Flexibility of a biotinylated ligand in artificial metalloenzymes based on streptavidin – an insight from Molecular Dynamics simulations with classical and *ab initio* force fields

Jarosław J. Panek[1*], Thomas R. Ward[2], Aneta Jezierska-Mazzarello[1,3], Marjana Novič[3]

[1]University of Wrocław, Faculty of Chemistry, ul. F. Joliot-Curie 14, 50-383 Wrocław, Poland
[2] University of Basel, Department of Chemistry, Spitalstrasse 51, CH-4056 Basel, Switzerland
[3] National Institute of Chemistry, Hajdrihova 19, SI-1001 Ljubljana, Slovenia

***Corresponding author:** Jarosław J. Panek, e-mail: jarek@elrond.chem.uni.wroc.pl, phone: +48 71 3757 246, Fax: +48 71 3282348

ONLINE RESOURCE 1

ADDITIONAL FORCE-FIELD PARAMETERS FOR THE COFACTOR AMBER9 FRCMOD FILE FORMAT

ATOMIC COORDINATES OF THE COFACTOR STRUCTURE OPTIMIZED AT THE DFT B3LYP/SDDAll LEVEL

VALIDATION OF THE COFACTOR PARAMETERS AGAINST DFT AND MP2 DATA: STRUCTURES AND ENERGIES OF COFACTOR-WATER MOLECULE COMPLEXES WITH HYDROGEN BONDS

ADDITIONAL FORCE-FIELD PARAMETERS FOR THE COFACTOR
AMBER9 FRCMOD FILE FORMAT

```
Ru-loaded cofactor
MASS
Fe 101.00        0.000              ATTN: mass corresponds to Ru

BOND
Fe-cl  200.0    2.478       QM-based parameter
Fe-ca  200.0    2.260       QM-based parameter
Fe-n2  200.0    2.097       QM-based parameter
Fe-n3  200.0    2.157       QM-based parameter

ANGLE
n -c2-o    71.411     117.460   Calculated with empirical approach
c3-c2-n    65.581     114.875   Calculated with empirical approach
ca-s6-n2   38.644     100.845   Calculated with empirical approach
ca-Fe-ca   30.000      77.300   QM-based parameter
cl-Fe-ca    5.000     115.000   QM-based parameter
Fe-ca-ha   30.000     125.000   QM-based parameter
Fe-ca-ca   30.000      73.000   QM-based parameter
hn-n3-Fe    5.000     106.000   QM-based parameter
n3-Fe-cl   60.000      80.000   QM-based parameter
n3-Fe-ca    5.000     115.000   QM-based parameter
c3-n3-Fe   30.000     111.120   QM-based parameter
c3-n2-Fe   30.000     111.460   QM-based parameter
n2-Fe-cl   60.000      92.200   QM-based parameter
n2-Fe-ca    5.000     120.000   QM-based parameter
n2-Fe-n3   30.000      78.400   QM-based parameter
s6-n2-Fe   30.000     120.000   QM-based parameter

DIHE
ca-Fe-ca-ca   1    1.500        60.000          3.000      Generalized
ca-Fe-ca-ha   1    1.500       180.000          2.000      Generalized
cl-Fe-ca-ha   1    0.050       180.000          2.000      Generalized
cl-Fe-ca-ca   1    0.050       180.000          2.000      Generalized
hn-n3-Fe-n2   1    1.500       120.000          3.000      Generalized
hn-n3-Fe-cl   1    0.050       120.000          3.000      Generalized
hn-n3-Fe-ca   1    0.050       180.000          6.000      Generalized
n3-Fe-ca-ca   1    0.050       180.000          6.000      Generalized
n3-Fe-ca-ha   1    0.050       180.000          6.000      Generalized
c3-n3-Fe-n2   1    1.500       180.000          2.000      Generalized
c3-n3-Fe-ca   1    0.050       180.000          6.000      Generalized
c3-n3-Fe-cl   1    0.050       120.000          6.000      Generalized
c3-n2-Fe-n3   1    1.500       180.000          2.000      Generalized
c3-n2-Fe-ca   1    0.050       180.000          6.000      Generalized
c3-n2-Fe-cl   1    0.050       120.000          6.000      Generalized
n2-Fe-ca-ca   1    0.050       180.000          6.000      Generalized
n2-Fe-ca-ha   1    0.050       180.000          6.000      Generalized
s6-n2-Fe-cl   1    0.500       180.000          6.000      Generalized
s6-n2-Fe-ca   1    0.050       180.000          6.000      Generalized
s6-n2-Fe-n3   1    0.050       180.000          6.000      Generalized

IMPROPER
n -n -c2-o          1.1          180.0         2.0          default value
c3-n -c2-o          1.1          180.0         2.0          default value
c2-ca-n -hn         1.1          180.0         2.0          General impr.
