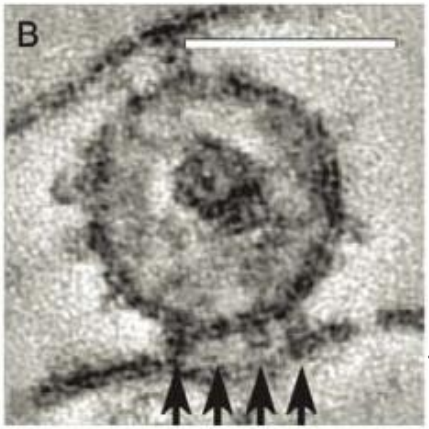
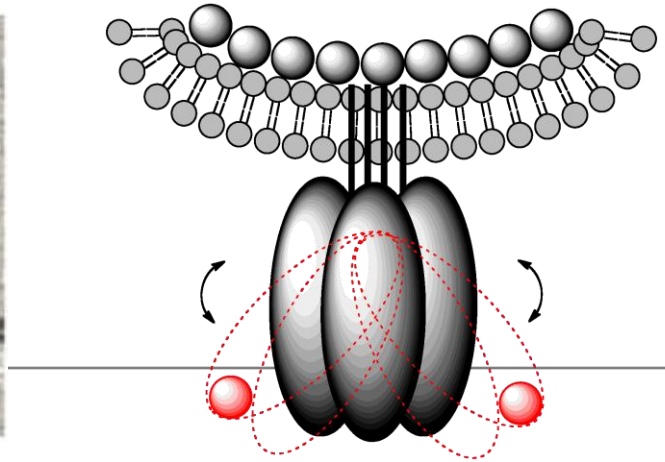
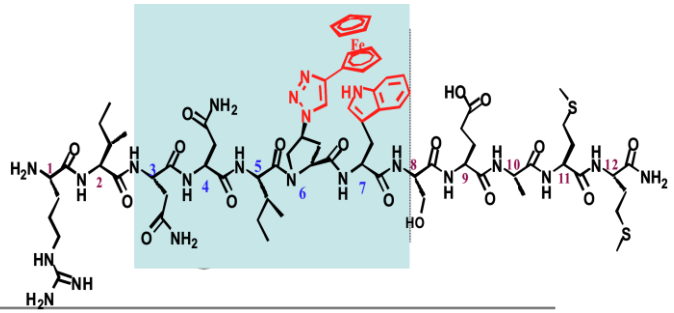
Attacking the HIV-1 Entry Machine

