

Molecular Epidemiology of Hepatitis B Virus in Pregnant Women: Genotype Distribution in a Central India Tertiary-Care Hospital

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Abstract

Background: Through describing the genetic diversity and geographical distribution of circulating viral strains, HBV genotyping is important for molecular epidemiology. During pregnancy, molecular characterization applies to complement serological screening and HBV DNA detection tests and relies on information characterizing the viral population present in infected women. **Material and Methods:** In Madhya Pradesh (MP), India, the serum HBsAg of 350 pregnant women was screened for Hepatitis B. Samples positive for HBsAg were tested for HBeAg/HBeAb and HBV DNA. The thesis laboratory protocol defines the genotypic characterization of HBV DNA positive specimens. **Results:** Eighteen of 350 women were HBsAg-positive. HBV DNA was found in 10 out of 18 HBsAg-positive samples. Genotype D was detected in 90% of DNA positive samples while genotype A was detected only in 10% of DNA positive samples. None other genotype was identified among the successfully genotyped ones. The findings of predominance of genotype D are supportive of several Indian population studies and the identification of the genotype A in the current study confirms that the genotype A also circulates in Central India. **Conclusion:** In this tertiary-care cohort, HBV DNA-positive pregnant women predominantly had genotype D. The research provides new pregnant-specific molecular information from Central India; however, the low number of genotyped samples limits this. There's a need for bigger multicentre studies that incorporate sequencing. Further, this should incorporate subgenotyping and phylogenetic analysis. Most importantly, these studies should define the molecular epidemiology of HBV in the region.

Keywords: Hepatitis B virus; genotyping; genotype D; genotype A; pregnancy; molecular epidemiology; HBV DNA; Central India.

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INTRODUCTION

The genome of HBV which is about 3.2kb contains a partially double-stranded circular DNA and is genetically diverse. Viral strains have been classified into genotypes based on nucleotide-sequence differences which are geographically distributed. Molecular characterization can complement standard serology with information on circulating viral lineages and genetic diversity.^[1,2]

In various regions of India along with genotypes A and D, HBV genotype E has also been spotted. Molecular studies from the northern, western, eastern and central regions show geographical variation and, in some instances, multiple subgenotypes or mixed infections.^[3-7]

Pregnant women are an important population for molecular surveillance as maternal HBV infection can cause MTCT. The current investigation aimed to characterize HBV genotypes detected among the HBV-DNA-positive pregnant women attending a tertiary care-indore hospital, thus providing the region-specific molecular evidence from Central India.

MATERIALS AND METHODS

The design and setting of the study took place at the Index Medical College Hospital and Research Centre, Indore,

Madhya Pradesh.

Participants: Three hundred and fifty pregnant women attending antenatal services were enrolled. Selected specimens were HBsAg-positive only for HBsAg.

Serological and molecular testing: HBsAg was assessed by ELISA, a serological test. HBeAg and HBeAb were focused on as additional markers. Real-time PCR detected HBV DNA. The samples that tested positive for HBV DNA were then examined for genotype using the molecular approach which is part of the protocol of this thesis.

Interpretation: Genotype frequencies were calculated among the successfully genotyped HBV DNA-positive samples. Genotype percentages were not extrapolated to Central Indian population, as available sample size for genotyping was only 10 specimens and were seen to be descriptive.

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RESULTS

Of the 350 pregnant women screened, 18 were HBsAg positive. Of the 18 specimens tested, 10 showed detectable HBV DNA by molecular testing. Characterization of the

samples showed that genotype D was the most predominant as it was identified in nine of the ten samples that were successfully genotyped. One identified a sample of genotype A. The findings showed that 90% of the isolates belonged to genotype D and 10% to genotype A.

Table 1: ?

Molecular parameter	Result
Pregnant women screened	350
HBsAg-positive	18 (5.1%)
HBV DNA-positive among HBsAg-positive	10/18 (55.6%)
HBV DNA-negative among HBsAg-positive	8/18 (44.4%)
Genotype D	9/10 (90.0%)
Genotype A	1/10 (10.0%)
Other genotype reported	None

DISCUSSION

Genotype D was the predominant molecular finding in HB-DNA-positive pregnant women.

Several Indian studies have shown that genotype D is an important or predominant circulating genotype which corroborates with this finding. The predominance of genotype D was reported among patients with chronic liver disease in New Delhi, India by Kumar et al., with similar work from North India also reporting genotype D in various clinical categories.^[3,4]

Research conducted in western India shows genotype D to be highly dominant. Chavan et al. noted that genotype D was found in 77.55% of patients infected with HBV, and genotypes C and A supplemented the rest. Research on the genomic sequencing of HBV, mainly on genotype A, has shown that genotype D is also present, indicating a diverse Indian HBV epidemiology.^[6]

The detection of genotype A in one pregnant woman is noteworthy from the perspective of Central India. As reported by Pacholi et. al, genotype A1 was documented from Central India, providing independent evidence for the circulation of genotype A in this region. This present observation thus supports regional evidence, but a single genotype-A isolate cannot be used to estimate this population prevalence.

The design of the study should be considered when interpreting the findings. There were only ten HBVDNA positive specimens available for genotyping, and all came from a single tertiary care centre. The 90% genotype-D proportion obtained is for the successfully genotyped specimens. It should not be interpreted as a precise estimate for Central India. Furthermore, without examination sequencing, no establishment of specific distribution, phylogenetic relationship or clinically relevant mutation at the subgenotype level is possible.

Molecular surveillance nevertheless has a lot of value. Genotype determination could help in mapping regional HBV diversity and may provide a basis for future sequencing-based investigations. Molecular methods used today, primarily sequencing and phylogenetic analysis of strains, are able to identify subgenotypes and characterize nucleotide variation above and beyond that afforded by simple serological tests.^[1,2,8]

Genotype surveillance and pregnancy-specific HBV prevention are worthwhile to consider. Assessment of maternal viral burden is not replaced by Genotyping. In terms of clinical decisions for MTCT prevention, virological and clinical criteria remain established and continue to guide decisions, particularly maternal HBV DNA where available and with appropriate neonatal immunoprophylaxis. As such, it must be regarded as complementary – not a substitute – to antenatal screening and viral-load assessment.^[9,10]

Strengths and Limitations: The main strength was the combination of antenatal HBsAg screening with HBV DNA detection and genotype characterization generating pregnancy-specific molecular data from Central India. A main limitation was the small molecular sample size because only ten specimens that were HBV DNA-positive were genotyped. The lack of external generalizability is further limited by its single-centre design and a lack of wider sequencing/subgenotyping. The distribution of genotypes must be considered as a first regional observation that needs to be confirmed in larger studies.

CONCLUSION

Genotype D was found in 90% while genotype A was detected in 10% pregnant women with HBV DNA-positive samples a tertiary care hospital in Indore India. These results contribute to the limited pregnancy-specific molecular evidence from Central India, which broadly corroborates the recognized prominence of genotype D in several Indian populations. Bigger multicenter studies utilizing sequencing, subgenotyping, and phylogenetic analysis are needed to better define the regional molecular landscape of HBV.

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Conflicts of interest

There are no conflicts of interest.

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