HBV-Host Junctional Sequences: A Novel Urine Tumor Marker for Recurrent Hepatitis B-Associated Hepatocellular Carcinoma with Low Alpha-Fetoprotein

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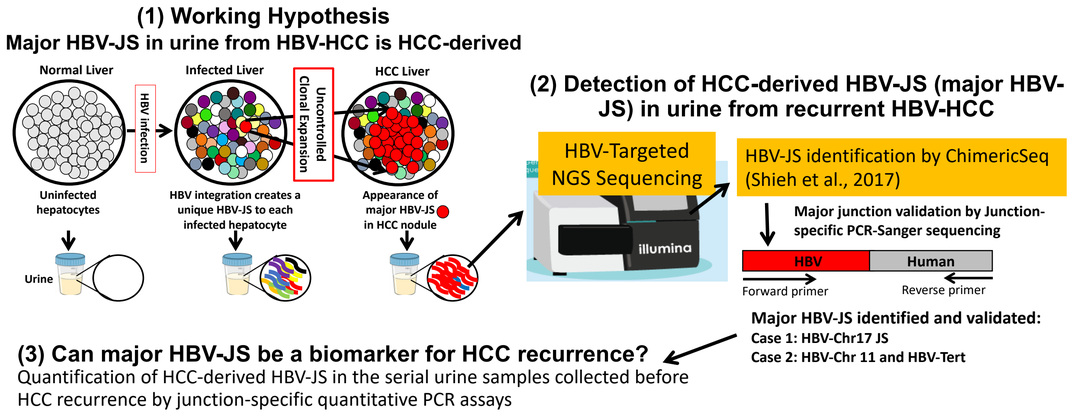
Introduction

- Hepatocellular carcinoma (HCC) is one of the most common and deadly cancers worldwide.
- A common risk factor for liver cancer is chronic infection with the hepatitis B virus (CHB).
- HCC screening and surveillance is limited by diagnostic accuracy of current testing modalities.
- Prior studies suggest the potential of urine-based biomarkers for HBV-associated HCC (HBV-HCC), as DNA fragments from both virus and tumor have been detected in urine from those with CHB and HCC.
- As most HBV-HCC tumors contain integrated components of the HBV genome, unique HBV-host junctional sequences (HBV-JS) represent a viable molecular signature to identify HCC.
- **This study evaluated a novel urine-based biomarker platform, utilizing HBV-JS DNA as a possible marker of tumor recurrence in patients with HBV-HCC.**

Methods

Urine sample from HBV-HCC patients with HCC recurrence confirmed by MRI was obtained. HBV-JS were detected by an HBV-targeted NGS assay (JBS Science Inc., Doylestown, PA) followed by ChimericSeq for junction detection. The most abundant NGS-detected junction sequences were then validated by PCR-Sanger sequencing. Quantitative junction-specific PCR assays were developed to track dynamic changes of HBV-JS in the urine specimens as summarized in the study outline below. HBV-JS sequences were detected from 2 cases of HBV-HCC with tumor recurrence **(Figure 1)**

Study Outline



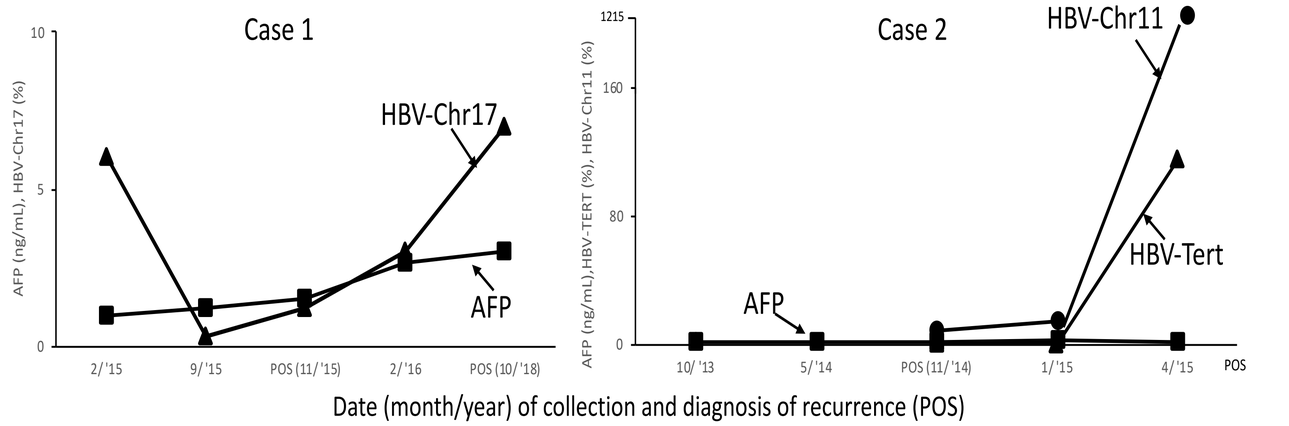
Case 1

A 78-year-old female with HBV-related cirrhosis was diagnosed with HCC in 2015. After microwave ablation, follow-up MRI revealed a new LI-RADS 3 lesion 1 year later. Subsequent imaging remained radiographically indeterminate until 2018 when the lesion was classified as definite HCC (LI-RADS 5). While serial AFP levels were negligible and MRI results variable, the unique HBV-JS DNA, HBV-Chr17, steadily increased from initial diagnosis to HCC recurrence.

Case 2

A 74-year-old male with HBV-related cirrhosis was diagnosed with HCC in 2014, which recurred with a LI-RADS 5 lesion after 1 year despite loco-regional therapy. While AFP levels were negligible, two HBV-JS DNA, HBV-Chr11 and HBV-Tert, rapidly increased after initial HCC diagnosis.

Figure 1: Detection of HBV-Host Junction DNA in Urine of Patients with Recurrent HCC



Discussion

- Unique HBV-JS sequences were detectable in the urine of patients with HBV- HCC.
- These sequences were detectable at increasing levels prior to diagnosis of recurrence of HCC by MRI imaging or AFP elevation.
- Together, our data suggest that HBV-JS DNA in urine maybe a further biomarker for the detection of HCC recurrence in patients with HBV-HCC.

References

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2. Lin SY, Zhang A, Lian J, et al. Recurrent HBV Integration Targets as Potential Drivers in Hepatocellular Carcinoma. *Cells*. 2021; 10(6):1294.

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