Determination of CCR5-Δ32 and CCR2-64I Gene Polymorphisms in HIV-1 Exposed Infants in Nairobi Province, Kenya

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ABSTRACT

An estimated 600,000 children per year are infected with Human Immunodeficiency Syndrome (HIV-1), majority of them through mother-to child transmission (MTCT). The chemokine co-receptor 5 (CCR5) and the δ-chemokine co-receptor (CX3CR4) are the major chemokine receptors involved in the binding and entry of non syncytium inducing (NSI or M- trophic) HIV-1 strains. The chemokine co-receptor 2 (CCR2) is used as a secondary co-receptor for the HIV-1 entry into the cell. It has been reported that different polymorphisms in the regulatory regions of CCR5 and in the CCR2 gene of the chemokine receptors are associated with altered levels of HIV infection and progression to AIDS. The CCR5-Δ32 and CCR2-64I gene polymorphisms have been associated with protection against HIV infection as well as slowing down the rate of progression of AIDS. Information on the significance of CCR5-Δ32 and CCR2-64I gene polymorphisms in vertical transmission of HIV-1 in African infants is scanty or non-existent. The main objective of this study was to determine the existence of CCR5-Δ32 and CCR2-64I gene polymorphisms in HIV-1 exposed infants in Nairobi Province. To achieve this, two hundred and forty (240) blood samples from exposed infants less than eighteen (18) months of age were obtained from different hospitals and health centers within Nairobi. Half of these samples were obtained from HIV positive infants while the other half were obtained from HIV negative infants. Both Dried Blood Spots (DBS) and whole blood were used. Genomic DNA was isolated using QIAGEN® DNA isolation kit for the DBS and DNAzol was used for isolation of genomic DNA from whole blood. The CCR5 and CCR2 genes were amplified using PCR with gene specific primers targeting the two gene regions. The amplified products were analyzed by gel electrophoresis and visualized under UV light. Since the CCR5-Δ32 is a 32 base pair deletion

mutation, mutant alleles were directly visualized at 147 base pairs (bp) while the wild type allele (WT) was visualized at 179 bp on the gel than the non- mutated alleles. BsaBI restriction enzyme was used to restrict the CCR2-64I mutant alleles. The restricted products were then analyzed by gel electrophoresis and visualized under ultraviolet (UV) light. From the results, no infant was found to possess the homozygous CCR5 allele. Four infants (1.7%) exhibited the heterozygous state (CCR5/ CCR5-Δ32 genotype). One (0.4%) of these heterozygous infants was HIV positive while the other three (1.25%) were HIV negative. The wild type (WT) allele (CCR5/CCR5 genotype) accounted for 236 (98%) of all the infants studied. For the CCR2 gene, 63 (26.25%) of all the infants studied had the CCR2-64I gene polymorphism. Of these, 53 (22.1%) possessed heterozygous allele (CCR2/CCR2-64I genotype). Twenty-three (23) of these infants representing 9.6% were HIV positive infants whereas 30 (12.5%) of the infants were HIV negative. Ten (4.2%) of the infants under study carried homozygous CCR2-64I allele (CCR2-64I/CCR2-64I genotype). Of these CCR2 homozygous, 3 (1.25%) were HIV positive while the other 7 (2.9%) were HIV negative. The results of this study reveal the presence of CCR5-Δ32 and CCR2-64I in the population studied and explain the different levels of infection and the slow progression of HIV-1 in infants possessing these polymorphisms.