MECHANISM OF VIROLYSIS INDUCED BY PEPTIDE TRIAZOLE THIOLS



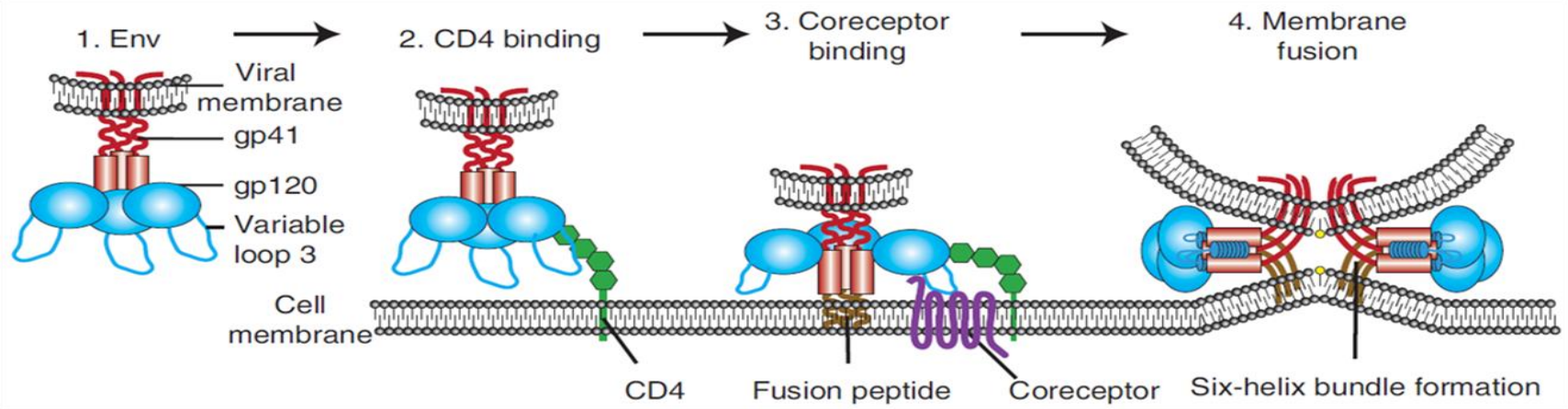




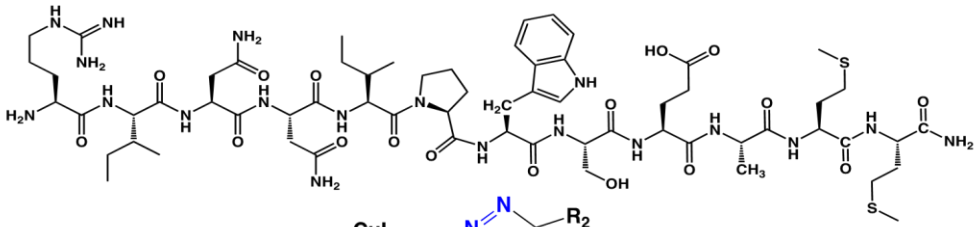
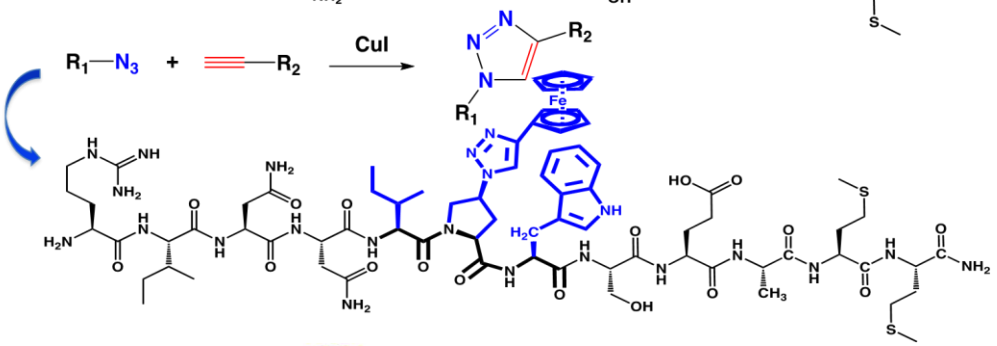
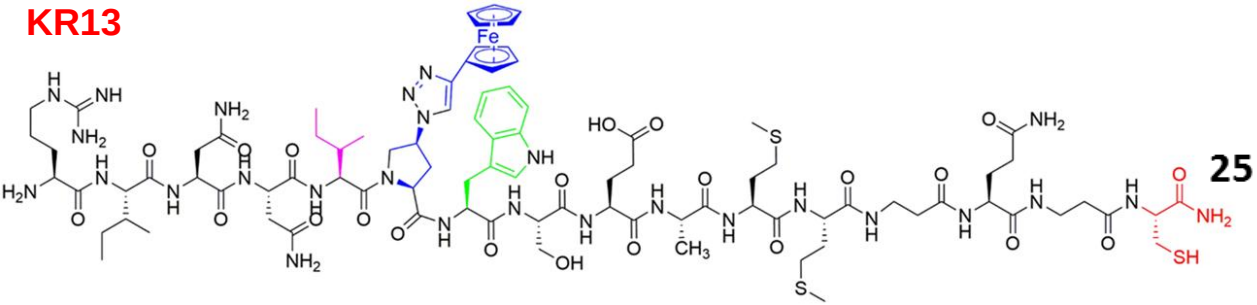
Drexel UNIVERSITY

HIV-1 Uses Dual Receptor Encounter and Metastability To Effect Cell Entry and Infection

