Title: Occult Hepatitis B and Risk of Reactivation after Hepatitis C Treatment with Direct-Acting Antivirals-reply

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We read the comments by Ozaras et al with great interest and would like to clarify a few issues raised on our post-marketing observational study on hepatitis due to hepatitis B virus (HBV) reactivation among Chinese with chronic hepatitis C (CHC) infection, treated with pan-oral direct-acting antivirals (DAAs) agents. In our study, occult HBV infection (OBI) was defined as negative hepatitis B surface antigen but positive serum HBV DNA by nested polymerase chain reaction (PCR). To determine OBI, pre-treatment serum samples of all patients were tested by nested PCR for the pre-S/S (S), precore/core (Core), and X viral regions according to the methods previously described by our group. Serum samples reactive by at least two of the three PCR assays were considered HBV DNA positive and diagnosed to have OBI. The sensitivity of this nested PCR assay was 10 copies/mL (approximately 1.8 IU/ml) as determined by serial 10-fold dilutions of cloned HBV DNA with known amount (10^8 copies/ml). There was no difference in sensitivity between S, Core, and X gene PCR. For those with OBI, serum HBV DNA levels were further quantified using the COBAS® AmpliPrep/COBAS® TaqMan® HBV Test, v2.0 (Roche Diagnostics, Branchburg, NJ), with a lower limit of detection of 6 IU/mL and a broad linear range from 20 – 1.7 x 10^8 IU/mL. Among 327 Chinese with CHC infection, 127 (40.0%) of the 317 hepatitis B surface (HBsAg) negative patients had OBI. None of the patients with OBI had quantifiable serum HBV DNA by COBAS® AmpliPrep/COBAS® TaqMan® HBV Test assay.

In our study, only those who developed hepatitis (defined as more than 2-fold increase of serum ALT on two consecutive determinations at least five days apart, from the nadir during direct-acting antiviral agents and follow-up till 12 weeks after end-of-treatment) were further tested for HBsAg and HBV DNA, from serial serum samples stored at -70°C. Hence, not all serial samples of those with OBI was tested for HBV DNA and HBsAg. We agree that we cannot define the incidence of HBV reactivation among occult HBV infected CHC patients. In contrary to other reports that DAAs treatment can lead to hepatitis due to HBV reactivation in CHC patients with OBI, none of our patients with OBI suffered from hepatitis due to HBV reactivation. Hence, we doubt whether there is any added value to test for HBV DNA from liver samples, which can only be obtained by invasive procedure. Unlike our study which included only Chinese, all the reported cases of hepatitis due to HBV reactivation in CHC with OBI occurred in non-Chinese. It remains unclear whether host immunogenetic could be an important factor and further studies will be required to clarify this issue.
References