Format for ANSWERING REVIEWERS

January 08, 2015

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 15770-review.doc).

Title: Platelet to lymphocyte ratio as a novel prognostic tool for gallbladder carcinoma

Author: Qing Pang, Ling-Qiang Zhang, Rui-Tao Wang, Jian-Bin Bi, Jing-Yao Zhang, Kai Qu, Su-Shun Liu, Si-Dong Song, Xin-Sen Xu, Zhi-Xin Wang, Chang Liu

Name of Journal: *World Journal of Gastroenterology*

ESPS Manuscript NO: 15770

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

Thanks for providing us a chance to revise our paper.

We have carefully read the suggestions by the reviewers and the editor, and then we tried our best to improve our manuscript according these suggestions. As the classification of our manuscript language quality evaluation was B in the initial manuscript, we carefully conducted a linguistic revision, again and again, in this revised manuscript. Then we sent our manuscript to AJE, one of the polishing companies that ***WJG*** specifies, to further improve our language. Now, we have provided the "language certificate" which could verify that our revised manuscript have reached Grade A in language. Hope the revised version will meet the requirements of ***WJG***.

Answer reviewers:

Reviewer (1): 2527871

In this manuscript, Pang et al. retrospectively summarized preoperative serum level of platelet and

lymphocyte count from 316 surgical GBC patients and calculated platelet to lymphocyte ratio (PLR). PASW Statistics 18.0 software was used to analyze the association of PLR and clinical characteristics, univariate analysis and multivariate analysis which suggested PLR was a novel prognostic factor for GBC. The study is well-supported by the data but lack of novelty which has been reported in several other cancers. Some important issues should be addressed before acceptance: 1. Is platelet or lymphocyte alone as a predictive factor in GBC? 2. What is the possible mechanisms underlying its prognostic role in GBC? Please make a description in Discussion part. 3. First paragraph of Discussion part should be transferred to Introduction part. (3)??

Comments 1: Is platelet or lymphocyte alone as a predictive factor in GBC?

Answer: First, we would like to express our appreciation to the reviewer for providing us a chance to revise our manuscript. Both platelet and lymphocyte count alone were ever been showed to be associated prognosis in GBC. However, in our cohort with 316 GBC patients, univariate analysis showed that neither platelet nor lymphocyte count alone was a significant predictive factor in GBC (Table 3 in our revised manuscript), no matter binary or continuous variables were used for the two parameters. However, in our cohort, PLR was firstly verified as an independent predictor for the survival in GBC. As suggested by the reviewer, we further highlighted this description in the part of Discussion in the revised manuscript, they were, "Recent studies have shown that abnormal PLT or lymphocyte count alone is predictive of poor survival in patients with GBC. However, in our cohort, neither the PLT count alone nor the lymphocyte count alone was able to significantly predict survival. However, the PLR, which is a simple combined index, strengthened both the role of PLTs and the significance of lymphocytes and was a powerful independent predictor. We confirmed the prognostic significance of the PLR both when it was expressed as binary data and when it was used as a continuous variable. "

Comments 2: What is the possible mechanisms underlying its prognostic role in GBC? Please make a description in Discussion part.

Answer: Thanks very much for this constructive suggestion. To explore the possible mechanisms, we retrieved several relevant literatures. None previous studies ever explained the possible mechanisms underlying the prognostic role of PLR in GBC. We thought the mechanisms could involve two factors, platelet and lymphocyte counts. One the one hand, a high level of platelet has been previously reported to significantly increase the risks of death from various cancers, including GBC. Moreover, an elevated serum level of platelet has been positively correlated with tumor size. *In vitro*, PLTs accelerate the growth and invasion of tumors. On the other hand, a lymphocyte count of less than 1,000/μl is a predictor of poor outcome in GBC. Furthermore, lymphocyte count has been found to be negatively correlated with TNM stage. In addition, lymphocytes have a cancer immune-surveillance role, by which lymphocytes can prevent tumor development. In our cohort, we showed the positive associations between the PLR and the CA-125 level as well as the TNM stage, which might further help us to understand the possible mechanisms. As suggested by the reviewer, we have made a description to explore the possible mechanisms and increased the above relevant contents in the Discussion part.

Comments 3: First paragraph of Discussion part should be transferred to Introduction part.