<u>Can the Two Host Cell Receptors Be Suppressed Coordinately?</u>



Origin of Peptide Triazole HIV-1 Env gp120 Inhibitors

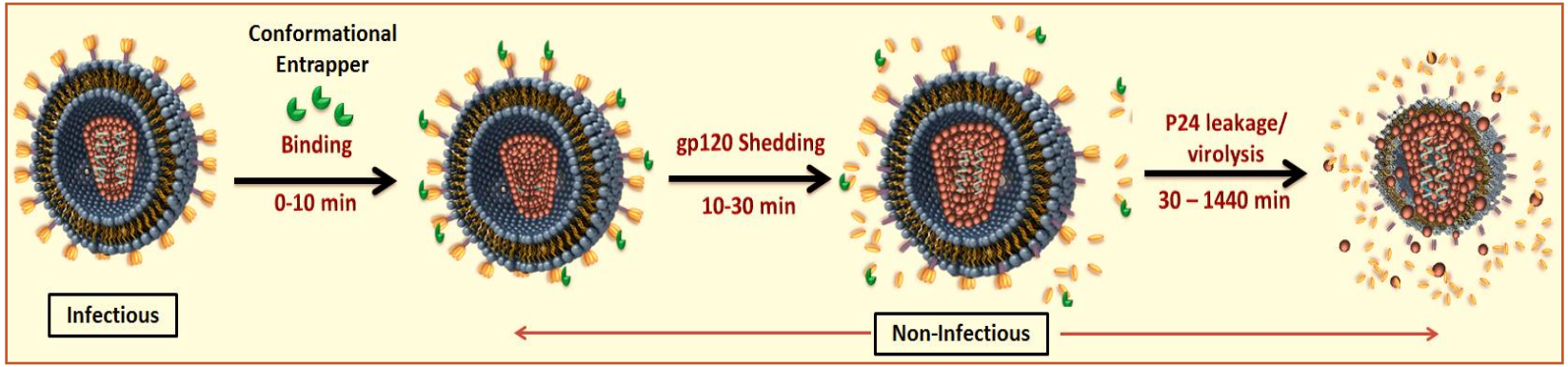
		IC_{50} (CD4) (nM)	IC_{50} (17b) (nM)	HIV-1_{BaL} EC_{50} (nM)
12p1		5400	7200	48000
HNG156		94	137	960
KR13		25	45.3	25

