

Supplementary material

Figure S1 – Analysis of the parameters from duplicates of 20 ns molecular dynamic simulations of the structural models of the complexes formed with HST (A) and HSD (B) determined by AutoDock program. At the top, the RMSDs of non-hydrogen atoms of the flavonoids (black and green line) flavonoids and backbone atoms of the M₂₋₁ protein (red and blue line) from the starting structure (AutoDock model) for the complexes are reported as a function of time. In the middle, number of contacts between atoms of M₂₋₁ and flavanones for distances < 0.6 nm. At the bottom, distance from Zn²⁺ atom in the zinc-finger domain of the protein to the rigid root of the flavanones.

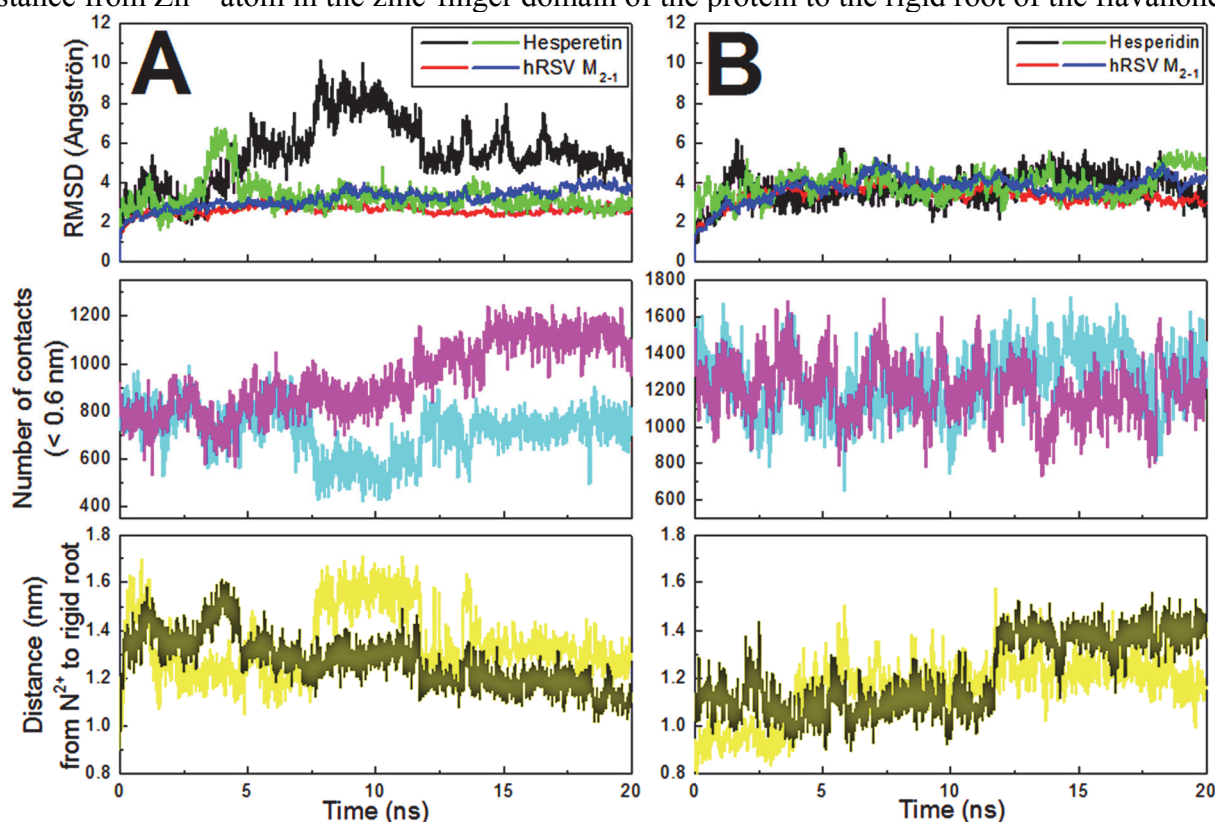


Figure S2 – Comparison of the positions of HST (A) and HSD (B) before and after MD simulations for the structural models of the M₂-1/flavonoid complexes. The protein is denoted as a cartoon and the flavonoids as sticks. The green color depicts the position before MD, while cyan and magenta indicate the positions after 20 ns simulations (duplicate). The spheres represent the Zn²⁺ atom in the zinc finger domain of the protein.

