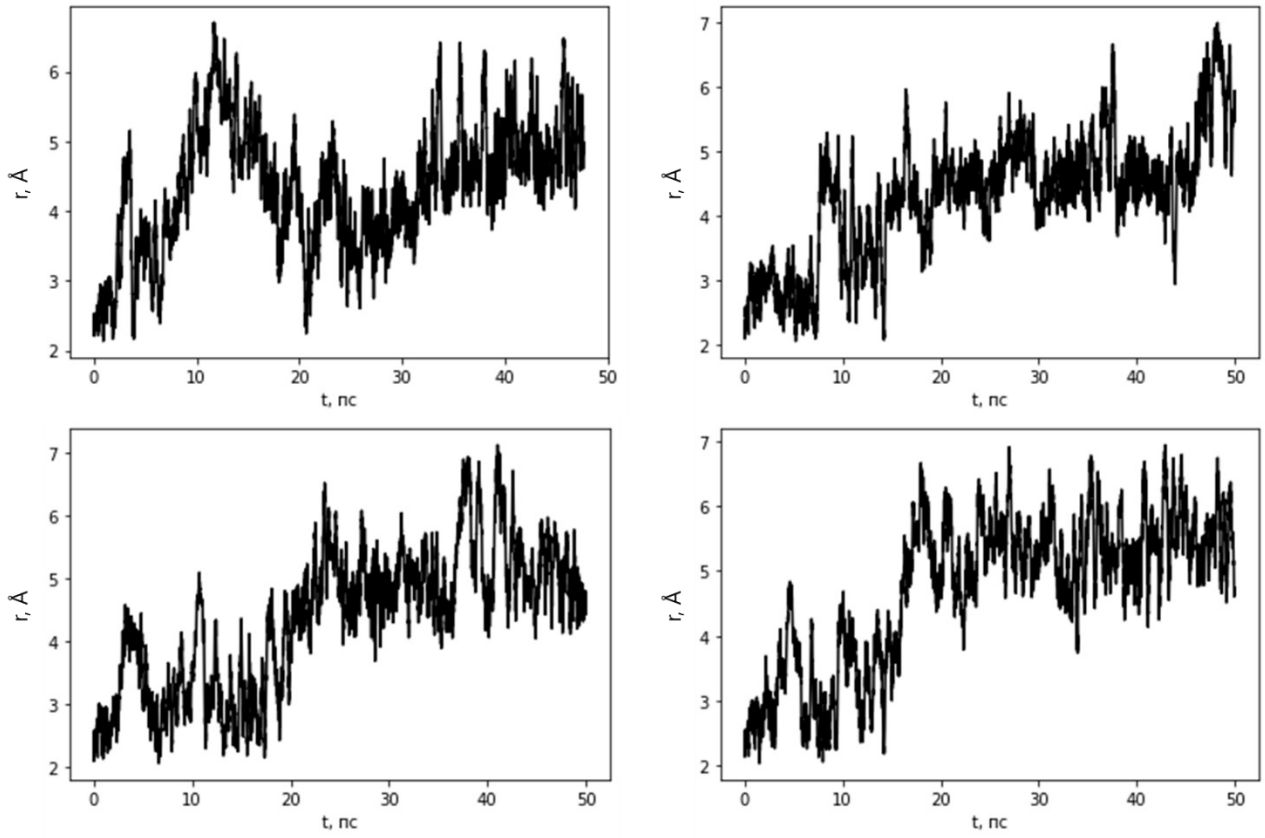
O(70)…H(123)



14 O(118)…N(99)

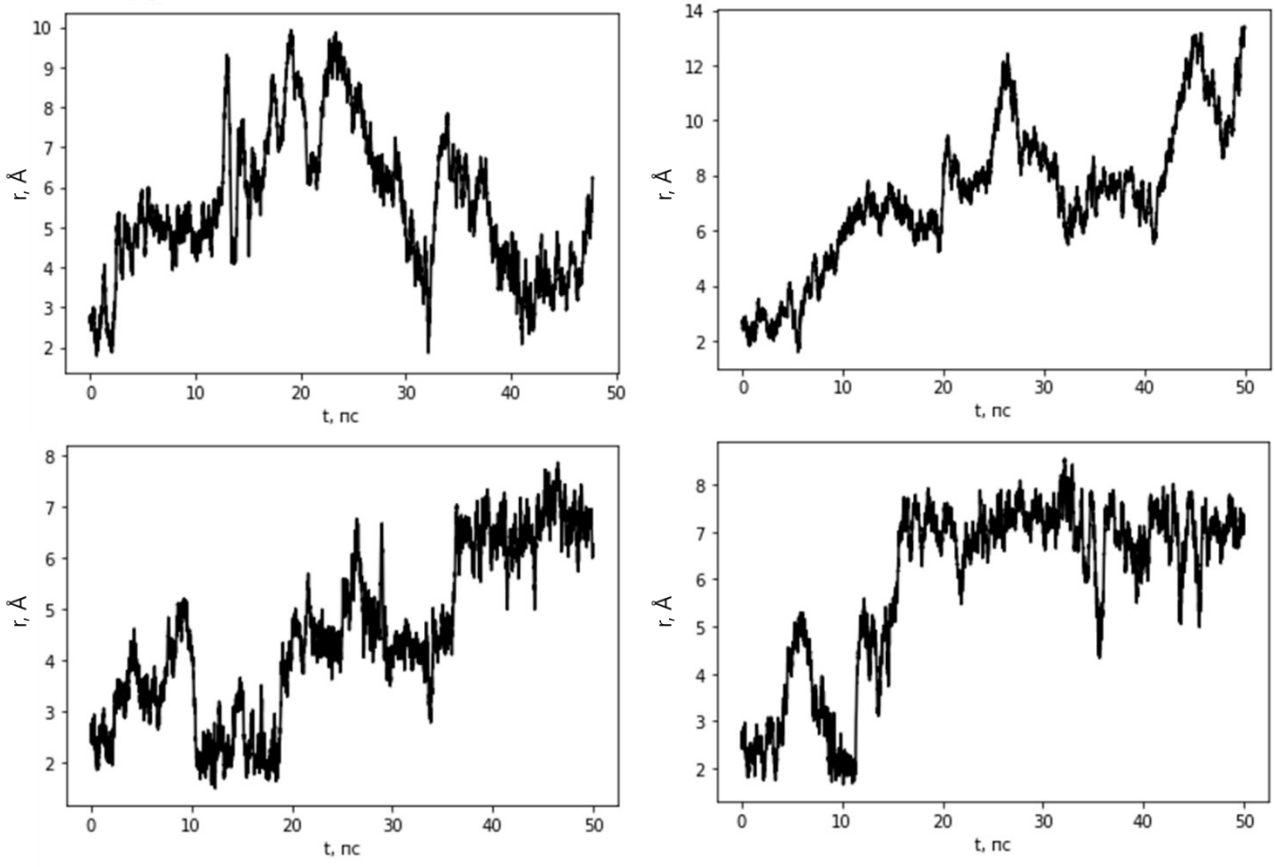


15 O(116)...H(14)



16

O(128)…H(36)