Ferrer & Harrison, JVI 1999 *Gopi et al., ChemMedChem 2006* *Bastian et al., ChemMedChem 2010*

Viral Inactivation by Peptide Triazoles

Preventative vaccine
Therapeutic vaccine

(Residual Virion with antigenic gp41)

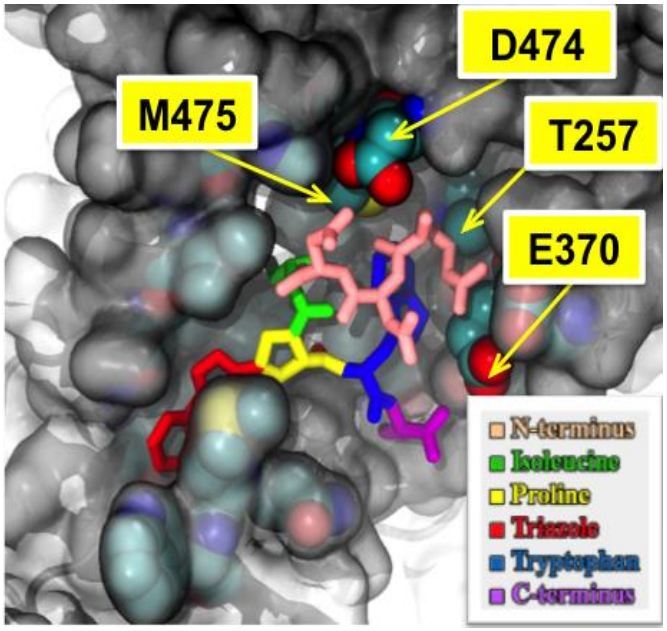


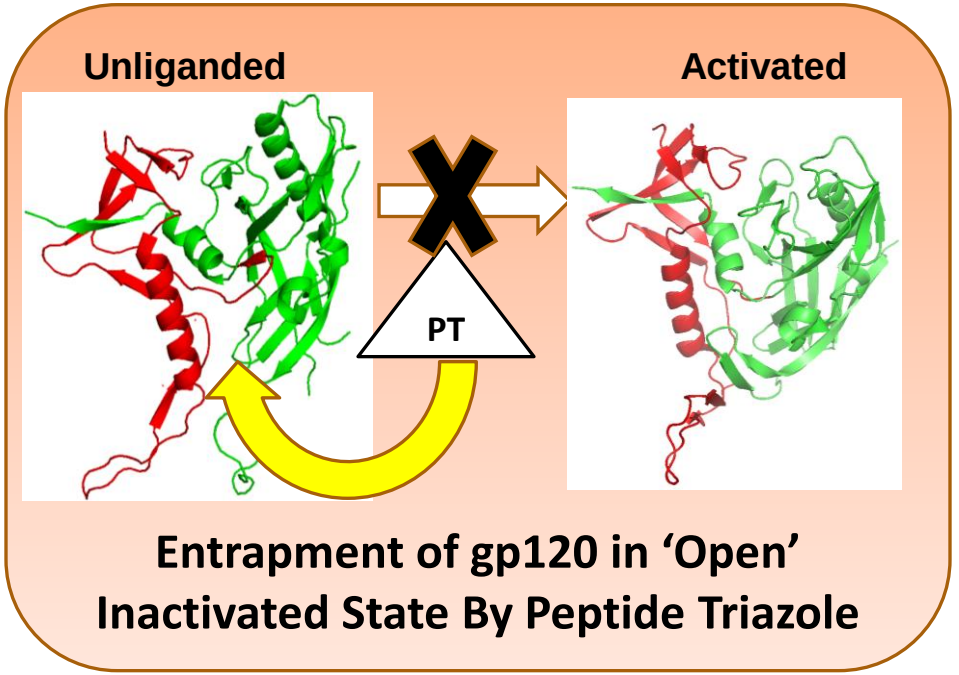
(Inactivation before host cell encounter)

Preventative microbicide

Bastian et al., Retrovirology 2013

Peptide Triazole Binding in a Conserved Site of gp120 and the Conformational Entrapment Model of gp120 Antagonism



