Dear Prof. Lian-Sheng Ma,

On behalf of my co-authors, we are very grateful to you for giving us an opportunity to revise our manuscript. We appreciate your positive and constructive comments and suggestions on our manuscript entitled **"The immune responses of STEAP4 functions as a novel biomarker in gastric cancer" (ID: 85687).** We have studied reviewers' comments carefully and tried our best to revise our manuscript according to the comments. The following are the responses and revisions we have made in response to the reviewers' questions and suggestions on an item-by-item basis. Our Manuscript was also polished by a native English speaker with biological background to make it easy understanding to readers. The revised portions are highlighted in yellow in the paper. Thank you again for the hard work of the editor and reviewers.

With best wishes

Jing Liu

The main corrections are in the manuscript and the responds to the reviewers' comments are as follows point-to-point.

To Reviewer #1:

1. This data is only retrospectively assessed by a single institute analysis.

Response: Thank you for your critical comments. According to the results of our study, the correlation between STEAP4 and immune cell infiltration in gastric cancer was analyzed from the mRNA level, protein level and clinical analysis, providing a theoretical basis for STEAP4 as a prognostic immune marker for gastric cancer in the future. In the follow-up study, we will use different institutional data for further in-depth research.

2. Statistically, this study is performed by a univariate, not multivariate analysis.

Response: Thank you for your professional suggestions and comments. According to the results of this study, in order to explore the potential of STEAP4 as a therapeutic target for gastric cancer, we analyzed the expression levels of tissue-chip cancer and para-cancer, as well as the clinicopathological parameters of gastric cancer. These results suggest that STEAP4 may be an important prognostic therapeutic target for gastric cancer in clinic.

3. There is no data concerning the chemotherapeutic regimens and their

effects.

Response: Thank you for the reviewer's critical and professional comments. Chemotherapy can extend the life of cancer patients to some extent. However, because there are few studies on STEAP4 in gastric cancer, there are not many studies on chemotherapy. We will follow up on this research in the following studies.

4. A total of 96 individuals are enrolled in this study, →I respect the authors' efforts of collecting the samples. However, the sample size is not enough. Cut off level of STEAP4, the authors should disclose how the cut off level was determined. Please show the ROC, etc. Table2, in order to evaluate the clinicopathological parameters in gastric cancer (GC) patients, at least, serum CEA, serum CA19-9 and Ki67 labeling index, etc. are needed in this table. STEAP4 should be evaluated comparing with the conventional useful biomarkers. The prognostic value of STEAP4 in GC Survival analysis, the authors should disclose the number, the methods, the immunoreactivity, chemotherapy regimens, backgrounds, etc. It is difficult to read. Above all, this manuscript is interesting. However, the present manuscript needs some revisions for the publication of "World Journal of Gastroenterology".

Response: Thank you for your in-depth analysis and professional comments. First of all, we use the online calculator (http://www.powerandsamplesize.com/) to calculate sample size. For the convenience of analysis, the parameters were set as Power = 0.8, Type I error rate = 5%, and the calculated sample size was 67. According to the tissue chip, there were 96 patients with gastric cancer, 84 of whom had corresponding adjacent tissues, which provided an adequate sample size for this study. Secondly, according to the study, the cut-off value of STEAP4 expression was divided according to the immunohistochemical score, that is, the sum of the degree of staining and the proportion of positive cells. In order to make it more convenient for readers to read, we have added the demonstration of STEAP4 immunohistochemical staining in the text (Page 6). In the follow-up researches, we will pay attention to this aspect and timely supplement the lack of this content.

To Reviewer #2:

该研究整体设计一般, STEAP4 并非胃癌的特异性标志物, 可能在其他的肿瘤中也有高表达, 对于胃癌特异性不强，由此研究团队得出结论过于片面，不推荐优先发表。

Response: First of all, we thank the expert for the recognition and rigorous comments. According to the study, STEAP4 acts as a novel

biomarker to play an immune response in gastric cancer. The expression of STEAP4 in gastric cancer and its correlation with immune cell infiltration were analyzed in multiple databases, combined with gastric and para-cancer expression levels in clinical samples and clinicopathological parameters. The association of STEAP4 with immune and stromal responses suggests that it may play an important role in the regulation of the tumor immune microenvironment. This study provided a theoretical basis for the potential of STEAP4 as a prognostic biomarker for gastric cancer.

To Reviewer #3:

1. Abstract. Incorrect sentence: "The expression level of STEAP4 in 96 GC patients and adjacent non-cancerous samples was characterized by immunohistochemistry." Perhaps the authors meant that "The expression level of STEAP4 was characterized by immunohistochemistry in tumor and adjacent non-cancerous samples in 96 GC patients".

Response: Thank you for your valuable and professional suggestions. According to the reviewer's suggestion, I have changed the content in the Abstract to "The expression level of STEAP4 was characterized by immunohistochemistry in tumor and adjacent non-cancerous samples in 96 GC patients." (Page 3).

2. Materials and methods considering that the analysis is based on the stratification of cases with high and low expression of STEAP4, the methodology for assessing the expression of the marker should be given in its entirety. In this case, it is not entirely correct to provide a link to an earlier work (Fang ZX, 2022). It is also necessary to indicate what sum of points was used to divide the samples into cases with high and low expression of the marker. It would be optimal if the authors illustrated all variants of the STEAP4 expression level (no, light yellow, brown yellow, and dark brown).

Response: Thank you for your in-depth analysis and practical comments. According to this study, the immunohistochemical expression score of STEAP4 in gastric cancer was obtained according to the sum of the degree of staining and the proportion of positive cells. In order to make reading easier for readers, specific scoring criteria have been listed in the Materials and Methods (Page 6).

3. In addition, it is not entirely clear if the authors observed dark brown staining (was scored as 3) in the samples, why the image with light brown yellow coloration was chosen as a representative image of high expression.

Response: Thank you reviewers for your professional and critical comments.

We changed the representative image of high expression in Figure 1.

4. Table 2. "Lymph node invasion", the string "Yes" - the amount of interest must be equal to 100.

Response: Thank you very much to the reviewer for your valuable and professional comments. To make it more convenient for readers to read, we adjusted the position of N/M pathological parameters, and added the grading of N0 and N1-N3 of N in Table 2.