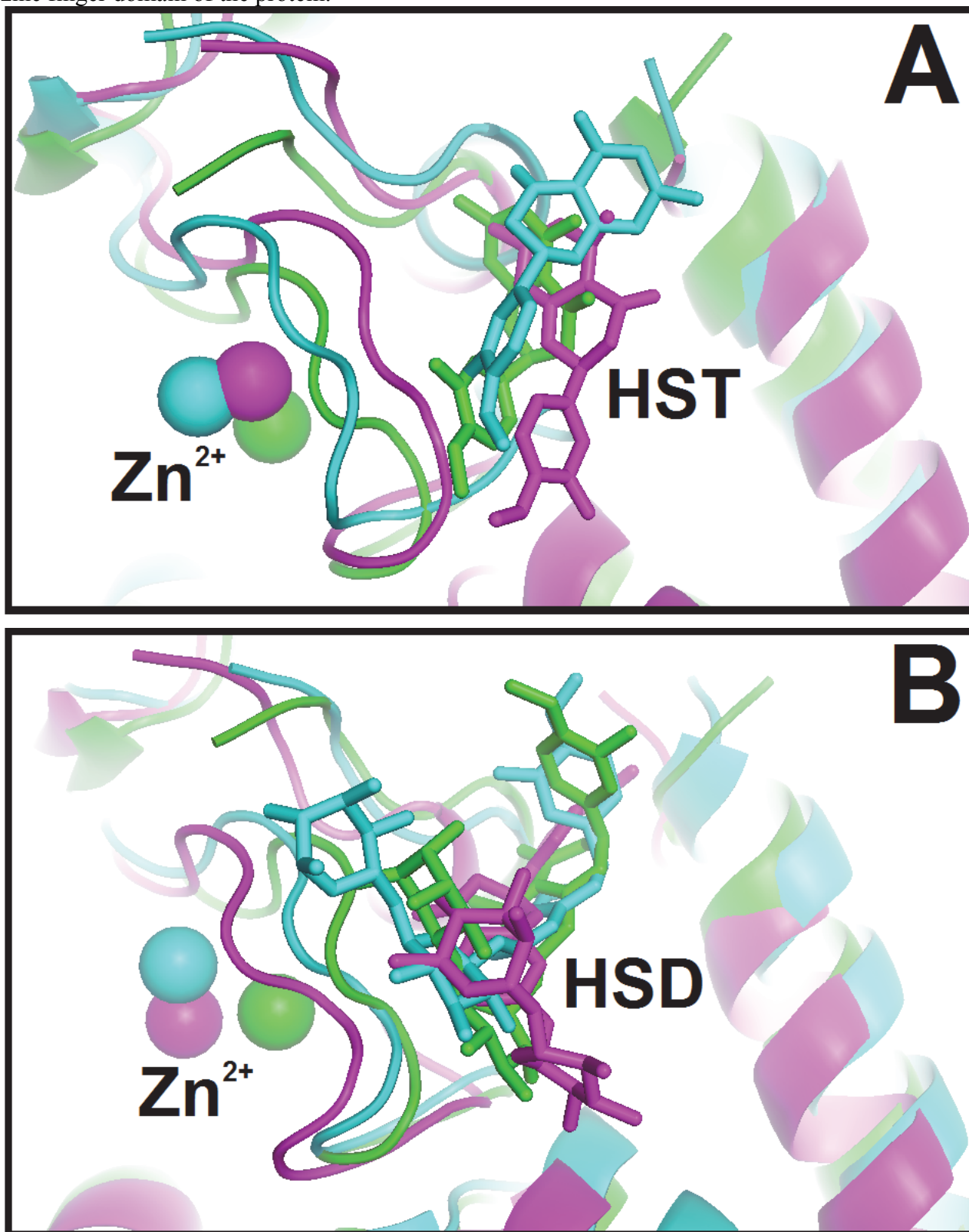


Figure S3 – RMSD values of non-hydrogen atoms of the flavonoids (black line) flavonoids and backbone atoms of the M₂₋₁ protein (red line) calculated from 80 ns MD simulation of the structural models of the complexes formed with HST (A) and HSD (B) determined by AutoDock program.

