February 8 , 2017

World Journal of Gastroenterology

Re: ESPS Manuscript No. 32025

Dear Dr. Qi:

My co-authors and I respectfully submit the accompanying revised manuscript, "Early hepatitis B viral DNA clearance predicts treatment response at week 96," for consideration of publication in the *World Journal of Gastroenterology*.

Our point-by-point response to the reviewers is attached. We have attempted to respond fully to the reviewer's comments and criticism, and we are grateful for their input, which has helped us improve the manuscript.

We will be grateful for your careful consideration of our article, and we look forward to receiving your reply.

Sincerely,

Encl:

Point-by-point response to reviewers' comments

Reviewer 0921008

1. **The title can be more clear.**
   We feel the title is concise and includes the principal features of the paper. The title is at the word limit (12 words) imposed by the World Journal of Gastroenterology, *so we cannot add more words.*

2. **In the abstract, once the ROC abbreviation was defined, do not use the full term again.**
   We have omitted the abbreviation from the abstract.

3. **Please elaborate on the design type in the abstract.**
   We have clarified that the study is a Prospective observation study.

4. **Observational studies differ from clinical trials. But I see the paper is registered as a clinical** trial at the journal's website. Please clarify.
   It is a Prospective observation study rather than clinical trail. We are so sorry that it is our error to choose the wrong type of article when submitting the manuscript.

5. **Please clarify if patients were administered any antivirals in the initial treatments?**
   We have stated, Lines 142-152, that the patients had "no previous use of any anti-HBV drugs or other antiviral agents."

6. **If so, did you account for the dose and types and duration of medications administered in the initial treatment? (in the M&M section, I saw you had excluded such patients. It would be nice to report this in the abstract too --optional).?????**
   The dose, types, and duration of antiviral drugs we used are listed in the M and M section, lines 142-152. We have mentioned the excluded patients in the abstract, lines 22, 23.

7. **In the introduction, please correct "Other therapy strategy " to "Other therapy strategies "**
   Correction has been made.

8. **Please make sure the text is free of grammar errors.**
   The paper has been proofread again to check for errors.

9. **"As this is clinical outcome " should become "As this clinical outcome "**
   Correction has been made.

10. **In the M&M section, how did you decide which patients should receive which treatments?**
    Follow the recommendations in the guideline, all patients voluntarily received the

antiviral drugs according to their condition and economic situation. Lines 145-7. "Patients were given individual conventional doses of one of the treatments for 96 weeks: etc."

11. **I see different interventions have been administered. So this study seems to be a clinical trial rather than an Observational study (as stated in the abstract). Please double check and make corrections.**
We have stated in the abstract and the material and methods (Line 130) that it is a clinical trial study.

12. **Was the intervention randomized?**
The intervention was not randomized.

13. **Was it blinded?**
No, it was not blinded.

14. **The conclusion at the end of the text should be expanded considerably. There are more valuable points to be reported as a take-home message.**
We have added several points to the conclusion, lines 340-6.

15. **Results, supplementary info, tables, and figures are nice. I suggest you to also publish your raw data**.
We ensure to protect all patients' privacy and interest. Data could be available only with the consent of patients.

16. **Discussion is currently more of an interpretation of the results. It has 4 paragraphs. The first one is interpretations. The second one describes that there were no previous similar reports and briefly compares few aspects of this study with previous ones. The third one is more suited for the Introduction. The fourth one is the limitations. The discussion can be expanded, taking into account more comparisons with previous studies, and explaining the reasons for controversies and agreements. It is accepted that the lack of very similar studies can shorten the discussion, but still many other aspects can be compared with previous studies. Good luck**

We have added text about the risk of HCC in unresolved treatment of HBV infection (Lines 302-310) and text concerning recurrent HBV infection (Lines 311-326). See reviewer No. 02860911, Point no. 4 and no. 5, below.

Reviewer 02860911

1. **The compliance issue shall be evaluated in three arms of patients.**

We are not sure what the reviewer wants us to add. We did not formally evaluate the compliance of patients in the three arms. However, we have no reason to believe that compliance was different among the patient groups.

2. The male to female ratio in the 3rd arm is not the same proportion to the other two arms, is gender be a factor for poor viral suppression by week 24?

