

Supplemental Information

Simultaneously Targeting the NS3 Protease And Helicase Activities For More Effective Hepatitis C Virus Therapy

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Figure S1: Alignment of the various HCV genotypes used in this study. The sequence of each replicon used was aligned with the sequence of the recombinant scNS4A-NS3 construct previously used for crystallization (PDB file 1CU1). Replicon sequences were aligned using Clustal W, and the result was aligned with the 1CU1 sequence using UCSF Chimera. Numbers refer to positions in the NS3 protein. Conserved amino acids are colored by chemical class, and non-conserved residues are colored black. Amino acids unique to genotype 2a are highlighted in yellow in the 2a sequence, and they are highlighted as spheres in Figure 2D. Amino acids within 5Å of the putative HPI binding-site are highlighted in green on the 1CU1 sequence.

1b(con1) - - - - - I T A Y S Q Q T R G L L G C I I T S L T G R D R N Q V E G E V Q V V 36
2a(JFH-1) - - - - - I T A Y A Q Q T R G L L G A I V V S M T G R D R T E Q A G E V Q I L 36
3a_S52 - - - - - I T A Y A Q Q T R G L L G T I V T S L T G R D K N I V T G E V Q V L 36
4a_ED43 - - - - - I T A Y A Q Q T R G L F S T I V T S L T G R D T N E N C G E V Q V L 36
1CU1_monomer_noH.pdb G S V V I V G R I I L S G S G S I T A Y S Q Q T R G L L G C I I T S L T G R D K N Q V E G E V Q V V 36

1b(con1) S T A T Q S F L A T C V N G V C W T V Y H G A G S K T L A G P K G P I T Q M Y T N V D Q D L V G W Q 86
2a(JFH-1) S T V S Q S F L G T T I S G V L W T V Y H G A G N K T L A G L R G P V T Q M Y S S A E G D L V G W P 86
3a_S52 S T A T Q T F L G T T V G G V M W T V Y H G A G S K T L A G A K H P A L Q M Y T N V D Q D L V G W P 86
4a_ED43 S T A T Q S F L G T A V N G V M W T V Y H G A G A K T I S G P K G P V N Q M Y T N V D Q D L V G W P 86
1CU1_monomer_noH.pdb S T A S F L A T C V N G V C W T V Y H G A G S K T L A G P K G P I T Q M Y T N V D Q D L V G W Q 86

1b(con1) A P P G A R S L T P C T C G S S D L Y L V T R H A D V I P V R R R G D S R G S L L S P R P V S Y L K 136
2a(JFH-1) S P P G T K S L E P C K C G A V D L Y L V T R N A D V I P A R R R G D K R G A L L S P R P I S T L K 136
3a_S52 A P P G A K S L E P C A C G S A D L Y L V T R D A D V I P A R R R G D S T A S L L S P R P L A C L K 136
4a_ED43 A P P G V R S L A P C T C G S A D L Y L V T R H A D V I P V R R R G D T R G A L L S P R P I S T L K 136
1CU1_monomer_noH.pdb A P P G A R S L T P C T C G S S D L Y L V T R H A D V I P V R R R G D S R G S L L S P R P V S Y L K 136

1b(con1) G S S G G P L L C P S G H A V G I F R A A V C T R G V A K A V D F V P V E S M E T T M R S P V F T D 186
2a(JFH-1) G S S G G P V L C P R G H V V G L F R A A V C S R G V A K S I D F I P V E T L D V V T R S P T F S D 186
3a_S52 G S S G G P V M C P S G H V A G I F R A A V C T R G V A K A L Q F V P V E T L S T Q A R S P S F S D 186
4a_ED43 G S S G G P L L C P M G H A A G I F R A A V C T R G V A K A V D F V P V E S L E T T M R S P V F T D 186
1CU1_monomer_noH.pdb G S S G G P L L C P S G H A V G I F R A A V C T R G V A K A V D F V P V E S M E T T M R S P V F T D 186

1b(con1) N S S P P A V P Q T F Q V A H L H A P T G S G K S T K V P A A Y A A Q G Y K V L V L N P S V A A T L 236
2a(JFH-1) N S T P P A V P Q T Y Q V G Y L H A P T G S G K S T K V P V A Y A A Q G Y K V L V L N P S V A A T L 236
3a_S52 N S T P P A V P Q S Y Q V G Y L H A P T G S G K S T K V P A A Y V A Q G Y N V L V L N P S V A A T L 236
4a_ED43 N S T P P A V P Q T Y Q V A H L H A P T G S G K S T K V P A A Y A A Q G Y K V L V L N P S V A A T L 236
1CU1_monomer_noH.pdb N S S P P A V P Q S F Q V A H L H A P T G S G K S T K V P A A Y A A Q G Y K V L V L N P S V A A T L 236

1b(con1) G F G A Y M S K A H G I D P N I R T G V R T I T T G A P I T Y S T Y G K F L A D G G C S G G A Y D I 286
2a(JFH-1) G F G A Y L S K A H G I N P N I R T G V R T V M T G E A I T Y S T Y G K F L A D G G C A S G A Y D I 286
3a_S52 G F G S F M S R A Y G I D P N I R T G N R T V T T G A K L T Y S T Y G K F L A D G G C S G G A Y D V 286
4a_ED43 G F G V Y M S K A Y G I D P N I R S G V R T I T T G A P I T Y S T Y G K F L A D G G C S G G A Y D I 286
1CU1_monomer_noH.pdb G F G A Y M S K A H G I D P N I R T G V R T I T T G A P V T Y S T Y G K F L A D G G C S G G A Y D I 286