ca-ca-ca-n          1.1          180.0         2.0          default value
ca-ca-ca-ha         1.1          180.0         2.0          General impr.
ca-ca-ca-s6         1.1          180.0         2.0          default value

NONBON
  Fe          1.1000  0.0125             ATTN: taken from Zn
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ATOMIC COORDINATES OF THE COFACTOR STRUCTURE OPTIMIZED AT THE DFT B3LYP/SDDAll LEVEL

ATOM	1	N1	CMN	1	10.053	-2.433	0.303	1.00	0.00
ATOM	2	H1	CMN	1	10.776	-2.810	0.892	1.00	0.00
ATOM	3	C1	CMN	1	10.335	-1.458	-0.632	1.00	0.00
ATOM	4	O1	CMN	1	11.451	-0.952	-0.878	1.00	0.00
ATOM	5	N2	CMN	1	9.138	-1.170	-1.262	1.00	0.00
ATOM	6	H2	CMN	1	9.105	-0.520	-2.028	1.00	0.00
ATOM	7	C4	CMN	1	7.992	-1.929	-0.765	1.00	0.00
ATOM	8	H4	CMN	1	7.562	-2.544	-1.566	1.00	0.00
ATOM	9	C2	CMN	1	8.648	-2.837	0.357	1.00	0.00
ATOM	10	H3	CMN	1	8.542	-3.901	0.111	1.00	0.00
ATOM	11	C3	CMN	1	7.983	-2.564	1.720	1.00	0.00
ATOM	12	H5	CMN	1	7.129	-3.226	1.885	1.00	0.00
ATOM	13	H6	CMN	1	8.686	-2.663	2.548	1.00	0.00
ATOM	14	S1	CMN	1	7.356	-0.767	1.680	1.00	0.00
ATOM	15	C5	CMN	1	6.858	-1.062	-0.158	1.00	0.00
ATOM	16	H7	CMN	1	5.946	-1.669	-0.122	1.00	0.00
ATOM	17	C6	CMN	1	6.581	0.249	-0.894	1.00	0.00
ATOM	18	H8	CMN	1	7.477	0.880	-0.855	1.00	0.00
ATOM	19	H9	CMN	1	6.397	0.014	-1.953	1.00	0.00
ATOM	20	C7	CMN	1	5.371	1.028	-0.344	1.00	0.00
ATOM	21	H10	CMN	1	4.488	0.375	-0.356	1.00	0.00
ATOM	22	1H1	CMN	1	5.556	1.288	0.707	1.00	0.00
ATOM	23	C8	CMN	1	5.079	2.307	-1.148	1.00	0.00
ATOM	24	2H1	CMN	1	5.959	2.962	-1.133	1.00	0.00
ATOM	25	3H1	CMN	1	4.881	2.053	-2.195	1.00	0.00
ATOM	26	C9	CMN	1	3.868	3.097	-0.604	1.00	0.00
ATOM	27	H14	CMN	1	4.028	3.348	0.452	1.00	0.00
ATOM	28	H15	CMN	1	3.780	4.044	-1.150	1.00	0.00
ATOM	29	C10	CMN	1	2.562	2.339	-0.799	1.00	0.00