A study has shown that gender is not correlated with the efficacy of nucleoside anti-viral drugs: Wan MB. The expert panel symposinm on treatment for chronic hepatitis B with Lamivudine. Chin Hepatol,2009,14:167-169. *Also, we performed a correlation analysis of gender and low viral DNA load at 24 wk, and found no correlation (p=0.833). Finally, the male:female ratio among our three patient groups was not statistically significantly different. Please see manuscript lines 212-215.*

3. Does different potent Nuc have different viral suppression result?

In new Table 5, referred to in text page 11, we have listed anti-viral efficacy of various nucleoside drugs. Among 54 patients with HBV DNA <10 IU/ml at 24 wk, 5 (5/16=31.25%) received LAM+ADV, 9 (9/33=27.27%) received telbivudine, and 21 (21/59=35.59%) received entecavir tablets; 20 (20/64=31.25%) were entecavir dispersible tablets. No significant differences among the drug treatments, as determined by HBV DNA below detection limits, ALT return-to normal rate, rate of conversion to HBeAg-negative status, and HBeAg conversion rate at 94 weeks were found (P=0.127). See lines 235-243.

Table 5. Treatment efficacies of various antiviral therapies

Virological parameters at 96 week	LAM+ADV	Telbivudine	Entecavir tablets	Entecavir dispersible tablets	P
HBV DNA below detection (<1000 IU/ml as a reference)	68.75%	66.67%	79.66%	78.13%	0.089
ALT return-to-normal rate	81.25%	75.76%	86.44%	85.94%	0.096
Rate of conversion to HBeAg negative status	31.25%	36.36%	35.59%	34.38%	0.615
HBeAg seroconversion rate	25%	33.33%	32.2%	31.25%	0.203

LAM, lamivudine; AVD, adefovir; ALT, alanine aminotransferase

How's the viral suppression result if separate the patients into ETV/TDF and others?

We did not use TDF because it was not on the market in China when we started this study. However, we did divide the patients into an ETV group (entecavir tablets and entecavir dispersible tablets, 49 cases) and non-ETV group (LAM+ADV and telbivudine, 123 cases). In the 54 patients with HBV DNA <10 IU/ml at 24 wk, 41 (41/123=33.3%) were in the ETV group, and 14 (14/49=28.57%) were in the non-ETV group. Thus, viral suppression was slightly lower in the non-ETV group, but the difference was not statistically significant (P=0.737), perhaps because of the low number of cases in that group.

4. Does the inadequate viral suppression leads to increase incidence of HCC during follow-up? since mutated virus has been reported to be carcinogenic effect

In our study, drug resistance occurred in some patients with inadequate viral suppression. Drug resistance can lead to HCC in chronic hepatitis B patients, and high HBV DNA load also can increase the risk of HCC (Chan HL, Tse CH, Mo F, et al. High viral load and hepatitis B virus carcinoma. J Clin Oncol, 2008 Jan 10,26(2):177-182). *If HBV DNA becomes negative or decreased after treatment, the risk of HCC decreases, and the risk of HCC is lower with lower HBV DNA loads* (Ikeda K, Arase Y, Kobayashi M, et al. Hepatitis B virus-related hepatocellular carcinogenesis and its prevention. Intervirology 2005 Jan-Feb,48(1):29-38. *We have added this subject and references to the discussion (Lines 312-320).*

5. What's the suggestion derived from this finding? Change to other treatment?

Based on our results, when the HBV DNA load is lower in the early stages of anti-viral treatment, later outcome is better, and the risk of drug resistance is lower. Research shows that, if patients are treated with standard anti-viral medications according to guidelines, and treatment is stopped after successful viral response, 44% will have virological recurrence (Fung J, Lai CL, Chan SC, et al. Correlation of liver stiffness and histological features in healthy persons and in patients with occult hepatitis B, chronic active hepatitis B, or hepatitis B cirrhosis. Am J Gastroenterol. 2010;105:1116–1122), *and 50% will have clinical recurrence* (Liang Y, Jiang J, Su M, et al. Predictors of relapse in chronic hepatitis B after discontinuation of anti-viral therapy. Aliment Pharmacol Ther. 2011;34:344–352.

The reason for recurrence is not known, but an important possibility is that, at the end of treatment, HBV DNA is not suppressed to an adequate level. We believe that the lower the HBV DNA load, the better the prognosis. High-sensitivity HBV DNA detection is useful in predicting anti-viral efficacy as well as in monitoring viral replication and recurrence after cessation of treatment. Patients whose HBV DNA is ≥10 IU/ml should be closely monitored, and drug-resistant loci tested when necessary, so that the treatment regimen can be adjusted at an appropriate time. Please see manuscript lines 321-336).