CLINICAL SIGNIFICANCE OF CIRCULATING CIRCADIAN PROTEINS BMAL1 AND PERIOD2 IN PATIENTS WITH HEPATITIS C VIRUS-RELATED LIVER DISEASE

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INTRODUCTION

- Hepatitis C virus (HCV) infection is a major cause of chronic liver disease worldwide.
- The circadian clock (CC) is an endogenous timekeeping system that synchronizes 24-hr oscillations of behavioral and biological processes.
- The CC machinery forms a feedback timing circuit and is composed of a series of genes.
- Brain and muscle Arnt-like protein 1 (BMAL1) and Period 2 (PER2) genes are crucial components of the CC.
- Disruption of the circadian rhythms is linked to a variety of diseases including viral infections, liver diseases, metabolic derangements, and cancer.

AIM of the Work

- The present study was conducted to assess the clinical significance of circulating circadian proteins BMAL1 and PER2, positive and negative regulators of the CC, in patients with HCV-related liver disease.

SUBJECTS

- 60 patients with chronic HCV infection {20 patients with chronic hepatitis C (CHC), 20 cirrhotic patients without hepatocellular carcinoma (HCC), 20 cirrhotic patients with HCC] and 20 healthy controls.
- Exclusion criteria: other causes of chronic liver disease; infections; inflammatory disorders; or malignancy; cardiac, respiratory or renal disease; previous antiviral or HCC treatment.

METHODS

- Anthropometric measurements; Body mass index (BMI) and Waist-to-hip ratio.
- Liver test profile, lipid profile, fasting plasma glucose (FPG), fasting serum insulin (FSI) and Homeostatic model assessment for insulin resistance (HOMA-IR).
- Serum alpha-fetoprotein (AFP).
- Serum BMAL1 and PER2 levels using ELISA kit.
- Child-Pugh classification and Model for End Stage Liver Disease Sodium (MELDNa) score.
- HCC stage using Barcelona Clinic Liver Cancer (BCLC) staging. System.
- Liver fibrosis scores: Aspartate aminotransferase-to-platelet ratio index (APRI) and Fibrosis-4 index (FIB-4).
- Acoustic radiation force (ARFI) impulse elastography to assess liver stiffness by measuring liver shear wave velocity (LSWV). Cirrhosis was highly suggestive if LSWV > 1.8 meter/sec (m/s).

RESULTS

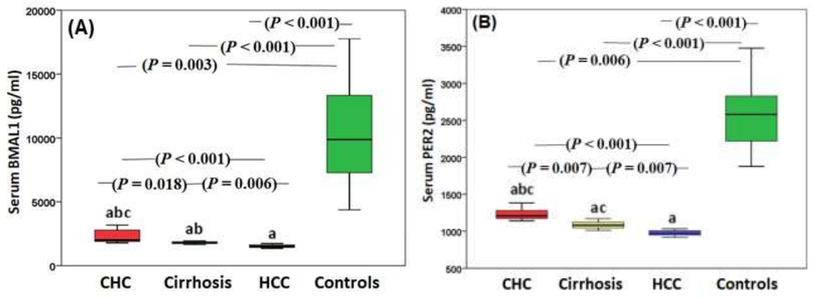

Figure 1: Statistical comparisons between patients with CHC, cirrhotic patients without HCC, cirrhotic patients with HCC and healthy controls as regards serum (A) BMAL1 and (B) PER2 levels (pg/ml) ($P < 0.001$).

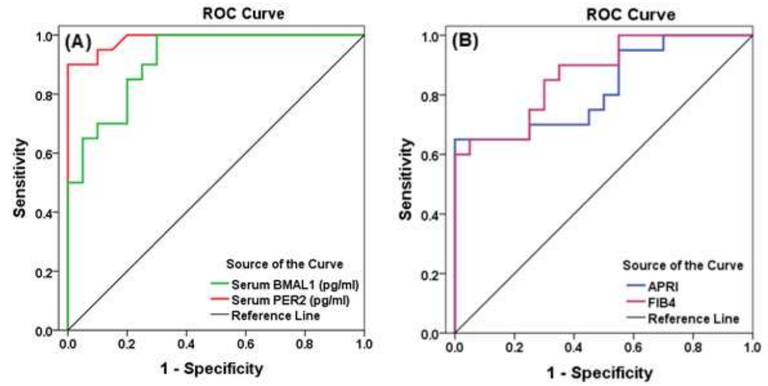

Figure 2: Diagnostic accuracy of (A) serum BMAL1 and PER2 (pg/ml), (B) APRI and FIB-4 in discriminating patients with cirrhosis from patients with CHC (AUC = 0.915, 0.986, 0.823 and 0.870 respectively, $P < 0.001$).

