

February 04, 2014

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 7669_Review.doc).

Title: Recent Advances in Molecular Diagnostics of Hepatitis B Virus (Topic Highlight)

Author: Sibnarayan Datta, Soumya Chatterjee, Vijay Veer

Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 7669

The manuscript has been revised and improved according to the suggestions of reviewers. A point to point answer to the comments by the reviewers is also attached with this letter for your kind perusal.

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology*.

Sincerely yours,



Dr. Sibnarayan Datta,
Scientist, Defence Research Laboratory,
Post bag No. 2, Tezpur, Assam, PIN-784001, INDIA.
email: *sndatta1978@gmail.com*
Telephone: +91-3712-258508; **Fax:** +91-3712-258534

Reviewer ID# 00052899

Reviewer's Comment: In this review manuscript, the authors clearly and systematically described recent advancements in the field of HBV molecular diagnostics, especially a number of isothermal amplification assays, such as LAMP, TMA, LCR, and RCA. However, there are some problems that should be considered.

Our Response: We thank the reviewers for their appreciation of our efforts.

Reviewer's Comment 1: the authors should go ahead to the introduction of HBV diagnostics instead of too many descriptions of HBV distribution, genome, and pathogenesis and so on.

Our Response: We have now omitted the above-mentioned portions as per the reviewer's suggestions.

Reviewer's Comment 2. The paper made a detailed analysis of recent advancements of each single molecular diagnostic assay, but lacked necessary comparison of advantages, disadvantages and applicable conditions among different techniques, which was recommended to be presented in the form of figures or tables to take place of lengthy descriptions in words.

Our Response: We have now included a table incorporating a comparison among these technologies.

Reviewer's Comment 3. The length of the manuscript and the number of reference should be shortened. More attention should be focused on the description, comparison and reasonable choice of various diagnostic methods.

Our Response: We have now revised the manuscript accordingly. We have included brief discussion on choice of diagnostic methods in the conclusion.

Reviewer's Comment 4. There are some typos that should be corrected prior to publication.

Our Response: We have now revised the manuscript carefully.

Reviewed by 02453015

Reviewer's Comment 1. According to the topic, this review paper should start from the section of "HBV Diagnosis".

Our Response: We have removed the portions as per the reviewer's suggestions.

Reviewer's Comment 2. As a review paper, Figures and/or tables comparing the mechanisms, benefits and differences of the diagnosis methods are highly recommended.

Our Response: We have now included a table incorporating a comparison among these technologies. We have also included brief discussion on choice of diagnostic methods in the conclusion.

Reviewer's Comment 3. The length and number of references should be reduced. Please pay special attention to the application of each method under different circumstances. A clear guide to choice of diagnosis methods is more beneficial than irrelevant commonly known HBV basics. Please also follow the requirement of Manuscript Type (Topic Highlights).

Our Response: We have now revised the manuscript according to the reviewer's suggestions.

Reviewer's Comment 3. English still needs to be improved.

Our Response: We have now revised the manuscript carefully.

Reviewed by 02822399

Reviewer's Comment: Datta and his colleagues did a good job and illustrate their idea clearly.

Our Response: We thank the reviewers for their appreciation of our efforts.

Reviewer's Comment 1- Too long introduction outside the scope of the review. It occupied about one third of the review. You have to go directly to your idea about molecular measurements. No need for HBV distribution, pathogenesis, life cycle and so on.

Our Response: We have removed the portions as per the reviewer's suggestions.

Reviewer's Comment 2- Again your review about molecular diagnosis and contain over than three pages about HBsAg.

Our Response: We have now removed the portions related to HBsAg, as per the reviewer's suggestions.

Reviewer's Comment 3- Starting from page 12 (polymerase chain reaction based strategies), you need to make subtitles for clarity and to organize your ideas.

Our Response: We have now modified the manuscript according to the reviewer's suggestions.

Reviewer's Comment 4- Your review focuses mainly on the isothermal amplification strategies. However, many other techniques are present like TAIL and others.

Our Response: We completely agree with the opinion of the Reviewer, that the present review is primarily aimed at isothermal nucleic acid amplification techniques. Isothermal amplification methods are preferred over PCR based methods now a days due to the simplicity of instrumentation and possibility of integration into point-of-care devices. We also acknowledge that apart from the methods described herein, a number of other amplification methods are also available, such as SDA, tHDA, TAIL-PCR, etc. Since it was not possible to include all of them within the limits of the article, we in this review, tried to incorporate only those methods, which have been reported for HBV diagnosis. Although there are almost no credible reports on use of SDA and tHDA for HBV diagnosis, we have now included these two methods in the revised manuscript as having considerable potential for application in HBV diagnostics.

However, with due regards, we wish to mention here that we have not included TAIL, since this method is primarily PCR based, and is predominantly aimed at amplifying large DNA fragments or unknown flanking sequences. Further, TAIL-PCR involves complex thermal cycling parameters, tricky standardization procedures and relatively higher time that limit its use in diagnostics.

Reviewer's Comment 5- You have to make comparisons between different techniques and the advantages of one over the others depending on time of detection, specificity, sensitivity and so on.

Our Response: We have now included a table incorporating a comparison among these technologies.

Reviewer's Comment 6- Abbreviations in your review needs to be revised carefully: a- Many abbreviations appear in your abstract without explanation. b- In pages 8 and 12 the same sentence is doubled for explanation of LAMP, NASBA and TMA. c- Many abbreviations appeared in your document without explanation: CHB, WHO, LC, RFLP, ... d- Try to make a table of abbreviation.

Our Response: We have now included a list of abbreviations at the end of the text.

Reviewer's Comment 7- You need English check for your document for example: a- page 3 line 11: and an estimated 1 million b- page 12 line 8: relatively simpler, requirement of c- page 17 line 13: IU/ml IU/mL d- page 20 line 19: developed to into an array-based. e- page 21 line 4: gen f- page 23 line 25: naïve

Our Response: We have now rectified these errors and have also revised the article thoroughly.

Reviewed by 00054317

Reviewer's Comment: This is a well written HBV molecular diagnostics overview that blends the perfect amount of referenced review with excellent description that is useful for the novice and 'jaded' expert alike!

Our Response: We are very much thankful to the reviewer for his/her nice words of appreciation. However, we wish to inform the reviewer that we had to remove some parts of the review article, as per the suggestions of the other three reviewers to make the article more focused.