OPEN PEER REVIEW

Open Peer Review (OPR)

As a new feature for progressing towards transparency, we decided to open a new window for all of our editors as well as authors titled "**Open Peer Review**". We hope by this new facility, our reviewers will be more motivated and authors will be more satisfied with the review process. We believe that publishing our peer review reports could make a transparent and clear environment for all our efforts within a journal, but not all reviewers tend to publish their comments.

What is "Open Peer Review" process?

An "Open Peer Review" process is making the details of all review process (including reviewers, associate editors, and EICs comments) as "Public" as it is agreed by EIC, Authors, and reviewers.

Advantages of "Open Peer Review" process

- More transparency, constructiveness and tactful comments of the peer review process: leads to an increase in the quality of reviews
- More motivations for all involved roles in the review process
- Authors' satisfactions from the review process: Increases honesty between authors and reviewers
- Education of both authors and new students
- Prevents reviewers from following their individual agendas and leading to the detection of reviewers' conflicts of interests

You can find out more at:
https://brief.land/briefland/knowledgebase/category/tree.html#opr.html

Jundishapur Journal of Microbiology
Article DOI: 10.5812/jjm.100038
Published in: Jundishapur Journal of Microbiology: 13(2); e100038

Peer Review Report for "The Prevalence of Hepatitis B and D Viruses and Evaluating YMDD Mutation in HBV-Suspected Patients in Qom Province, Iran"

Author(s): Ali Reza Azad, Mohsen Zargar, Mohammad Reza Zolfaghari, Abolfazl Mohammadbeigi

Review Timeline:

Submit Date: 8 Dec 2019
Revised Date: 17 Mar 2020
Accepted Date: 18 Mar 2020

Revision (0)

Here, you can see the **Reviewers**, **Associate Editors** and **EICs'** comments from the beginning to the end of the revision process.

REFEREE: Reviewer | **Revision** (0)

4 Jan 2020

Dear AE,

The manuscript entitled" The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran" contains valuable information about the prevalence of HBV, HDV, and drug resistance mutations, but due to some mistakes, the manuscript could be accepted after major revisions by the authors.

1. All parts of the manuscript need grammatical and punctuational corrections.
2. Keywords should be written on the basis of MeSH.
3. The introduction part is incomplete; the authors should notice the prevalence of HBV and HDV in Iranian populations.
4. What are the inclusion and exclusion criteria of this study?
5. Please mention the gene accession number of designed primers.
6. What is your Justification for the Same results in ELISA and PCR methods?
8. In paragraph two, is it the real prevalence of HBV in Tehran?

9. The discussion is too short, please rewrite it.
10. The conclusion isn't Comprehensive, please correct all the text.
11. Some references are very old, the authors must substitute them with new references.
Thanks.

REFEREE: Reviewer | **Revision** (0) 1 Feb 2020

Dear AE,
Comments for the authors:
1. Please write the title in AMA style
2. You wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.
- Introduction
3. In the introduction section you have written: There are eight known genotypes (A-H). Please use new papers and information about HBV genotypes. There are ten known genotypes (A-J)
4. You have written: Recently, 350 million individuals are chronically infected with HBV around the world (3, 5).
What do you mean by recently?
- Material and Methods
5. I think it is no need that you to write a calculation formula
6. Material and Methods part was not written as a scientific paper, therefore you write this part again.
7. The kits that you used for testing are not universally accepted and reliable
For example, DNA extraction kit (Sinaclon, Iran), and also ELISA kit (Pishtazteb, Iran).
8. On the other hand why do you use “a genomic DNA extraction kit (Sinaclon, Iran). You should use a viral DNA extraction kit, not a genomic DNA extraction kit.
9. You should not use the table for writing: the instruction for the first and second rounds for amplification of the HBs gene in Table 2. You should write it in the text of your manuscript.
10. You didn’t write anything about the method of statistical analysis, and also you didn’t write complete information about your used version of SPSS and Microsoft Excel software.
Results
11. You should write the mean age of the patients with SD and the range of the age. You don’t write the age of patients with median: (32-38 age median).
12. How many HBsAg-positive patients were HBV DNA positive?
Were all the HBsAg-positive patients were positive for HBV DNA, this is impossible. You didn’t write anything about that.
13. What method did you use to determine the type of mutation in the YMDD region of the HBV Virus?
14. You haven't written about the type of mutations that you found. This was your main goal.
To find mutations in this area you could use the RFLP assay or sequencing methods and easily detect the type of mutation.
15. Phylogenetic tree for HDV has a problem and you should draw it again.
16. Why didn't you register the sequences of the amplified gene of HDV at the Gene Bank

(NCBI)?
If you did, write their access number in your manuscript.
17. Please write the discussion part again. This part is a really important section and needs complete revision. This part is very messy and unclear. I think there are a lot of errors in this part. For instance: you have written that the average prevalence of hepatitis B antigen is 37.5% in Tehran province????????????
If this prevalence is for the general population or, ….??????
As I have written before, you wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.
Thanks.

