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Title: Integrated analysis of microRNA and mRNA expression profiles in HBx-expressing hepatic cells

Dear Editors and Reviewers,

Thank you for your letter and for the reviewers' comments concerning our manuscript entitled "Integrated analysis of microRNA and mRNA expression profiles in HBx-expressing hepatic cells". Those comments are all valuable and very helpful for revising and improving our paper, as well as the important guiding significance to our researches. We have studied comments carefully and have made correction, which we hope meet with approval. Revised portion are marked in red in the manuscript. The main corrections in the paper and the point-by-point response to the reviewer's comments are as following:

Reviewer #1:

Comment 1: One is that authors only contemplate unidirectional regulation microRNA to mRNA, and also they didn't mention any possible regulation between genes without the participation of microRNAs.

Response: Thank you for your nice suggestion. The interaction network of miRNA and mRNA is very complicated inside the cells and it is worthy of in-depth research. In this study, we mainly focus on the regulation of miRNA to mRNA, however we will try to address the complex interaction between microRNA and mRNA in the future work as you have suggested.

Comment 2: The second concern is that all the relationship between microRNA and mRNA are in- silico predictions they didn't perform any corroborations of their results or mention if their predictions (some of them) had been validated by other groups.

Response: Thank you for your question. Our study is indeed majorly based on the results of RNA-seq analysis of the total RNA sample via gene chips. Besides, we also performed q-PCR to confirm the relative mRNA expression of the 9 selected genes and 4 selected microRNAs in which we are interested for further study (Please see Figure 1C and Figure 3B). Some of our predictions have been validated by other studies or groups, which we have mentioned in the discussion part (Please see the third paragraph of Discussion in manuscript).

Comment 3: The last concern is that authors did not mention if DNA methylation by HBx could play a role in the hepatocarcinogenesis.

Response: Thank you. As you have mentioned, amounting evidence has demonstrated that HBx could promote DNA methylation via upregulation of DNA methyltransferases activity, thus playing an important role in hepatocarcinogenesis [1]. It's very important for us to explore the role of HBX and methylation as well as other posttranslational modification in the future study.

Reviewer #2:

Comment 1: Their approach to clarify the association of miRNA and mRNA in HBx-carcinogenesis using softwares seems interesting. However, they need to show whether the knockout of sopme Hub miRNA result in the upregulation of target mRNA shown in Fig.4 in order to verify the data.

Response: Thank you for your nice suggestion. We will go on study the role of hub miRNAs in hepatocarcinogenesis in our future work, including knockout the expression of some hub miRNAs to investigate the change of target mRNAs and biological behaviors of HCC cells.

Reference

1. Zhang XD, Wang Y, Ye LH. Hepatitis B virus X protein accelerates the development

of hepatoma. Cancer biology & medicine **2014**; 11:182-90.