Answer: We agree with this commendable suggestion. Thanks very much for carefully reading our manuscript. We have transferred the first paragraph of Discussion to the Introduction part in the revised manuscript.

We appreciated the reviewer 2527871 very much for giving us so many valuable suggestions. We have improved our manuscript in the revised version according to all these constructive suggestions.

Reviewer 2: 3002166

The authors confirmed the predictive power of PLR in a rare malignancy at first time in the literature. The findings were supported with adequate statistical analysis. In the conclusion they formulate themselves one of the inadequacies of the manuscript: the disproportionateness due to the stage distribution. It would make the manuscript more valuable if the therapeutic methods behind the follow-up data would be known.

Answer: Thanks very much for your careful reading, positive comments, and nice suggestions. All our included patients received surgical therapy, either radical operation or non-radical operation. The median survival time of the patients after surgical resection was 9 months, with 1-, 3-, and 5-year OS probabilities of 37.1%, 18.9%, and 11.8%, respectively. To our regret, the authors (Qu K, Song SD and Xu XS) who followed up these patients failed to completely collect the therapeutic methods behind the follow-up data. That means, only part of the 316 patients had the information of therapeutic methods behind the follow-up data. Even so, as assessed, of the patients with the information of therapeutic methods behind the follow-up data, most of them did not receive further treatments or only receive symptomatic treatment after discharge. Other patients received chinese herbs or chemotherapeutics, while the effect was unsatisfied. Nearly two thirds of the 316 included patients died within one year, indicated that patients and their relations might not be satisfied with operative treatment.

Comment 1: Some questions to the authors: - The authors wrote: "Patients who had been previously treated for GBC and who had metastasis were excluded." How is it then possible that so many patients with stage IV have been enrolled? - How was the histopathological distribution? - How were the patients treated? As most patients were of advanced stage how many patients underwent radical operation? How many cases were unresectable? How many patients received neoadjuvant chemotherapy?

Answer: In the beginning, we planned to exclude the patients who had metastasis. Later, to obtain a larger sample size, we decided to include the patients with TNM IVB stage, and stratified the 316 patients according to TNM stage in the subsequent prognostic analysis. To our regret, we forgot to omit the words "and who had metastasis were excluded". Please forgive our carelessness. In the revised manuscript, we have removed these words.

We only included the patients who received surgical therapy in our institute, either with radical operation (n=238) or with non-radical operation (n=78). We have increased the no. of patients with radical or non-radical operation in our revised paper. Cases of patients with each TNM stage were showed in Table 1 in the revised manuscript.

Comment 2: - Were any patients excluded due to other causes such as ongoing inflammation

(pneumonia, cholecystitis, Crohn's disease etc.) or anemia?

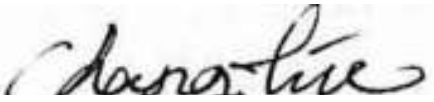
Answer: Thanks for this commendable advice. In the initial manuscript, our description for the selection criterion was incomplete. In the revised manuscript, we have improved selection criterion. Generally, the patients who had potential diseases (or recently took relevant drugs) that influence the values of platelet count and (or) lymphocyte count were all excluded in advance. To be specific, the patients who had concomitant diseases suspected of increasing/reducing the serum platelet count, including severe hypertension, splenic disease and blood coagulation disorders, and the patients who used aspirin or other acetylsalicylic acid drugs one month before the surgery were excluded. Similarly, the patients who had autoimmune diseases (Crohn's disease, rheumatoid arthritis, autoimmune hepatitis, et al), leukemia, viral infection-related diseases or other diseases that influence lymphocyte count were also excluded. In addition, of the included 316 included patients, 160 and 30 patients had histories of gallbladder stone and diabetes respectively. We have added the above criterions in the first paragraph of Method part.

Thanks very much for giving us so many constructive suggestions. We have improved our manuscript in the revised version according to all these suggestions.

3 References and typesetting were corrected

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

Sincerely yours,



Chang Liu, MD, PhD

Dept. of Hepatobiliary Surgery

The First Affiliated Hospital of Medical College

Xi'an Jiaotong University

Xi'an 710061, China

Fax: +86-29-82654746

E-mail: liuchangdoctor@163.com