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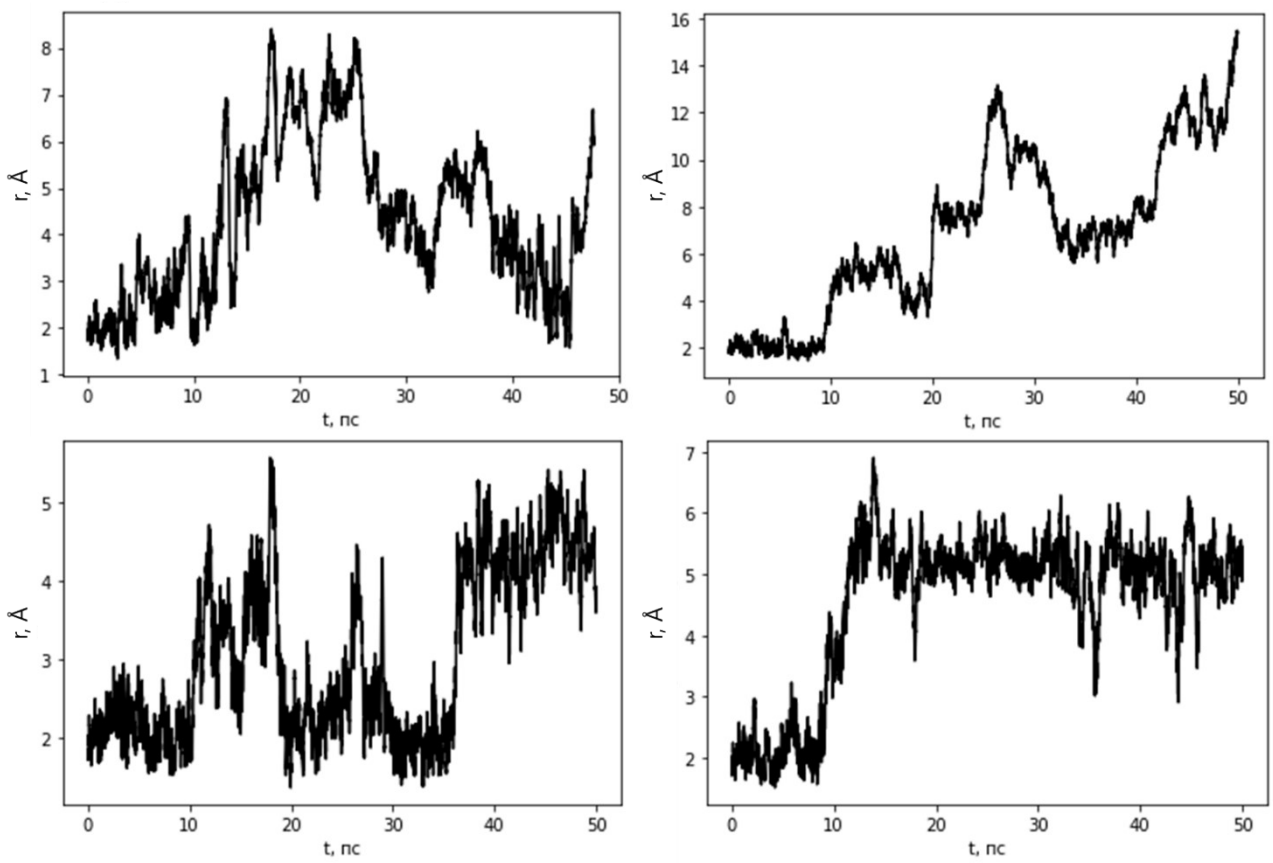
O(119)…H(36)



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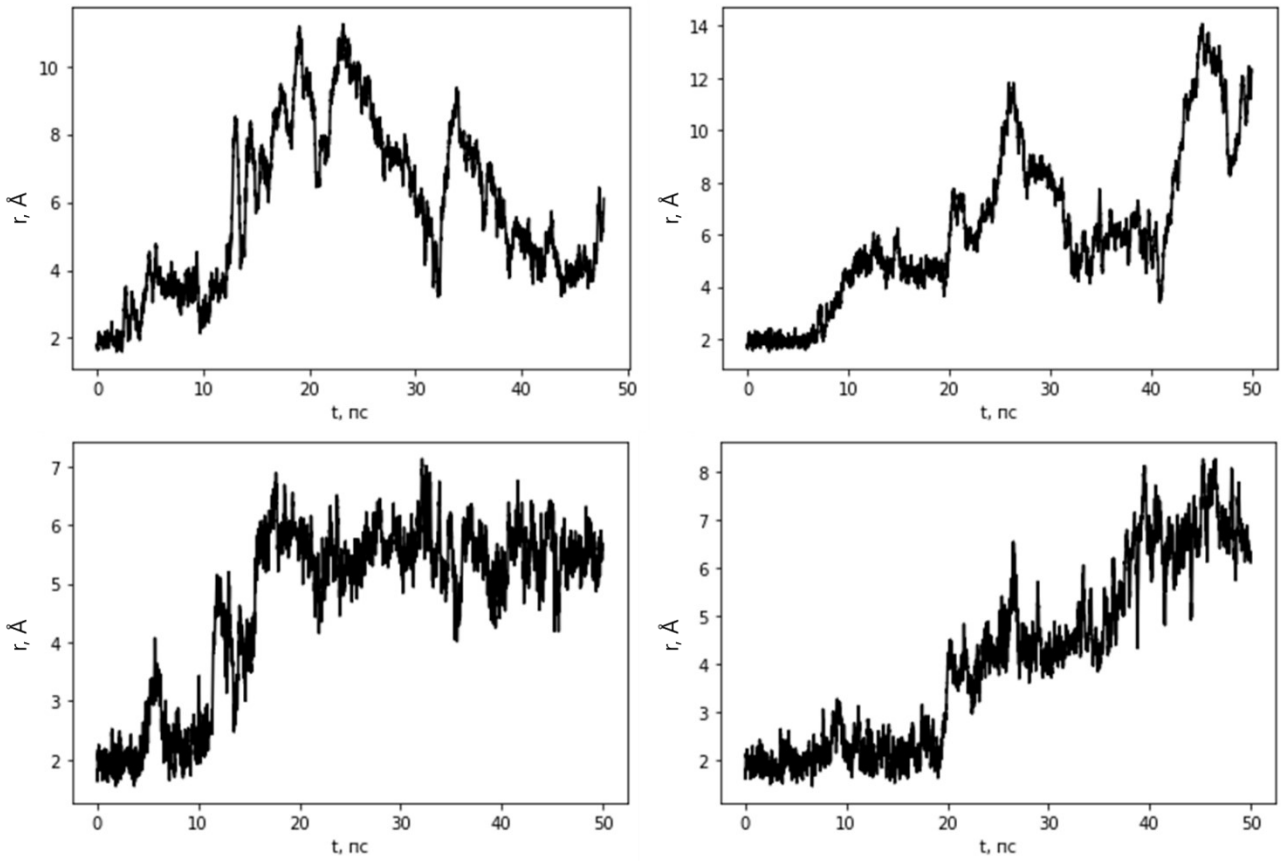