REFEREE: Associate Editor | **Revision** (0) 1 Feb 2020

Dear EIC,
The manuscript entitled "The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran" could be published after major revision.
Thanks.

REFEREE: EIC | **Revision** (0) 10 Feb 2020

Dear Authors,
Thank you for submitting your manuscript to JJM. I have completed the review of your manuscript and a summary is appended below (Pleases find attached files). The reviewers recommend reconsideration of your paper following major revision. I invite you to resubmit your manuscript after addressing all reviewer comments. Please answer point by point to reviewer's comments on a separate sheet under WORD format and write your modifications in the revised version (WORD format) in colored or highlighted letters. Your answers to reviewers' queries are not only for the Editorial Board but for the readers and should be included, when appropriate, in the manuscript for improvement and a better understanding. When resubmitting your manuscript, please carefully consider all issues mentioned in the reviewers' comments, outline every change made point by point, and provide suitable rebuttals for any comments not addressed. In addition JJM INSTRUCTION attached to mail, Pleases find it.
Note:
Please add ORCID number into you and your collages account, then automatically appear in your published papers in JJM both in HTML and pdf. Pleases follow below link for the process. http://single.demo.pub/en/knowledgebase/category/tree.html#orcid.html
Thanks.

OPEN PEER REVIEW

Revision (1)

Reply to Reviewers

Ideally, the reviewing process can significantly improve the submitted manuscripts by allowing the authors to take into account the advice of reviewers. Author(s) must reply to all reviewers' comments in a separate Word file, point by point. A "**Reply to Reviewers**" document is submitted along with revised manuscript during submission of revised files, summarizing the changes that the authors made in response to the reviewers' comments. The responses to reviewers' comments specifies how the authors addressed each comment the reviewers made.

You can read the authors' responses to the reviewers' comments in the next page.

Thank you for your attention. Highlights sentences with blue are the answer to reviewers' comments. Also, edited sentences determined with blue highlight in the manuscript.

Reviewer 1:

The manuscript entitled" The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran" contains valuable information about the prevalence of HBV , HDV and drug resistance mutations, but due to some mistakes, the manuscript could be acceptable after major revisions.

1. All parts of manuscript needs to grammatical and punctuational corrections.
The manuscript edited by a good English writer.

2. Key words should be written on the basis of MeSH.
The Key words editted. (see line 25-26 in the manuscript).

3. The introduction part is incomplete; the authors should notice the prevalence of HBV and HDV in Iranian populations.
The prevalence of HBV and HDV in Iranian populations added in the introduction part and more details has explained in discussion. (see line 45, 61 &157-165).

4. What is the inclusion and exclusion criteria of this study?
In this cross-sectional study aimed to evaluate the prevalence of HBV and HDV infections, and to investigate the presence of YMDD motif in HBV-infected patients. The current study was performed for the first time in Qom province, Iran. Our results showed that 180 (11.25%) out of 1600 patients were HBsAg positive and 11 patients (6.1%) were found to be positive for dual infection (HBV/HDV). The prevalence was relatively high.

5. Please mention the gene accession number of designed primers.
The primers was design based on the *HBs* gene of HBV using NCBI site, GenRunner and CLC sequence viewer 6. We used the BLAST program to determine specificity of our primers. GenBank searches indicated complete identity (E value ≤1.1) of PCR primers only with the target gene sequences.

6. What is your Justification about the Same results in ELISA and PCR methods?
First, HbsAg-positive patients detected by ELISA method and in order to confim the existence of HBV DNA HbsAg-positive patients was applied nested PCR technique. There are not false positive sample.

8. In paragraph two, is it the real prevalence of HBV in Tehran?
An average prevalence of hepatitis B antigen was 37.5% in Tehran Province in 2016 according to 17 reference (Rahimyan K., et al.) (see line 243 in the manuscript).