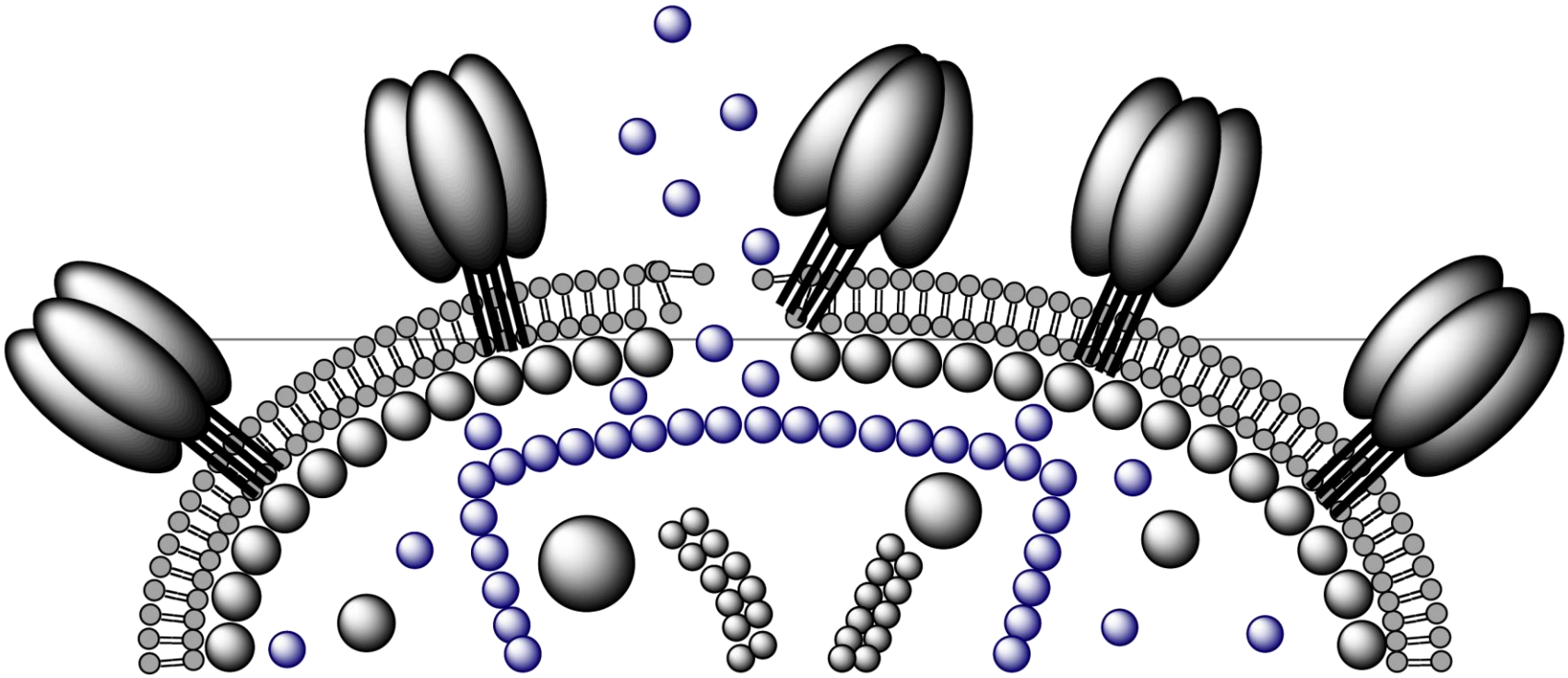


Tuzer et al., Proteins 2013

Emileh et al., Biochemistry 2013

p24 Release Infers Virus Env Disruption

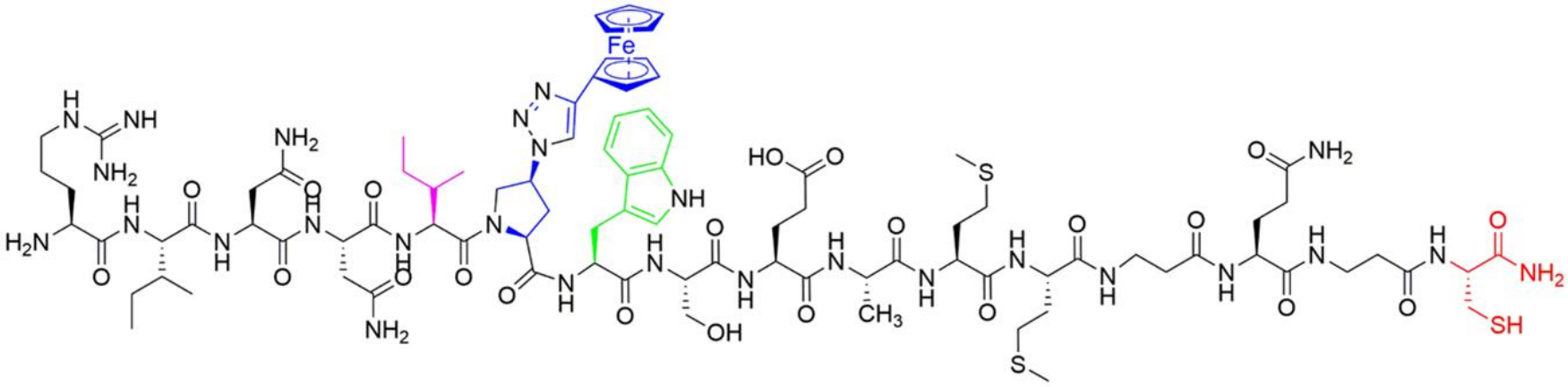
What's the mechanism?