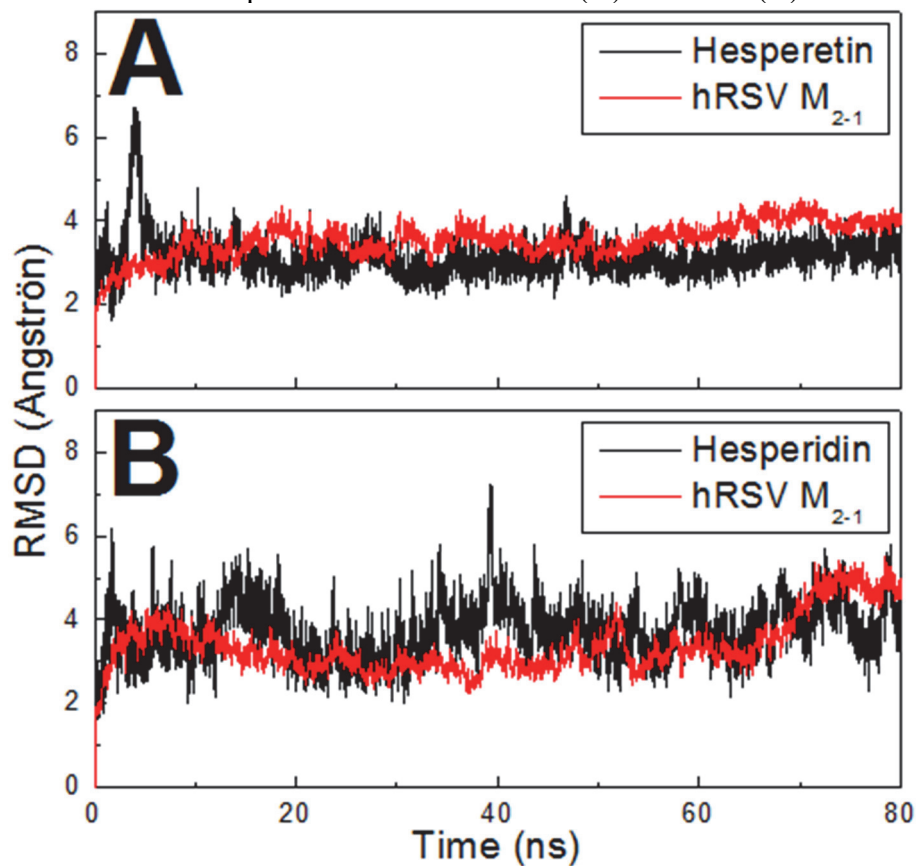


Figure S4: The lowest energy pose from the largest cluster for HST (A) and HSD (B) determined by blind docking (blue) and final docking with reduced grid box (green). The orientation of AMP molecule (yellow) in the binding site of the M₂₋₁ protein was obtained from PDB ID 4CS9 [1]. The protein is represented as a cartoon with the monomers colored in different colors (cyan and gray). The flavonoids and AMP are denoted as sticks and the zinc atom as magenta spheres.

