Evaluation of miR-122 levels in chronic HBV and liver cirrhosis patients

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Abstract

Background: MicroRNA is a type of small RNA of about 22 nucleotide length. Most popular miRNAs are found in the liver and are extensively important in determining the biological and clinical functions. MicroRNA, in host cells, may impact the replication of viruses either positively or negatively. In this study the miR-122 expression was examined and compared in three groups, two sample groups of patients with chronic hepatitis and hepatitis B virus-associated cirrhosis, and a healthy control group.

Materials and Methods: In this study, 108 samples of whole blood were taken from each participant. Then the miRNA expression evaluation was conducted through relative real time PCR.

Results: The results indicated that miR-122 expression was elevated in patients with chronic hepatitis B and HBV related cirrhosis about 1.8 times (P<0.05) more than control group which is statistically significant.

Conclusion: According to the results of this study, measuring the miR-122 expression levels may be used as a biomarker and an indicator of the disease progression of chronic hepatitis B to HBV related cirrhosis to HCC, but needs more investigations and more samples.

Keywords: microRNA, chronic hepatitis B, liver cirrhosis, miR-122

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Introduction

Hepatitis B Virus (HBV) is the most popular infectious disease throughout the world (1). Hepatitis B infection is an inflammatory and liver necrosis disease, and still remains an important global issue (2). MicroRNA is a type of small RNAs of about 22 nucleotide length; it is expressed in most eukaryotes and acts as a regulating key of gene expression by attaching to the end of 3'mRNA target (3). Viral mRNA targeted by cellular mir-RNA, includes all cells involved in cell survival, replication and immune responses, through which cell cycle of viruses improves in a host cell environment (4). Viruses can affect the microRNA expression profiles positively to promote replication potential and pathogenesis. (5). The sequence of miRNA-122 includes a single genomic locus on chromosome 18 in humans derived from 85 coding exons. Reduced miR-122 expression in HBV patients causes enhanced replication and simulation of the virus, indicating Cyclin G1 expression suppression by miR-122. (6) In this study the miR-122 expression was examined and compared in three groups, two sample groups of patients with chronic hepatitis and hepatitis B virus-associated cirrhosis, and a healthy control group.

Methods

In this study 108 participants (54 men and 54

women), including patients with chronic hepatitis B, HBV related cirrhosis and the control group were examined. The samples of whole blood (5ml) treated with EDTA, were taken from participants.

The RNA extraction was done using 2ml whole blood by RNX-PLUS kit following the manufacturer guidelines.

To evaluate the miRNA expression, relative real time PCR by Parsgenome, Iran mirAmp kit used. The Real time PCR by $\Delta\Delta$ CT was used to measure miRNA expressions. Finally, the collected data was processed with T-Test.

Results

The results of this study indicated that miRNA-122 expression in patients with chronic hepatitis B and HBV related cirrhosis was 1.8 times higher than the control group which is statistically significant (P < 0.05). Figure 1 shows the results of the study in all three groups.

Discussion

The target miRNAs act as a new basic key in controlling gene expression of cells. To predict the risk of disease progression in patients with chronic hepatitis B is of considerable importance. The reports state that the level of miR-122 relates to HBs antigen directly, which is an indicator for viral translation; miR-122 levels can be an important indicator for progressing stages of the disease (7).

In this study, two groups of patients with chronic hepatitis B and HBV related cirrhosis showed an increase in HBsAg and miR-122 levels. The most popular miRNAs are found in liver and are extensively important in determining the biological and clinical functions (8). In 2010, some reports by TaqMan format real time PCR indicated that miR-122 contains a suppression effect in the expressing of HBV genes (9).

Jian Zhou et al, aimed to examine the effect of HBV related HCC on miRNAs and indicated that miRNA expressions in four sample groups (healthy, chronic hepatitis B, cirrhosis, and HBV related HCC) through microarray to measure the miR-122 expression levels, which showed a significant increase in patients with HCC compared to the



Figure 1. The miR-122 expression levels in three groups by T- test method (P value = 0.047)

healthy individuals. But measuring miR-122 expression by qRT-PCR method showed a decrease in HCC group compared to the healthy individuals (10).