9. The discussion is too short, please rewrite it.
We tried that the discussion improved.

10. The conclusion isn't Comprehensive, please correct all the text.
The conclusion corrected.

11. Some references are very old, the authors must substitute them with new references.
Some references substitute with new references.

Reviewer 2:

Manuscript Full Title: The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran

Dear colleague

Comments for authors:

1. You wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.
A total of 1600 blood specimens were collected from HBV-suspected patients who were referred to diagnostics center before and after lamivudine therapy. The number of HBsAg positive patients was 180 among HBV-suspected patients.

Introduction

2. In introduction section you have written: There are eight known genotypes (A-H) . Please use new papers and information about HBV genotypes. There are ten known genotypes (A-J).
It editted. (see line 36-38).

3. You have written: Recently, 350 million individuals are chronically infected with HBV around the world (3, 5). What do you mean by recently?
According to recent statistics, 350 million individuals have been chronically infected with HBV worldwide.

Material and Methods

4. I think it is no need that you write calculation formula.

calculation formula deleted.

5. Material and Methods part was not written as a scientific paper, therefor you write this part again.
It editted.

6. The kits that you used for testing are not universally accepted and reliable For example, DNA extraction kit (Sinaclon, Iran), and also ELISA kit (Pishtazteb, Iran).
The positive control DNA extracted using Vivantis kit. DNA quantify and assess purity of samples measured by he NanoDrop Spectrophotometer.
we are proud to have achieved many accomplishments such as being given the title of The exemplary exporter for several years and being declared as a Knowledge-Based Company by the Organization of Vice-Presidency for Science and Technology. In addition, our innovative thinking has enabled us to achieve quality management certificates of ISO 13485: 2016, FDA QRS 21 CFR 820 (cGMP), ISO 10002:2018 and CE mark accreditation for most of our products.

7. On the other hand why you used "a genomic DNA extraction kit (Sinaclon, Iran). You should use viral DNA extraction kit, not a genomic DNA extraction kit.
Our aim was total DNA extraction.

8. You should not use table for writing: the instruction the first and second rounds for amplification of HBs gene in Table 2. You should write it in the text of your manuscript.
The instruction the first and second rounds for amplification of *HBs* gene and *HDAg* gene was written in Table 2 due to prevention of repetition sentences.

9. You didn't write anything about method of statistical analysis, and also you didn't write complete information about your used version of SPSS and Microsoft Excel software.
SPSS version 22.0 and Microsoft Excel version 2016 were used for data analysis. The level of significance in t-test was set at $P < 0.05$.

Results

10. You should write the mean age of the patients with SD and range of the age. You don't write the age of patients with median: (32-38 age median).
It editted.

11. How many of HBsAg positive patients were HBV DNA positive? Were all the HBsAg positive patients were positive for HBV DNA, this is impossible. You didn't write anything about that.

First, HbsAg-positive patients detected by ELISA method and then, in order to confim the existence of HBV DNA HbsAg-positive patients was applied nested PCR technique. There are not false positive sample.

12. What method did you use to determine the type of mutation in the YMDD region of the HBV Virus?
All specimens were tested by PCR method to detect YMDD motif. The type of mutation in the YMDD region of the HBV Virus was not determineded.

13. You haven't written about the type of mutations that you found. This was you main goal. To find mutations in this area you could use the RFLP assay or sequencing methods and easily detect the type of mutation.
The type of mutations was not main goal. The present of mutation was our aim.

14. Phylogenetic tree for HDV has problem and you should draw it again.
Phylogenetic tree editted. (figure 4).

15. Why didn't you register the sequences of the amplified gene of HDV at the Gene Bank (NCBI)?If you did, write their access number in your manuscript.
The sequences of the amplified gene of HDV at the Gene Bank will register.

16. Please write the discussion part again. This part is really important section and needs complete revision. This part is very messy and unclear. I think there are a lot of errors in this part. For instance: you have written that an average prevalence of hepatitis B antigen is 37.5% in Tehran province???????????? If this prevalence is for general population or, …..??????

As I have written before, you wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.
We tried that the discussion improved.
An average prevalence of hepatitis B antigen was 37.5% in Tehran Province in 2016 according to 17 reference (Rahimyan K., et al.) (see line 243 in the manuscript).
HBV-suspected patients (age range: 20-67 years), who were referred to the diagnostic centers before and after lamivudine therapy of Qom Province. This Province is a major destination for travelers and immigrants, most of whom come from different provinces of Iran or African and Asian countries, which are endemic to many diseases and have poor healthcare systems.