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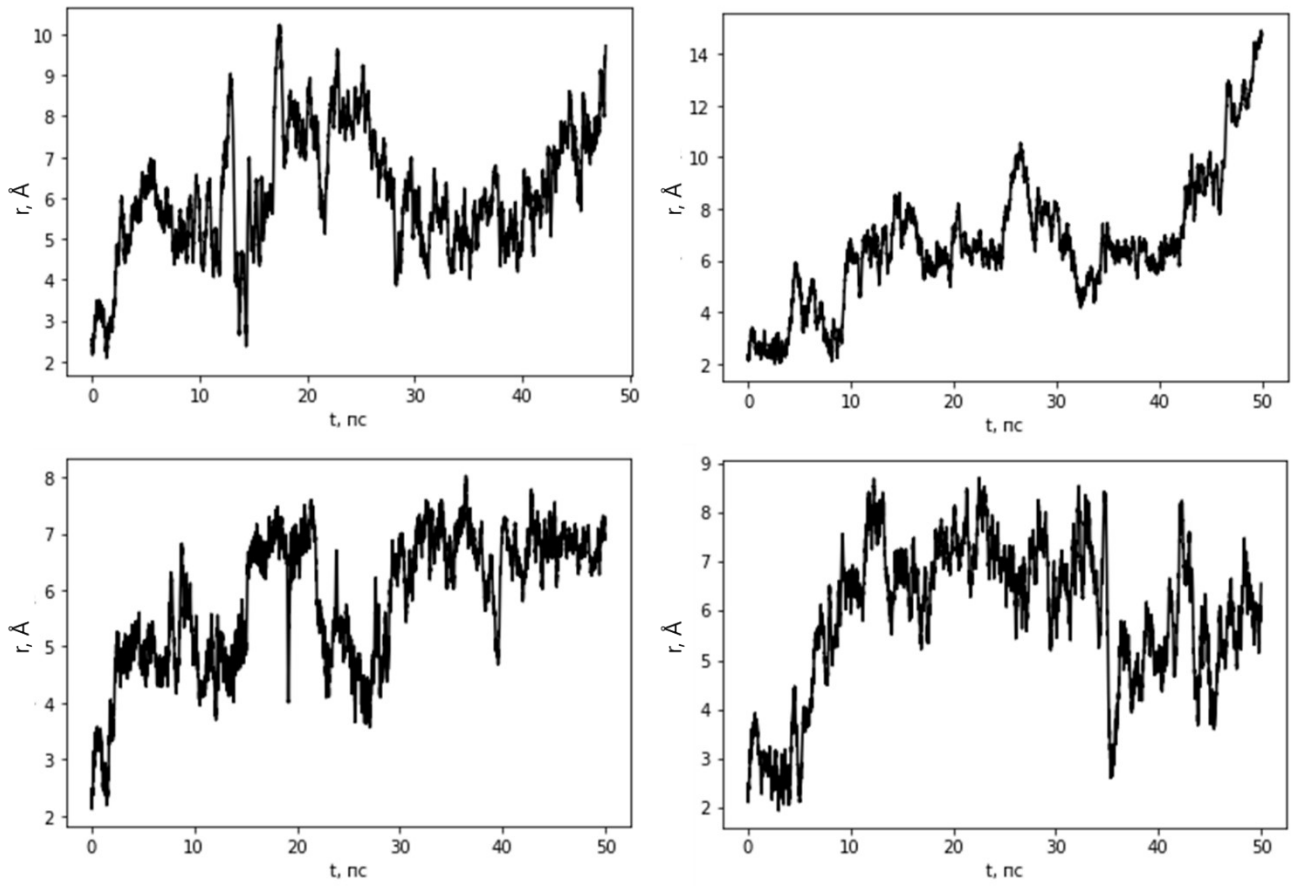