ATOM	30	O2	CMN	1	2.236	1.870	-1.914	1.00	0.00
ATOM	31	N3	CMN	1	1.779	2.218	0.332	1.00	0.00
ATOM	32	H16	CMN	1	2.156	2.624	1.178	1.00	0.00
ATOM	33	C11	CMN	1	0.516	1.593	0.483	1.00	0.00
ATOM	34	C14	CMN	1	-0.068	1.632	1.764	1.00	0.00
ATOM	35	C15	CMN	1	-1.310	1.041	1.996	1.00	0.00
ATOM	36	H20	CMN	1	-1.775	1.047	2.974	1.00	0.00
ATOM	37	H19	CMN	1	0.453	2.121	2.583	1.00	0.00
ATOM	38	C12	CMN	1	-0.154	0.949	-0.576	1.00	0.00
ATOM	39	H17	CMN	1	0.302	0.916	-1.555	1.00	0.00
ATOM	40	C13	CMN	1	-1.397	0.353	-0.340	1.00	0.00
ATOM	41	H18	CMN	1	-1.918	-0.155	-1.142	1.00	0.00
ATOM	42	C16	CMN	1	-1.954	0.418	0.933	1.00	0.00
ATOM	43	S2	CMN	1	-3.611	-0.427	1.279	1.00	0.00
ATOM	44	O3	CMN	1	-3.455	-2.003	0.741	1.00	0.00
ATOM	45	O4	CMN	1	-3.864	-0.187	2.913	1.00	0.00
ATOM	46	N4	CMN	1	-4.741	0.428	0.054	1.00	0.00
ATOM	47	C23	CMN	1	-4.767	1.891	0.371	1.00	0.00
ATOM	48	C24	CMN	1	-5.597	2.491	-0.756	1.00	0.00
ATOM	49	N5	CMN	1	-6.917	1.769	-0.780	1.00	0.00
ATOM	50	H29	CMN	1	-7.412	1.911	-1.659	1.00	0.00
ATOM	51	H30	CMN	1	-7.508	2.061	0.006	1.00	0.00
ATOM	52	3H2	CMN	1	-5.745	3.568	-0.617	1.00	0.00
ATOM	53	H24	CMN	1	-5.106	2.317	-1.716	1.00	0.00
ATOM	54	1H2	CMN	1	-5.222	2.106	1.344	1.00	0.00
ATOM	55	2H2	CMN	1	-3.754	2.315	0.350	1.00	0.00
ATOM	56	Fe1	CMN	1	-6.671	-0.317	-0.289	1.00	0.00
ATOM	57	Cl1	CMN	1	-7.494	0.545	1.884	1.00	0.00
ATOM	58	C19	CMN	1	-7.347	-2.411	0.201	1.00	0.00
ATOM	59	H26	CMN	1	-7.447	-2.790	1.210	1.00	0.00
ATOM	60	C18	CMN	1	-6.104	-2.517	-0.455	1.00	0.00

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ATOM     61  H27 CMN     1      -5.250  -2.937   0.060  1.00  0.00
ATOM     62  C17 CMN     1      -5.906  -1.922  -1.755  1.00  0.00
ATOM     63  C26 CMN     1      -4.575  -2.030  -2.444  1.00  0.00
ATOM     64 1H3  CMN     1      -4.500  -2.995  -2.962  1.00  0.00
