**X-ray Structure for an RNase H Inhibitor Bound to HIV-1 Reverse Transcriptase**

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We have determined a 3.0 Å resolution X-ray crystal structure of HIV-1 reverse transcriptase (RT) complexed with DHBNH, an RNase H inhibitor (RNHI). HIV-1 RT uses two enzymatic activites, a polymerase and an RNase H, to convert the viral genomic single-stranded RNA into double-stranded DNA suitable for integration into the host genome [1]. RNase H is essential for virus replication; however, very few small molecule inhibitors targeting this function have been reported, and there are no crystal structures of HIV RT in a complex with an RNase H inhibitor. DHBNH is an N-acyl hydrazone derivative that inhibits RNase H with an $IC_{50}$ of 0.5 μM but does not inhibit the RT polymerase ($IC_{50}$>20 μM). Despite this specificity, the inhibitor binds more than 40 Å away from the RNase H active site, at a novel binding site in the palm of the p66 subunit, between the primer grip and the polymerase active site. The inhibitor partially overlaps the non-nucleoside RT inhibitor (NNRTI) binding pocket. The inhibitor appears to interact with the conserved residues Asp186 and Trp229, as well as with Tyr188, Lys223, Asp224, Pro226, Phe227, and Leu228. Certain substitutions on DHBNH can enhance interactions in the NNRTI binding pocket, resulting in "dual inhibitors" that inhibit both the polymerase and RNase H activities of HIV-1 RT. Our results are consistent with the view that binding of DHBNH alters the trajectory of the nucleic acid substrate, affecting the RNase H activity. Knowledge gained from this study provides new opportunities for structure-based drug design.

[1] Coffin J. M., Hughes S. H., Varmus H. E., *Retroviruses*, Cold Spring Harbor Laboratory, Plainview, NY., 1997.

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