Crystallography as a Tool to Identify the Best Inhibitor in a Complex Mixture

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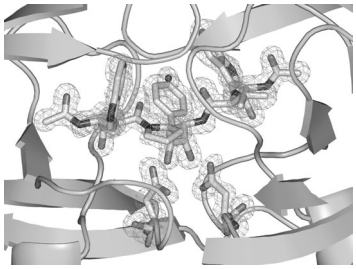

In this study we crystallized the HIV-1 aspartic protease using an equimolar mixture of four stereoisomeric inhibitors. Fourier maps obtained by high resolution diffraction data (up to 1.3 Å) from synchrotron radiation, clearly show that the catalytic site is fully occupied by a single ordered molecule (see Figure). This permitted unambiguously the identification of nature and stereochemistry of the bound inhibitor. Furthermore, the clear electron density map, without residuals, suggests that the inhibition constant of this compound should be at least one order of magnitude lower than the constants of the other compounds. The full occupancy of the site indicates that its value is less than 1μM. This biocrystallographic study has allowed a first assessment of inhibition properties without the purification of the mixture and the classic activity assays that are normally conducted on each compound. The co-crystallization strategy could be applied in conjunction with combinatorial chemistry synthesis to discover, by self selection, new potent inhibitors.

Keywords: single-crystal structure analysis, inhibitor binding, isomers