

Supplementary Information

Computational Screening of Newly Designed Compounds Against Coxsackievirus A16 and Enterovirus A71

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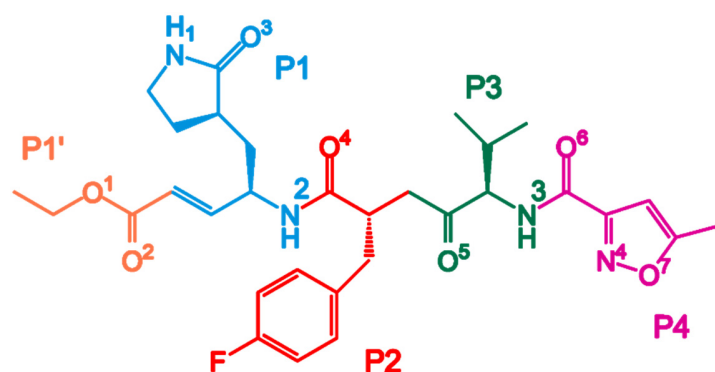
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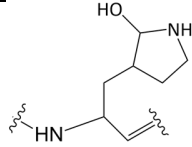
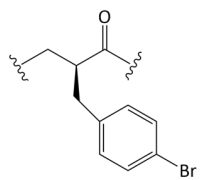
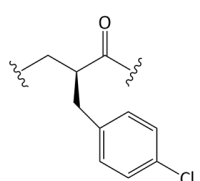
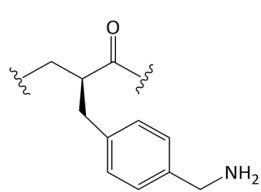
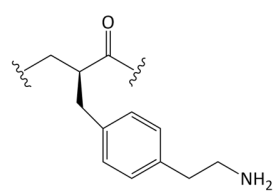
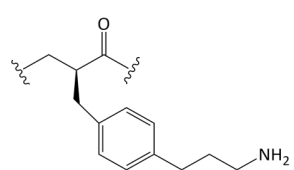
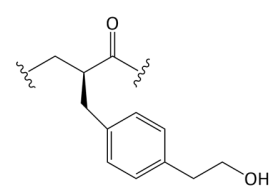
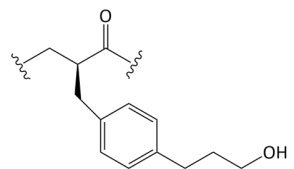
Table of Content

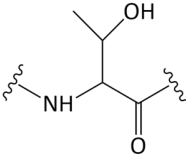
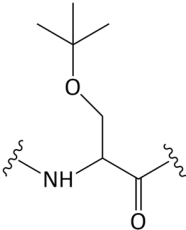
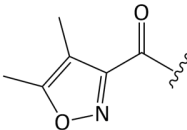
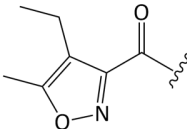
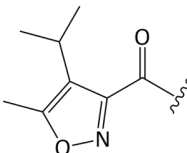
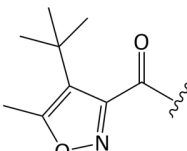
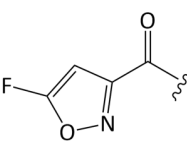
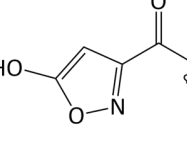
1	Table S1. Chemical structures of rupintrivir and its 50 analogs	S3
2	Table S2 binding free energy based on the MM/PB(GB)SA method for rupintrivir and P2-m3 binding to EV-A71 and CV-A16 3Cpro	S10
3	Figure S1 Rupintrivir in the active site of EV-A71 3Cpro	S10
4	Figure S2 Superimposition of ligand at the binding site derived from the last 50 ns of run1 simulation	S11
5	Figure S3 The solubility of rupintrivir and P2-m3 from ADMET property	S12
6	Figure S4 Sequence alignment of EV-A71 and CV-A16 3C proteases	S12