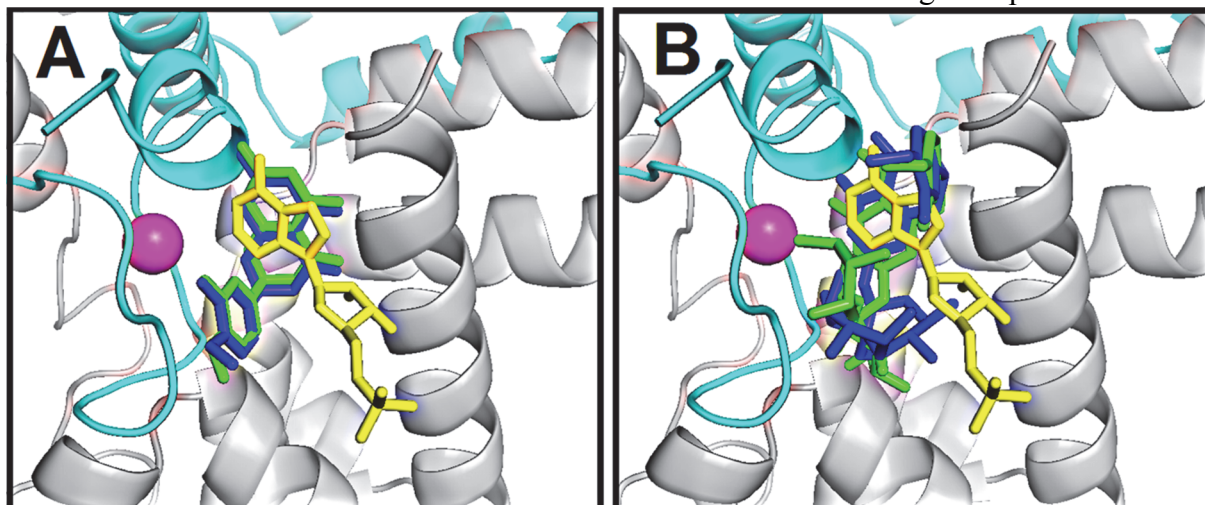


Table S1 – The percentage occupancies (%Occ.) of hydrogen bonds formed between M₂₋₁ and HST during 20ns MD trajectory were obtained for the simulation duplicates. The amino acid residues of the protein involved in hydrogen bonds with average values (%Ave.) of accumulated percentage occupancy higher than 10% (bold) were considered as a significant non-covalent interaction for the molecular stabilization of the M₂₋₁/HST complex.

First MD simulation				Second MD simulation				>10%				
Donor ^a	Acceptor ^a		%Occ	%Acc. _b	Donor ^a	Acceptor ^a		%Occ	%Acc. _b	Res. ^c	Atm. _d	%Ave
K16 ₉	NZ	HST O3	0.55	0.7	K16 ₉	NZ	HST O4	1.549	27.036	K8	N	70.315
K16 ₉	NZ	HST O2	0.15		K16 ₉	NZ	HST O3	25.287		N17₄	O2	69.815
N17 ₄	N	HST O4	3.998	11.794	K16 ₉	NZ	HST O2	0.2	22.789	N17	ND2	26.762
N17 ₄	N	HST O3	7.796		N17 ₄	N	HST O4	2.499		K8	NZ	25.812
N17 ₄	ND ₂	HST O4	0.2	44.428	R4	NH ₁	HST O3	0.2	25.737	R20	O	14.643
N5	ND ₂	HST O5	7.596		R4	NH ₂	HST O3	0.15		K16₉	NZ	13.868
K8	N	HST O5	6.097	51.174	K8	N	HST O5	96.202	6.947			
K8	N	HST O4	0.25		K8	NZ	HST O4	0.45				
K8	N	HST O3	32.484	6.447	F9	N	HST O5	0.1	9.845			
K8	N	HST O6	5.597		N17	ND ₂	HST O1	0.15				
K8	NZ	HST O5	0.25	30.735	N17	ND ₂	HST O2	22.639	3.199			
K8	NZ	HST O4	29.135		R20	NE	HST O1	3.548				
K8	NZ	HST O3	21.789	1.05	R20	NE	HST O2	22.189				
F9	N	HST O4	0.1		R20	NH ₁	HST O1	0.35				
F9	N	HST O3	6.347	1.0	R20	NH ₁	HST O2	6.597				
N17	ND ₂	HST O6	0.4		R20	NH ₂	HST O1	1.099				
N17	ND ₂	HST O1	0.05	0.05	R20	NH ₂	HST O2	8.746				
N17	ND ₂	HST O2	30.285		H22	N	HST O1	0.45				
R20	NH ₂	HST O1	0.05	1.0	H22	N	HST O2	2.749				
H22	N	HST O1	0.05		H22	NE2	HST O3	0.05				
H22	N	HST O2	1.0	89.455	HST	O4	N17 ₄ O1	1.0				
H22	NE2	HST O2	0.15		HST	O4	N17 ₄ O2	89.455				
F23	N	HST O2	0.5	9.895	HST	O2	E81 OE1	30.385				
HST	O5	I173 O	0.05		HST	O2	E81 OE2	9.895				
HST	O5	N5 OD ₁	0.5	1.949	HST	O2	N17 OD ₁	0.5				
HST	O5	N5 O	5.547		HST	O2	R20 O	1.949				
HST	O5	P6 O	0.55	9.345	HST	O2	H22 ND ₁	9.345				
HST	O4	N17 ₄ O1	11.894									
HST	O4	N17 ₄ O2	50.175									
HST	O2	N17 OD ₁	6.797									
HST	O2	N17 O	0.05									

HST	O2	G18	O	0.05
HST	O2	R20	O	27.336
HST	O2	H22	ND 1	3.348

^a The column on the right denotes the amino acid residue and the other on the left, the atom.

