

POSTER PRESENTATIONS

IDENTIFICATION OF CONSERVED GENOMIC REGIONS FOR DIAGNOSTIC ASSAYS IN LOCAL HEPATITIS B VIRUS ISOLATES

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Chronic hepatitis B virus (HBV) infection remains a major global public health concern, frequently leading to severe liver pathologies including cirrhosis and hepatocellular carcinoma (with WHO estimating approximately 254 million chronic carriers worldwide based on 2024 reports). Uzbekistan is classified as a region with moderate-to-high endemicity, where genotype D (subgenotypes D1–D3) predominates in 85–96% of cases. Due to the lack of proofreading (3'-5' exonuclease) activity in the viral reverse transcriptase, HBV exhibits high genetic variability. Point mutations, insertions, and deletions within primer- and probe-binding sites can diminish the sensitivity of commercial diagnostic kits, increasing the risk of false-negative results. The present study aimed to identify the most conserved genomic regions of local HBV isolates through multiple sequence alignment to design next-generation PCR primers and fluorescent probes.

Blood plasma samples collected from patients with confirmed HBV infection were investigated. HBV DNA was extracted using the commercial "SOREX-DNA" kit via the silica-based sorption method. Target open reading frames (ORFs S, P, C, and X) were amplified using real-time PCR (TaqMan) format with specific primers designed independently by the authors. Obtained sequences were aligned using CLUSTAL W and MAFFT algorithms in MEGA 11 and BioEdit 7.2 software. Position-specific stability was evaluated based on Shannon entropy and polymorphism frequency, setting conservation criteria at a variability rate of <1–2% (conservation index >98–99%) with continuous conserved block lengths of 18–24 bp for primers and 20–30 bp for probes.

This methodological approach enables the rational design of in-house real-time PCR assays and fluorescent probes tailored to the genetic profile of local genotype D isolates, facilitating the localization of diagnostic test systems (import substitution). Detailed quantitative results of this study will be published following the release of the main research article.

Acknowledgement. This work was carried out within the framework of applied grant A-FA-2021-418 of the ICPS, Academy of Sciences of the Republic of Uzbekistan.