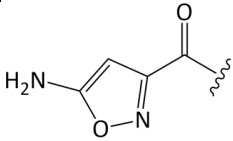
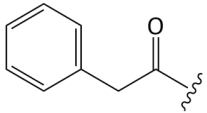
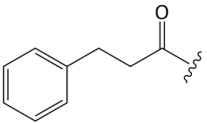
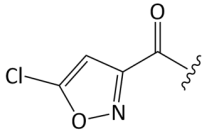
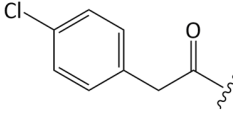
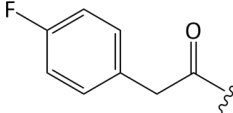
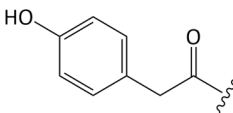
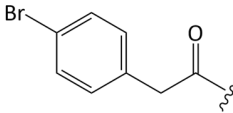
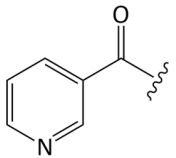
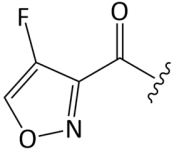
1. Table S1 Chemical structures of rupintrivir and its 50 analogs

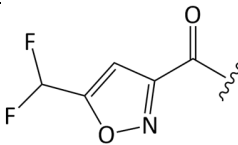
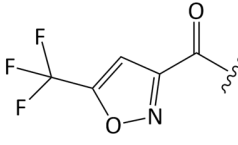
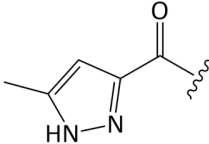


Code	P1'	P1	P2	P3	P4
rupintrivir					
P1'-1					
P1'-2					
P1-1					
P1-2					
P1-3					
P1-4					

Code	P1'	P1	P2	P3	P4
P1-5					
P2-p1					
P2-p2					
P2-p3					
P2-p4					
P2-p5					
P2-p6					
P2-p7					

Code	P1'	P1	P2	P3	P4
P3-3					
P3-4					
P4-1					
P4-2					
P4-3					
P4-4					
P4-5					
P4-6					

Code	P1'	P1	P2	P3	P4
P4-7					
P4-8					
P4-9					
P4-10					
P4-11					
P4-12					
P4-13					
P4-14					
P4-15					
P4-16					

Code	P1'	P1	P2	P3	P4
P4-17					
P4-18					
P4-19					

2. Table S2 Binding free energy based on the MM/PB(GB)SA method of rupintrivir and P2-m3

Compound		EV-A71		CV-A16	
		MM/PBSA	MM/GBSA	MM/PBSA	MM/GBSA
Rupintrivir	Run1	-10.1±1.5	-11.9±0.6	-7.8±0.5	-9.5±0.8
	Run2	-7.1±1.2	-9.1±1.0	-6.0±0.4	-8.5±0.9
	Run3	-6.4±1.0	-8.6±0.7	-8.9±0.9	-11.1±1.2
	Mean±SD	-7.9±2.0	-9.9±1.4	-7.6±1.1	-9.7±1.7
P2-m3	Run1	-17.2±0.6	-17.4±0.5	-11.8±0.3	-13.0±0.4
	Run2	-15.1±0.6	-15.6±0.3	-10.4±0.5	-12.7±0.3
	Run3	-15.0±0.8	-15.4±0.6	-10.1±0.7	-12.9±1.0
	Mean±SD	-15.8±1.2	-16.1±0.8	-10.8±0.9	-12.9±1.1

