Response to Reviewers' Comments

March 4, 2016

Prof. Damian Garcia-Olmo
Prof. Stephen C Strom
Prof. Andrzej S Tarnawski

Editors-In-Chief
World Journal of Gastroenterology

Dear Sir,

On behalf of my co-authors, I am submitting the revised version of the manuscript authored by Behairy *et al*. "24105". All the authors have revised the manuscript and approved its contents. Response to reviewers' comments was addressed carefully point-by-point in the following pages and changes in the manuscript are highlighted yellow.

Regarding **CrossCheck analysis**, we tried to used the website, but we got the feedback of associate memebership manager that we can not use the service as it is not available to individuals, but available for publishers only.

Please find enclosed the edited manuscript in Word format (file name: 24105-revision.docx).

Title: Transient Elastography Compared to Liver Biopsy and Morphometry for Predicting Fibrosis in Pediatric Chronic Liver Disease: Does Etiology Matter?

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Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 24105

The manuscript has been improved according to the suggestions of reviewers:
1 Format has been updated

2 The manuscript has been revised for proper English language by a specialized office for scientific writing services.

3 Revision has been made according to the suggestions of the reviewer point-by-point

Reviewer # 00742209

Comments to Authors

General:

Comment 1:
Please review for clarifications and typographical errors (table 1).

Response 1:
Revised and corrected

Comment 2:
The major limitation is small sample sizes in study groups by etiologies. The authors are encouraged to focus on the data analysis for the entire study population and not on the comparison among etiologies for this study which used a cross-sectional design (see Tables 3 and 4).

Response 2:
We agree with the reviewers that the small number in individual groups is a limitation in the study. For that, we removed the significance analysis (P-value) from table 3 and 4 and presented that data as descriptive.

In discussion, last three paragraphs in page 14 and the first paragraph in page 15 concerning the analysis of individual etiological groups (In the HCV group ….; In the AIH group……; In the Wilson disease group…….) were removed to amend the discussion and focus only on the results of the entire population.

Abstract

Comment 1:

Were the correlations (r) for FAF and Ishak statistically compared to support the statement that Ishak was better than FAF? If a statistical comparison was not performed, state "appeared better." Provide units for LSM

Response 1:

No statistical comparison was made, so we changed the statement to "appeared better". The measure units for LSM was provided as instructed.

Comment 2:

State "appears reliable" in the conclusion because this was a cross-sectional study design. Would like to confirm these findings using a longitudinal cohort with a large sample size of patients.

Response 2:

The statement was changed to "appears reliable" in both the abstract (page 5) and discussion (page 16).

Methods

Comment 1:

Clarify the method used to estimate kPa using the ultrasound How were the cutoffs in the ROC determined?

Response 1:

Liver stiffness is measured through a device that is called FibroScan which is composed of an ultrasound transducer probe mounted on the axis of a vibrator. Vibrations of mild amplitude and low frequency are transmitted by the transducer, inducing an elastic shear wave that propagates through the underlying tissues. Pulse echo ultrasound acquisition is used to follow the propagation of the shear wave and to measure its velocity, which is directly related to tissue stiffness: the stiffer the tissue, the faster the shear wave propagates. [1]

Note: The information was added to methodology section, page 9, subtitle "Transient Elastography" lines 3-10

The cut-off values for optimal clinical performance (optimal sensitivity and specificity simultaneously) were determined from the ROC curves (The upper most left point).

Note: The information was added to Methodology section, page 10, subtitle "statistical analysis" lines 2-4 from below.

Results

Comment 1:

Review rationale to compare LSM among etiologies of liver disease by stages of fibrous because 28.5% of the cells were incomplete in the dataset (paras 2 and 3, Table 4)

Response 1:

We agree with the reviewer, so we removed the significance analysis (*P*-value) and we presented the data as descriptive only in table 4.

Comment 2:

Table 1. Clarify n(%) for activity grade and fibrosis category

Response 2:

n(%) was added in the table next to both activity grade and fibrosis category and with other ualitative variables.

Comment 3:

Table 3. See heading in table 2 for additional information that defines diagnostic accuracy

Response 3:

In hand with the reviewer recommendation to focus on the data of the entire population and not the individual groups, we found it is not acceptable to calculate the sensitivity, specificity, PPV and NPV for each cut-off value in the different etiological group because of the small number. So, we modified the title of table 3 to be "Area under receiver operating characteristic curve for liver stiffness in predicting individual Ishak fibrosis stages in the different etiological groups".

Comment 4:

Table 4. Provide units for values in the table

Response 4:

Provided as indicated

Comment 5:

Table 5. What does the 95CI represent in the multiple regression? Provide the r2 for the multiple regression model. The "NI" entry under the beta coefficient is confusing in the table.

Response 5:

Sorry for the mistake, the 95% CI is for the unstanderdized coefficient, which is not presented in the results, so we removed it. To avoid any confusion with the "NI" entry in the table, we removed the data of regression analysis from the table and integrated it in the text of "Results" section, page 12, paragraph 1, line 6 together with the R^2 value.

Discussion

Comment 1:

Review for consistency the statements about the ability to discriminate early stages of fibrosis (F0) for all patients (see para 2 in discussion and para 2 in results)

Response 1:

The statement in the "Results" section is consistent, so we modified the statement in the "Discussion" section to match that in the results. "Our results demonstrated that LSM could significantly discriminate individual stages of fibrosis even the earlier stages (≥F1) from absent fibrosis (F0).", page 12, paragraph 4, lines 1-2.