(non-Core CA p24 in virus: Briggs et al. Nature Struct Biol 2004)

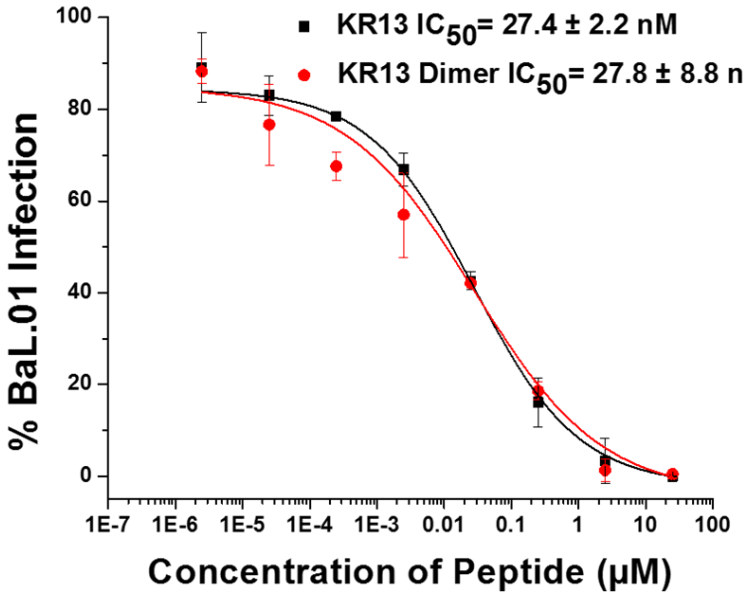
Lysis Requires Free Thiol of KR13

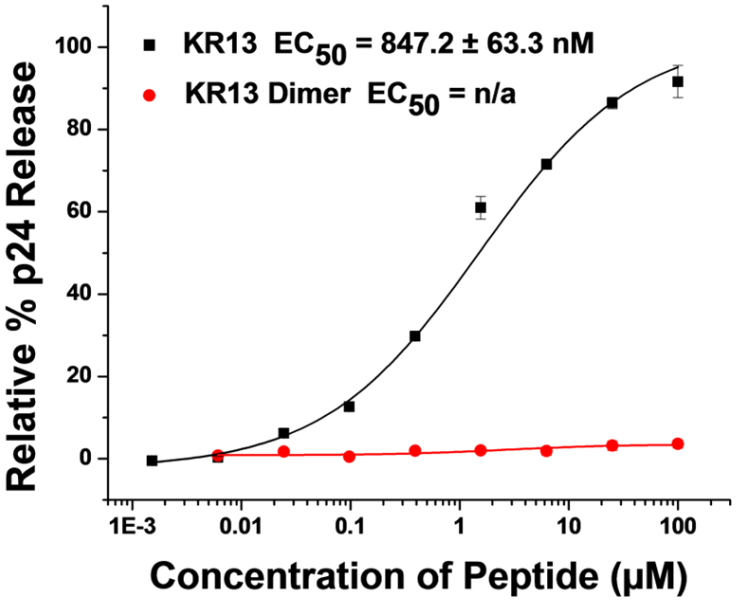
KR13



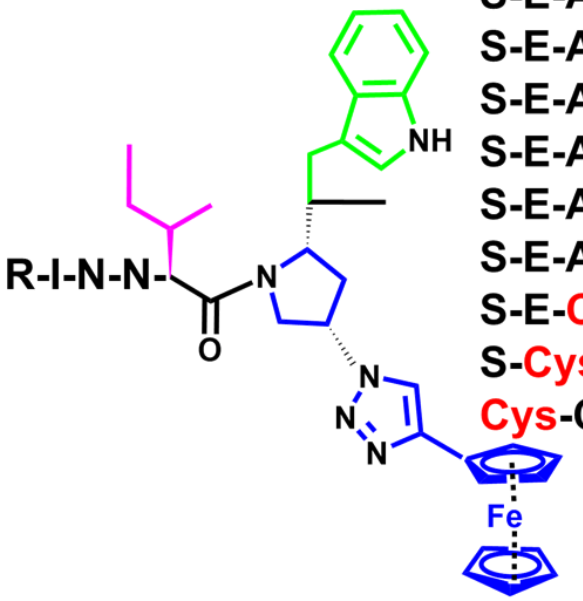
KR13 Dimer

Mechanism of Lytic Inactivation KR13 vs KR13 Dimer



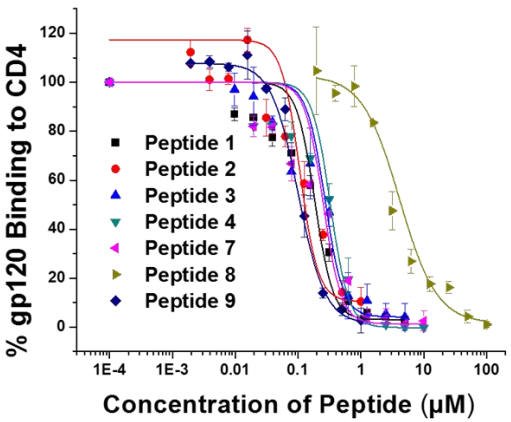


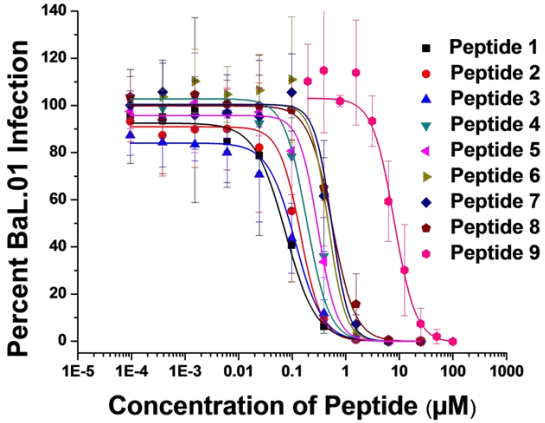
Peptide Triazole Thiols (PTT) Truncates

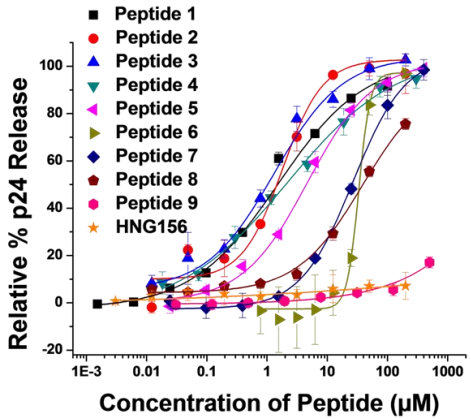


S-E-A-M-M-βA-Q-βA-Cys CONH2	Peptide 1
S-E-A-βA-Q-βA-βA-Cys-CONH2	Peptide 2
S-E-A-βA-Q-βA-Cys-CONH2	Peptide 3
S-E-A-βA-Q-Cys-CONH2	Peptide 4
S-E-A-βA-Cys-CONH2	Peptide 5
S-E-A-Cys-CONH2	Peptide 6
S-E-Cys-CONH2	Peptide 7
S-Cys-CONH2	Peptide 8
Cys-CONH2	Peptide 9