ATOM     65 2H3  CMN     1      -4.442  -1.236  -3.185  1.00  0.00
ATOM     66 3H3  CMN     1      -3.768  -1.977  -1.708  1.00  0.00
ATOM     67  C22 CMN     1      -6.997  -1.226  -2.335  1.00  0.00
ATOM     68  H28 CMN     1      -6.868  -0.741  -3.297  1.00  0.00
ATOM     69  C21 CMN     1      -8.283  -1.171  -1.683  1.00  0.00
ATOM     70  H25 CMN     1      -9.089  -0.631  -2.162  1.00  0.00
ATOM     71  C20 CMN     1      -8.486  -1.765  -0.418  1.00  0.00
ATOM     72  C25 CMN     1      -9.814  -1.745   0.321  1.00  0.00
ATOM     73  H34 CMN     1      -9.574  -1.602   1.381  1.00  0.00
ATOM     74  C28 CMN     1     -10.749  -0.595  -0.095  1.00  0.00
ATOM     75  H35 CMN     1     -11.640  -0.599   0.539  1.00  0.00
ATOM     76  H36 CMN     1     -10.254   0.372   0.030  1.00  0.00
ATOM     77  H37 CMN     1     -11.086  -0.696  -1.134  1.00  0.00
ATOM     78  C27 CMN     1     -10.515  -3.119   0.160  1.00  0.00
ATOM     79  H38 CMN     1     -11.432  -3.145   0.759  1.00  0.00
ATOM     80  H39 CMN     1     -10.785  -3.298  -0.888  1.00  0.00
ATOM     81  H40 CMN     1      -9.870  -3.940   0.489  1.00  0.00
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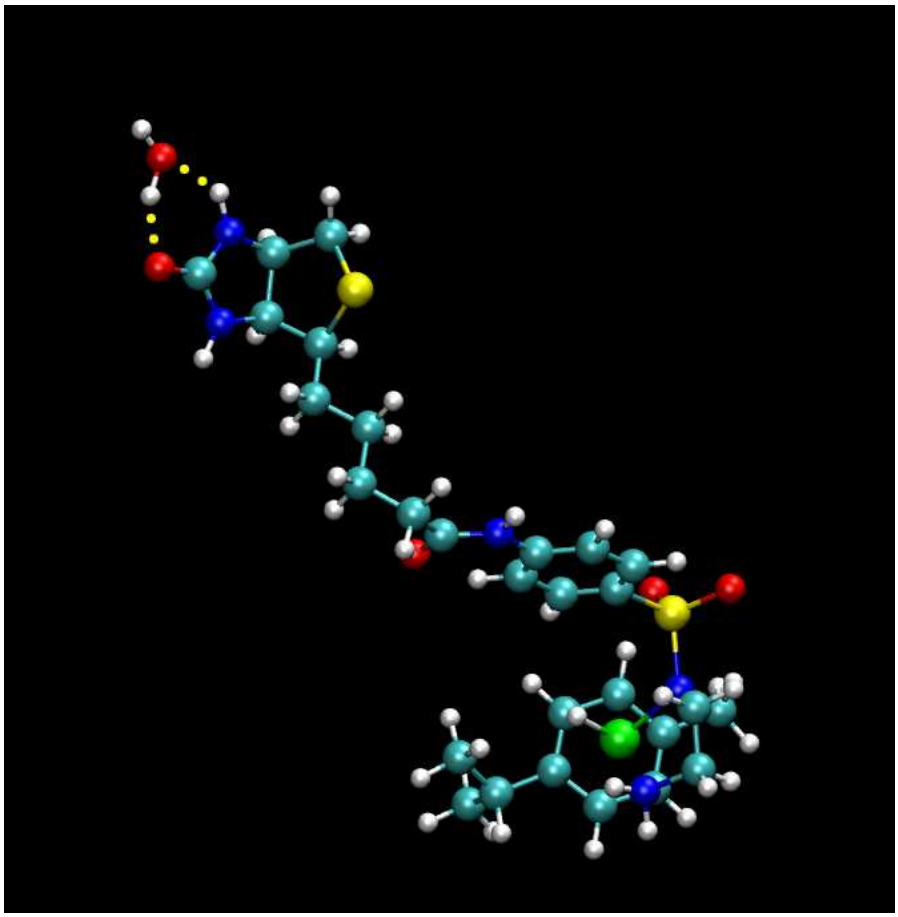
VALIDATION OF THE COFACTOR PARAMETERS AGAINST DFT AND MP2 DATA: STRUCTURES AND ENERGIES OF COFACTOR-WATER MOLECULE COMPLEXES WITH HYDROGEN BONDS

Note to the validation:

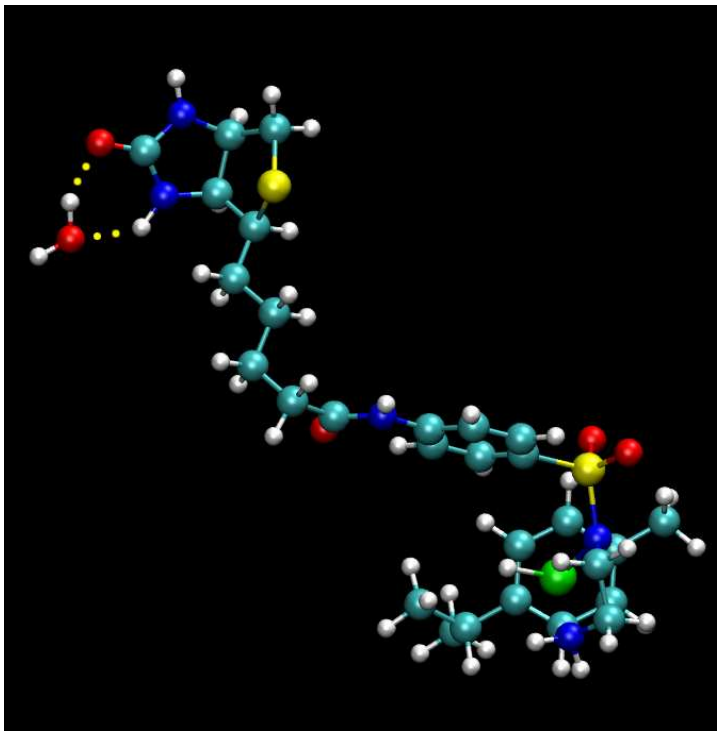
- Only those bond and angle parameters were created form the QM data, which were missing from the GAFF extension to the standard AMBER ff99SB force field. Dihedrals were adapted from the GAFF force field by analogy. Properties of the AMBER suite necessitated substituting iron (Fe symbol) instead of ruthenium in the topology files.