^b %Acc. is the accumulated percentage occupancy of hydrogen bonds for a type atom of a residue.

^c Res. denotes an amino acid residue.

^d Atm. depicts a type atom of a residue.

Table S2 – The percentage occupancies (%Occ.) of hydrogen bonds formed between M₂₋₁ and HSD during 20 ns MD trajectory were obtained for the simulation duplicates. The amino acid residues of the protein involved in hydrogen bonds with average values (%Ave.) of accumulated percentage occupancy higher than 10% (bold) were considered as a significant non-covalent interaction for the molecular stabilization of the M₂₋₁/HSD complex.

First MD simulation				Second MD simulation				>10%		
Donor ^a	Acceptor ^a	%Occ	%Acc. _b	Donor ^a	Acceptor ^a	%Occ	%Acc. _b	Res. _c	Atm. _d	%Ave
K16 ₉	NZ	HSD O4	0.05	K16 ₉	NZ	HSD O15	0.25	R20	O	22.56
N17 ₄	N	HSD O4	4.298	K16 ₉	NZ	HSD O14	0.5	H22	ND1	17.04
R4	NE	HSD O15	0.05	K16 ₉	NZ	HSD O8	1.0	K8	N	14.79
R4	NE	HSD O14	0.1	K16 ₉	NZ	HSD O6	0.05	F9	N	10.49
R4	NE	HSD O13	0.05	N17 ₄	N	HSD O4	1.699			
R4	NH ₁	HSD O15	0.15	N17 ₄	N	HSD O3	1.199			
R4	NH ₁	HSD O14	0.85	N17 ₄	ND ₂	HSD O4	2.149			
R4	NH ₁	HSD O13	0.35	N17 ₄	ND ₂	HSD O3	1.949			
R4	NH ₂	HSD O15	0.05	K8	N	HSD O2	3.998			
R4	NH ₂	HSD O14	0.45	K8	NZ	HSD O4	1.749			
R4	NH ₂	HSD O13	0.1	K8	NZ	HSD O3	0.9			
N5	N	HSD O15	0.1	F9	N	HSD O2	11.294			
N5	ND ₂	HSD O15	0.2	N17	ND ₂	HSD O9	1.199			
N5	ND ₂	HSD O14	0.3	N17	ND ₂	HSD O8	8.796	10.245		
N5	ND ₂	HSD O13	0.05	N17	ND ₂	HSD O6	0.25			