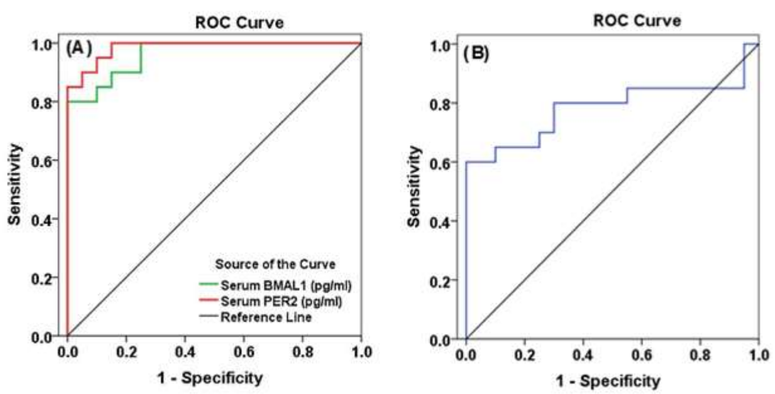

Figure 3: Diagnostic accuracy of (A) serum BMAL1, PER2 (pg/ml), and (B) AFP (ng/ml) in discriminating cirrhotic patients with HCC from those without HCC (AUC = 0.963, $P < 0.001$; 0.985, $P < 0.001$ and 0.783, $P = 0.002$ respectively).

RESULTS

Table 1: Statistical correlations between serum levels of BMAL1 and PER2 (pg/ml) and other parameters in patients with chronic HCV infection.

Parameters	Serum BMAL1 (pg/ml)		Serum PER2 (pg/ml)	
	r value	*P*-value	r value	*P*-value
Serum AST (U/L)	-0.756	< 0.001*	-0.660	< 0.001*
Serum ALT (U/L)	-0.703	< 0.001*	-0.623	< 0.001*
HCV RNA (x10³ IU/ml)	–0.262	0.043*	-0.345	0.007*
Child-Pugh score	-0.544	< 0.001*	-0.592	< 0.001*
MELDNa score	-0.443	0.004*	-0.438	0.005*
APRI	-0.765	< 0.001*	-0.718	< 0.001*
FIB-4	-0.726	< 0.001*	-0.717	< 0.001*
LSWV (m/s)	-0.819	< 0.001*	-0.852	< 0.001*
BMI (kg/m²)	-0.449	< 0.001*	-0.399	0.002*
Waist-to-hip ratio	-0.458	< 0.001*	-0.391	0.002*
Serum TC (mg/dl)	-0.299	0.020*	-0.283	0.028*
HDL-C (mg/dl)	0.469	< 0.001*	0.405	0.001*
LDL-C (mg/dl)	-0.404	0.001*	-0.392	0.002*
Serum TG (mg/dl)	-0.281	0.020*	-0.322	0.012*
HOMA-IR	-0.557	< 0.001*	-0.557	< 0.001*

TC, Total cholesterol; HDL-C, High density lipoprotein-cholesterol; LDL-C, Low density lipoprotein-cholesterol; TG, Triglycerides.

Table 2: Statistical correlations between serum levels of BMAL1 and PER2 (pg/ml) and tumor progression parameters in cirrhotic patients with HCC.

Parameters	Serum BMAL1 (pg/ml)		Serum PER2 (pg/ml)	
	r value	*P*-value	r value	*P*-value
Serum AFP (ng/ml)	-0.599	0.005*	-0.628	0.003*
HCC diameter (cm)	-0.642	0.002*	-0.588	0.006*
BCLC stage	-0.591	0.006*	-0.594	0.006*

CONCLUSIONS

- Decreased circulating BMAL1 and PER2 during chronic HCV infection signify a dysfunction of the circadian rhythm and may play a role in liver disease progression, associated metabolic derangements and pathogenesis of HCC.
- Serum BMAL1 and PER2 could be potential biomarkers for the progression to cirrhosis and development of HCC in patients with HCV-related liver disease.