

Geographic and Genetic Diversity of Hepatitis B

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Abstract

Hepatitis B Virus (HBV) infection is a global health problem and may lead to chronic hepatitis, liver cirrhosis and hepatocellular cancer. The ability of HBV to adapt to the host environment by genetic variation has led to the evolution of 8 established (A-H) and 2 putative genotypes (I-J), each corresponding to a rather well-defined geographical distribution. The genotypes have clinical impact on natural course of infection, prognosis and treatment outcome. Genotypes A-D were identified in 1988, E-F in 1993, G in 2000 and H in 2002. The last decade the study of HBV phylogeny has been focused on identifying subgenotypes and recombinants, and their geographic distribution. The relatedness to non-human HBV has also become in focus but has remained unknown. Molecular methods have become increasingly used for epidemiological investigations of HBV infections, but the most appropriate genetic regions for such applications have not been established.

The aims of this thesis were to investigate the genotypes and genetic variants of HBV from different geographic areas in order to better understand the evolution and genetic variability of HBV and how it can be analysed.

HBV strains from Vietnam, Mongolia and Australia were amplified by polymerase chain reaction, subjected to direct sequencing and classified after phylogenetic analysis. In Vietnam 77% of all strains were of genotype B (mainly B4), 22% were of subgenotype C1 and one strain was a X/C recombinant (putative genotype I). Southeast Asian genotype C with C-1858, a variant that rarely develops the precore stop codon that abolishes synthesis of HBeAg, was shown to constitute a clade with two phylogenetic subgroups. Subgenotype D1 was found in strains from Mongolia, suggestive of a closer relation to Middle East and the Mediterranean area than with China. HBV from Australian Aborigines were found to represent a subgenotype (C4), with an S region that may originate from early recombination with an unknown genotype. Molecular clock rates were estimated by calculations based on genetic distances and data for human migration and were compared with mutations rates observed in patients. These rates, ranging from 2×10^{-5} to 10^{-6} substitutions per site per year suggest that HBV genotypes separated 2,000 – 40,000 years ago, while subgenotypes and the X/C recombinant would have evolved 1,000 – 20,000 years ago.

The HBV-D3 strains causing acute hepatitis B in Swedish injection drug users has changed at a low rate since the 1970ies, but three clades were shown to have circulated since 1975, and they were shown to be distinguishable by their pattern at 3 residues in the *a* determinant part of the S region. The S region appears to be favourable as primary target for subgenomic molecular epidemiology of HBV-D3.

In summary, results of this thesis contribute to explain the evolution of HBV and have a clinical impact on molecular epidemiology of acute HBV infections.

Keywords: hepatitis B virus, genotype, molecular epidemiology, phylogeny, sequence analysis, mutation

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- I. Alestig E, Hannoun C, Horal P, Lindh M.**
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- II. Alestig E, Hannoun C, Horal P, Lindh M.**
Hepatitis B virus genotypes in Mongols and Australian Aborigines. *Archives of Virology* 2001 Dec;146(12):2321-9.
- III. Thuy PTB, Alestig E, Liem NT, Hannoun C, Lindh M.**
Genotype X/C recombinant (putative genotype I) of hepatitis B virus is rare in Hanoi, Vietnam - genotypes B4 and C1 predominate. *Journal of Medical Virology* 2010 Aug;82(8):1327-33.
- IV. Alestig E, Söderström A, Norkrans G, Lindh M.**
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