OPEN ACCESS

ISSN: 2835-9496

Mourtzikou A, et al. (2025): 1246
Ameri J Clin Med Re, 2025; 5(9): 100238
DOI: 10.71010/2835-9496/ajcmr-e238

Research Article

American Journal of Clinical and Medical Research

Evaluation of Novel Biomarkers in HBV DNA Positive Patients: Preliminary Results

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Citation: Mourtzikou A, Seitopoulou C, Stamouli M, Kalliora G, Kimouli M (2025) Evaluation of Novel Biomarkers in HBV DNA Positive Patients: Preliminary Results. Ameri J Clin Med Re: AJCMR-238.

Received Date: 03 September, 2025; **Accepted Date:** 09 September, 2025; **Published Date:** 15 September, 2025

Abstract

Chronic hepatitis B infection (HBV) is a public health problem worldwide, due to the increased likelihood of progression to cirrhosis and hepatocellular carcinoma (HCC) [1-3]. In addition to the relevant biochemical and molecular markers, such as ALT, AST, TBIL, ALP, GGT and HBV-DNA, new biomarkers and hepatokines have recently been identified, such as FABP1, FGF-19, ANGPTL3, ANGPTL4, ANGPTL6/AGF and HGF, which are associated with hepatic function, inflammation and liver injury [4-7]. FABP1 levels are elevated in patients with high HBV viral load, and show positive correlations with ALT, AST, and other markers of liver damage [4, 8-10]. FGF-19 exhibits a consistent increase from chronic HBV infection to cirrhosis and HCC, correlating with cholestasis markers and viral load [11-15]. HGF also appears to be elevated, although further investigation in HBV positive patients is required [16-17]. Although evidence regarding ANGPTL3, ANGPTL4, and ANGPTL6/AGF in HBV remains limited, they represent promising markers for metabolic regulation and liver function [16-21].

The aim of this study is to provide a concise review of novel biomarker levels in the context of HBV patients with positive HBV-DNA.

Positive correlations were observed between AST, ALT, ALP, GGT, TBIL and CA 19-9 with ANGPTL3, ANGPTL4 and FGF-19. Moreover, AFP correlated with FGF-21 and FGF-23.

The results showed positive correlations of ANGPTL3 and ANGPTL4 with FABP1, as well as of FGF-21 with FGF-23 were also observed. Viral load correlated with GGT, Total protein and AFP. Correlations regarding HGF and ANGPTL6 were not observed.

The biomarkers under study constitute a significant prognostic and diagnostic value in HBV patients with positive viral load, complementing the information obtained by biochemical and oncological markers.

Keywords: cirrhosis, hepatocellular carcinoma, hepatokines, viral load.

Introduction

Chronic hepatitis B virus (HBV) infection remains a major cause of chronic liver disease, with increased risk of cirrhosis and hepatocellular carcinoma, particularly in patients with high viral load [1]. Despite advances in antiviral therapies, there is an unmet need for additional biomarkers to enhance risk assessment and disease activity evaluation [5, 22, 23].

Traditional markers, such as AST, ALT, GGT, as well as HBV-DNA, remain essential in disease management, with defined thresholds guiding therapy decisions [24- 29].

Other factors, such as age >30 years, family history of HCC or cirrhosis, and elevated AST/ALT ratio (>2× ULN), further support the decision for treatment initiation or closer evaluation.

In this context, novel biomarkers and hepatokines are gaining increasing importance.

FABP1 is a lipid-binding isoform, elevated in HBV patients with positive viral load, positively correlating with AST, ALT, TBIL, GGT, ALP, and inversely with ALB, indicating inflammation and liver injury [30-32].

FGF-19 shows progressive elevation with disease progression from HBV to cirrhosis and HCC. Moreover, it correlates with markers of cholestasis, liver function, and fibrosis [12].

HGF is elevated in conditions with impaired liver function and represents a promising candidate in HBV patients [16].

ANGPTL3, ANGPTL4 and ANGPTL6/AGF are angiopoietin-like proteins involved in metabolic and lipid regulation. Although data in HBV remain scarce, they are considered as promising biomarkers for future studies [17].

Aim of the study

The purpose of this study is to summarize the most recent and emerging data regarding these novel biomarkers in relation to viral load and disease activity in HBV-DNA positive patients, within the framework of current guidelines [1, 23]. The observed findings correspond to preliminary results of a prospective study.

The study included 49 patients (25 males and 24 females, aged 25–87 years) with chronic HBV infection and positive viral load.

