Novel hepatitis B virus genotype a subtyping assay that distinguishes subtype Aa from Ae and its application in epidemiological studies

Abstract:

The eight genotypes of hepatitis B virus (HBV) have different geographical distributions, virological characteristics, and clinical manifestations. A unique subtype of HBV genotype A (HBV/A) was reported in sub-Saharan Africa, raising the possibility that patients infected with this subtype (HBV/Aa ["a" for African and Asian]) may have different clinical outcomes than other HBV/A isolates (HBV/Ae ["e" for European]). Comparison between 30 HBV/Aa and 30 HBV/Ae isolates indicated that almost all HBV/Ae isolates had G at nucleotide (nt) 1809 and C at nt 1812, whereas HBV/Aa isolates had T1809/T1812. Taking advantage of these two single nucleotide polymorphisms (SNPs), a novel subtype-specific PCR assay in the X/precore/core region was developed. This assay was combined with a restriction fragment length polymorphism assay using BglII in a different region (nt 1984 to 1989), which has a SNP distinguishing HBV/Aa from HBV/Ae, resulting in 100% specificity for the combined assay. Application of the subtyping assay using sera from 109 paid donors in the United States indicated significantly different distributions of HBV/A subtypes among races; African-Americans, Caucasians, and Hispanics had HBV/Ae, whereas Asians had mainly HBV/Aa, suggesting that the HBV/Aa isolates may have been imported by recent immigration from Asia. In conclusion, the specificity and sensitivity of the combined subtyping assay were confirmed, and its usefulness was demonstrated in a practical context.