OPEN PEER REVIEW

Revision (1)

Here, you can see the **Reviewers**, **Associate Editors** and **EICs'** comments from the beginning to the end of the revision process.

REFEREE: EIC | **Revision (1)** 16 Mar 2020

Dear Authors,
Please specify the location of each answer in your text. Please indicate the number of the line.
Thanks.

OPEN PEER REVIEW

Revision (2)

Reply to Reviewers

Ideally, the reviewing process can significantly improve the submitted manuscripts by allowing the authors to take into account the advice of reviewers. Author(s) must reply to all reviewers' comments in a separate Word file, point by point. A "**Reply to Reviewers**" document is submitted along with revised manuscript during submission of revised files, summarizing the changes that the authors made in response to the reviewers' comments. The responses to reviewers' comments specifies how the authors addressed each comment the reviewers made.

You can read the authors' responses to the reviewers' comments in the next page.

Thank you for your attention. Highlights sentences with blue are the answer to reviewers' comments. Also, edited sentences determined with blue highlight in the manuscript.

Reviewer 1:

The manuscript entitled" The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran" contains valuable information about the prevalence of HBV , HDV and drug resistance mutations, but due to some mistakes, the manuscript could be acceptable after major revisions.

1. All parts of manuscript needs to grammatical and punctuational corrections.
All parts of manuscript edited by a good English writer.

2. Key words should be written on the basis of MeSH.
The Key words editted. (see line 25-26 in the manuscript).

3. The introduction part is incomplete; the authors should notice the prevalence of HBV and HDV in Iranian populations.
The prevalence of HBV and HDV in Iranian populations added in the introduction part and more details has explained in discussion. (see line 45, 61 &157-167 in the manuscript).

4. What is the inclusion and exclusion criteria of this study?
In this cross-sectional study aimed to evaluate the prevalence of HBV and HDV infections, and to investigate the presence of YMDD motif in HBV-infected patients. The current study was performed for the first time in Qom province, Iran. Our results showed that 180 (11.25%) out of 1600 patients were HBsAg positive and 11 patients (6.1%) were found to be positive for dual infection (HBV/HDV). The prevalence was relatively high.

5. Please mention the gene accession number of designed primers.
The primers was design based on the *HBs* gene of HBV using NCBI site, GenRunner and CLC sequence viewer 6. We used the BLAST program to determine specificity of our primers. GenBank searches indicated complete identity (E value ≤1.1) of PCR primers only with the target gene sequences. (see line 97 in the manuscript).

6. What is your Justification about the Same results in ELISA and PCR methods?
First, HbsAg-positive patients detected by ELISA method and in order to confim the existence of HBV DNA HbsAg-positive patients was applied nested PCR technique. There are not false positive sample.

8. In paragraph two, is it the real prevalence of HBV in Tehran?

An average prevalence of hepatitis B antigen was 37.5% in Tehran Province in 2016 according to 17 reference (Rahimyan K., et al.). (see line 244 in the manuscript).

9. The discussion is too short, please rewrite it.
We tried that the discussion improved.

10. The conclusion isn't Comprehensive, please correct all the text.
The conclusion corrected. (see line 189-193 in the manuscript).

11. Some references are very old, the authors must substitute them with new references.
Some references substitute with new references. 4, 7, 10, 15, 20 references added.

Reviewer 2:

Manuscript Full Title: The prevalence of hepatitis B and D viruses and evaluating YMDD mutation in HBV-suspected patients in Qom province, Iran

Dear colleague

Comments for authors:

1. You wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.
A total of 1600 blood specimens were collected from HBV-suspected patients who were referred to diagnostic centers before and after lamivudine therapy. The number of HbsAg-positive patients was 180 among HBV-suspected patients based on ELISA and nested PCR techniques. (see line 123-129 in the manuscript).

Introduction

2. In introduction section you have written: There are eight known genotypes (A-H) . Please use new papers and information about HBV genotypes. There are ten known genotypes (A-J).
It editted. (see line 36-38 in the manuscript).

3. You have written: Recently, 350 million individuals are chronically infected with HBV around the world (3, 5). What do you mean by recently?
According to recent statistics, 350 million individuals have been chronically infected with HBV worldwide. (see line 44-45 in the manuscript).

Material and Methods

4. I think it is no need that you write calculation formula.
calculation formula deleted.

5. Material and Methods part was not written as a scientific paper, therefor you write this part again.
Material and Methods part editted.