3. Figure S1 Rupintrivir in the active site of EV-A71 3Cpro

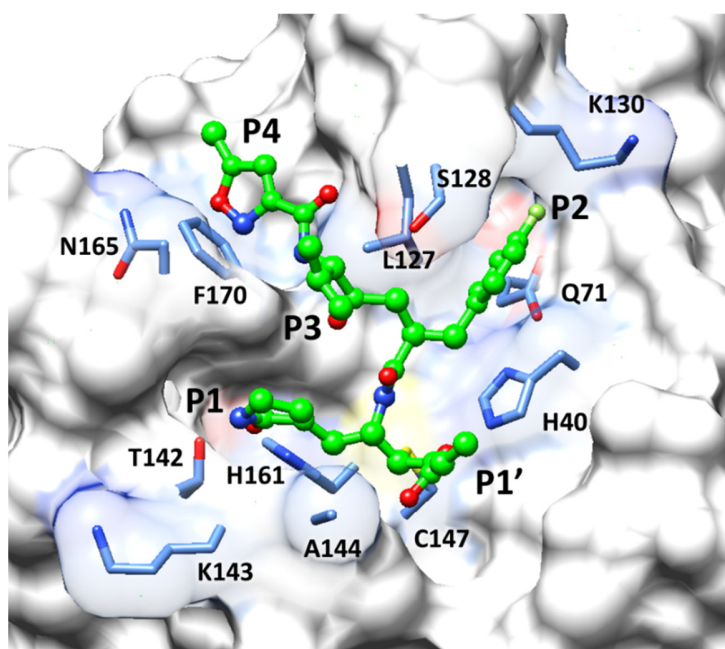


Figure S1 Rupintrivir (bond and stick model) binding at the active site of EV-A71 3Cpro, where its the surrounding residues (stick model) are shown and labeled

