Peer Review File

Article Information: Available at http://dx.doi.org/10.21037/tau-20-687

Response to Reviewer A:

Major points

Comment 1: The reviewer has big concerns regarding the clinical significance of the variable used for built this nomogram by the Authors also, if from a statistical point of view the methodology is correct. For example, the authors put in this model AST levels but not the TNM stage, which is an important driver factor regarding oncological outcomes. Which is the rationale for what AST levels could influence CSS and PFS?

Reply 1: We include these factors not only based on univariate and multivariate analysis but also previous studies. In up to 20% of cases of RCC, several paraneoplastic syndromes have been described, including erythrocytosis, thrombocytosis, hypercalcemia, and hepatic dysfunction (1,2). The term nephrogenic hepatic dysfunction syndrome was first proposed by Stauffer. Although the pathophysiology of Stauffer's syndrome has not been clearly elucidated, tumor overexpression of interleukin 6 (IL-6) is present in 50–80% of patients with RCC and has been suggested as a possible causative factor (3). Chang et al. (4) experimentally found out that colony stimulating factor (CSF) in the pathogenesis of Stauffer's syndrome. G4 RCC is more malignant and may release more of these biological factors than low grade RCC. Some

studies suggested that RCC patients with hepatic dysfunction have worse prognosis than those without it, despite successful tumor resection (5).

Bezan et al. first found AST/ALT ratio (De Ritis ratio) was a prognostic factor for metastasis-free survival and OS in localized RCC (6). Hakmin Lee et al. proposed that its prognostic value should be limited to ccRCC (7). In our cohort, De Ritis ratio failed to be a significant factor in univariate analysis. It is reasonable, given the presence of metastatic and non-ccRCC patients in our cohort. Hepatic dysfunction may cause an increase in AST level. Therefore, we explored the prognostic value of AST and proved AST was an independent prognostic factor of grade 4 RCC. Besides, we included some parameters of TNM stage (diameter and lymph node metastasis) in the model.

Comment 2: It makes no sense to the reviewer to put in the same basket all T and M stages together only because they all harbored FG4. Indeed, the survival of T4 M1 RCC patients will be very different from a T1 M0 patients also if they presented FG4.

Reply 2: In a study of 4063 patients with RCC from eight international institutions, the 10-year cancer-specific survival rate of Fuhrman 1-4 subgroups were 81.0%, 56.6%, 30.1%, and 18.8%, respectively (8). Previous studies found out that Asian patients with low grade (grades I-II) tumors had a higher 5-year cancer specific survival (CSS) and recurrence free survival (RFS) rates than those with high grade (grades III-IV) tumors in T1N0M0 group. Patients with higher Fuhrman grade have high risk of tumor recurrence and death (9). Therefore, we included FG4 RCC patients in the present cohort. We also think the prognosis of patients in the present

cohort varies greatly. Thus, we developed the nomograms to predict the prognosis.

Comment 3: Does the metastasis variable take into account only visceral metastasis or also node metastasis?

Reply 3: The metastasis refers to bone and visceral metastasis. We have corrected it in the revised manuscript (page 7, line 132).

Comment 4: The Authors put in the nomogram tumor size. It is the pathological or the clinical one? Moreover, within patients with more than 7 cm tumor size, which is the maximum diameter? Did u choose a cut off size? The Authors cannot include tumor of 14 cm if they are outliers.

Reply 4: The tumor size refers to the pathological tumor size (page 5, line 89). We collected the data directly from the pathological report. The maximum diameter is 19cm. Among the 135 patients, only 4 patients had tumors > 14 cm in diameter. Besides, we divided the diameter into two groups bounded by 7cm rather than include it directly into the survival analysis.

Comment 5: According to WHO 2016 guidelines. Fuhrman grade should be used only for clear cell and papillary histological subtype. However, the Authors included also collecting duct or unclassified histology. Please explain.

Reply 5: We collected data from patients diagnosed with RCC between January 2013 and October 2018. The Fuhrman grade was obtained from pathological reports of our institute directly. Fuhrman grade was used for other types of RCC in the department of pathology of our hospital before 2016. Forty patients were diagnosed with G4 RCC after 2016 in our cohort. The histological subtype of them are all clear cell and

papillary histological subtype.

Comment 6: What is mixt histology? Please explain.

Reply 6: Sorry, we made a mistake. We classified renal cell carcinoma with more than one pathological feature as mixt histology wrongly. The mixt histology in the present cohort refers to clear cell/papillary histological subtype. We have corrected the mistakes in the revised manuscript (Please see in Table 1).

Minor comments

Comment 1: Please improve the quality of the images.

Reply 1: We have improved the quality of the figures.

Comment 2: There are some typing and grammar errors.

Reply 2: We have corrected all typing and grammar mistakes and our manuscript has been polished by native English speakers.

Comment 3: Please number lines through all the manuscript to make corrections easier.

Reply 3: We have numbered lines through all the manuscript.

Response to Reviewer B:

Comment 1: The number of patients included was too small to develop nomograms for the prediction of postoperative survival.

Reply 1: Of 4043 patients in our cohort, 171 (3.9 %) grade 4 RCC patients were identified between January 2013 and October 2018. It is difficult to expand the sample size for us.

We included the factors of the nomograms based on univariate and multivariate analysis. Besides, the C-indexes of the nomograms for predicting OS, CSS and PFS were 0.729 (95% CI, 0.659-0.799), 0.725 (95% CI, 0.654-0.796) and 0.702 (95% CI, 0.626-0.778). All C-index > 0.7 and all calibration plots revealed excellent predictive accuracy of the models. Still, external validation by long-term follow-up is needed to test the predictive accuracy of the models.

Comment 2: IMDC risk criteria can be used to evaluate the prognosis of patients with metastatic renal cell carcinoma. However, this study included patients without metastasis at the time of curative surgery. Therefore, IMDC risk scores should be removed from the analyses.

Reply 2: IMDC score has been used to predict the prognosis of metastatic RCC. We explored the prognostic value of IMDC score in grade 4 RCC, considering grade 4 RCC is more prone to metastasis than the other three types. It turned out that IMDC is an independent prognostic factor of survival in grade 4 RCC. Moreover, the nomograms performed well with IMDC risk group included.

Comment 3: As for the OS analysis, only 51 patients died during the study period. However, 9 factors were incorporated into the multivariate analysis shown in Table 2, which might be inappropriate. Please refer to a statistician for advice.

Reply 3: We included the factors of univariable analysis based on previous studies and clinical experience. Only significant variables in the single factor analysis were included in the multivariable analysis. The statistician of our institute thinks it is acceptable, considering there was no significant collinearity among the factors included

in our analysis and it is inappropriate to abandon some significant variables in univariable analysis subjectively.

Reference

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