139

MKC-442, a Highly Potent and Specific Inhibitor of HIV-1

M. Baba,[1] S. Shigeta,[1] S. Yuasa,[2] M. Ubasawa,[2] H. Tanaka,[3] T. Miyasaka,[3] R.T. Walker,[4] and E. De Clercq[5] *Department of Microbiology, Fukushima Medical College, Fukushima 960-12,*[1] *Mitsubishi Kasei Corporation Research Center, Yokohama 227,*[2] *Department of Synthetic Chemistry, Showa University, Tokyo 142,*[3] *Japan; University of Birmingham, Birmingham B15 2TT, Great Britain*[4]*; and Rega Institute for Medical Research, Katholieke Universiteit Leuven, B-3000 Leuven, Belgium*[5]

MKC-442, 6-benzyl-1-ethoxymethyl-5-isopropyluracil (I-EBU), has recently been identified as a highly potent and specific inhibitor of human immunodeficiency virus type 1 (HIV-1) reverse transcriptase (RT). Since the compound has favorable pharmacokinetic and toxicity profiles *in vivo*, we have evaluated MKC-442 for its inhibitory effect on the replication of HIV-1 in various cell cultures including human peripheral blood lymphocytes (PBLs) and monocyte-macrophages (MΦs). The 50% and 90% effective concentrations (EC_{50} and EC_{90}) for HIV-1 ($HTLV\text{-}III_B$) replication in MT-4 cells were 15 and 98 nM, respectively. MKC-442 was also inhibitory to HIV-1 replication in PBLs and MΦs as determined by the production of p24 antigen in the culture supernatant. Fluorescence-activated cell sorter (FACS) analysis revealed that MKC-442 was equally active against AZT-resistant mutants and AZT-sensitive strains. Furthermore, combinations of MKC-442 with either 3'-azido-3'-deoxythymidine, 2',3'-dideoxycytidine, or 2',3'-dideoxyinosine synergistically inhibited the replication of HIV-1. Thus, MKC-442 was considered as a candidate for clinical efficacy studies.

140

Effects of combinations of HIV-1 specific RT inhibitors with complementary resistance profiles on the time and type of drug-resistant HIV-1 development.

R. Pauwels[1], M.P. de Béthune[1], K. Andries[2], P. Stoffels[2], J. Desmyter[1], P.A.J. Janssen[2] and E. De Clercq[1]
[1]Rega Institute for Medical Research, Katholieke Universiteit Leuven, Leuven, Belgium and [2]Janssen Research Foundation, Beerse, Belgium

Recently several chemically distinct families of compounds have been described that exhibit a unique specificity for HIV-1. These include the TIBO, HEPT, nevirapine, pyridinone, BHAP, TSAO and α-APA derivatives. Despite their chemical diversity, they all are selective and potent inhibitors of the HIV-1 reverse transcriptase (RT). The stringent structure-activity relationship within each class of compounds points to a highly specific interaction with a putative receptor site ("pocket") on the target enzyme. Upon serial passage in cell culture, these compounds rapidly select for drug-resistant HIV-1 variants whose mutations in the RT gene cluster around the drug-binding pocket. As some of these mutations (e.g. $Y^{181}C$) were observed for a number of these agents, it created the notion that the various nonnucleoside RT inhibitors all behave similarly in that the mutants selected by these compounds display cross-resistance to other compounds of this class. However, some of these mutants can display large differences in sensitivity for different compounds. This is seen with the $Leu^{100}Ile$ RT mutant which is highly resistant to TIBO R82913 and R86183 but very sensitive to the α-APA derivative R89439. We have also found that the 8-chloro TIBO derivative R86183 was active against an HIV-1 strain containing the$Y^{181}C$ mutation which is selected for by the α-APA derivative R89439. R86183 also selected for a new mutation ($M^{230}T$) indicating that the shift of the chlorine from the 9- to the 8-position resulted in a different resistance profile. In a series of virus breakthrough experiments with constant drug pressure, the combination of R86183 and R89439, which have an apparent complementary sensitivity pattern and differential resistance profile, was found to delay the resistance development but generated other mutations than those observed for the individual drugs. These and several other combinations, including 2 or more nucleoside and non-nucleoside RT inhibitors, will be discussed in the context of the current knowledge of the HIV-1 RT drug binding site(s) and new strategies to combat HIV infection.