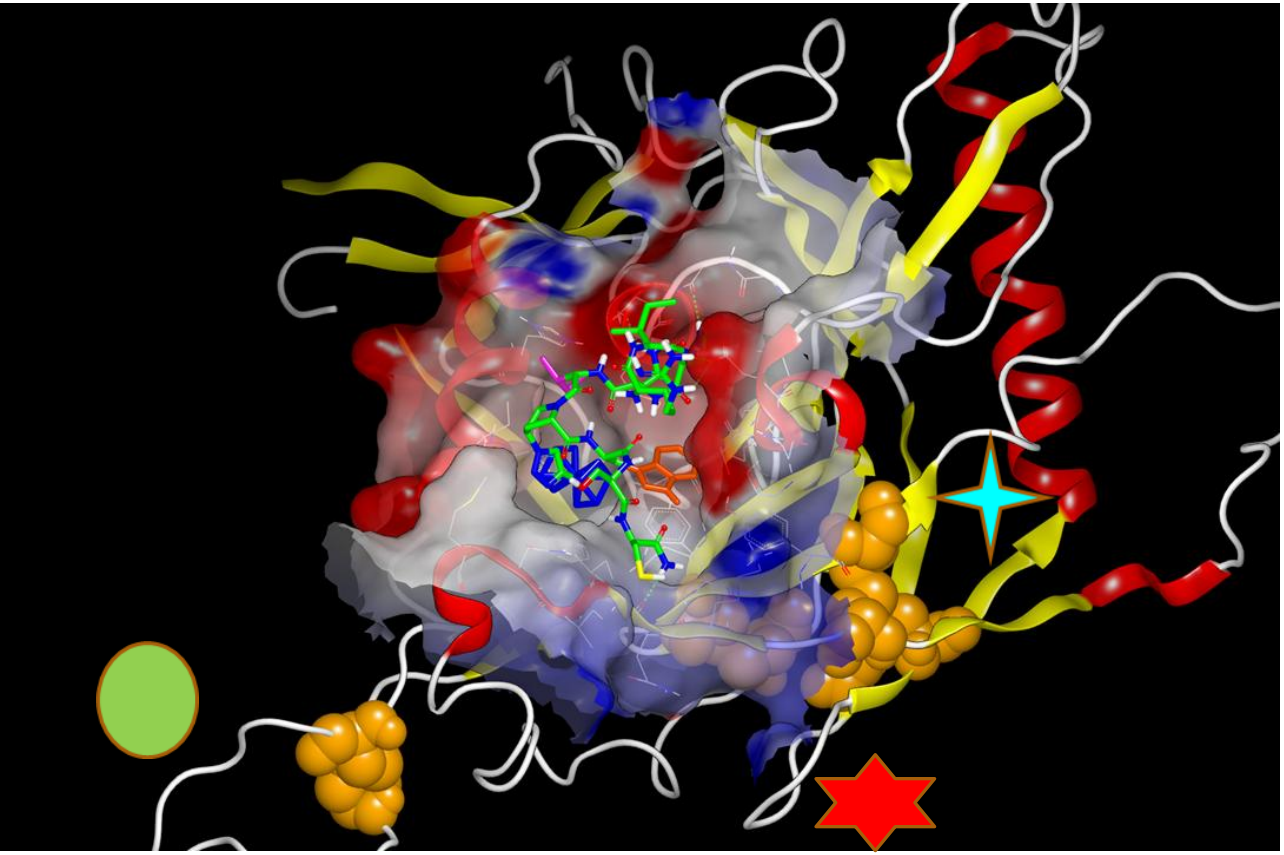
Functional Characterization of PTT Truncates







