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Update on Hepatitis B: Role of Hepatitis B Virus Mutations In Liver Disease

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Abstract

Chronic hepatitis B virus (HBV) infection may result in a wide spectrum of liver diseases. Differences in activity of HBV-induced liver disease have been attributed to difference in host immune response, but viral factors may also play a role. The mutations in HBV genome (pre-core and core regions) and liver disease, with special reference to Chinese patients will be discussed.

Chronic hepatitis B virus (HBV) infection may result in a wide spectrum of liver disease. HBV is generally regarded to be non-cytopathic, HBV-induced liver disease being mediated by cytotoxic T lymphocyte (CTL) lysis of infected hepatocytes. Thus, differences in activity of HBV-induced liver disease have been attributed to differences in host immune response. However, activity of HBV-induced liver disease may also be related to viral factors. With the advent of modern molecular biology techniques, it is possible to study the relationship between mutations in HBV genome and liver disease.

The most extensively studied HBV mutation is the pre-core stop codon variant: The pre-core region consists of a stretch of 87 nucleotides encoding 29 amino acids just upstream of the core gene. Translation of the pre-core/core region produces a precursor protein which is processed to the secretory hepatitis B e antigen (HBeAg). Seroconversion from HBeAg to HBe antibody (anti-HBe) is usually associated with cessation of HBV replication and remission of liver disease. However, a small proportion of anti-HBe positive patients remain viremic with active liver disease. Several studies, mostly from Japan and Mediterranean countries, found that most of these patients have a mutation that creates a stop codon in codon 28 of the pre-core region of HBV, thus preventing HBeAg production. This pre-core stop codon variant (M2) is mainly found in HBeAg negative patients with chronic active hepatitis as well as patients with fulminant hepatitis suggesting that it may cause more active liver disease. The reason for the frequent detection of this mutation is postulated to be due to immune selection. HBeAg may serve as a target antigen on HBV-infected hepatocytes. Failure to produce a target antigen may be a means to evade immune clearance. Some of these patients have an additional mutation: gly-asp substitution at codon 29 (M4).

In our studies on Chinese patients, we found in addition to M2 and M4, two new mutations in the pre-core HBV region: M1 - pro-ser substitution in codon 15, and M3 - gly-ser substitutionin codon 29; as well as a silent mutation: MO in codon 15. We demonstrated that while Ml was usually present at the onset of infection and did not appear to affect HBeAg secretion, M2 tended to emerge or was selected from wild type (WT) sequence shortly before or at the time of HBeAg seroconversion. Most (95%) HBeAg positive but only

46% HBeAg negative patients with M2 had active liver disease. Thus, M2 on its own may not be the cause of active liver disease, but the immune process by which WT sequence is eliminated and M2 is selected may be the responsible factor. We also found that Mland M2 were mutually exclusive, and that M3 was only found in association with Ml. The invariable coexistence of certain mutations in these two non-contiguous regions is explained by the fact that part of the pre-core/core region can fold back as a stem-loop structure thus placing codon 15 and codon 28 and 29 opposite each other. The mutations that we described create additional base-pairing in the stem. This stem loop structure has been recently identified to be the cis-acting pre-genome encapsidation signal that regulates packaging of the pre-genomic RNA into the core particle: a crucial step in the replication of HBV. Thus, the primary function of the mutation in the pre-core region of HBV may be to stabilize the secondary structure of the pregenome encapsidation signal to ensure perpetuation of HBV replication.

Since the major target for CTL attack is the hepatitis B core antigen (HBc-Ag), changes in level of HBcAg expression and/or structure of HBcAg epitopes may have more direct impact on the activity of HBV-induced liver disease. Recent studies from Japan showed that mutations in the core gene were more frequently found in patients with chronic active hepatitis or fulminant hepatitis. In addition, most of these mutations were clustered in three regions. The authors suggested that these mutations may change the immune recognition sites of HBcAg, thereby eliciting or evading immune clearance.

In our studies on Chinese patients, we found that mutations in the core gene were rare during the early phase (first 1-3 decades) of HBV infection. Most of the mutations appeared around the time of HBeAg seroconversion. Patients who developed M2 in the pre-core region were also more likely to develop mutations in the core gene. The mutations were clustered in the same regions that had been previously identified by the Japanese investigators. Further studies are under way to determine whether these mutations preceed or follow HBeAg seroconversion and to elucidate whether these mutations are the cause or consequence of increased activity of liver disease.

Love seeketh not Itself to please, Nor for itself hath any care. But for another gives it ease, And builds a Heaven in Hell's despair. Love seeketh only Self to please, To bind another to its delight, Joys in another's loss of ease, And builds a Hell in Heaven's despite. -- William Blake