

October 13, 2014

**Dear Editor,**

Please find enclosed the edited manuscript in Word format (file name: 13148-review.doc).

**Title:** High neutrophil-lymphocyte ratio indicates poor prognosis for ACLF after liver transplantation. A Research Report.

**Author:** Bingyi Lin, Lin Zhoua, Lei Geng, Zhiyun Zheng, Junjun Jia, Jing Zhang, Jia Yao, Shusen Zheng

**Name of Journal:** World Journal of Gastroenterology

**ESPS Manuscript NO:** 13148

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

1 Format has been updated

2 Revision has been made according to the suggestions of the reviewer

Thank you for your letter and for the reviewers' professional comments concerning our manuscript entitled "High neutrophil-lymphocyte ratio indicates poor prognosis for ACLF after liver transplantation" (ID: 13148). Those comments are all valuable and very helpful for revising and improving our study, as well as the important guiding significance to our researches. We have studied comments carefully and have made correction which we hope meet with approval. Revised portion are marked in red in the paper. The main correction in the paper and the responds to the reviewers' comments are as flowing:

Responds to the reviewer's comments:

Reviewer#1:

1. Response to comment: Identifying PBC subsets (CD4/CD8<sup>+</sup>, N1 N2 for neutrophils etc.) is now days important, in order to assess the basic mechanisms governing the phenomena observed by the gastroenterologists.

Response: It is really true as Reviewer suggested that PBC subsets is very helpful to explore the mechanism underlying the phenomena observed by the transplant surgeons. CD4<sup>+</sup>/CD8<sup>+</sup> lymphocyte ratio is a marker of infection, and CD4<sup>+</sup> Treg or CD8<sup>+</sup> Treg even is the major effector to inhibit the graft rejection. Regrettably, the PBC subsets is not tested in the blood routine test. In addition, the NLR is not only an effective marker of infection, but also is a reflection for disorder of the immune response and severity of diseases. Therefore,

we chose NLR to predict the prognosis of liver receivers with ACLF. The limitation for this study is demonstrated in revised manuscript as following:

Notably, there were several inevitable limitations for the present study. First, only 24 (15.7%) liver receivers with high Scr were enrolled in this cohort. The further exploration and larger sample size are needed. In addition, lymphocyte and neutrophil subsets were not routine measured and the data of their alteration before LT were not available. Hence, the basic mechanisms needs to be further assessed.

2. Response to comment: Could you please give the same info after 1 month? Are the values maintained?

Response: The NLR value at one month after transplantation is available. However, in the liver transplantation cohort, patients are given high dose of immunosuppressants which mainly inhibit the lymphocytes proliferation. Hence, the unsteadily post-transplant NLR is deprived of the predictive superiority for outcomes. We try our best to preclude the disturbance of infection and different pathologies, the pre-transplant NLR is relatively stable and credible.

3. Response to comment: English needs to be improved and the paper shortened just to focus on the specific issues addressed by the authors

Response: We are very sorry for our incorrect writing. The paper have been sended to American Journal Experts to revise our English. Moreover, we cut out a part of concents according to the Reviewer's suggestion.

Reviewer#2:

1. Response to comment: does this knowledge affect treatment of patients?

Response: High NLR is a reflection of immune response dysfunction. In addition, in the study cohort, we found that ACLF patients with high NLR presented worse clinical conditions such as high MELD score and more hepatic encephalopathy attack. Although previously published study suggested that short-course steroids and artificial liver support can't improve the survival outcomes of ACLF patients, our results suggested that ACLF patients taking a combination of a variety of therapies potentially improve the survival outcomes of LT.

2. Response to comment: Is there any clinical use for this knowledge?

Response: Except for LT, a plenty of methods possibly alleviate the clinical symptoms of ACLF patients but can not rescue their lives. In China, because of a great many patients with hepatitis B, liver donation is far away from filling in the need of liver transplantation. Therefore, improving the prognosis of LT is a hot issue. However, the criteria of LT for ACLF is according to ALF, which remains about 20% liver receivers with poor survival outcomes. The pre-transplant high NLR is a reflection of suboptimal patient conditions and immune response disorder, which could precisely predict the prognosis of LT. This result potentially be applied to select appropriate candidates with well outcomes for LT and even improve the current criteria of LT for ACLF.

3. Response to comment: Do both factors combined give further improvement in clinical testing?

Response: we are very sorry for our negligence of the analysis for combining both factors. We have added the analytic result in results and discussion part according to your good suggestion as following:

Patients who presented normal NLR combining normal Scr showed pretty favorable survival outcomes. The 1-, 3- and 5-year OS rates were up to 98.4%, 96.4% and 96.4% respectively. Reversely, liver receivers with high NLR and increased Scr who presented extremely adverse prognosis. The 1-, 3- and 5-year OS rates were down to 60.0%, 60.0% and 50.0% respectively (Figure 3).

Moreover, liver receivers who showed high NLR and increased Scr presented a worst survival outcomes. Although there was poor sensitivity of the serum creatinine in determining the definite extent of renal dysfunction[39, 40], the result revealed that the markers of pre-transplant extrahepatic organ dysfunction potentially affect the prognosis of LT and combining NLR would present an effectively predictive value.

3 References and typesetting were corrected

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

Sincerely yours,

Shusen Zheng

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Dear editor,

we have revised our manuscript No.13148 according to the professional comments of the Editor-in-Chief. we replaced reference 17 with Kayadibi H, Sertoglu E, Uyanik M, Tapan S. Neutrophil-lymphocyte ratio is useful for the prognosis of patients with hepatocellular carcinoma. World J Gastroenterol. 2014 Jul 28;20(28):9631-2. doi: 10.3748/wjg.v20.i28.9631, and added Li X, Chen ZH, Ma XK, Chen J, Wu DH, Lin Q, Dong M, Wei L, Wang TT, Ruan DY, Lin ZX, Xing YF, Deng Y, Wu XY, Wen JY. Neutrophil-to-lymphocyte ratio acts as a prognostic factor for patients with advanced hepatocellular carcinoma. Tumour Biol. 2014 Aug 6. as reference 18.

In addition, we are sorry for liver "receivers".we have revised it with "Liver recipients".

**Please inform us if there are any other inappropriate iterns in the manuscript.** Thank you very much!

Best regards,

-Shusen Zheng

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