- The tables and figures below show different contacts between probe molecule (water) and the metal-bearing cofactor. Interaction energies were calculated at the B3LYP/SDDAll, MP2/SDDAll quantum-chemical levels and at the classical force field level. Donor-acceptor distances are given in Å, and the energies in kcal/mol.

TEST 1 – WATER AND UREIDO MOIETY, SIDE 1



ENERGY			DFT		AMBER	
B3LYP	MP2	AMBER	OW-H...O	N-H...OW	OW-H...O	N-H...OW
-15.18	-14.70	-10.49	2.669	2.764	2.694	2.930

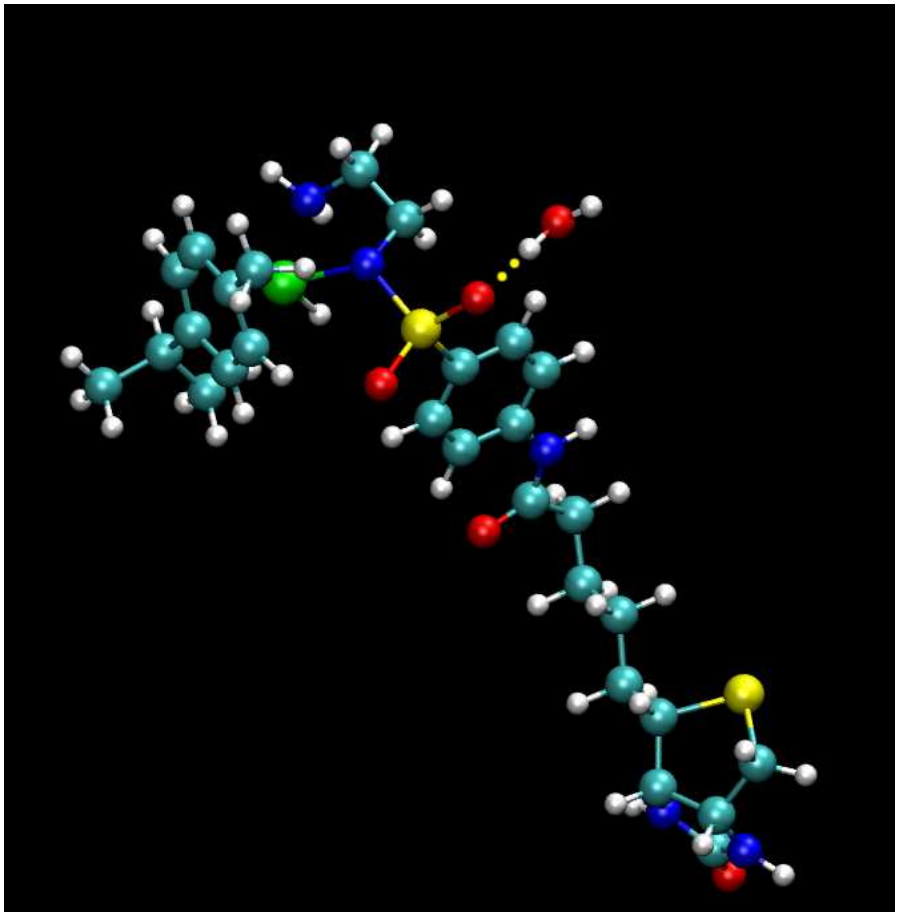
TEST 2 – WATER AND UREIDO MOIETY, SIDE 2



ENERGY			DFT		AMBER	
B3LYP	MP2	AMBER	OW-H...O	N-H...OW	OW-H...O	N-H...OW
-15.55	-14.98	-12.03	2.665	2.765	2.693	2.946

Donor-acceptor distances are given in Å, and the energies in kcal/mol.