Our findings indicated that the rate of miR-122 expression in patients with chronic hepatitis B and HBV related cirrhosis was 1.8 times more than the healthy control group which is statistically significant (P < 0.05).

The results of this study indicated that patients with liver cirrhosis exhibited higher levels of miR-122. In this investigation, we focused on limited miRNAs role in interactions between host and the virus, especially in hosts with HBV. Various studies revealed that miRNA plays a key role in viral physiologic and pathologic processes.

Comparing the results with previous findings, shows conflicting results; the results of Jian Zhou et al. (10) reported a decrease in miR-122 expression levels in Hepatitis B patients, however our results indicates that miR-122 expression increases in hepatitis B patients, this difference in results can be described by the fact that previous investigations were on patients with HCC, but the sample group in this study only included patients with HBV related cirrhosis.

This study shows that the increased miR-122 expression compared to the control group can be used as a biomarker to indicate chronic hepatitis B and HBV related cirrhosis before progressing to Hepatocellular carcinoma.

Conflicts of Interest

The authors of this paper declare no competing interests.

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References

1. Robinson SW: Hepatitis B virus and hepatitis D virus. In: Mandell GL, Bennet JE, and Dolin R, (eds.), Principle and practice of Infectious Disease. New York: Churchill Livingstone, pp. 1406-1439, 2000.

2. Wooddell CI, Rozema DB, Hossbach M, John M, Hamilton HL, Chu Q, at al. Hepatocyte-targeted RNAi therapeutics for the treatment of chronic hepatitis B virus infection. Mol Ther 2013; 21: 973-985.

3. Thomas F Baumert, Robert Thimme, Fritz von Weizsacker . Pathogenesis of hepatitis B virus infection. World J Gastroenterol 2007 January 7; 13(1): 82-90.

4. Xia J, Tatsuo K, Shuang Wu, Masato N, Tatsuo M, Shingo N, at al. Regulation of microRNA by hepatitis B virus infection and their possible association with control of innate immunity. World J Gastroenterol 2014 June 21; 20(23): 7197-7206.

5. Jian Zh, Lei Yu, Xue G, Jie Hu, Jiping W, Zhi D, at al. Plasma

MicroRNA Panel to Diagnose Hepatitis B Virus–Related Hepatocellular Carcinoma. Journal of Clinical Oncology, Volume 29, Number 36, December 2011, 4781-4788.

6. Coulouarn C, Factor VM, Andersen JB, Durkin ME, Thorgeirsson SS. (2009). Loss of miR-122 expression in liver cancer correlates with suppression of the hepatic phenotype and gain of metastatic properties. Oncogene; 28(40):3526-36.

7. Muriel G, Emmanuel J, Arnold M, Stanislas L, Alexandra H. miR-122, a paradigm for the role of microRNAs in the liver. Journal of Hepatology 48 (2008) 648–656.

8. Xiaoyong Zh, Jinlin H, Mengji Lu. Regulation of hepatitis B virus replication by epigenetic mechanisms and microRNAs. Frontiersin Genetics, Epigenomics and Epigenetics. October 2013, Volume4, Article 202.

9. L Qiu, H Fan, W Jin, B Zhao, Y Wang, Y Ju, L Chen. miR-122induced down-regulation of HO-1 negatively affects miR-122mediated suppression of HBV. Biochemical and Biophysical Research Communications 398 (2010) 771–777.

10. Jian Zh, Lei Yu, Xue G, Jie Hu, Jiping W, Zhi D, at al. Plasma MicroRNA Panel to Diagnose Hepatitis B Virus–Related Hepatocellular Carcinoma. Journal of Clinical Oncology, Volume 29, Number 36, December 2011, 4781-4788.