

## Supplementary Information

### Identification of fragments binding to SARS-CoV-2 nsp10 reveals ligand-binding sites in conserved interfaces between nsp10 and nsp14/nsp16.

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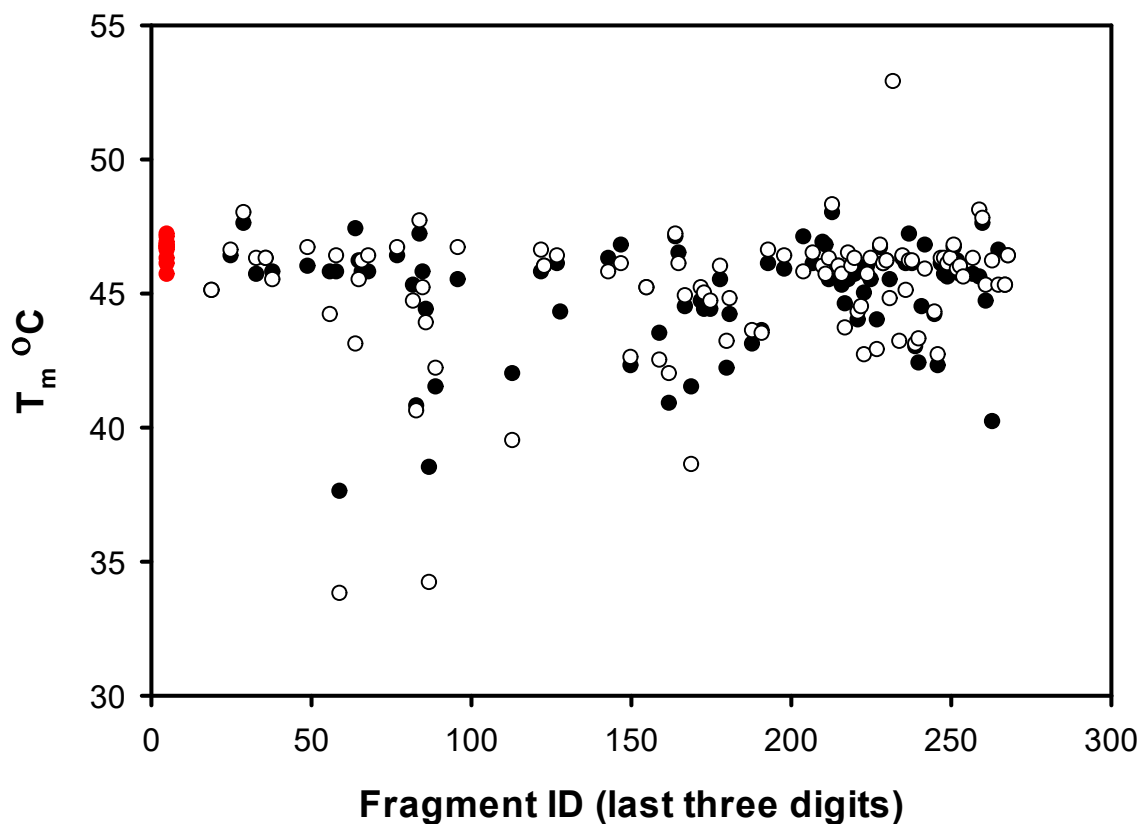
**Table S1. List of fragments from the FragMAX library used in nsp10 nanoDSF experiments.** The  $T_m$  values of nsp10 in both nanoDSF experiments are given (empty in case atypical curves were obtained). The average  $T_m$  of nsp10 without any ligand under the same assay conditions was determined to be  $46.6 \pm 0.4^\circ\text{C}$  ( $n = 11$ ). The two fragments that showed a  $T_m$  equal or above the cut-off of  $47.8^\circ\text{C}$  are marked in yellow. The four fragments that were not tested in XFS are marked in light blue. The four fragments that were co-crystallised with nsp10 are marked in light green.

Fragment ID	SMILE	$T_m$ °C (run 1)	$T_m$ °C (run 2)
VT00019	<chem>O=S1(=O)CC(CN1)C1=CC=CC=C1</chem>	45.1	45.1
VT00022	<chem>N1C=NC(=C1)C1=CC=CC=C1</chem>		
VT00025	<chem>OC1CN(C1)C(=O)C1=CC=CC=C1</chem>	46.4	46.6
VT00029	<chem>OC(=O)CN1CCCCC1=O</chem>	47.6	48.0
VT00033	<chem>CC1=C(C=CC(N)=C1)C(N)=O</chem>	45.7	46.3
VT00034	<chem>NC1=CC2=C(CCOC2)N=C1</chem>		
VT00036	<chem>CC(=O)NC1CCNCC1</chem>	46.3	46.3
VT00038	<chem>O=C1NC=C(C=C1)S(=O)(=O)N1CCCC1</chem>	45.8	45.5
VT00048	<chem>ClC1=CC=C2NC(=O)OC2=C1</chem>		
VT00049	<chem>NC1=CC=NN1CC1=CC=NC=C1</chem>	46.0	46.7
VT00056	<chem>NC1=NC(=CO1)C(F)(F)F</chem>	45.8	44.2
VT00058	<chem>COC1=CC=NC(=C1)C(N)=O</chem>	45.8	46.4
VT00059	<chem>CNC(=O)C1=CC=CS1</chem>	37.6	33.8
VT00064	<chem>COC1=CC(N)=CC(F)=C1</chem>	47.4	43.1