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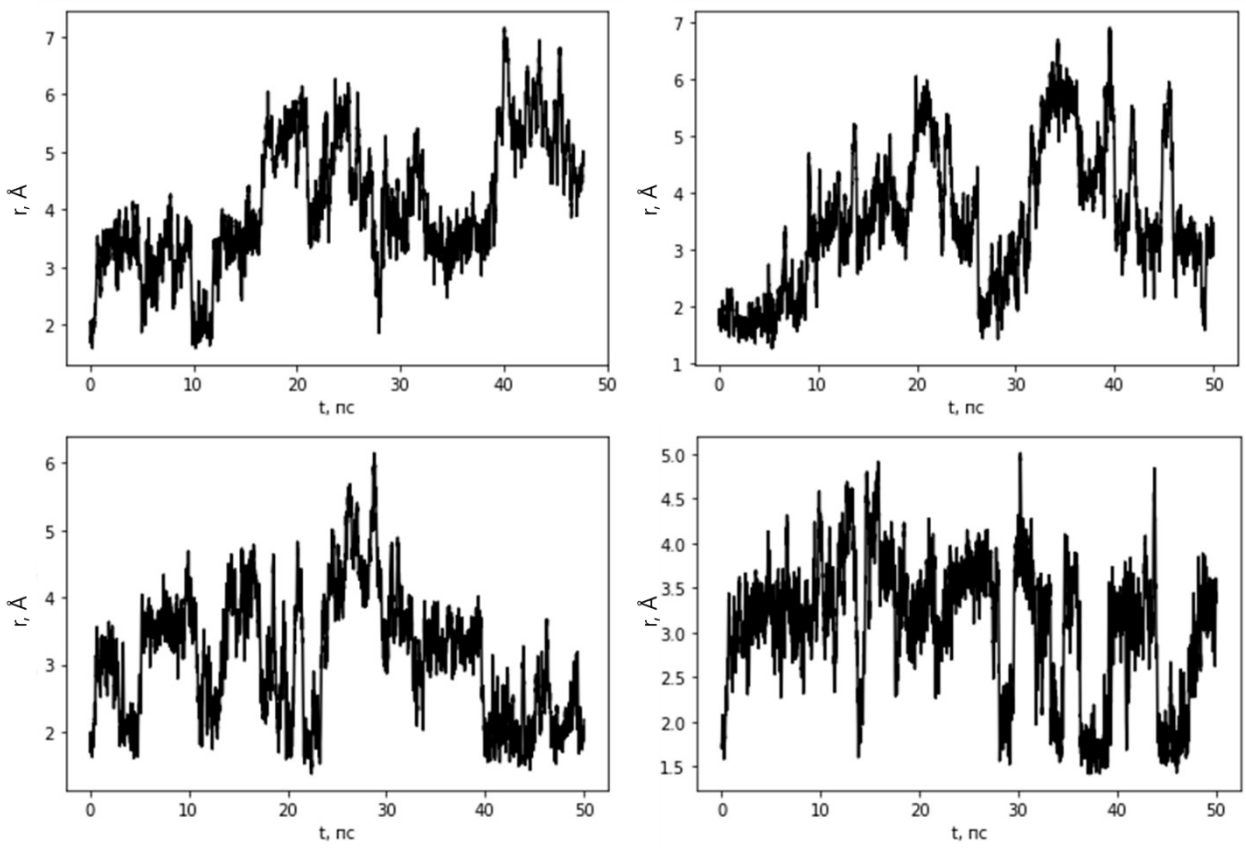
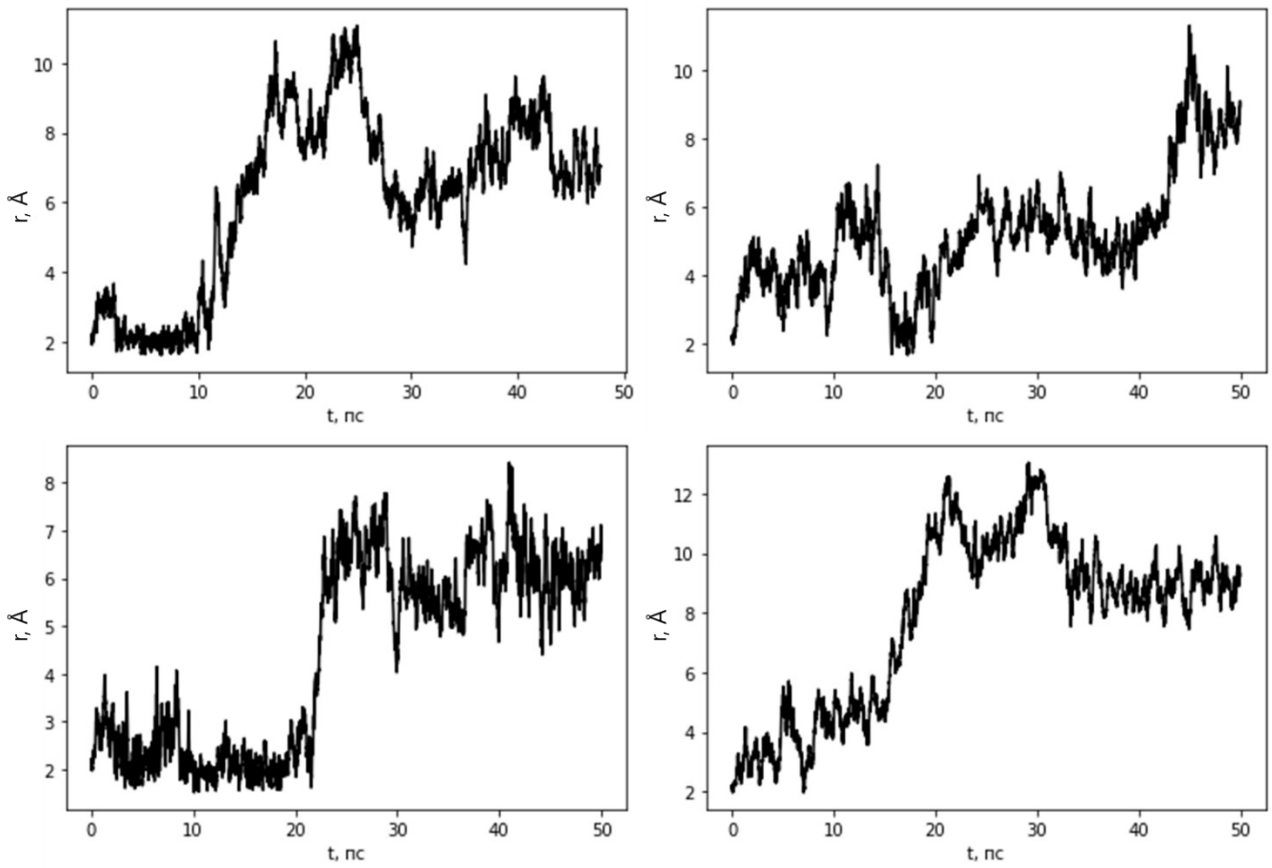


