Crystal structures of a number of complexes of HIV-1 RT with non-nucleoside inhibitors (NNIs), including nevirapine, α-APA, 1051U91, HEPT, 9-Cl-TIBO, MKC-442 and TNK-651, have been determined at high resolution (Ren et al. 1995 Nature Struct. Biol. 2, 293-302; Enouf et al. 1995 Nature Struct. Biol. 2, 303-308; Ren et al. 1995 Structure 3, 915-926; Hopkins et al. J. Med. Chem. in the press). All the inhibitors we have studied bind at the same pocket formed between two β-sheets of the β6α6, some 10 Å from the polymerase active site. The internal surface of the pocket is predominantly hydrophobic being constructed mainly from leucine, valine, tryptophan and tyrosine residues. The structures reveal a common mode of binding for these chemically diverse compounds. Each compound has its own particular structural characteristics and there is sufficient plasticity in certain regions of the surrounding protein to allow some unfavourable contacts to be relieved without changing the overall binding mode. The volume of the pocket varies with the inhibitors, ranging between some 600 and 700 Å3, of which the inhibitors occupy in the order of 250-350 Å3. However, much of the pocket lining remains very similar in all cases so that there is a very marked matching of shape in those compounds that occupy this volume. In some cases this is achieved by conformational rearrangement of the compound from its lowest energy structure in solution. These results allow us to understand the structural basis of the potency of the inhibitors and suggest possible modifications which should improve interactions with the enzyme.

A variant of influenza virus with 1000-fold less sensitivity in vitro to the anti-influenza drug 4-guanidino-Neu5Ac2en has been isolated by multiple passage of influenza virus NWS/G70C with the N9 subtype neuraminidase in MDCK cells in the presence of 4-guanidino-Neu5Ac2en. The crystal structure of this variant of influenza virus NWS/G70C neuraminidase, has been determined to 2 Ångstrom resolution for the native enzyme and a complex with 4-guanidino-Neu5Ac2en. Our laboratory used high weight polyethylene glycol precipitations at acidic pH to produce crystalline ternary complexes of thrombin with active site inhibitors and an exosite blocking agent. This inhibitor design effort will be described in the context of the overall thrombin effect in the pharmaceutical community. In our case, the most important contributions were to examine those structures that did not fit the expected structure-activity relationships. It was by analyzing these structures that we were able to provide the chemists with new and useful design information.

Human thrombin, an important enzyme in the blood coagulation response, has been crystallized in the presence of a variety of high affinity, synthetic active-site inhibitors. X-ray crystallography has been used to determine the orientations of the inhibitors in the thrombin active site and the modes of inhibitor binding. Knowledge of the specifics of inhibitor binding proved useful in the design of successively more potent inhibitors.

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The switch region peptide binds to stromelysin in nearly identical manner to that observed for the switch region peptide is opposite to that observed in stromelysin-1 (Fotouhi et al, 1994). Comparison of high resolution crystal structures of a truncated form of prostromelysin and of a complex formed between active stromelysin and a tight binding cyclic peptide demonstrated that the switch region peptide binds to stromelysin in nearly identical manner in prostromelysin and the cyclic peptide. In both prostromelysin and the inhibitor complex a mixed beta-sheet structure is formed between the RCGV peptide and the protein. Furthermore, the orientation of this peptide is opposite to that observed with peptides bound in a substrate-like mode. Additionally, analysis of these two crystal structures revealed that the zinc-bound cysteine side chain only partially fills the S1' substrate binding site. The remainder of this large cavity, which extends through the protein core, is occupied by solvent molecules. Additionally, but smaller binding cavities from the S1-S3 and S2-S4 subsites. The binding properties of the large S1' binding site have been exploited in the development of inhibitors derived from a cysteine or iso-cysteine building block and incorporating aromatic moieties. The analysis of several crystal structures will be described, and will include descriptions of enzyme conformational changes induced by different ligands and as well as changes in ligand interactions resulting from ligand modifications.