VT00065	CNCC1=NC=CS1	46.2	45.5
VT00066	CN1C=CN=C1C(O)C(F)(F)F	45.8	46.2
VT00068	CC1=NC=C(N1)C(F)(F)F	45.8	46.4
VT00077	CC1=CSC(CC(N)=O)=N1	46.4	46.7
VT00079	COC1=C(C=CC=C1)C(N)=O		
VT00082	OC(=O)C1=CC2=C(NC(=O)C2)C=C1	45.3	44.7
VT00083	CC1=CC=CN2C(N)=CN=C12	40.8	40.6
VT00084	CC1=CC=C(C=C1)S(=O)(=O)N1CCC[C@H]1C(O)=O	47.2	47.7
VT00085	NC1=CC=CC2=NC=CC=C12	45.8	45.2
VT00086	NC(=O)C1CCC2=CC=CC=C2N1	44.4	43.9
VT00087	NC1=NC(=CN=C1)N1CCCC1	38.5	34.2
VT00089	NC(=O)C1=CC2=C(C=NN2)C=C1	41.5	42.2
VT00096	NC1=CC(=NO1)C1CC1	45.5	46.7
VT00113	NC1=CN=C(N=C1)N1CCOCC1	42.0	39.5
VT00118	COC1=CC=CN=C1N		
VT00122	O=C1CC(CN1)C1CC1	45.8	46.6
VT00123	O=C1CC(CN1)C1=CC=NC=C1	46.0	46.0
VT00127	O=C1NN=CN1C1=CC=CC=C1	46.1	46.4
VT00128	OC1=CC=CC(=N1)C(F)(F)F	44.3	
VT00143	NC1=CC(=CC=N1)C#N	46.3	45.8
VT00147	CC(=O)N1CCCCC1CO	46.8	46.1
VT00150	COC1=CC=C(C=N1)C#N	42.3	42.6
VT00154	NCC1=CN2C=CC=CC2=N1		
VT00155	O=C1CCC2=CC=CC3=C2N1CC3	45.2	45.2
VT00159	O1N=CC=C1C1=CC=CC=C1	43.5	42.5
VT00162	N1C2=C(C=CC=C2)N=C1C1=CC=CN=C1	40.9	42.0
VT00164	NC(C1=CC=CN1)C(F)(F)F	47.1	47.2
VT00165	NCC1=CC2=C(CCC2)S1	46.5	46.1
VT00167	NC(=O)C1=C(C=CS1)C1=CC=CC=C1	44.5	44.9
VT00169	CC1=CN2C=C(N)C=CC2=N1	41.5	38.6
VT00172	N#CC1=C2C=CNC2=NC=C1	44.7	45.2
VT00173	NS(=O)(=O)C1=CN=CC=C1	44.4	45.0
VT00175	O=C1NC2=CC=CC=C2C=C1	44.4	44.7
VT00178	NC1CCN(C1=O)C1=CC=CC=C1	45.5	46.0
VT00180	NC(=O)C1=CNC2=CC=CC=C12	42.2	43.2
VT00181	CCS(=O)(=O)NC1=CC=CN=C1	44,2	44,8
VT00188	FC(F)(F)C1=CC(=O)NC=C1	43,1	43,6
VT00190	CC1=NC2=CC=CC=C2N1CCO		
VT00191	COC(=O)C1=C2C=CC=CN2C=N1	43,6	43,5
VT00193	CS(=O)(=O)N1CCOCC1	46,1	46,6
VT00198	CNC(=O)C1=CC=CC=C1	45,9	46,4
VT00204	COC(=O)C1CN(C)C(=O)C1	47.1	45.8
VT00207	COC(=O)CN1C=CC(N)=N1	46.1	46.5
VT00210	CN1CCCCS1(=O)=O	46.9	46.0
VT00211	OCC1CCCCO1	46.8	45.7
VT00212	CC(=O)CC1=NC(=NO1)C1=CC=C(C)C=C1	45.5	46.3

