Binding Pocket Mutational Analysis of HIV-1 Protease Crystal Structures

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Introduction

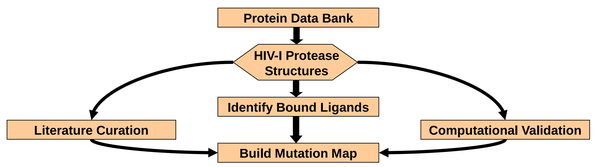
- HIV (Human Immunodeficiency Virus) is a retrovirus that can lead to Acquired Immune Deficiency Syndrome (AIDS).
- HIV-1 protease inhibitors are a class of major antiretroviral drugs designed to inhibit the activity of HIV-1 protease to prevent cleavage of nascent proteins into active viruses.
- The HIV proteases develop resistances to the inhibitors through mutations in its DNA sequence, resulting in an enzyme structure that decreases the binding affinity of inhibitors while maintaining catalytic efficiency.
- Mutations occur frequently outside the binding pocket, while the amino acids that form the binding pocket tend to be conserved in a natural environment.
- In order to develop a better understanding of drug induced mutations, an accurate mutation map of mutations in the binding pocket that occur in HIV-1 protease must be developed.
- In this study, we extend the mutation map by incorporating newly released PDB structures since our last presentation (ACS Western Regional Meeting, October 2007) and focus our study on the binding pocket mutations.

PDB (Approx 42,000) → HIV Proteins (400) → HIV Proteases (200) → HIV-1 Proteases Complexed with FDA Approved Drugs (52)

Methodology

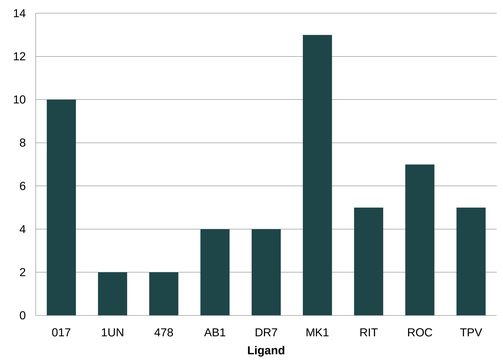
- The Protein Data Bank (PDB) houses experimentally derived crystal structures of more than 42,000 biological macromolecules [1].
- Crystal structures of HIV-1 proteases complexed with FDA approved protease inhibitors were studied.
- Mutational information for each structure was determined through the corresponding literature as well as through a computational sequence comparison of the wild type sequence (HXB2 isolate) with the sequence reported in the PDB files [2].
- Using the DeepView Swiss-PdbViewer application, the amino acids found to be within 6Å of the bound ligand were noted to form the binding pocket [3].

Flowchart of Mutation Map Development



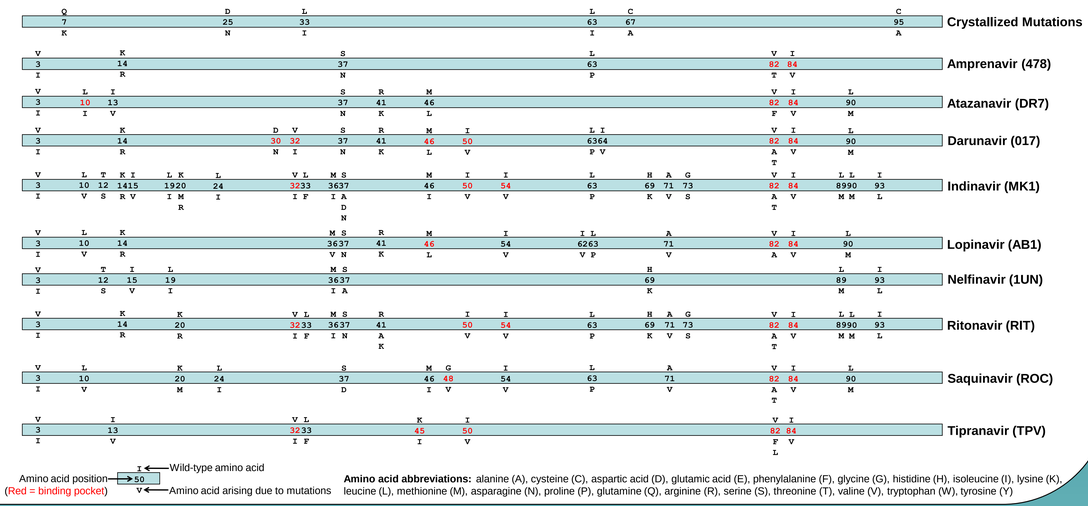
Results

Number of PDB Structures Complexed With FDA Approved Protease Inhibitors



Ligand abbreviations: Darunavir (017), Nelfinavir (1UN), Amprenavir (478), Lopinavir (AB1), Atazanavir (DR7), Indinavir (MK1), Ritonavir (RIT), Saquinavir(ROC), Tipranavir (TPV)

HIV-1 Protease Crystal Structures Mutation Map



Amino acid abbreviations: alanine (A), cysteine (C), aspartic acid (D), glutamic acid (E), phenylalanine (F), glycine (G), histidine (H), isoleucine (I), lysine (K), leucine (L), methionine (M), asparagine (N), proline (P), glutamine (Q), arginine (R), serine (S), threonine (T), valine (V), tryptophan (W), tyrosine (Y)

Discussion

- In this study, the mutation map was extended by incorporating all mutations in the PDB crystal structures into mutation maps according to their bound ligand and highlighted all mutations that occur within the binding pocket of HIV-1 protease.
- The crystallized mutations are performed to minimize autoproteolysis (Q7K, L33I, L63I), prevent cysteine-thiole oxidation (C67A, C95A), and to complex the protease with the substrate without cleaving (D25N).
- Mutations at the 82nd and 84th position are common for all drugs except Nelfinavir (1UN), for which the deposited crystal structures (1OHR and 2R5Q) have no mutations in the binding pocket.
- Nelfinavir has a unique binding pocket mutation at the 48th position (G48V).
- Darunavir has a unique binding pocket mutation at the 30th position (D30N).
- Atazanavir has a unique binding pocket mutation at the 10th position (L10I).

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Conclusions and Future Work

- In this study, we have improved the mutation map by incorporating all mutations in the PDB crystal structures and highlighted mutations that occur within the binding pocket.
- Our future plans on this project include:
  - Extending crystal structure mutation dataset with HIV-1 protease structures from the NIST HIV Structural Database [5, 6].
  - Performing cluster analysis to determine mutation trends among FDA approved protease inhibitors.
  - Building HIV-1 protease structural visualization models with binding pocket mutations highlighted.
  - Performing data mining analysis of chemical descriptors derived from the crystal structures coupled with mutational information for future HIV-I protease smart drug design.

References

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