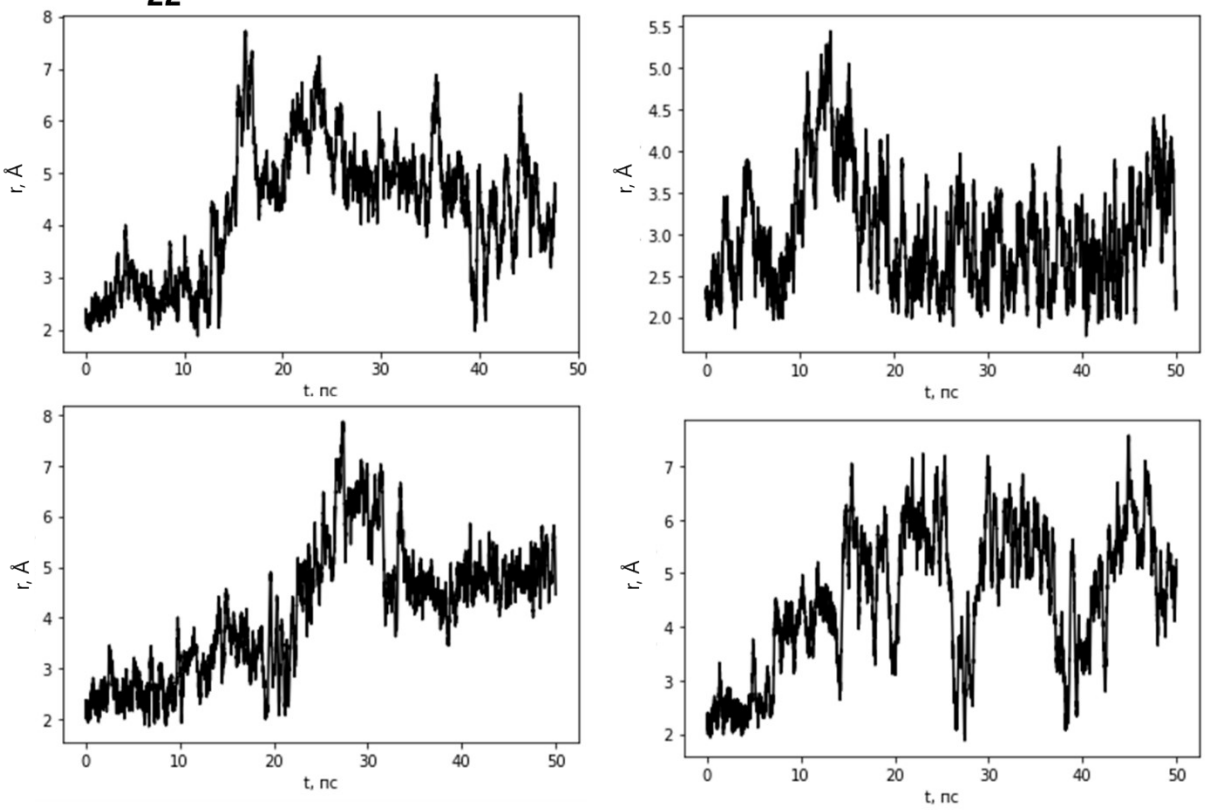
21

O(118)...H(88)



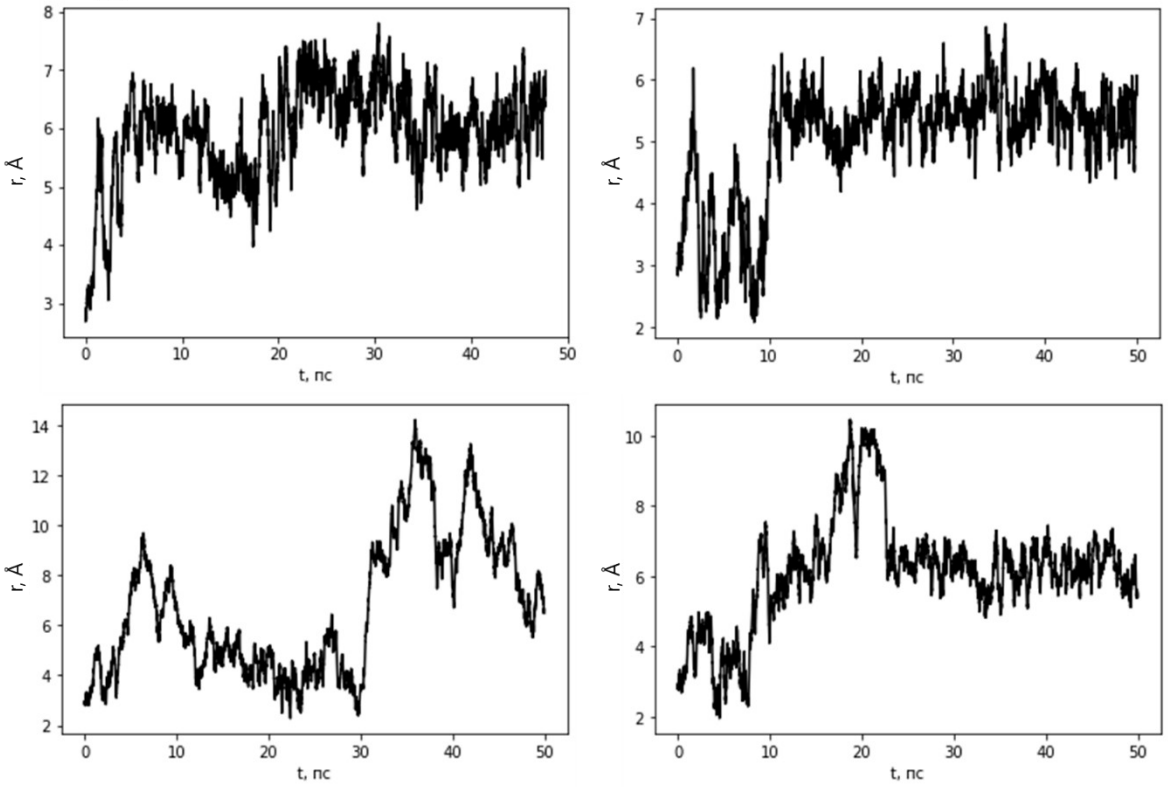
22

O(120)...H(74)