gp120 Protein Ligands Used to Deduce Likely Disulfides Targeted

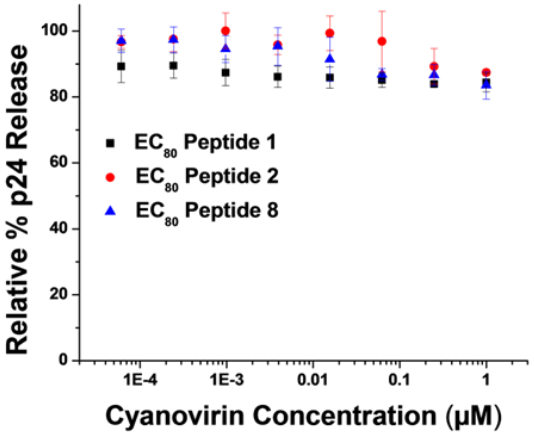


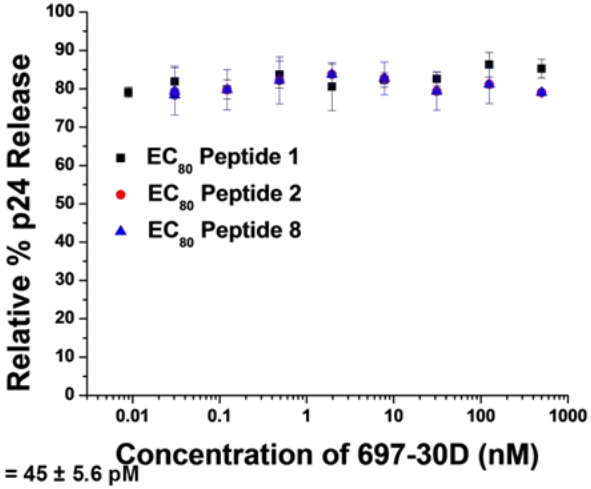
2G12

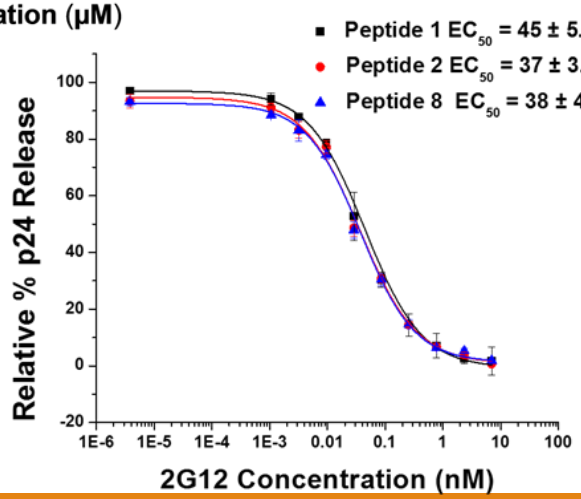
697-30D

Cyanovirin

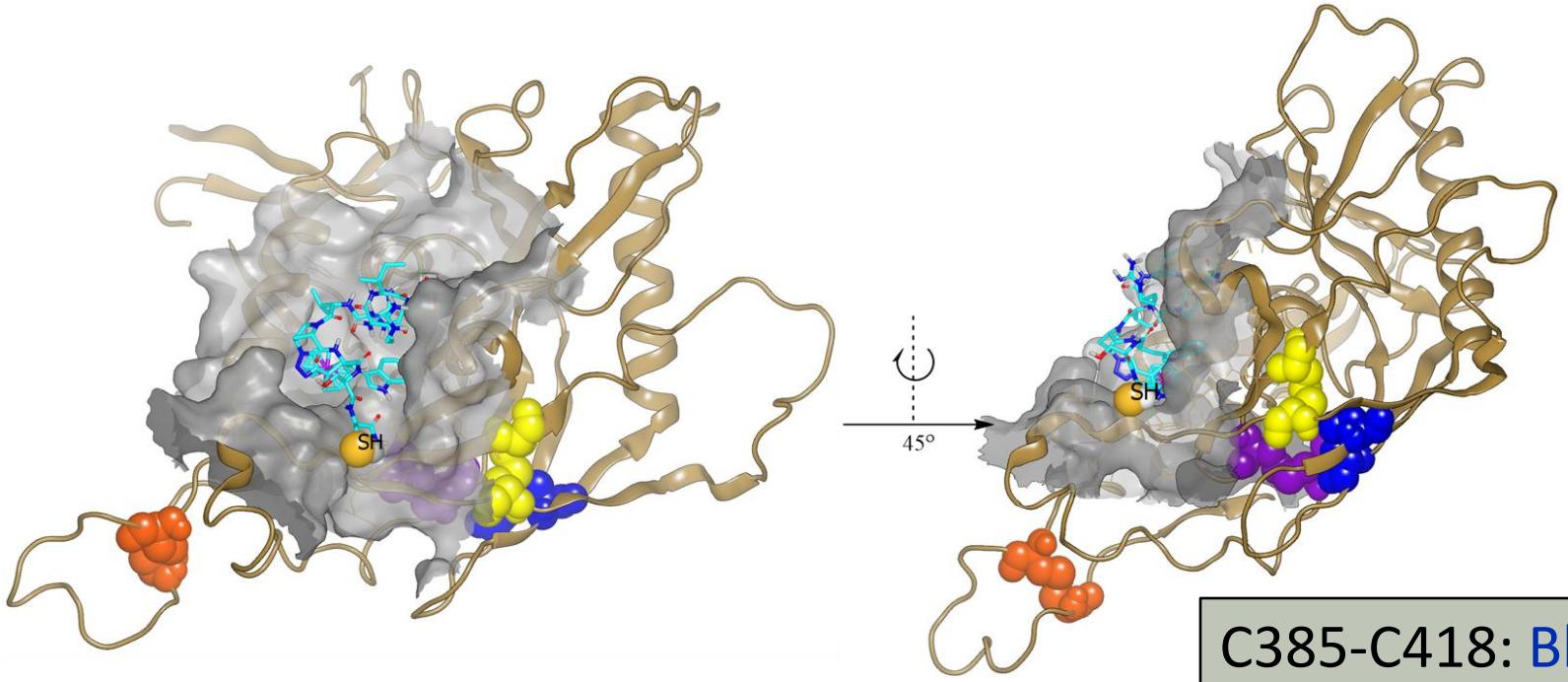
Effect of protein ligands on p24 release by PTTs







Molecular Docking Simulation



C385-C418: Blue
C378-C445: Yellow
C296-C331: Purple
C119-C205: Orange

Conclusions:

- There is a discontinuous relationship between virolysis and peptide length, indicating that a minimal length is required for efficient virolysis.

- The minimum linker length between PT pharmacophore and PT-SH appears needed to enable contact between SH and disulfides of the envelope protein.

- V3 loop disulfide is implicated in the disulfide exchange process.

- Our results support the hypothesis that thiol exchange occurs between the peptide C-terminal thiol group and a specific CD4 exposed disulfide cluster in gp120.

- Key goals going forward are to refine the pharmacophore, identify the specific Env protein disulfides involved, investigate the disulfide exchange process of virolysis.

Thank you

Chaiken Lab

- Dr. Rachna Aneja
- Dr. Arangassery Rosemary Bastian
- Dr. Mark Contarino
- Caitlin Duffy
- Dr. Ali Emileh
- Andrew Holmes, PhD Candidate
- Dr. Kantharaju Kamanna
- Dr. Huiyuan Li
- Dr. Adel A. Rashad
- Ramalingam Venkat Kalyana Sundaram, PhD Candidate
- Dr. Ferit Tuzer