Author Response

Dear Editors and Reviewers:

Thank you for your letter and for the reviewers' comments concerning our manuscript ID TAR-19-173 entitled "The clinical value of suPAR in diagnosis and prediction for patients with chronic obstructive pulmonary disease: a systematic review and metaanalysis". Those comments are all valuable and very helpful for revising and improving our paper, 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 yellow in the paper. The main corrections in the paper and the responds to the reviewer's comments are as following.

Reviewer(s)' Comments to Author:

Reviewer 1

Comments to the Author

This systematic review regarding the unspecific biomarker of disease suPAR in COPD is performed according to guidelines for systematic reviews/meta-analysis. results are interesting but I find that the conclusions of the review are much more positive than results warrant. There are no state-of-the-art cohort studies regarding this biomarker. No studies have been performed using validation cohorts and no studies have a long follow-up allowing for assessment of risk of AECOPD.

Response:

Thanks for your kind suggestions and comments.

As suggested by Reviewer, we adjusted the statement in the conclusion section, and supplemented it accordingly in the discussion limitations section.

Specifically, in the discussion section, we adjusted the statement from "*In conclusion, from the results of this systematic review and meta-analysis, suPAR is a promising biomarker for early diagnosis of COPD and AECOPD prediction. The level of suPAR is closely associated with the state of COPD. It can effectively indicate the early state COPD and help aiding clinical decision.*" to "*From the results of this systematic review and meta-analysis, suPAR as a novel biomarker has the potential of early diagnosis of COPD and prediction of AECOPD. There is a potential correlation between the level of suPAR and the state of COPD, which may also indicate the early state of COPD. With further clinical research, the application of suPAR will contribute to clinical decision-making."*

In the last paragraph of Section Discussion, we added the following statement in limitations, *"Third, only four studies reported the accuracy of suPAR in the diagnosis of COPD. Although this can also indicate the trend of suPAR diagnosis superiority, more original research is still needed. In addition, future studies in a larger series of patients and a control group composed of healthy population may reflect the inflammatory process of COPD patients based on plasma suPAR levels. Therefore, before being used in clinic, further study including more patients are needed to assess the suPAR level of COPD patients and AECOPD patients.".*

Major comments:

1. Can the authors please explain quality criteria for the included studies (only 4 are mentioned in Table S3.

Response:

Thank you very much for your constructive comments.

We are sorry for our unclear statement. Although we expounded the quality assessment in the Materials and Methods section, the expression was not accurate. We have carefully examined and revised these misleading statements. The corresponding quality and bias assessment scales should be used according to the type of study. The types of studies included in this systematic review and meta-analysis included *case control studies, cohort studies, and cross-sectional design studies*. The NewcastleOttawa Scale (NOS) was used to assess the quality and bias of case control and cohort studies, while the Agency for Healthcare Research and Quality (AHRQ) was used for the cross-sectional design. The results of these quality and bias assessments are presented in Table S1 and Table S2 of the supplementary appendix. Since the four included studies reported the results of the diagnostic value of suPAR for COPD and the 2×2 contingency table, the Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2) was required to further assess the quality and bias of the four articles including the diagnostic results. The results are showed in Table S3.

As Reviewer suggested, we recognize that inappropriate statements may lead to misunderstandings among readers and obscure some of the key points of this study. To this end, we have carefully revised and further clarified the statements in Data Extraction and Quality Assessment section. We adjusted the statement to *"The quality and bias of the included studies were assessed by two independent authors (M Zhang and J Lu). The Newcastle-Ottawa Scale (NOS) was used to assess the quality and bias of case control and cohort studies (Table S1). As for the cross-sectional design, the Agency for Healthcare Research and Quality (AHRQ) was used for the quality and bias assessment (Table S2). If the included study contained the results of the diagnostic value of suPAR for COPD and the 2×2 contingency table, the Quality Assessment of Diagnostic Accuracy Studies-2 (QUADAS-2) was conducted to further assess the quality and bias (Table S3)".*

2. From Pubmed it is not possible to find these included studies: Portelli 2011; Liu 2018; Hu 2017 and Loukeri 2016 seems to refer to an unpublished abstract. I don't think that unpublished data or papers published in journals that are not peerreviewed should be included.

Response:

Thanks for Reviewer's kind suggestions and comments.