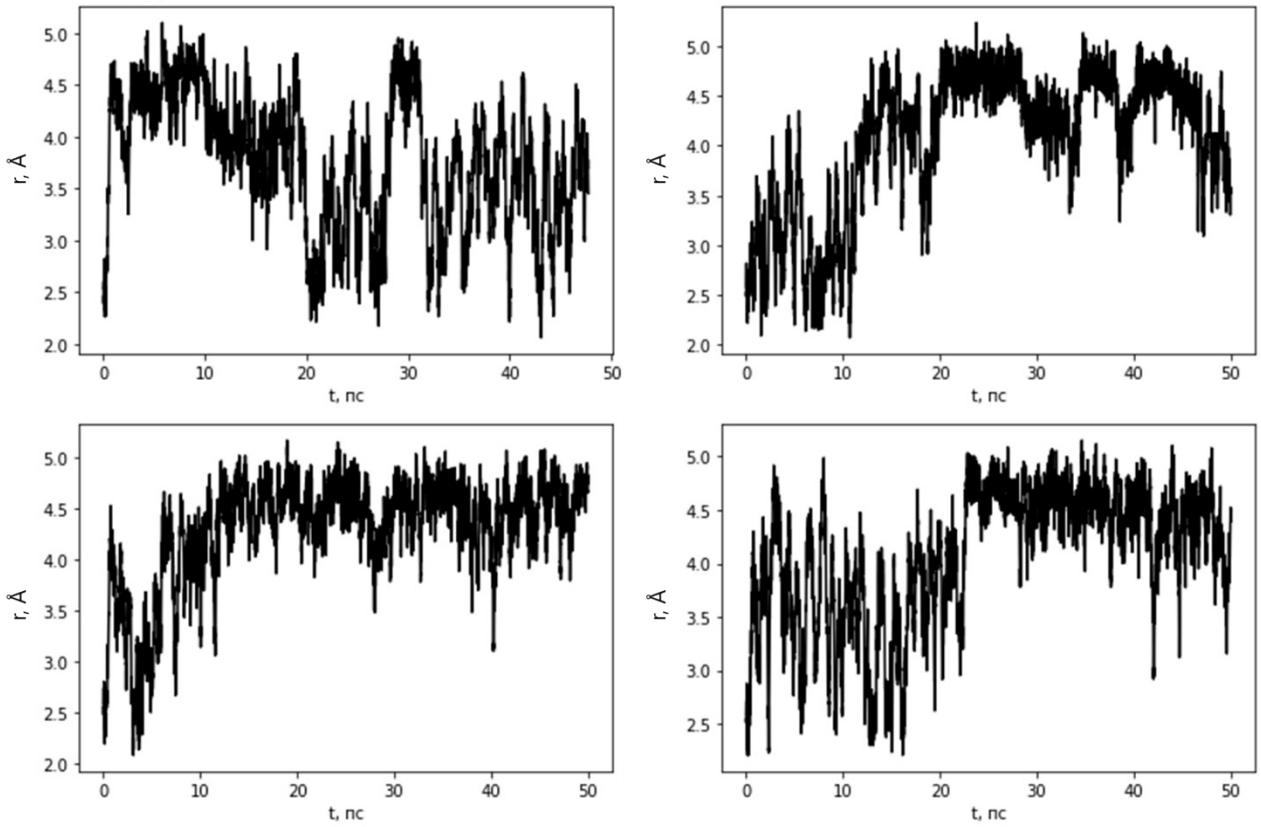
23

O(2)...H(55)