VT00213	<chem>CCC1=NOC(C1)C(O)=O</chem>	48.0	48.3
VT00215	<chem>COC(=O)C1=C(C)NN=C1C</chem>	46.0	46.0
VT00216	<chem>CN1C=CC=C1C(N)=O</chem>	45.3	45.7
VT00217	<chem>O=C1OCCC2=CC=CC=C12</chem>	44.6	43.7
VT00218	<chem>C1CCC2=NN=CN2C1</chem>	45.5	46.5
VT00219	<chem>CN(CC1=CC=CC=C1)C(N)=O</chem>	46.0	46.0
VT00220	<chem>CN1N=C(C=C1C(N)=O)C(F)(F)F</chem>	45.7	46.3
VT00221	<chem>NC1=CC=C2C=CC=CC2=N1</chem>	44.0	44.3
VT00222	<chem>CN(C(C)=O)C1=CC=CC=N1</chem>	45.9	44.5
VT00223	<chem>CN(C)S(=O)(=O)C1=CC=CC=C1F</chem>	45.0	42.7
VT00224	<chem>NC(=O)C1=CC=NC(C1)=C1</chem>	46.0	45.7
VT00225	<chem>CC1=NNC(=C1)C(F)(F)F</chem>	45.5	46.3
VT00226	<chem>NC1=CC=C(C=N1)C(F)(F)F</chem>		
VT00227	<chem>NC1=NC(=CO1)C1=CC=CC=C1</chem>	44.0	42.9
VT00228	<chem>COC1=NC=CC=C1C(O)=O</chem>	46.7	46.8
VT00229	<chem>OCC1=CN(CC2=CC=CC=C2)N=N1</chem>	46.1	46.1
VT00230	<chem>NC1=C(C=NN1)C1=CC=C(F)C=C1</chem>	46.2	46.2
VT00231	<chem>NCC1=CC=CN=C1N1CCCC1</chem>	45.5	44.8
VT00232	<chem>C1=CN(N=C1)C1=CC=CC=N1</chem>		52.9
VT00233	<chem>CNC1=NC=CC=C1OC</chem>		
VT00234	<chem>OCC1=CN=C(N1)C1=CC=CC=C1</chem>		43.2
VT00235	<chem>CC1=CC=C(C1)N=N1</chem>	46.3	46.4
VT00236	<chem>COC(=O)C1=CC=NN=C1</chem>	46.1	45.1
VT00237	<chem>CC(O)C1=NC(C)=CS1</chem>	47.2	46.2
VT00238	<chem>OC1=NOC2=CC=CC=C12</chem>	46.1	46.2
VT00239	<chem>CN(C)C1=CC=NC(N)=C1</chem>	43.0	43.1
VT00240	<chem>N1C=CC2=CC=CN=C12</chem>	42.4	43.3
VT00241	<chem>OC1=CC=C(C1)N=C1</chem>	44.5	
VT00242	<chem>CC1=CC(C)=NC=N1</chem>	46.8	45.9
VT00243	<chem>CN1C=NC2=C(C1)N=CN=C12</chem>		
VT00245	<chem>CC1=CC=C(S1)C1=CC(=NN1)C(O)=O</chem>	44.2	44.3
VT00246	<chem>C1=NN2C=CC=CC2=N1</chem>	42.3	42.7
VT00247	<chem>CC1=NN2CCC(=O)NC2=C1</chem>	46.1	46.3
VT00248	<chem>CS(=O)(=O)N1C=CC=C1</chem>	45.7	46.3
VT00249	<chem>O=C1N[C@@H](CO1)C1=CC=CC=C1</chem>	45.6	46.1
VT00250	<chem>O=C1NC2CCC1CC2</chem>	45.9	46.3
VT00251	<chem>O=C1CNCCN1C1=CC=CC=C1</chem>	46.7	46.8
VT00252	<chem>O=C1NCCN2CCCC12</chem>	46.2	45.9
VT00253	<chem>CC1CCC2(CC(=O)N2)CC1</chem>	45.7	46.0
VT00254	<chem>O=C1NC(=CS1)C1=CC=CN=C1</chem>	45.7	45.6
VT00257	<chem>CNS(=O)(=O)C1=C(C)ON=C1C</chem>	45.7	46.3
VT00258	<chem>COC1=C2NN=CC2=CC=C1</chem>		
VT00259	<chem>COC(=O)C1=NNC2=C1CCCC2</chem>	45.6	48.1
VT00260	<chem>CCS(=O)(=O)N1CCCC(C1)C(O)=O</chem>	47.6	47.8
VT00261	<chem>N#CC1=CC=CN=C1N1CCOCC1</chem>	44.7	45.3
VT00263	<chem>CN1CCCN(C)C1=O</chem>	40.2	46.2
VT00264	<chem>COC1=CC=C2NC(C)=NC2=C1</chem>		

VT00265	<chem>NC1=C2N=CNC2=CC=C1</chem>	46.6	45.3
VT00267	<chem>CN1CCCC1CO</chem>	45.3	45.3
VT00268	<chem>C1=CC=C(C=C1)C2=CSC(=N2)N</chem>	46.4	46.4



**Figure S1. Thermal shift assays of nsp10 in the presence of fragments.** Black circles are from the first experiment and empty circles from the second one. To the left, the  $T_m$  values of nsp10 without any ligand under the assay conditions (average:  $46.6 \pm 0.4^\circ\text{C}$  ( $n=11$ )) are shown as red dots. Fragments that lead to atypical curves (Table S1) are not included in this plot.