In order to be able to include data more comprehensively, the search strategy of this systematic review and meta-analysis did not limit the language, status and publication date. Among them, Liu 2018 and Hu 2017 are two peer-reviewed papers published in Chinese journals. The corresponding DOI number and links of these two papers are as follows:

Liu 2018:

DOI: 10.3969 /j. issn.1000-8535.2018.02.013;

Link:

http://gzyy.cbpt.cnki.net/WKC/WebPublication/paperDigest.aspx?paperID=7359ae19-87b4-4c6f-ab0c-f9a754251a1b.

Hu 2017:

DOI: 10.3969 /j.issn.1671-7562.2017.01.008;

Link: http://www.xdyx.org.cn/Magazine/Show.aspx?Id=3386.

As for Portelli 2011 and Loukeri 2016, these two papers are conference abstracts. Although the abstracts contain the results and data we need, it is really true as reviewer suggested that unpublished data without peer-reviewed should not be included, thus we exclude these two articles and relevant data. We reviewed and adjusted the number of included studies and population and the related results.

Loukeri 2016 reported the result of diagnosis value of suPAR in patients with COPD. After excluding the result of Loukeri 2016, the direction of the result of diagnosis value of suPAR in patients with COPD (predicted FEV1%≥80%) did not change, which still demonstrated that elevated suPAR levels were effectively associated with high risk of COPD.

Portelli 2011 reported the result of predictive role of suPAR in COPD patients. After excluding the result of Portelli 2011, the direction of the result of predictive role of suPAR in COPD patients (AECOPD vs. COPD) did not change, which still demonstrated that the further increase in suPAR levels effectively indicates the high risk of AECOPD and reflects the status of COPD.

3. From Fig 2 it seems that the study by Wang h et al (2016, ref.no 27) also contributes positively to the association with COPD; however in this study only the biomarker PAI-1 was associated with COPD, whereas elevated suPAR levels were not. Can the authors please explain this.

Response:

Thank you very much for your constructive comments and suggestions.

As prompted by Reviewer, we carefully examined and reviewed the full text of Wang H 2016, and found that the suPAR-related results were not the main results in Wang's research, and Wang et al. did not report the complete results of the association between suPAR and COPD. Fortunately, their study provided the raw data on suPAR related results, which can be incorporated into meta-analysis only after data calculation and consolidation. For example, they reported the serum PAI-1 levels in two groups (COPD patient group and healthy control group), whereas the result of serum suPAR levels was reported in four groups, including healthy nonsmokers, healthy smokers, COPD nonsmokers and COPD smokers. Therefore, the data of suPAR needs to be consolidated. Previously, we simply added the data and obtained results with statistical differences. After being reminded by Reviewer, we re-selected a new and more suitable method of data consolidation.

Based on the data characteristics of continuous variables, we use the following methods to merge the data of subgroup: suppose that the sample size of subgroup A is N_1, the mean is M_1, and the standard deviation is SD_1, the sample size of subgroup B is N_2, the mean is M_2, and the standard deviation is SD_2, then for the combined sample size, we have

Sample size: $N = N_1 + N_2$,

Mean: $M = (N_1 M_1 + N_2 M_2)/(N_1 + N_2)$,

Standard deviation:

$$SD = \sqrt{\frac{(N_1 - 1)SD_1^2 + (N_2 - 1)SD_2^2 + \frac{N_1 N_2}{N_1 + N_2}(M_1^2 + M_2^2 - 2M_1 M_2)}{N_1 + N_2 - 1}}$$

We adopt the above calculation method and recombined the data. After recalculating the result of Wang H 2016, the direction of the result of diagnosis value of suPAR in patients with COPD (predicted FEV1%≥80%) did not change, which still demonstrated that the elevated suPAR levels were effectively associated with a higher risk of COPD. Meanwhile, after excluding the data of Loukeri 2016 and recalculating the data of Wang H 2016, we found that the heterogeneity increased to 47.1%. Therefore, we supplemented the sensitivity analysis (Figure S3) to explore the source of heterogeneity, and added analysis of heterogeneity in Section Discussion. We found that the main source of heterogeneity was the data from Wang H 2016. When the study by Wang et al. was excluded from the analysis, the I 2 decreased to 0%. Then, we carefully rechecked the 11 included studies and found that found that only Wang et al. derived the reported data into subgroups of smokers and non-smokers, which needed to be merged.

