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Antiviral Activity of P-1946, a Novel Anti-HIV Protease Inhibitor

G Sévigny¹, B Tian¹, A Dubois¹, B Stranix¹, G Sauvé⁴, C Petropoulos², Y Lie², N Hellmann², B Conway³ and J Yelle¹

¹Pharmacor Inc., Laval, Canada; ²ViroLogic, Inc. South San Francisco, USA, ³UBC, Department of Pharmacology & Therapeutics, Vancouver, Canada, ⁴INRS-Institut Armand-Frappier, Laval, Canada.

Background Protease inhibitors (PIs) have significantly improved the treatment of HIV-infected individuals and provide continued hope for the long term clinical management of AIDS. Unfortunately, the emergence of HIV strains resistant to currently available antiretroviral (ARV) drugs compromises current treatment regimens. Therefore, novel PIs with distinct resistance profiles are needed.

Methods Protease inhibitors derived from an L-lysine were prepared using a simple, straightforward synthesis scheme developed in our laboratory. Antiviral activity (EC₅₀) of the compounds was determined using MT-4 cells by measuring the inhibition of HIV-induced cytopathic effect with an MTT colorimetric assay or the PhenoSense HIV assay (ViroLogic). The threshold for phenotypic resistance to the test compound was arbitrarily defined as fold-resistance (EC_{50test}/EC_{50wt}) > 4.0 (if not arbitrary, the criteria should be defined, current PIs loose activity at less than 4 fold reductions in susceptibility).

Results A novel family of L-lysine derivatives was readily obtained in relatively few synthetic steps using classic chemistry. Five compounds (P-1933, P-1935, P-1939, P-1946, P-1999) with the highest antiviral activity on the wild type HIV-1 NL4.3 virus (EC₅₀<400 nM) retained activity against two PI resistant strains carrying mutations 48/90 (saquinavir-resistant strain) and 10/46/63/82/84 (strain 4596). Compound P-1946 was selected as a prototype for this family of inhibitors and was further characterized. P-1946 displayed good antiviral activity against wild type strain NL4.3 (EC₅₀=150 nM). Cytotoxicity (CCIC₅₀) of P-1946 was 40 µM and is within the same range as FDA-approved PIs. In order to define the resistance profile of P-1946, we analyzed the antiviral activity in the presence of protease mutations at amino acids 10, 46, 48, 63, 82, 84 and 90, using three HIV strains obtained from the NIH. In addition to saquinavir-resistant and 4596 strains, P-1946 was active (fold-resistance < 4) against an V82A/I84V variant that exhibits resistance to several PIs, including cyclic urea. Eight additional viruses were used to further define the resistance profile of P-1946. The compound retained full activity against nelfinavir resistant (D30N) and lopinavir resistant (I50V) viruses. Resistance to P-1946 appears to require several mutations in the protease gene (can't say it requires six with this limited virus panel...there could be many mutation profiles consisting of less than six mutations that confer resistance, but were not tested in this study).

Conclusion P-1946 is an amino acid derivative representative of a new family of protease inhibitors. Antiviral activity of P-1946 makes it a good lead compound for the development of new potent protease inhibitors that retain inhibitory activity against HIV strains resistant to current PIs.