All samples were positive for HBsAg, anti-HBc, and anti-HBe. Liver biochemistry markers (AST, ALT, GGT, ALP, TBIL, total serum proteins, albumin) and tumor markers (AFP, CA 19-

9) were measured, along with ANGPTL3, ANGPTL4, ANGPTL6/AGF, FABP1, FGF-19, FGF-23 and HGF.

AST, ALT, GGT, ALP, TBIL, total serum proteins and albumin measurements were performed by the KONELAB 60 biochemistry analyzer, AFP, CA 19-9 by Cobas Roche e801 immunology analyzer and HBV-DNA by Roche Taqman 48 Light Cycler. ANGPTL3, ANGPTL4, ANGPTL6/AGF, FABP1, FGF-19, FGF-23 and HGF were measured by Luminex® assay (Luminex xMAP® technology).
Statistical analysis was performed by SPSS Version 25.0 and statistical significance was set at 0.050.

Results

Descriptive statistics of the biomarkers under study are presented in the following table.

Descriptive Statistics				
	Minimum	**Maximum**	**Mean**	**Std. Deviation**
VIRAL LOAD	7.10	3650000.00	78212.44	521050.04
SGOT	11.60	2299.70	74.57	326.12
SGPT	11.00	1521.50	63.28	217.29
TBIL	0.19	10.77	0.88	1.48
GGT	8.70	127.30	21.29	17.48
TPROT	5.21	8.59	7.47	0.51
ALB	3.46	5.17	4.49	0.35
ALP	35.40	186.70	64.44	25.43
CA 19-9	0.10	460.00	22.48	70.64
AFP_2	0.14	18.98	2.20	3.78
ANGPTL3	3.66	227.16	12.18	31.73
ANGPTL4	63.36	553.25	146.40	72.14
ANGPTL6/AGF	20.57	172.88	77.81	38.74
FABP1	0.27	13.46	1.01	2.01
FGF-19	0.04	0.59	0.21	0.12
FGF-21	0.01	0.48	0.07	0.10
FGF-23	0.02	0.21	0.06	0.04
HGF	0.04	0.51	0.19	0.10

Table 1. Descriptive statistics of the biomarkers under study.

Statistically significant differences in the mean values of all measured biomarkers according to gender (p-value=0.425) and age (p-value=0.219) were not observed.

Positive correlations were observed between AST, ALT, ALP, GGT, TBIL and CA 19.9 with ANGPTL3, ANGPTL4 and FGF-19. Moreover, AFP correlated with FGF-21 and FGF-23. Positive correlations of ANGPTL3 and ANGPTL4 with FABP1, as well as of FGF-21 with FGF-23 were also observed. Viral load correlated with GGT, Total protein and AFP. Correlations regarding HGF and ANGPTL6 were not observed. For all the aforementioned correlations p-values ranged from 0.000 to 0.020.

Discussion

This study included 49 patients with chronic HBV infection and positive viral load. All patients were positive for HBsAg, anti-HBc, and anti-HBe.

Statistically significant correlations of ANGPTL3 and ANGPTL4 with liver function enzymes and CA 19.9 were observed. ANGPTL3 and ANGPTL4 are angiopoietin-like proteins, *implicated in metabolic dysfunction and inflammatory processes in the liver, indicating potential prognostic value in HBV infection. The positive correlations between ANGPTL3 and ANGPTL4 with FABP1 further support the metabolic nature of HBV infection, as FABP1 has been shown to associate with liver injury and ALT/AST levels [17, 32-37]. Of particular interest, viral load was positively correlated with GGT, total proteins, and AFP, potentially reflecting the degree of disease activity and the risk of progression to more severe hepatic injury [1]. Moreover, the correlation of AFP with FGF-21 and FGF-23, as well as the interrelationship between FGF-21 and FGF-23, underscore the importance of fibroblast growth factors as regulators in chronic HBV infection [5, 23, 32, 37]. The correlation between liver function enzymes and FGF-19 is consistent with recent reports highlighting FGF-19 as a biomarker of progression from HBV to cirrhosis and HCC [12].

Conclusions

The present study highlights the prognostic and diagnostic importance of novel biomarkers, including ANGPTL3/4/6, FABP1, FGF-19, FGF-23, and HGF, in patients with chronic HBV infection and positive viral load. These biomarkers complement biochemical and oncological markers, offering additional insights into liver inflammation, metabolic regulation, cholestasis, and neoplastic progression. Although based on a small sample size, this study contributes to the growing body of evidence that these biomarkers could be integrated to laboratory diagnostic and prognostic algorithms.

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