1b(con1) I I C D E C H S T D S T T I L G I G T V L D Q A E T A G A R L V V L A T A T P P G S V T V P H P N I 336
2a(JFH-1) I I C D E C H A V I D A T S I L G I G T V L D Q A E T A G V R L T V L A T A T P P G S V T T P H P D I 336
3a_S52 I I C D E C H A Q D A T S I L G I G T V L D Q A E T A G V R L T V L A T A T P P G S I T V P H S N I 336
4a_ED43 I I C D E C H S T D S T T I L G I G T V L D Q A E T A G V R L T V L A T A T P P G S V T T P H S N I 336
1CU1_monomer_noH.pdb I I C D E C H S T D S T T I L G I G T V L D Q A E T A G A R L V V L A T A T P P G S V T V P H P N I 336

1b(con1) E E V A L S S T G E I P F Y G K A I P I E T I K G G R H L I F C H S K K K C D E L A A K L S G L G L 386
2a(JFH-1) E E V G L G R E G E I P F Y G R A I P L S C I K G G R H L I F C H S K K K C D E L A A A L R G M G L 386
3a_S52 E E V A L G S E G E I P F Y G K A I P I A L L K G G R H L I F C H S K K K C D E V A A K L R G M G L 386
4a_ED43 E E V A L P T T G E I P F Y G K A I P L E L I K G G R H L I F C H S K K K C D E L A R Q L T S L G L 386
1CU1_monomer_noH.pdb E E V A L S N T G E I P F Y G K A I P I E A I R G G R H L I F C H S K K K C D E L A A K L S G L G I 386

1b(con1) N A V A Y Y R G L D V S V I P T S G D V I V V A T D A L M T G F T G D F D S V I D C N T C V T Q T V 436
2a(JFH-1) N A V A Y Y R G L D V S I I P A Q G D V V V V A T D A L M T G Y T G D F D S V I D C N V A V T Q A V 436
3a_S52 N A V A Y Y R G L D V S V I P T T G D V V V C A T D A L M T G F T G D F D S V I D C N V A V E Q Y V 436
4a_ED43 N A V A Y Y R G L D V S V I P T S G D V V V C A T D A L M T G F T G D F D S V I D C N T S V I Q T V 436
1CU1_monomer_noH.pdb N A V A Y Y R G L D V S V I P T I G D V V V V A T D A L M T G Y T G D F D S V I D C N T S V I Q T V 436

1b(con1) D F S L D P T F T I E T T T V P Q D A V S R S Q R R R G T G R G R M G I Y R F V T P G E R P S G M F 486
2a(JFH-1) D F S L D P T F T I T T Q T V P Q D A V S R S Q R R R G T G R G R Q G T Y R Y V S T G E R A S G M F 486
3a_S52 D F S L D P T F S I E T R T A P Q D A V S R S Q R R R G T G R G R L G T Y R Y V A P G E R P S G M F 486
4a_ED43 D F S L D P T F S I E T T T V P Q D A V S R S Q R R R G T G R G R L G T Y R Y V T P G E R P S G M F 486
1CU1_monomer_noH.pdb D F S L D P T F T I E T T T V P Q D A V S R S Q R R R G T G R G R R G I Y R F V T P G E R P S G M F 486

1b(con1) D S S V L C E C Y D A G C A W Y E L T P A E T S V R L R A Y L N T P G L P V C Q D H L E F W E S V F 536
2a(JFH-1) D S V V L C E C Y D A G A A W Y D L T P A E T T V R L R A Y F N T P G L P V C Q D H L E F W E A V F 536
3a_S52 D S V V L C E C Y D A G C S W Y D L Q P A E T T V R L R A Y L N T P G L P V C Q D H L D F W E S V F 536
4a_ED43 D T A V L C E C Y D A G C A W Y E L T P A E T T T R L K A Y F D T P G L P V C Q D H L E F W E S V F 536
1CU1_monomer_noH.pdb D S S V L C E C Y D A G C A W Y E L T P A E T S V R L R A Y L N T P G L P V C Q D H L E F W E S V F 536

1b(con1) T G L T H I D A H F L S Q T K Q A G D N F P Y L V A Y Q A T V C A R A Q A P P P S W D Q M W K C L I 586
2a(JFH-1) T G L T H I D A H F L S Q T K Q A G E N F A Y L V A Y Q A T V C A R A K A P P P S W D A M W K C L A 586
3a_S52 T G L T H I D A H F L S Q T K Q Q G L N F S F L T A Y Q A T V C A R A Q A S P P S W D E T W K C L V 586
4a_ED43 T G L T H I D G H F L S Q T K Q S G E N F P Y L V A Y Q A T V C A K A L A P P P S W D T M W K C L I 586
1CU1_monomer_noH.pdb T G L T H I D A H F L S Q T K Q A G D N F P Y L V A Y Q A T V C A R A Q A P P P S W D Q M W K C L I 586

1b(con1) R L K P T L H G P T P L L Y R L G A V Q N E V T T T H P I T K Y I M A C M S A D L E V V T 631
2a(JFH-1) R L K P T L A G P T P L L Y R L G P I T N E V T L T H P G T K Y I A T C M Q A D L E V M T 631
3a_S52 R L K P T L H G P T P L L Y R L G P V Q N D I C L T H P V T K Y I M A C M S A D L E V T T 631
4a_ED43 R L K P T L H G P T P L L Y R L G S V Q N E V V L T H P I T K Y I M A C M S A D L E V V T 631
1CU1_monomer_noH.pdb R L K P T L H G P T P L L Y R L G A V Q N E V T L T H P I T K Y I M A C M S A D L E V V T 631