The relevant results have been revised in the manuscript, specifically: In the penultimate paragraph of Section Discussion (Heterogeneity analysis part), we added the following statement to explain the potential influencing factors of heterogeneity. *"As the heterogeneity of the predicted FEV1% ≥80% subgroup was I2=47.1%, we further conducted a sensitivity analysis to explore the source of heterogeneity. From the result of sensitivity analysis, it showed that the main source of heterogeneity derived from the study by Wang et al. (Figure S3). When the study by Wang et al. was excluded from the analysis, I2 decreased to 0%. Compared with other included studies, Wang's study further divided COPD population and healthy population into smokers and nonsmokers subgroups. However, they did not clearly indicate the inclusion criteria for the subgroup, so the inclusion population of this study may be potentially different from other studies, which may lead to the increasing heterogeneity."*

Minor comments.

1. In the Discussion the authors refer to the study of Moberg et al (ref 45, p. 12, l. 23); however this study did not measure suPAR.

Response:

Thank you very much for pointing out our negligence carefully. We are very sorry for the misunderstanding caused by this statement. Due to our negligence in processing the document, the statement should have been deleted. We have deleted the relevant expressions and added the due expressions. *"This is consistent with the conclusion of the review conducted by Can et al, which demonstrated that suPAR has the potential in the follow-up of COPD treatment. Indicating inflammation in COPD and assessing suPAR levels can play a key role in the evaluation of the inflammatory process in COPD."*

2. In the Methods, p.6 it is stated which of the authors performed different parts of the review. In the last paragraph, l. 43 it reads: ".... were assessed by two independent authors." Who were they? Are they in the author group?

Response:

Thank you for your careful reminder.

The quality and bias of the included studies were assessed by Mengmeng Zhang and Jiaju Lu (M Zhang and J Lu), and both of whom belonged to the author group of this study. We identified the author's name in this part of the statement as *"The quality and bias of the included studies were assessed by two independent authors (M Zhang and J Lu)."*

Reviewer: 2

Comments to the Author

A biomarker to identify patients who are suffering an AECOPD would be extremely useful.

Some major comments regarding the paper:

1. - I suggest a careful review of English language.

Response:

As the Reviewer suggested, we have revised the WHOLE manuscript carefully and tried to avoid any grammar or syntax error. In addition, we have asked several colleagues who are skilled authors of English language papers to check the English. We believe that the language is now acceptable for the review process.

2. - The term suPAR should be explained in the main text body.

Response:

Thank you very much for your kind suggestion.

As Reviewer suggested, we added the explanation of the term suPAR in the second paragraph of Section Introduction, as shown below.

"Soluble urokinase-type plasminogen activator receptor (suPAR) is a soluble form of the urokinase plasminogen activator receptor (uPAR), which is released by membrane-bound plasminogen activator and is positively correlated with the activation of the immune system. suPAR is found in various body fluids, including blood, urine, and cerebrospinal fluid 8. It is expressed by endothelial cells, macrophages, monocytes, neutrophils, lymphocytes, and fibroblasts, and upregulated by infection and proinflammatory cytokines. The suPAR can promote plasminogen activation, cell adhesion, chemotaxis, and immune cell activation."

3. - In Discussion Section: Paragraph 2, line 43 ended by (are needed to prove it.) it is important to add the reference of this paragraph.

Response:

As Reviewer suggested, we have added references to this section.

In the study conducted by AboEl-Magd et al, they demonstrated that more clinical studies, including more patients, are needed to assess the diagnostic value of serum suPAR compared to other known COPD markers. In the study conducted by Kurtipek et al, they also pointed out that future studies with larger patient series and control group consisting of healthy patients may reflect the inflammatory process in COPD patients according to the plasma suPAR levels. However, further clinical studies, including more patients, are needed to evaluate suPAR in COPD patients before using it like CRP. The abovetwo studies can support the viewpoints of this study, as shown below.

25. AboEl-Magd GH and Mabrouk MM. Soluble urokinase-type plasminogen activator receptor as a measure of treatment response in acute exacerbation of COPD. J Bras Pneumol 2018; 44: 36-41. 2018/03/15. DOI: 10.1590/s1806-37562017000000151.

29. Kurtipek E, Kesli R, Bekçi TT, et al. Assessment of soluble urokinase-type plasminogen activator receptor (suPAR) in chronic obstructive pulmonary disease. International Archives of Medicine 2015; 8.

Article. DOI: 10.3823/1623.