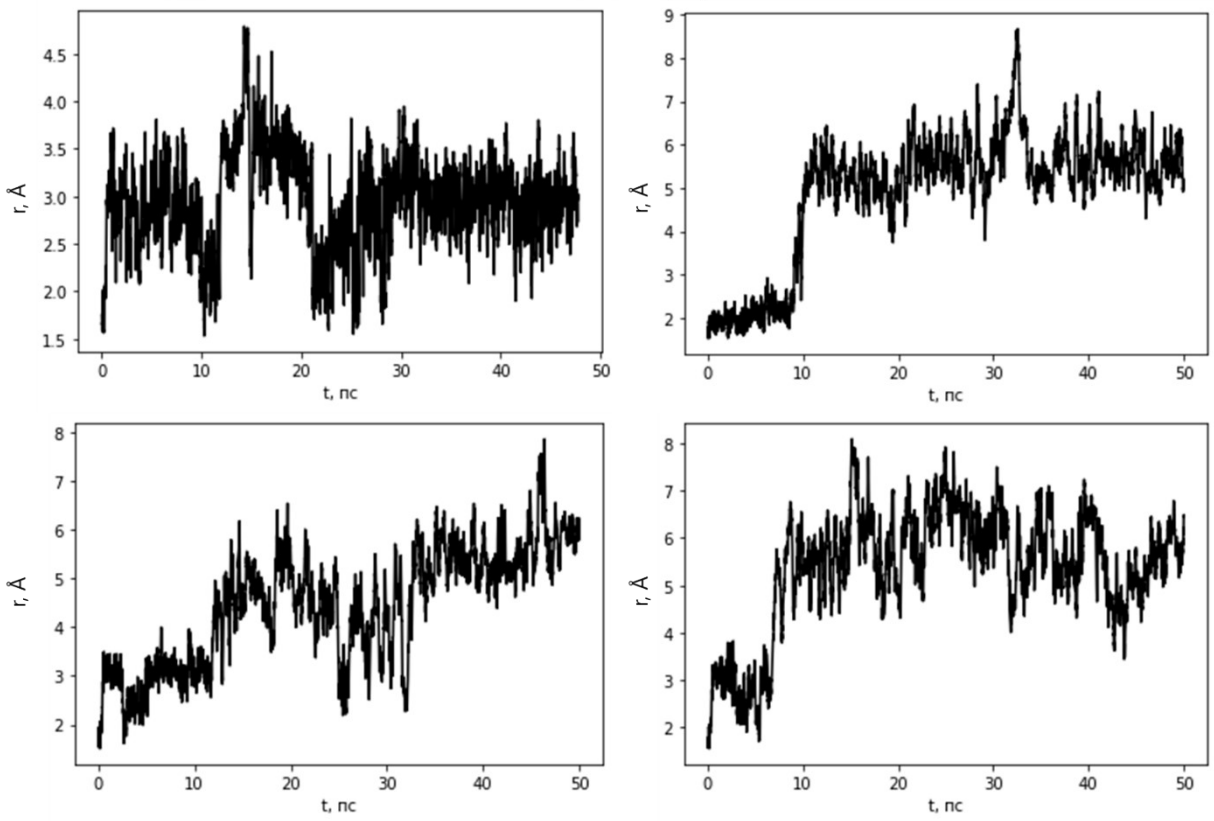


24

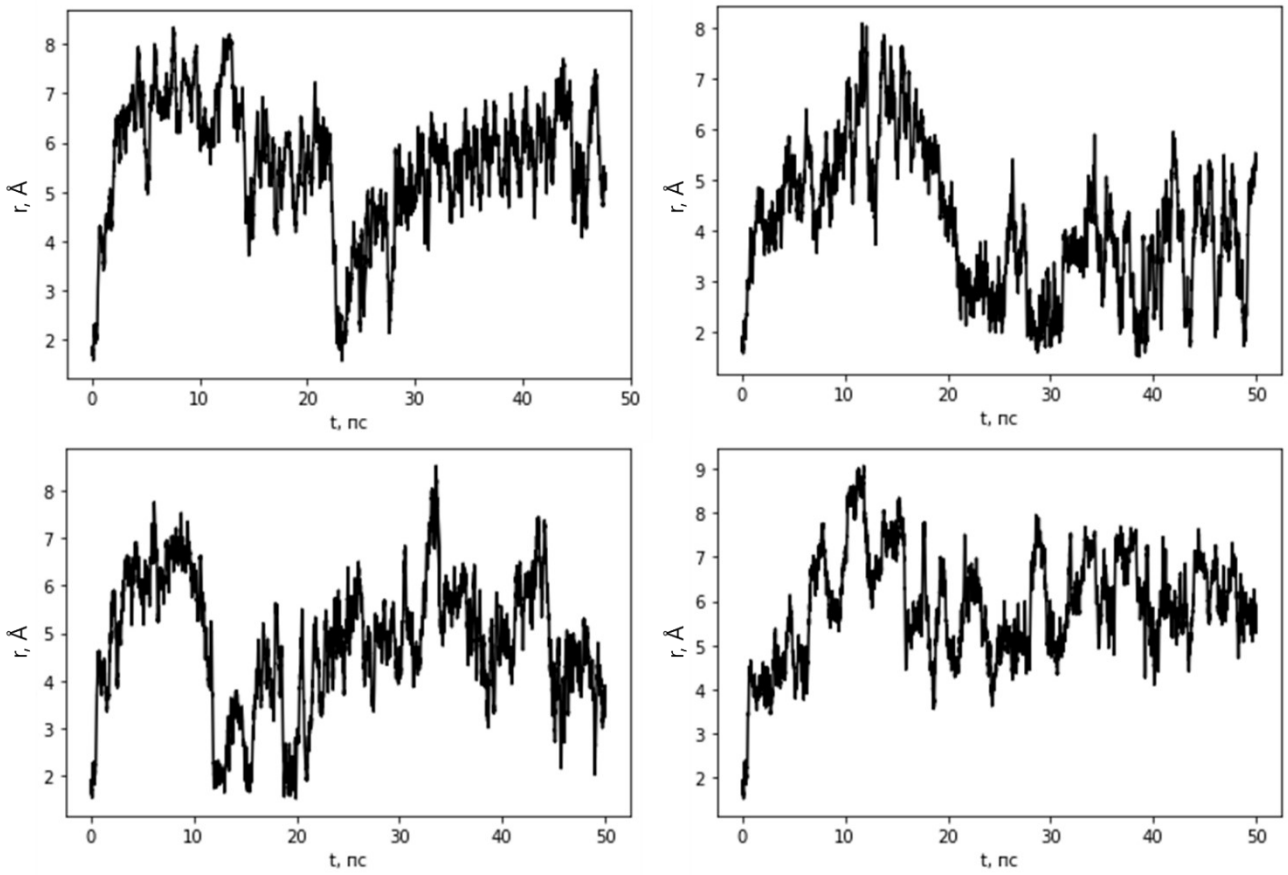
O(132)…H(52)



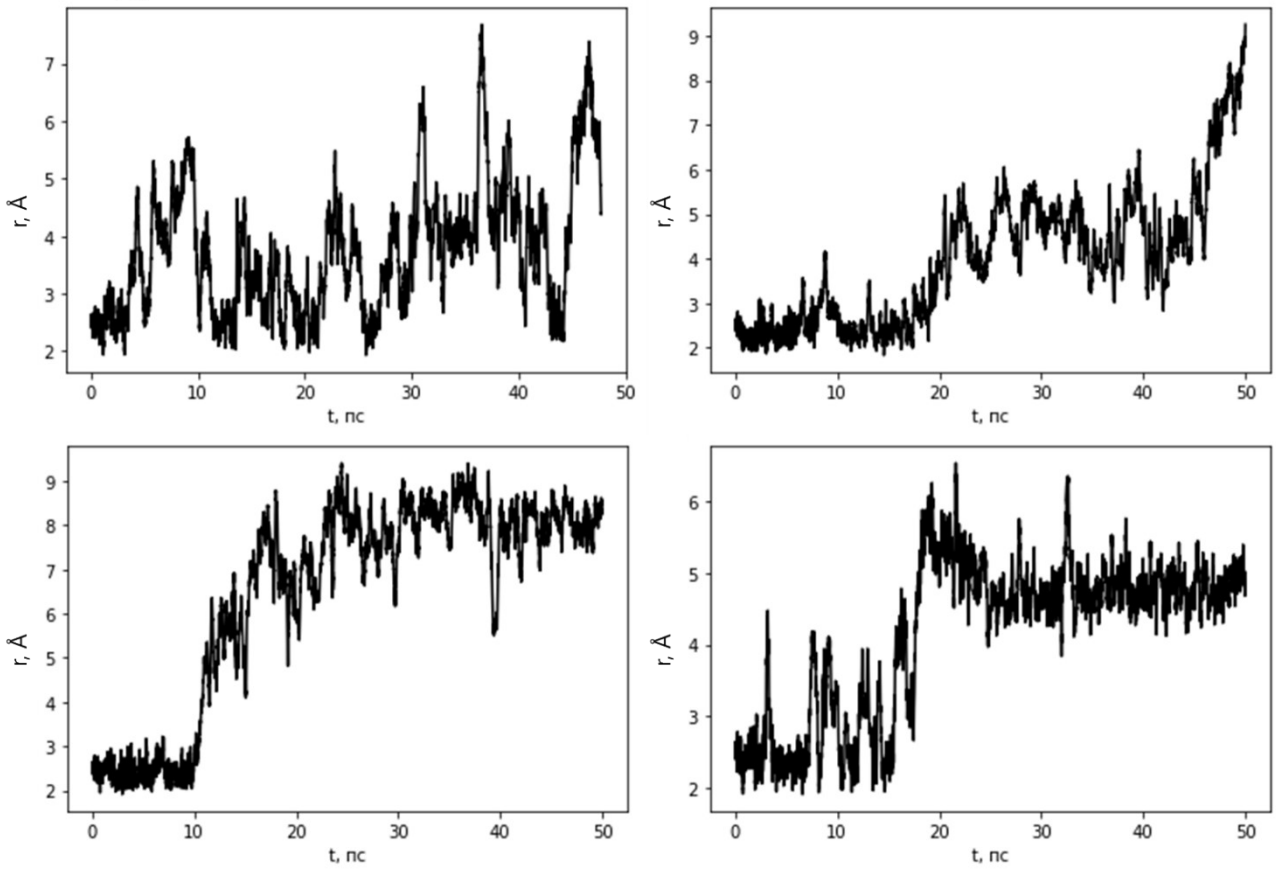
25 O(90)...H(64)



26 O(132)…H(65)



27 O(132)…H(34)



28

O(45)…H(34)