6. The kits that you used for testing are not universally accepted and reliable For example, DNA extraction kit (Sinaclon, Iran), and also ELISA kit (Pishtazteb, Iran).
The positive control DNA extracted using Vivantis kit. DNA quantify and assess purity of samples measured by he NanoDrop Spectrophotometer.
we are proud to have achieved many accomplishments such as being given the title of The exemplary exporter for several years and being declared as a Knowledge-Based Company by the Organization of Vice-Presidency for Science and Technology. In addition, our innovative thinking has enabled us to achieve quality management certificates of ISO 13485: 2016, FDA QRS 21 CFR 820 (cGMP), ISO 10002:2018 and CE mark accreditation for most of our products.

7. On the other hand why you used "a genomic DNA extraction kit (Sinaclon, Iran). You should use viral DNA extraction kit, not a genomic DNA extraction kit.
Our aim was total DNA extraction.

8. You should not use table for writing: the instruction the first and second rounds for amplification of HBs gene in Table 2. You should write it in the text of your manuscript.
The instruction the first and second rounds for amplification of *HBs* gene and *HDAg* gene was written in Table 2 due to prevention of repetition sentences. (see line 114 in the manuscript).

9. You didn't write anything about method of statistical analysis, and also you didn't write complete information about your used version of SPSS and Microsoft Excel software.
SPSS version 22.0 and Microsoft Excel version 2016 were used for data analysis. The level of significance in t-test was set at $P < 0.05$. (see line 120-121 in the manuscript).

Results

10. You should write the mean age of the patients with SD and range of the age. You don't write the age of patients with median: (32-38 age median).
It editted. (see line 124, 128 in the manuscript).

11. How many of HBsAg positive patients were HBV DNA positive?

Were all the HBsAg positive patients were positive for HBV DNA, this is impossible. You didn't write anything about that.

First, HbsAg-positive patients detected by ELISA method and then, in order to confim the existence of HBV DNA HbsAg-positive patients was applied nested PCR technique. There are not false positive sample. (see line 90, 93 in the manuscript).

12. What method did you use to determine the type of mutation in the YMDD region of the HBV Virus?

All specimens were tested by PCR method to detect YMDD motif. The type of mutation in the YMDD region of the HBV Virus was not determineded.

13. You haven't written about the type of mutations that you found. This was you main goal. To find mutations in this area you could use the RFLP assay or sequencing methods and easily detect the type of mutation.

The type of mutations was not main goal. The present of mutation was our aim. (see line 133-135 in the manuscript).

14. Phylogenetic tree for HDV has problem and you should draw it again.

Phylogenetic tree (Figure 4) editted. (see line 148 in the manuscript).

15. Why didn't you register the sequences of the amplified gene of HDV at the Gene Bank (NCBI)?If you did, write their access number in your manuscript.

The sequences of the amplified gene of HDV at the Gene Bank will register.

16. Please write the discussion part again. This part is really important section and needs complete revision. This part is very messy and unclear. I think there are a lot of errors in this part. For instance: you have written that an average prevalence of hepatitis B antigen is 37.5% in Tehran province???????????? If this prevalence is for general population or, ….??????

As I have written before, you wrote that the research was conducted on people suspected of having HBV infection. Please explain who you are in your study population because your results have a very high prevalence compared to people in our general population.

We tried that the discussion improved.

An average prevalence of hepatitis B antigen was 37.5% in Tehran Province in 2016 according to 17 reference (Rahimyan K., et al.) (see line 244 in the manuscript).

HBV-suspected patients (age range: 20-67 years), who were referred to the diagnostic centers before and after lamivudine therapy of Qom Province. This Province is a major destination

for travelers and immigrants, most of whom come from different provinces of Iran or African and Asian countries, which are endemic to many diseases and have poor healthcare systems.

OPEN PEER REVIEW

Revision (2)

Here, you can see the **Reviewers**, **Associate Editors** and **EICs'** comments from the beginning to the end of the revision process.

REFEREE: EIC | **Revision** (2) 18 Mar 2020

Dear authors
According to the associated editor report, your manuscript is accepted for publishing in JJM and send to the publisher for the next process.
Note:
Please add the ORCID number to you and your collages account, then automatically appear in your published papers in JJM both in HTML and pdf. Please follow the below link for the process. http://single.demo.pub/en/knowledgebase/category/tree.html#orcid.html
Thanks.