The University of Toledo *Translation Journal of Medical Sciences*
Infectious Diseases Abstract, Department of Medicine Research Symposium UTJMS 2023 May 5; **11**(1):e1-e1

Protease-Inhibitor Binding Reveals Conformational Transition Mechanism

D. Nguyen[1*], S. Level[1], J. Babula, PhD[1], J. Liu, PhD[1]

[1]Division of Infectious Diseases, Department of Medicine, The University of Toledo, Toledo, OH 43614

*Corresponding author: Dylan.Nguyen@rockets.utoledo.edu

Published: 05 May 2023

Human Immunodeficiency Virus Type-1 (HIV-1) protease is a flexible dimeric protein required for post-translational activation of the HIV Gag-ProPol polypeptide. In clinic, protease inhibitors are commonplace in the multi-drug regimens used by physicians to combat drug-resistant HIV strains with an unfortunate exchange for greater patient toxicity. To confront HIV-1 protease drug resistance, Ghosh and colleagues developed protease inhibitors modeled after FDA approved HIV-1 protease inhibitor, Darunavir. These new inhibitors, Inhibitor 3 (In3) and Inhibitor 4 (In4), differ from each other by a single atom; an oxygen in In3 instead of a carbon in In4. Surprisingly, In4 exhibited a >1,000-fold drop in enzymatic inhibition, and >500-fold loss in antiviral capacity compared to In3. We investigated the mechanism behind this reduction in affinity utilizing Molecular Dynamics (MD) simulations. We found that In3 locks the protease in the closed conformation while In4 does not. The Apo-protease simulations suggested Asp29 is part of a residue triad which plays a critical “switch” role in the protease conformational transition. The formation of an Asp29-Arg87 salt bridge contributed to the closed conformation’s stability, which was further enhanced by the hydrogen bond between the oxygen of In3 and the main chain nitrogen atom of Asp29, which was abrogated in In4 binding. Additionally, arginine stacking between Arg87 and Arg8 of the opposite chain stabilized the protease open conformation. These observations explain the tremendous drop in affinity of In4 compared to In3 and reveal the mechanism of HIV-1 protease conformational changes that are being validated by mutagenesis and Bio-SAXS in-vitro.

https://dx.doi.org/10.46570/utjms.vol11-2023-758