

Evaluation of intracellular and plasma pharmacokinetics, pharmacogenetics and clinical features in HBV e antigen-negative patients treated with entecavir

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Hepatitis B infection affects two billion people worldwide and 350 million of these are chronically infected. Chronic hepatitis B virus is one of the most important cause of mortality and morbidity worldwide. Treatment options include currently available pegylated interferon and nucleos(t)ide analogues: among these, entecavir is currently the most used. Despite this, not many information are currently available about its plasma pharmacokinetics and its intracellular disposition.

Vitamin D is an important immunomodulator and it has a role in inflammatory and metabolic liver diseases, including infection with hepatitis C virus (HCV). The relationship between vitamin D metabolism and chronic HBV infection remains unknown, but its levels result low in patients affected by this virus.

For these reasons, in this work we aimed to study entecavir plasma/intracellular pharmacokinetics and the impact of single nucleotide polymorphisms (SNPs) related to its metabolism/distribution and vitamin D pathway.

We have enrolled HBV e antigen-negative chronically infected adult patients treated with the standard dose of 0.5 mg/die of entecavir; we have evaluated plasma and peripheral blood mononuclear cells (PBMCs) entecavir exposure according to SNPs in the following genes: *CYP27B1*, *CYP24A1*, *VDR*, *UGT1A3*, *SLC28*, *SLC29*, *OAT1*, *OCT1*, *AK1*, *NT5C2*, *HNF4*, *ESR1*, with real-time PCR analyses. Drug plasma and intracellular concentrations were measured through an UPLC-MS/MS validated method.

36 patients infected with different HBV genotypes were enrolled; pharmacogenetic results suggested that plasma concentrations were influenced by AK1 rs1109374 CC (p=0.041), CYP24A1 rs2248359 TC/CC (p=0.028), CNT2 124 rs11854484 CT/TT (p=0.037), CNT2 225 rs1060896 CA/AA (p=0.037) genotypes; a significant association between CYP24A1 rs927650 CT/TT (p=0.020) and entecavir PBMCs levels was found. In the logistic regression analysis, CNT2 225 CA/AA (p=0.008) and CYP27B1-1260 GT/TT (p= 0.006) genotypes were factors able to predict the Δ HBV DNA at three months of therapy (drop of HBV DNA at three months of therapy compared to HBV DNA at baseline).

Concerning pharmacokinetics, according to the efficacy marker of HBV DNA reduction of 2 Log at three months of therapy, a PBMC/plasma ratio cut-off value of 276 was identified (p=0.028, sensitivity 50%, specificity 87%; AUROC= 0.741).

Pharmacogenetics and therapeutic drug monitoring could contribute to improve therapy optimization on the basis of interindividual genetic variability. Our work provides a new overview of entecavir pharmacogenetics and it adds information concerning intracellular exposure, which actually lacks in literature.