4. Figure S2 Superimposition of ligand at the binding site derived from the last 50 ns of run1 MD simulation

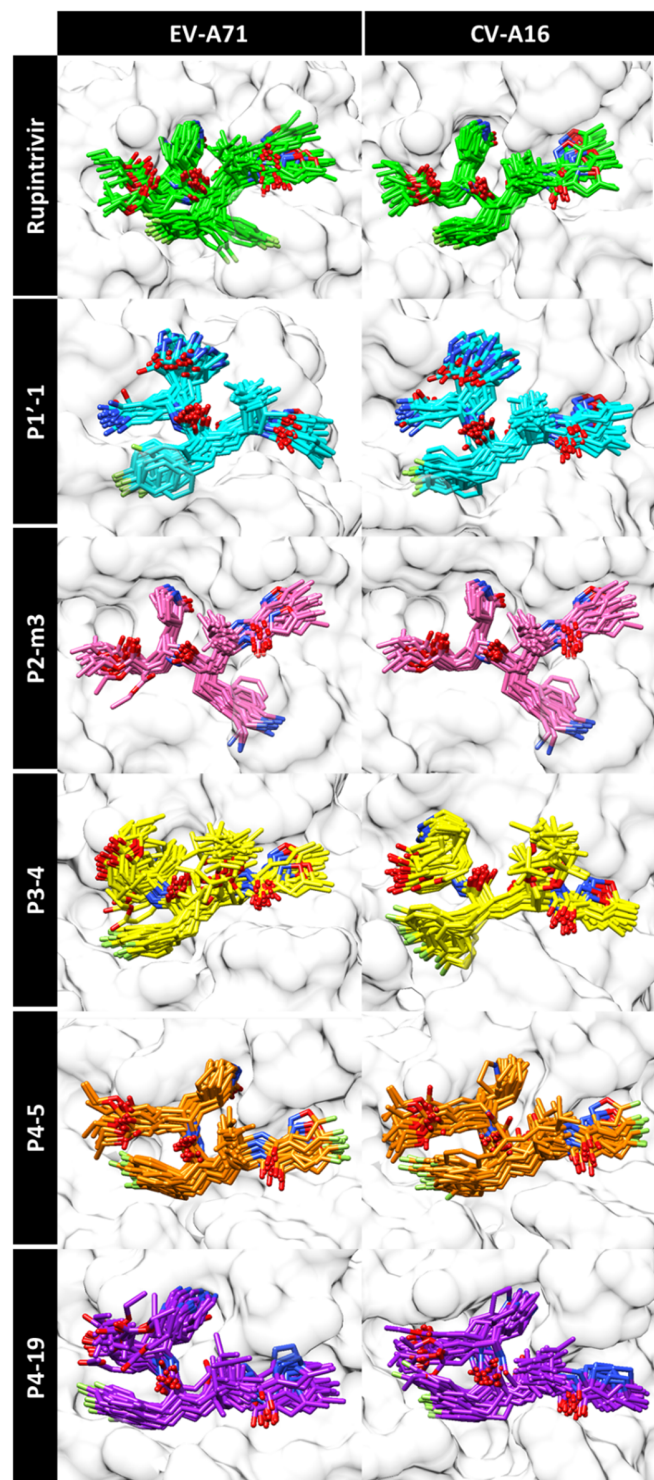


Figure S2 Superimposition of rupintrivir and 5 screened analogs at the binding pocket of EV-A71 and CV-A16 3Cpro derived from the last 50 ns of run1 MD simulation

5. Figure S3. The solubility of rupintrivir and P2-m3 from ADMET property

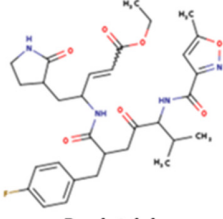
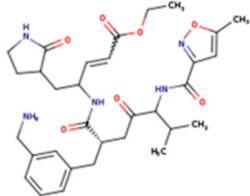
Compound	Solubility
 <p>Rupintrivir</p>	<p>Log S (ESOL) [?] -6.05</p> <p>Solubility 5.36e-04 mg/ml ; 8.95e-07 mol/l</p> <p>Class [?] Poorly soluble</p>
 <p>P2-m3 analog</p>	<p>Log S (ESOL) [?] -3.72</p> <p>Solubility 1.17e-01 mg/ml ; 1.92e-04 mol/l</p> <p>Class [?] Soluble</p>

Figure S3. The solubility from ADMET property of rupintrivir and P2-m3. P2-m3 showed better solubility than rupintrivir

6. Figure S4. Sequence alignment of EV-A71 and CV-A16 3C proteases

Consensus	1	11	21	31	41
Conservation	- - - M G P S L D F	A L S L L R R N i R	Q V Q T D Q G H F T	M L G V R D R L A i	L P R H S Q P G K T
3R0F.pdb	G S H M G P L D F	A L S L L R R N V R	Q V Q T D Q G H F T	M L G V R D R L A V	L P R H S Q P G K T
3SJI.pdb	- - - M G P S L D F	A L S L L R R N I R	Q V Q T D Q G H F T	M L G V R D R L A I	L P R H S Q P G K T
Consensus	51	61	71	81	91
Conservation	I W i E H K L i N V	L D A V E L V D E Q	G V N L E L T L i T	L D T N E K F R D i	T K F I P E t I s t
3R0F.pdb	I W I E H K L V N V	L D A V E L V D E Q	G V N L E L T L I T	L D T N E K F R D I	T K F I P E N I S T
3SJI.pdb	I W V E H K L I N V	L D A V E L V D E Q	G V N L E L T L V T	L D T N E K F R D V	T K F I P E T I T G
Consensus	101	111	121	131	141
Conservation	A S D A T L i I N T	E H M P S M F V P V	G D V V Q Y G F L N	L S G K P T h R T M	M Y N F P T K A G Q
3R0F.pdb	A S D A T L V I N T	E H M P S M F V P V	G D V V Q Y G F L N	L S G K P T G R T M	M Y N F P T K A G Q
3SJI.pdb	A S D A T L I I N T	E H M P S M F V P V	G D V V Q Y G F L N	L S G K P T H R T M	M Y N F P T K A G Q
Consensus	151	161	171	181	191
Conservation	C G G V V T S V G K	I I G I H I G G N G	R Q G F C A G L K R	s Y F A S E Q - - -	- - -
3R0F.pdb	C G G V V T S V G K	I I G I H I G G N G	R Q G F C A G L K R	S Y F A S E Q - - -	- - -
3SJI.pdb	C G G V V T S V G K	I I G I H I G G N G	R Q G F C A G L K R	G Y F A S E Q H H H H H H	- - -

Figure S4. Sequence alignment of EV-A71 (PDB code: 3SJI) and CV-A16 (PDB code: 3R0F) proteases