04-february-2025

Dear prof. Rowaiye,

You submitted manuscript AJGH-2412-1072, " Gene expression and histopathological analysis of the effect of Allium cepa on carbon tetrachloride (CCl4)-induced Anti-Hepatic steatosis in Wister rats," to the African Journal of Gastroenterology and Hepatology. The reviewer's comments (s) are included at the end of this email.

The reviewers have requested revisions to your manuscript.  Therefore, I invite you to respond to the reviewers' comments and revise your manuscript. Instructions on how to do this can be found at the bottom of this email.

Because we are trying to facilitate the timely publication of manuscripts submitted to the African Journal of Gastroenterology and Hepatology, your revised manuscript should be submitted as soon as possible.

Once again, thank you for submitting your manuscript to the African Journal of Gastroenterology and Hepatology. I look forward to your revision.

Sincerely,

Salem Y Mohamed

Chief Editor, African Journal of Gastroenterology and Hepatology

Please revise your manuscript using a word processing program and save it on your computer.  Please also highlight the changes to your manuscript within the document using the track changes mode in MS Word or using bold or colored text.

Once the revised manuscript is prepared, you can send it to me via email.

When submitting your revised manuscript, please provide your responses and details of changes made to each comment in an Author Response Letter.  To expedite the processing of the revised manuscript, please be as specific as possible in your response to the reviewers.

**Reviewer Comments to Author:**

**Reviewer1: (HR): pathology**

Excellent work.

**Reviewer 2: (SH): biochemistry and genetics**

Thanks for allowing me to revise the paper entitled

“Gene expression and histopathological analysis of the effect of Allium cepa on carbon tetrachloride (CCl4)-induced Anti-Hepatic steatosis in Wister rats”

This paper investigates the possible ameliorative effect of Allium cepa on CCl4-induced liver disease in rats by examining the molecular profile and histopathological picture.

The idea is good. However, many points should be addressed to be suitable for publication:

General points:

-The manuscript needs extensive language editing: many extra spaces, multiple font, and size of words, many grammar errors, too long sentences (e.g. the background in the abstract is a single sentence with many errors)

- References need updating (the most recent is 2020, which is 5 years ago)

-Abbreviations should be only used after the full name for the first time. After that, don’t repeat the full name. For example, the full name of AFP, TNF, and HMG-COA reductase appeared for the first time in the discussion.

- P<0.5 should be replaced by the actual value.

- Page numbers were missed.

- Unify the abbreviations all over the manuscript (e.g. KI67, HMG-COA)

Title:

I think it’s not suitable. I suggest using” Gene expression and histopathological analysis of the effect of Allium cepa on carbon tetrachloride (CCl4)-induced Anti-Hepatic steatosis in Wister rats

Introduction

- the authors stated that “NAFLD has now been recognized as a leading cause of chronic liver disease worldwide, although there is still inadequate knowledge regarding the disease (Maurice and Manousou, 2018)’. How come using it now for a reference 7 years ago?!

- the authors stated that “Recent epidemiology provided evidence showing that NAFLD has become the second largest liver disease after viral hepatitis, with an incidence of 20–30%, and the incidence of obesity in the population is up to 57.74% (Zhong et al,2020) ”. the authors didn’t mention the locality (e.g. Worldwide, Western, Eastern,…) and which population the meant. Also, by revising the reference, Zhong et al.,2020 was a review of experimental models of NAFLD and not an epidemiological study. Please refer to the original study.

- the authors mentioned, “This study aimed to assess the protective effects of Allium cepa, measure relevant biochemical markers,….” However, no biochemical markers were studied in the manuscript. This allows us to ask why we didn’t perform liver enzymes.

Methodology

- Preparation of onion extract and raw onion needs a suitable reference.

The preparation of CCL4 (solvent, e.g., olive oil) and dosage need further details and a suitable reference. CCL4 can be used to induce multiple liver diseases and even lung diseases. I think this dose can lead to more severe liver disease.

Were the extracts used prophylactic or therapeutic? According to the literature, the model takes 8 weeks, while you administer the treatment after 5 weeks only.

-It was difficult for me to understand the treatment in groups 6 and 7. What was meant by ratios and ratios of what? I need further explanation.

- The Declaration of Helsinki is used for human research, not animal research. Please correct it.

- “The experimental protocols were reviewed and approved by the Adekunle Ajasin University Animal Ethics Committee”. This sentence is repeated twice.

- The serial number and ethical committee approval date are missing. Please provide it

-In Table 1 of primers, please mention the accession number of genes or suitable references. Also, the name of primers in rats is written in small letters except for the first letter capitalized

- Further explanation of molecular methodology (steps, used kits, and instruments) and result calculation (fold expression delta ct or delta ct) are required.

Results

I think the results and discussion were merged together. This happens in research communication, not the original paper. I don’t know if the journal accepts such a type. If not, try to represent your results better in the results section.

-Add scale bar to figures.

- For comparison figures, it’s better to represent figures of all groups at the same magnification (e.g., group 2 needs x100). I think some magnification errors are present.

- Regarding gel image, in figure 1 lane 2 (represent onion control) and lane 5(represent 200mg onion extract) shows lower density band while graph shows similar expression to normal control.

-How gene results were calculated. if delta delta ct was used the control expression must equal 1

List of Figures

- Accompanied by numbers at the end of each sentence (refer to what?)

List of tables

The title of Table 1 is unsuitable; it refers to a primer sequence of the studied genes, not gene expression analysis.

- Please provide a table comparing gene results in the studied groups with significance test and P value.

**Reviewer 3: biostatistician(AM)**

Please provide point-by-point responses to comments in the attached files.

**Reviewer4:**

**good work**

**Editor Comments to Author:**

1. Please check the author names and affiliations included on your Title Page, mainly that the spelling of all authors' names is correct. They are cited in the order you wish them to appear in the final article. In addition, each author's affiliation details are correct.

2. Please include a 'Structured Abstract': not more than 250 words, broken down into, i.e., Aims, Patients & Methods/Materials & Methods, Results, and Conclusions. For authors presenting the results of clinical trials, the guidelines recommended by CONSORT should be followed when writing the abstract (http://www.consort-statement.org/), and the clinical trial registration number should be included at the end of the abstract, where available.

3. Please include up to 10 keywords in your revised manuscript (including the four keywords you selected as part of the submission process).

4. Please amend the references as per the author's guidelines:

a. References should be numerically listed in the reference section in the order they occur in the text.

b. References should appear as a number, i.e., [1, 2] in the text.

c. References should cite three authors et al.: It is our house style to list a maximum of six authors and, if there are more than this, three authors et al.

5. Please ensure all tables and boxes are titled and cited in the text. Three-line tables are preferred.

Please find a link to the African Journal of Gastroenterology and Hepatology Author Guidelines, which explains these sections in more detail: <https://ajgh.journals.ekb.eg/journal/authors.note>.

6. Please check the PDF file of your manuscript regarding plagiarism checking.

7. Please add the scale bar, annotations, magnifications, and program that generated these figures. Also, it is better to submit figures with high resolution and brightness.