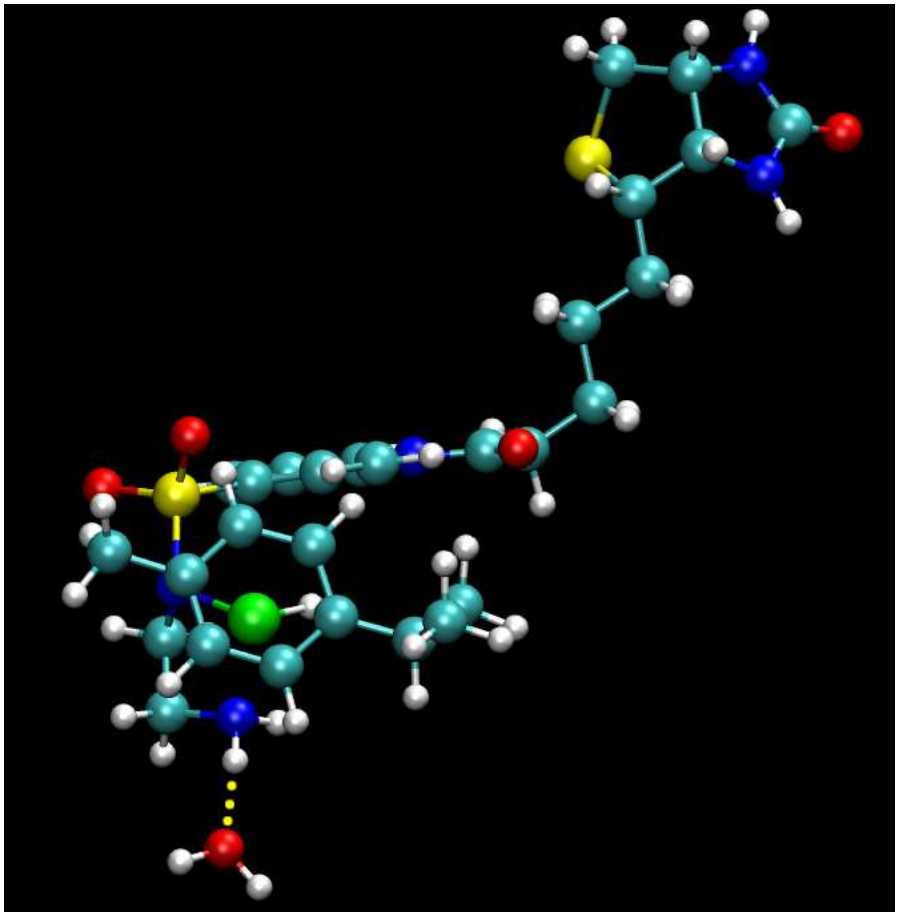
TEST 3 – WATER AND $–SO_2–$ MOIETY



ENERGY			DFT	AMBER
B3LYP	MP2	AMBER	OW-H...O=S	OW-H...O=S
-13.20	-15.11	-9.82	2.668	2.670

Donor-acceptor distances are given in Å, and the energies in kcal/mol.

TEST 4 – WATER AND $–NH_2–$ MOIETY AT THE METAL



ENERGY			DFT	AMBER
B3LYP	MP2	AMBER	OW-H...O=S	OW-H...O=S
-7.51	-8.94	-8.50	2.964	2.994

Donor-acceptor distances are given in Å, and the energies in kcal/mol.

SHORT ANALYSIS OF THE TESTS

The tests indicate that the force field reproduces very well structures of the hydrogen-bonded complexes with water, if we take into account limitations of the classical force fields. While O···O bonds are reproduced indeed well, those involving nitrogen are too long by ca. 0.2Å. Interaction energy values also indicate that the classical force field reproduces the strength of hydrogen bonding at least semi-quantitatively. MP2 calculations show that the DFT level used in this study provides sufficient accuracy to model the studied hydrogen bonds.