N5	ND 2	HSD	O4	0.7		G18	N	HSD	O9	0.3		
N5	ND 2	HSD	O3	1.449		G18	N	HSD	O8	0.5		0.8
K8	N	HSD	O2	24.338	25.587	R20	N	HSD	O8	0.05		
K8	N	HSD	O4	1.249		R20	NE	HSD	O12	0.75		
K8	NZ	HSD	O4	0.55	1.2	R20	NE	HSD	O11	0.35		
K8	NZ	HSD	O3	0.65		R20	NE	HSD	O10	0.6		
F9	N	HSD	O2	9.695		R20	NE	HSD	O9	0.35		2.1
N17	ND 2	HSD	O8	1.649		R20	NE	HSD	O8	0.05		
N17	ND 2	HSD	O6	0.05	1.699	R20	NH 1	HSD	O15	0.05		
N17	ND 2	HSD	O5	0.1		R20	NH 1	HSD	O12	2.399		
R20	NE	HSD	O10	0.05		R20	NH 1	HSD	O11	3.248		
R20	NE	HSD	O9	0.4	3.299	R20	NH 1	HSD	O10	4.698		8.946
R20	NE	HSD	O8	2.799		R20	NH 1	HSD	O9	0.4		
R20	NE	HSD	O6	0.05		R20	NH 1	HSD	O8	2.799		
R20	NH 1	HSD	O9	0.8		R20	NH 1	HSD	O7	0.05		
R20	NH 1	HSD	O8	3.698	4.998	R20	NH 2	HSD	O12	0.6		
R20	NH 1	HSD	O6	0.5		R20	NH 2	HSD	O10	1.049		2.149
R20	NH 2	HSD	O10	0.05		R20	NH 2	HSD	O9	0.4		
R20	NH 2	HSD	O9	0.45	0.85	R20	NH 2	HSD	O7	0.1		
R20	NH 2	HSD	O8	0.2		H22	N	HSD	O15	5.547		
R20	NH 2	HSD	O6	0.15		H22	N	HSD	O14	1.449		11.094
H22	N	HSD	O13	0.75	1.1	H22	N	HSD	O13	2.099		
H22	N	HSD	O7	0.35		H22	N	HSD	O12	0.35		
H22	NE2	HSD	O10	0.85		H22	N	HSD	O11	0.25		
H22	NE2	HSD	O8	0.1	1.0	H22	N	HSD	O7	1,399		
HSD	O15	H22	NE2	0.05		H22	NE2	HSD	O15	0,95		
F23	N	HSD	O15	0.05		H22	NE2	HSD	O14	0,45		
F23	N	HSD	O13	0.35	2.799	H22	NE2	HSD	O12	1.399		
F23	N	HSD	O5	2.399		H22	NE2	HSD	O11	1.999		4.898
HSD	O15	H22	ND 1	0.3		H22	NE2	HSD	O7	0.05		
HSD	O14	H22	ND 1	0.15		HSD	O13	H22	NE2	0.05		
HSD	O13	H22	ND 1	0.65	17.892	F23	N	HSD	O14	0.15		
HSD	O10	H22	ND 1	15.942		F23	N	HSD	O13	0.05		0.4
HSD	O8	H22	ND 1	0.85		F23	N	HSD	O5	0.2		
HSD	O15	H22	O	1.899		R4	NH 2	HSD	O15	0.15		
HSD	O14	H22	O	0.2	2.899	HSD	O15	H22	ND 1	10.545		
HSD	O13	H22	O	0.8		HSD	O14	H22	ND 1	2.749		16.193
HSD	O15	N5	OD	0.05	5.347	HSD	O13	H22	ND	2.149		

HSD	O14	N5	OD 1	0.1		HSD	O10	H22	ND 1	0.15	
HSD	O4	N5	OD 1	5.197		HSD	O5	H22	ND 1	0.6	
HSD	O13	R20	O	0.15	3.248	HSD	O15	R20	O	0.1	
HSD	O8	R20	O	3.098		HSD	O13	R20	O	0.3	
HSD	O8	N17	OD 1	0.8		HSD	O10	R20	O	0.15	41.83
HSD	O4	I173	O	0.05	HSD	O8	R20	O	6.647		
HSD	O4	N17 4	O1	1.549		HSD	O5	R20	O	34.633	
HSD	O4	N17 4	O2	2.849		HSD	O8	R20	NH 2	0.05	
HSD	O4	N5	O	2.299		HSD	O15	C21	N	0.05	
HSD	O4	P6	O	1.949		HSD	O15	H22	O	0.6	
						HSD	O14	H22	O	0.55	1.5
						HSD	O13	H22	O	0.35	
						HSD	O10	E81	OE1	0.05	
						HSD	O9	E81	OE1	17.191	35.382
						HSD	O8	E81	OE1	18.141	
						HSD	O9	E81	OE2	19.29	40.329
						HSD	O8	E81	OE2	21.039	
						HSD	O9	N17	OD 1	0.25	1.649
						HSD	O8	N17	OD 1	1.399	
						HSD	O8	G18	O	1.399	
						HSD	O4	K16 9	O	0.1	
						HSD	O4	T172	OG 1	0.05	
						HSD	O4	T172	O	4.598	
						HSD	O4	N17 4	O1	0.1	
						HSD	O4	N17 4	O2	3.698	
						HSD	O4	N5	OD 1	0.15	
						HSD	O4	P6	O	0.05	

^a The column on the right denotes the amino acid residue and the other on the left, the atom.

^b %Acc. is the accumulated percentage occupancy of hydrogen bonds for a type atom of a residue.

^c Res. denotes an amino acid residue.

^d Atm. depicts a type atom of a residue.

References

[1] Leyrat, C.; Renner, M.; Harlos, K.; Huiskonen, J.T.; Grimes, J.M. Drastic changes in conformational dynamics of the antiterminator M2-1 regulate transcription efficiency in *Pneumovirinae*. *eLIFE* **2014**, e02674.