June 13, 2016

Shui Qiu,
Science Editor
Editorial Office
World Journal of Hepatology

Dear Shui Qiu,

Herein you will find the Revised Manuscript No. 25637 entitled: ***CD36 genetic variation, fat intake and liver fibrosis in chronic HCV infection*** by authors Omar Ramos-Lopez, Sonia Roman, Erika Martinez-Lopez, Nora A. Fierro, Karina Gonzalez-Aldaco, Alexis Jose-Abrego and Arturo Panduro.

We have addressed point-by-point the observations made by the reviewers, which are underlined in the revised version of the manuscript. We hope that this manuscript is now appropriate for publication.

We guarantee that there is no conflict of interest in our paper and we have not submitted this paper elsewhere.

Best regards,

Arturo Panduro MD. Ph.D. FAASLD.
Department of Molecular Biology in Medicine
Civil Hospital of Guadalajara, "Fray Antonio Alcalde"
Hospital 278, Col. El Retiro
Guadalajara, Jalisco 44280 México
Phone/Fax: +52(33)36147743
E-mail: apanduro@prodigy.net.mx

June 13, 2015

Dear Editor,

Please find enclosed the edited manuscript in Word format (25637-Revised_manuscript.doc).

Title: CD36 genetic variation, fat intake and liver fibrosis in chronic HCV infection

Authors: Omar Ramos-Lopez, Sonia Roman, Erika Martinez-Lopez, Nora A. Fierro, Karina Gonzalez-Aldaco, Alexis Jose-Abrego and Arturo Panduro

Name of Journal: ***World Journal of Hepatology***

ESPS Manuscript NO: 25637

The manuscript has been improved overall according to the suggestions of reviewers.

I. Format has been updated

II. Documents uploaded:

25637-Revised_manuscript
25637-Answering reviewers
25637-Copyright assignment
25637-Audio core tip
25637-Institutional review board statement
25637-Informed consent statement
25637- Biostatistics statement
25637-Conflict-of-interest statement
25889-Data sharing statement
25637-Google Scholar
25637-CrossCheck
25637-Grant application form
25637-Language certificate

III. Revision has been made according to the suggestions of the reviewer and underlined in the text.

Reviewer No. 02860848

In the current manuscript, the authors Ramos-Lopez et al. suggest that CD36 genetic variation (therefore presumably CD36 expression is different) correlates with amount of fat intake and liver fibrogenesis. Although much more data are definitely needed for convincingly showing that elevated amount of fat intake, due to CD36 genetic variation, enhances liver fibrogenesis in patients with chronic HCV, the current study is potentially interesting.

MAJOR CONCERN:

1. Method for dietary assessment is absolutely not clear. In a previous publication form authors (Ramos-Lopez et al., Nutrition and Food Sciences 2015) dietary assessment method is not visibly mentioned. Since the data about fat intake is a key point in the presented manuscript, the authors should provide more detailed information (e.g. as a supplementary information).

Answer: A three-day food record was used as a tool to assess the patient's dietary intake, which provides accurate data concerning intake of food and nutrients [32]. Each subject was instructed on how to register the type, amount and mode of preparation of all foods consumed during two weekdays and one weekend day. Handling of the dietary data was performed by using food scales and models to increase the accuracy of the portion size. The food records were reviewed and coded by a trained registered dietitian using the Nutrikcal computer program (Nutrikcal VO®, Mexico), which is based on the Mexican System of Food and Equivalents [33]. This program calculated the total of calories, fat, protein and carbohydrates as well as the daily intake of food group servings such as sugars, meat, fruits, vegetables, fats, milk, legumes and cereals. Dietary data were averaged over the three-day food records and were compared with the recommended dietary intakes of the Mexican population [34].
This was added on Page 7.

2. Which guideline do authors follow?

Answer: Dietary data were compared with the recommended dietary intakes of the Mexican population, which were added to Tables 2 and 3.

3. There is a huge difference on fat intake between AA and GG genotypes, but CH and protein intake (%) is nearly identical.

Answer: We evaluated the consumption of several food groups by *CD36* genotype such as sugars, meat, fruits, vegetables, fats, milk, legumes and cereals. However, fats were the only food group associated with the *CD36* genotype (Table 3 and Page 10)

4. What do patients with GG genotype eat less?

Answer: The GG genotype carriers consumed less fat servings than the AA genotype carriers (Table 3).

