

POSTER 71

SEQUENCE VARIATIONS OBSERVED IN VIRAL LTR ENDS IN PLASMA HIV-1 FROM PATIENTS DO NOT AFFECT SUSCEPTIBILITY TO INTEGRASE INHIBITORS *IN VITRO*

D. Goodman¹, R. Hluhanich², D. McColl², M. Miller², K. Borroto-Esoda¹, and E. Svarovskaia¹

¹Gilead Sciences, Inc., Durham, NC; ²Gilead Sciences, Inc., Foster City, CA

Background: An interaction between the HIV-1 integrase enzyme and the viral DNA ends is required for integration of the virus. It has been previously suggested that changes in the terminal nucleotides of viral LTRs may affect the antiviral activity of integrase inhibitors (INIs). In this study, mutations observed in the U3 and U5 regions of HIV-1 LTR from INI treatment-naïve patient samples were analyzed for replication competence and susceptibility to INI.

Methods: ~300 bp fragments of both the ppt-U3 and U5-PBS junctions encoding the viral DNA termini were RT-PCR amplified and sequenced from 50 treatment experienced patients who were naïve to INI therapy. Based on mutations that were observed in the terminal 16 nucleotides, site-directed mutants were constructed in HXB2. A comparison of replication between mutant and wild-type virus was performed using a MAGI assay. Phenotypic analysis using a luciferase-based cell viability assay was used to measure susceptibility to the INIs elvitegravir (EVG) and raltegravir (RAL).

Results: Sequence variability in the 16 terminal bases was higher in the U3 region where 37% (17/46) of patient baseline samples had a change from the consensus sequence versus 10% (5/50) in the U5 region. No patient viruses had changes in the highly conserved last four bases of either U3 (ACTG) or U5 (CAGT) LTR. Only one mutation, a single A insertion, accounted for the sequence variability in U5. 10 different mutations were observed in the U3 region. No differences in viral titer were observed between any of the U3 or U5 site-directed mutants and wild-type virus using a MAGI assay. Susceptibility assays in MT2 cells with site-directed mutants revealed no change (<2 fold) in EC₅₀ compared to wild-type for both EVG and RAL.

Conclusions: Polymorphisms that are found in the U3 and U5 viral ends of HIV-1 from patients naïve to INIs appear to have no effect on viral replication or susceptibility to INIs.