

## **Abstract:**

The development of viral diversity during the course of human immunodeficiency virus type 1 (HIV-1) infection may significantly influence viral pathogenesis. The paradigm for HIV-1 evolution is based primarily on studies of male cohorts in which individuals were presumably infected with a single virus variant of subtype B HIV-1. In this study, we evaluated virus evolution based on sequence information of the V1, V2, and V3 portions of HIV-1 clade A envelope genes obtained from peripheral blood and cervical secretions of three women with genetically heterogeneous viral populations near seroconversion. At the first sample following seroconversion, the number of nonsynonymous substitutions per potential nonsynonymous site ( $d_n$ ) significantly exceeded substitutions at potential synonymous sites ( $d_s$ ) in plasma viral sequences from all individuals. Generally, values of  $d_n$  remained higher than values of  $d_s$  as sequences from blood or mucosa evolved. Mutations affected each of the three variable regions of the envelope gene differently; insertions and deletions dominated changes in V1, substitutions involving charged amino acids occurred in V2, and sequential replacement of amino acids over time at a small subset of positions distinguished V3. The relationship among envelope nucleotide sequences obtained from peripheral blood mononuclear cells, plasma, and cervical secretions was evaluated for each individual by both phylogenetic and phenetic analyses. In all subjects, sequences from within each tissue compartment were more closely related to each other than to sequences from other tissues (phylogenetic tissue compartmentalization). At time points after seroconversion in two individuals, there was also greater genetic identity among sequences from the same tissue compartment than among sequences from different tissue compartments (phenetic tissue compartmentalization). Over time, temporal phylogenetic and phenetic structure was detectable in mucosal and plasma viral samples from all three women, suggesting a continual process of migration of one or a few infected cells into each compartment followed by localized expansion and evolution of that population.