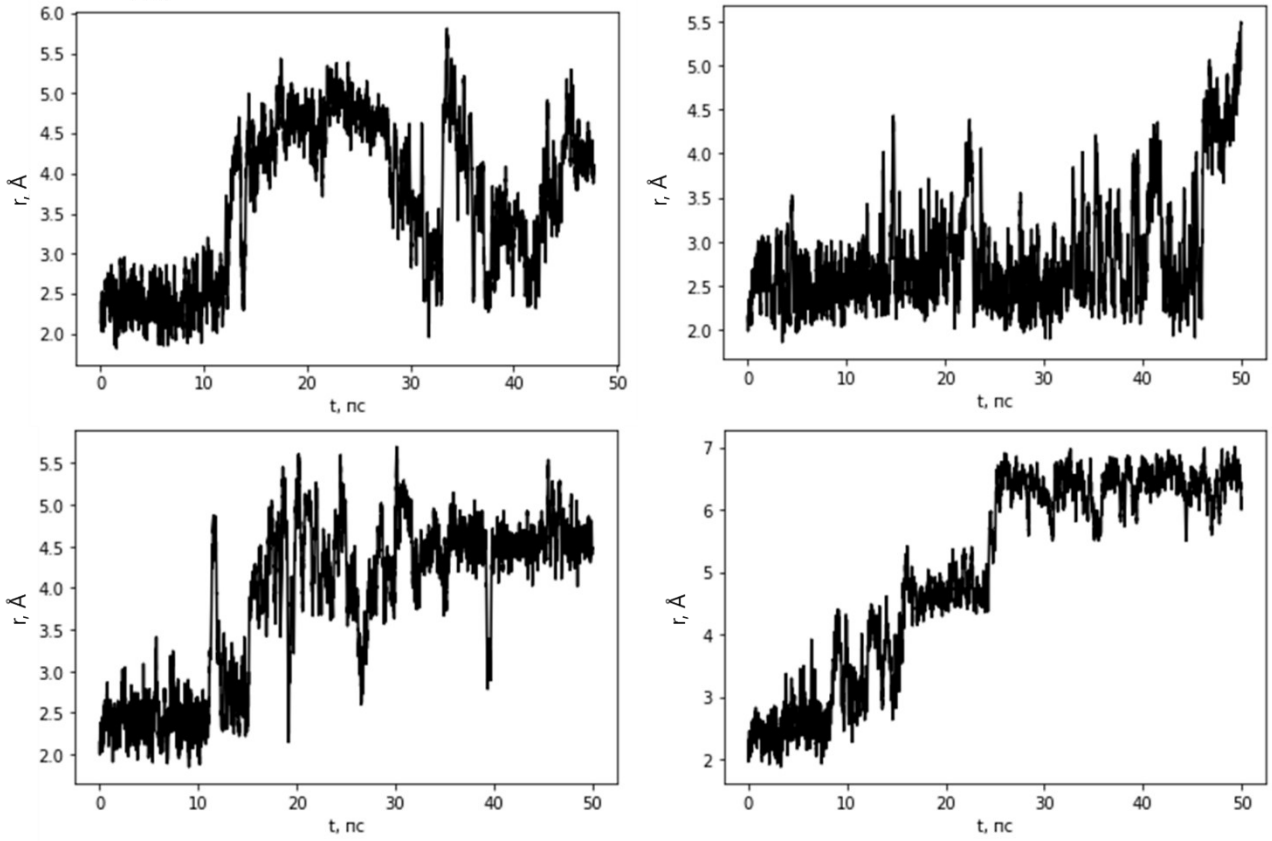
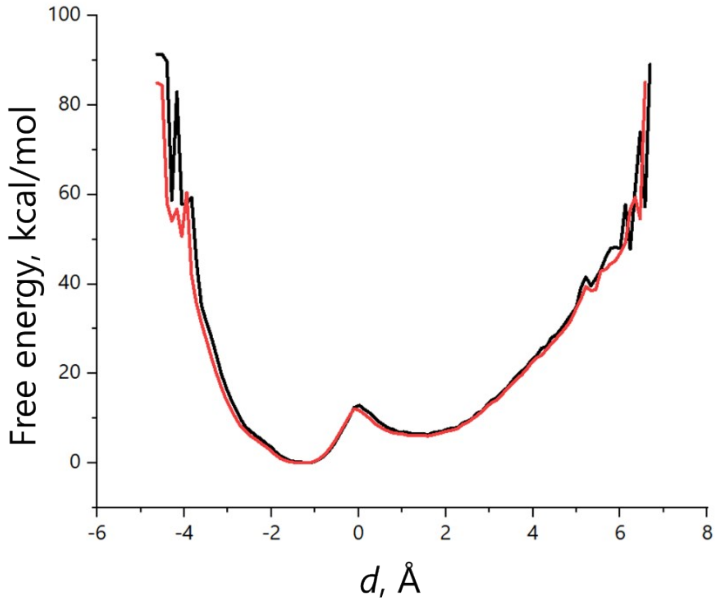


Figure S5. Free energy profiles along the reaction coordinate from well-tempered metadynamics simulation. Black line corresponds to free energy profile obtained after summation of gaussian hills[1]; red line corresponds to free energy profile obtained from reweighted histogram[2]. In both cases the zero free energy value is taken as a minimum value in the corresponding profiles.



Collective variable *d* is determined as follows:

$$d = r(\text{P-OEt}) - r(\text{P-O}^{2'}),$$

where *r*(P-OEt) – bond cleaved during the reaction; *r*(P-O$^{2'}$) – bond formed during the reaction. Thus, d < 0 correspond to reagent state, d > 0 correspond to product state.

To conduct metadynamics experiment we used the same methodology as described in the main text. The bias factor was set to 20. We added two harmonic potential type restraints with force constants 1.2 kcal·mol^{-1}·Å^{-1} on the relative positions of His12, His119 and model ligand and one harmonic potential type restraints with force constants 0.6 kcal·mol^{-1}·Å^{-1} to *r*(P-OEt) distance to sample configurational space more effectively

Root mean square deviation (RMSD) between the profiles is 1.3 kcal/mol in the range from −4 Å to 5 Å, corresponded to reliably sampled space. Free energy difference between the observable maximum (*d* = 0 Å) and first minimum (*d* = −1.1 Å) is 12.9 kcal/mol. Free energy difference between the two minima is 6.4 kcal/mol.

1 A. Barducci, G. Bussi and M. Parrinello, Well-tempered metadynamics: A smoothly converging and tunable free-energy method, *Phys. Rev. Lett.*, 2008, **100**, 1–4.

2 T. M. Schäfer and G. Settanni, Data Reweighting in Metadynamics Simulations, *J. Chem. Theory Comput.*, 2020, **16**, 2042–2052.