5. Authors suggest that increased fat intake in patients with AA genotype show no significant difference concerning glucose, TC, TG, HDL, LDL, etc. but only AST is significantly higher in AA. It is hard to understand how elevated fat intake for long time (presented data are from 3-day analysis but authors consider this is representative

of whole time food intake behavior) does not have any metabolic parameters or obesity and only AST?

Answer: These findings may be explained because chronic HCV infection is characterized by hypocholesterolemia and low levels of LDL-c, TG and apoB [54]. This statement was added on Page 12.

6.Do authors have biopsy materials? Steatosis? Inflammation?

Answer: In this study, the *CD36* AA genotype was associated with advanced liver fibrosis, which is a stage of the disease characterized by the presence of liver steatosis and persistent inflammation [4]. This was added on Page 12.

MINOR CONCERN:

1. How did the authors identify exact duration of infection for each patient?

Answer: Duration of infection (years) was estimated by the self-reported date of exposure to any known risk factor for HCV infection including history of surgeries, blood transfusions, hemodialysis, acupuncture, injection-drug use and tattooing [30]. This statement was added on Page 6.

2. Do patients have HCC?

Answer: No. We have documented that HCC is rare in Mexico due to genetic and environmental risk factors. Additionally, patients with liver cirrhosis are young and death occurs earlier than HCC development. (Roman et al., Hepatitis B Virus Genotype H and Environmental Factors Associated to the Low Prevalence of Hepatocellular Carcinoma in Mexico. Journal of Cancer Therapy Vol.4 No.2A, Pub. Date: February 28, 2013. DOI: 10.4236/jct.2013.42A044.

3. Data for ALT and AST are similar, but there is huge difference concerning significance.

Answer: This particular difference may be related to the greater predictive value of AST for the progression of liver fibrosis than other liver enzymes, such as ALT or GGT [49]. This statement was added on Page 12.

Reviewer No. 3536815

I read with great interest the manuscript titled "**CD36 genetic variation, fat intake and liver fibrosis in chronic HCV infection" By Omar Ramos-Lopez *et al.*** It explores the extremely interesting relationship between liver fibrosis, evaluated through non-invasive

ultrasound based technique, and genetics. Authors aimed to determine the association of the *CD36* (rs1761667) polymorphism with dietary intake and liver fibrosis in patients of West Mexico with chronic hepatitis C (CHC). Patients with the *CD36* AA genotype seemed to have a higher intake of calories, total fat and saturated fatty acids (SFA) than those with the AG and GG genotypes. Moreover patients with advanced liver fibrosis had a higher frequency of the *CD36* AA genotype than those with mild liver fibrosis (42.1% *vs.* 17.1%, p=0.002), respectively. And authors concluded that likewise, the AA genotype is associated with advanced liver fibrosis. The paper is fluent and well witten, the results clearly explained and the conclusions convincing.

I have only minor concerns about it:

1. Please add a brief mention of the limitation of the use of three-day food record in the dietary assessment.

Answer: In regards to food consumption, despite that the three-day food record may not be representative of the long-term food variety, these individuals had a fat intake that was >30% of the total energy consumed. The long-term consumption of a high-fat diet is a recognized risk factor for the development of metabolic disorders [46]. This point is discussed on Page 11.

2. According to journal guidelines the conclusion must include the contributions of the conclusions to the field and the weaknesses of the study, and provide future research directions.

Answer: In conclusion, our data suggest that the AA genotype of the rs1761667 *CD36* polymorphism is associated with higher fat intake and more instances of advanced liver fibrosis in CHC patients in West Mexico. To our knowledge, this is the first study that analyzes the role of *CD36* genetic variation on HCV-related liver disease in Mexican patients. However, further large-scale studies that involve distinct ethnic groups are needed to determine how *CD36* genetics may influence the onset and progression of liver fibrosis in chronic HCV infection. This was added on Page 14.

IV. Language polishing.

We attached the certificate of English language

V. References and typesetting were corrected

We have revised references and typesetting.

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

Best regards,

Arturo Panduro, M.D., Ph.D., FAASLD
Department of Molecular Biology in Medicine,
Civil Hospital of Guadalajara, "Fray Antonio Alcalde"
Guadalajara, Jalisco, 44280, Mexico
apanduro@prodigy.net.mx