Reduced growth ability and increased nuclear abnormality in HBV-infected human hepatocytes of humanized chimeric mouse liver

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Several studies performed in established cell lines and transgenic mice have reported that expression of hepatitis B virus (HBV) proteins, such as surface and X proteins, affect hepatocyte proliferation and induce growth of hepatocellular carcinoma. However, it has not been elucidated how HBV infection affects phenotypes of normal human hepatocytes (HHs), which are the natural host for HBV. In the present study, we examined the effects of HBV infection on the morphology and proliferation of HHs by using cDNA-uPA/SCID chimeric mice with humanized livers.[1)]

Seven-week-old chimeric mice were inoculated with HBV genotype C. At 12 and 20 weeks after infection, HHs were collected by collagenase perfusion. Compared with naïve HHs, HBV infection induced robust HH hypertrophy, and increased the number of binuclear HHs at 12 weeks after infection. At 20 weeks after infection, the HHs were more enlarged and many atypical nuclei were observed. Flow cytometric analysis and microscopic observation revealed that ploidy increased depending on the duration of infection, and pegylated interferon alpha-2a treatment for 8 weeks reduced the cell size and inhibited the ploidy increase caused by HBV infection. To examine the effects of HBV infection on HH proliferation, HHs were isolated from naïve and HBV-infected chimeric mice and transplanted into 3-week-old cDNA-uPA/SCID mice. At 16 weeks after transplantation, blood human albumin levels reached 9 mg/mL. On the other hand, maximum human albumin level was approximately 1 mg/mL in chimeric mice transplanted with HBV-infected human hepatocytes. Histological examination indicated that the repopulation ratio of chimeric mouse liver transplanted with naïve HHs was more than 80%, but that of chimeric mouse liver transplanted with HBV-infected HHs was less than 10%.

These results suggest that HBV infection not only induced hepatocyte hypertrophy and ploidy increase but also inhibited hepatocyte proliferation *in vivo*.

[1)]Tateno *et al*., Generation of novel chimeric mice with humanized livers by using hemizygous cDNA-uPA/SCID mice. *PLoS One*. 2015; 10: e0142145