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Prevalence and Molecular Characterization of Occult HBV Infection in Chronic Liver Disease Patients

1. Occult HBV infection was identified when two regions (Surface over lapping polymerase and Pre Core - Core region) of the HBV were amplified.
2. Occult HBV infection was found in 10%, 2.5%,5% and 28.8% of the cryptogenic liver disease patients, VBD's, HCW's and family contacts.
3. We could find 9.09%, 6%, 4.1% and 10.3% prevalence of the subjects with only single HBV region amplification (excluding patients with two region amplification).
4. Occult HBV infection was not only found in the IgG anti HBc positive patients, but also in the seronegative (IgG anti HBc negative) (27.2% in patients with cryptogenic liver disease, 40% in voluntary blood donors, 33.3% in health care workers and 3.5% in occult HBV infected family subjects.
5. Whole genome of 3.2 kb was amplified in 9.09% of the occult HBV infection positive cryptogenic liver disease patients, while in fragments in 63.6% of the occult HBV infection positive cryptogenic patients.
6. HBV DNA quantification was done by the both real time PCR and digene hybrid capture and was found to be low in most of the patients.
7. Full-length amplification was done in one patient, with high viral titer of 5418 IU/ml. Full length amplification was also successful in fragments with both high (2477, 1381,313 IU/ml) and low viral titer patients.
8. Liver biopsy was done in the occult HBV infected positive cryptogenic liver disease patients with high inflammation and fibrosis. Liver biopsy was done in the 5 occult HBV infected family contacts and in 13 chronic HBV infected patients.
9. Genotyping was done by the multiplex PCR and confirmed by sequencing in all the study and the control groups. Genotypes A and D were found in the occult HBV infected cryptogenic liver disease patients, voluntary blood donors, health care workers, family contacts. Sub genotype D3 was found

in occult HBV infected positive cryptogenic liver disease patients with full-length amplification.

10. Surface gene Mutation in the “a” determinant: C124F, A128V, T/S143M/L polymorphism, S114Q, T115H mutation, T143M, amino acid methionine (characteristics of genotype E instead of threonine at amino acid position 125) and P127S were observed in occult HBV infected positive cryptogenic liver disease patients, voluntary blood donors, health care workers, family contacts and in Chronic HBV infection patients. At amino acid position 223 (830 nucleotide), tryptophan (W) was known to be present in all the above study groups.

11. Mutations in full-length HBV studies: Naturally occurring mutations were found in all the four regions of the occult HBV infected cryptogenic liver disease patients with full-length HBV amplification. We found stop codon in the surface, pre-core, core and polymerase region with D3 sub-genotype showing genotype specificity.

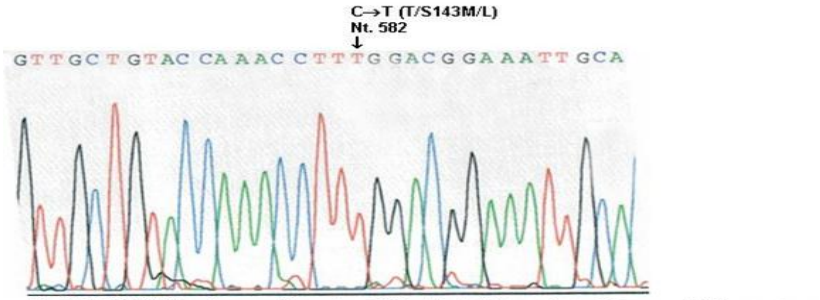
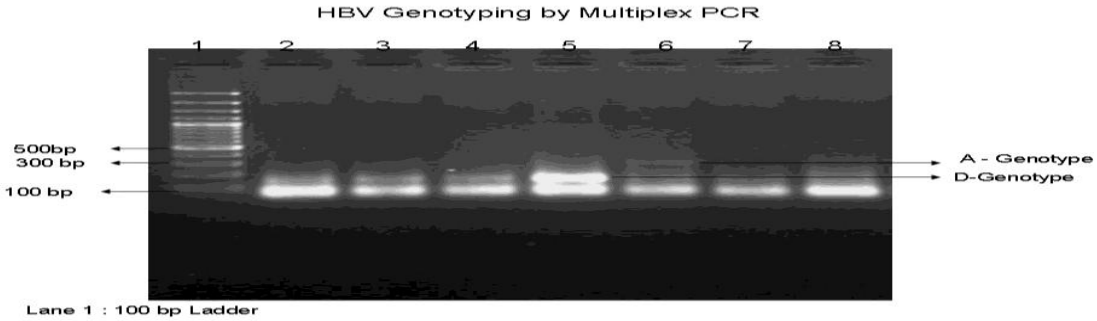


Fig 6.7 :T/S143M/L polymorphism at aa 143 in the “a”determinat of S gene with D -genotype



Lane 1 : 100 bp Ladder
Lane 2 : Positive Control
Lane 3 to 5 : D – Genotype
Lane 6 : A – Genotype
Lane 7 & 8 : Negative & Nested Negative control