Comment 2:

Clarify the contribution of the site of fibrosis (portal vs. central vein) to the apparent difference in correlation between LSM and indicators used to assess for fibrosis (para 7, discussion)

Response 2:

Sandrini et al [2] investigated what type of fibrosis influences LSM. They found that the area of portal–bridging fibrosis better correlated with the liver stiffness than did the area of whole fibrosis or the area of perisinusoidal fibrosis. In the early fibrosis stages, there was a significant increase of perisinusoidal fibrosis from F0 to F2 Metavir, more than that of portal–bridging fibrosis. In subsequent stages F3 and F4, the area of perisinusoidal fibrosis stabilized.[2] Ishak scoring system is based on the pattern and extension of portal fibrosis, while FAF evaluates the whole amount of collagen whether portal or perisinusoidal. This may explain the better correlation of LSM with Ishak scores in our study.

Note: The information was added to discussion section, page 14, paragraph 3.

Comment 3:

Consider performing a statistical comparison of the correlations between LSM and the PELD / MELD scores and not rely on the p values to determine strength of correlation (para 10, discussion)

Response 3:

We compared the two correlations as instructed using r to z test. There was no statistical significant difference (*P* value = 0.342), so we changed the statement to "better correlation"

Comment 4:

Clarify "the performance of LSM has been under estimated" in para 16 (discussion)

Response 4:

Staging of fibrosis with biopsy will always carry a risk, albeit low, of misclassification thus making the term ''best" standard more appropriate than ''gold" standard for liver biopsy.[3] As liver biopsy with its limitations[4] is used as a reference, a perfect surrogate will never reach maximal value.[3]

Note: the information was added to discussion section, page 15, last paragraph, and page 16, first paragraph.

Comment 5:

State "appears reliable" in para 18 (discussion)

Response 5:

Corrected as instructed in discussion section, last paragraph, line 1

Reviewer # 03498496

Comments to Authors

Summary of the paper: Behairy et al. reported correlation between histological fibrosis and liver stiffness assessed by transient elastography (TE) in pediatric patients with different chronic liver diseases. The authors demonstrated that Ishak fibrosis stage was the only independent variable associated with liver stiffness measurement assessed by TE. They concluded that TE may be useful for distinguishing different stages of liver fibrosis in pediatric patients with chronic liver diseases. Overall impression: The authors well demonstrated usefulness of TE for assessing liver fibrosis in pediatric patients with different chronic liver diseases. The readers of World J Gastroenterol may be interested in these findings. I have only some specific points:

Specific points:

Comment 1:

Table 5. The main purpose of this study was to examine whether TE can be used to predict histological liver fibrosis (Ishak score). Therefore, in this Table, the authors should assess whether clinical variables, including liver stiffness measurement assessed by TE can predict Ishak staging. That is, Ishak score should be defined as a dependent variable and liver stiffness measurement by TE should be included in clinical parameters (variables).

Response 1:

We clearly understand the reviewer′s opinion; meanwhile we would like to explain the aim and the hypothesis tested by the regression analysis in table 5. We aimed to test the influence of the different parameters as an independent variable on the values of LSM as a dependent variable, so we designed the analysis in such way.

Note: the regression analysis result was removed from table 5 and integrated in the text, "Results" section, page 12, paragraph 2, line 6 together with the R^2 value presented.

Comment 2:

On a related point, Y-axis should indicate Ishak score or fibrosis area fraction, and X-axis should indicate liver stiffness in Figures 1. Also, in Figure 4, Y-axis should indicate Ishak score, and X-axis should indicate PELD and MELD scores.

Response 3:

The hypothesis tested in figure 1 is how the change in Ishak score and and FAF as independent factors (represented on X axis) will affect the values of LSM as an independent variable (presented on Y axis). The same hypothesis apply to figure 4. Ishak score and FAF are the independent variables and the PELD/MELD is the dependent one.

Comment 3:

Statistical analysis: Results should be considered significant if P-value is <0.05.

Response 3:

Corrected as instructed

References

1 Ji D, Shao Q, Han P, Li F, Li B, Zang H, Niu X, Li Z, Xin S, Chen G. The frequency and determinants of liver stiffness measurement failure: a retrospective study of "real-life" 38,464 examinations. *PLoS One* 2014; **9**(8): e105183 [PMID: 25122123 DOI: 10.1371/journal.pone.0105183]

2 Sandrini J, Boursier J, Chaigneau J, Sturm N, Zarski JP, Le Bail B, de Ledinghen V, Cales P, Rousselet MC. Quantification of portal-bridging fibrosis area more accurately reflects fibrosis stage and liver stiffness than whole fibrosis or perisinusoidal fibrosis areas in chronic hepatitis C. *Mod Pathol* 2014; **27**(7): 1035-1045 [PMID: 24390214 DOI: 10.1038/modpathol.2013.225]

3 Bedossa P, Carrat F. Liver biopsy: the best, not the gold standard. *J Hepatol* 2009; **50**(1): 1-3 [PMID: 19017551 DOI: 10.1016/j.jhep.2008.10.014]

4 Regev A, Berho M, Jeffers LJ, Milikowski C, Molina EG, Pyrsopoulos NT, Feng ZZ, Reddy KR, Schiff ER. Sampling error and intraobserver variation in liver biopsy in patients with chronic HCV infection. *Am J Gastroenterol* 2002; **97**(10): 2614-2618 [PMID: 12385448]

We appreciate the careful review and would like to thank the reviewers for their comments and suggestions that were helpful in revising the manuscript. We believe that the manuscript has significantly improved with the changes made. We hope that our manuscript is now suitable for publication in the *World Journal of Gastroenterology*.

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

Sincerely yours,

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