## PEER REVIEW HISTORY

BMJ Open publishes all reviews undertaken for accepted manuscripts. Reviewers are asked to complete a checklist review form (**http://bmjopen.bmj.com/site/about/resources/checklist.pdf**) and are provided with free text boxes to elaborate on their assessment. These free text comments are reproduced below.

## ARTICLE DETAILS

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| **TITLE (PROVISIONAL)** | The Clinical Features, Bacteriology of Endotracheal Aspirates and Treatment Outcomes of Patients with Chronic Obstructive Pulmonary Disease and Community-acquired Pneumonia in An Intensive Care Unit in Taiwan with An Emphasis on Eosinophilia Versus Non-eosinophilia: a retrospective case-control study |
| **AUTHORS** | Huang, Wei-Chang; Lee, Ching-Hsiao; Wu, Ming-Feng; Huang, Chen-Cheng; Hsu, Cheng-Hui; Chen, Hui-Chen; Hsu, Jeng-Yuan; Huang, Chieh-Chen |

## VERSION 1 – REVIEW

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| **REVIEWER** | Dr. Shakti Shukla<br>The University of Newcastle, Australia |
| **REVIEW RETURNED** | 04-Dec-2017 |

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| **GENERAL COMMENTS** | The original research paper by Huang et al. entitled “The Clinical Features, Sputum Bacteriology and Treatment Outcomes of Patients with Chronic Obstructive Pulmonary Disease and Community-acquired Pneumonia in An Intensive Care Unit with An Emphasis on Eosinophilia Versus Non-eosinophilia: a retrospective, cross-sectional study” is yet another important study aimed to address whether blood eosinophilia could be a useful marker to predict the treatment outcomes in COPD patients, in particular COPD individuals presenting with life-threatening CAP. The paper is based on robust rationale and backed up by the availability of very useful clinical data. In my opinion, this manuscript could benefit from minor amendments, which are as follows;<br><br>1. Abstract (conclusion): Authors should mention the specific patient population to avoid generalisation (COPD patients presenting with CAP requiring mechanical ventilation/ICU admission).<br>2. Introduction (paragraph 3): The authors should also mention that blood eosinophilia in stable COPD patients is associated with an increased exacerbation risk, particularly in non-smoking (either ex-smoker/never smoker) patients on maintenance therapies (Price et al., European Respiratory Journal 2014 44: 4416). The majority of participants in this study are indeed either ex-smoker/never smoker.<br>3. I was surprised that approximately half of the participants were not on any maintenance therapy despite majority of the patients were classified as stage II or greater; the authors should discuss the potential clinical implications of this aspect in relation to already published studies exploring the steroid responsiveness in similar population (e.g., Pavord et al., The Lancet Respir Med. 2016:4(9):731–741). Moreover, please discuss the potential implications of these results in context of gender (majority of participants in this study are males). |

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| | 4. Discussion (page 21; line 8-10): The authors mention that the mechanisms linking eosinophilia and adverse clinical outcomes maybe complex, I suggest briefly outlining the possible explanation(s) to better inform the readers. For instance, sputum eosinophilia is considered as a reliable predictor of inhaled corticosteroid responsiveness in COPD patients (Liesker et al., Respir Med 2011; 105: 1853–60).<br>5. Conclusion: It would be worth mentioning how the results of this study should be utilised to further validate the potential of blood eosinophilia in diagnostic/clinical decision making in addition to predicting the clinical outcomes. Moreover, the results should be validated in larger cohorts (especially with a balanced m/f ratios).<br>6. Fig 1. Please correct the spelling of bacterial nomenclature, e.g., Haemophilus influenzae (instead of Hemophilus), Streptococcus pneumoniae (instead of pneumonia) |

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| **REVIEWER** | Aykut Cilli<br>Akdeniz University Hospital Department of Pulmonary Diseases, Antalya-Turkey |
| **REVIEW RETURNED** | 20-Jan-2018 |

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| **GENERAL COMMENTS** | Wei-Chang et al. reported in their current study that eosinophilia may be associated with better clinical outcomes in patients with COPD and CAP. Here is my comments about their article:<br>Methods:<br>1. Authors pointed out that patients with lung cancer were excluded from the study, but in the result section - in table 1- there are 62 patients (19%) with malignancy. Does this mean they included all cancer patients except lung cancer? This must be clearly stated in method section.<br>2. How were the microbiological samples collected? Trans-bronchial aspiration does not mean sputum! This is called endotracheal aspiraton! If the patients were intubated and recieved mechanical ventilation upon arrival, how could it be possible to collect sputum sample? Authors should not be used “sputum” for endotracheal aspiration.<br>Results:<br>1. Lower white blood count does not mean less inflammation.<br>2. Most of the COPD patients (n=170, 53%) were not on any drug treatments for COPD. This is highly interesting finding since all the patients were diagnosed as COPD before admission. Can authors make any comment on this issue?<br>3. How many of the patients were using antibotic while collecting microbiological samples?<br>4. Any patients received NIV?<br>5. Any patients received systemic corticosteroid treatment? If so, any difference between groups?<br>Discussion:<br>1. Limitation part must be a separate paragraph and includes low microorganism eradication rate, antibiotic use before admission may mask microbiological etiology, retrospective design, single center, etc…..<br>2. What is the possible underlying mechanism(s) between eosinophil level and clinical outcomes? This must be discussed, at least briefly.<br>3. Etiological diagnosis of pneumonia may be difficult in COPD because chronic colonization confounds the interpretation of sputum culture results. S. pneumoniae is the most common cause of CAP in COPD (Cilli A, Curr Infect Dis Rep 2015; 17:444). CAP caused by gram-negative bacilli and P. aeruginosa is an uncommon diagnosis for COPD patients, they are not leading pathogens. P.aeruginosa as |

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| | a CAP etiology in COPD patients has been shown to be associated with older age (84.9 vs 76.3, p<0.01), moderate to severe disease, and those patients who were treated regularly with oral corticosteroid (Pifarre, Respir Med 2007, Ko, Respir Med 2007). Therefore, there is a clear need to be explained by the authors that why is S. pneumoniae is the eight leading etiologic agent in their current study, and why is P. aeruginosa and other gram-negatives are leading pathogens?<br>4. Authors should also discuss the findings of etiologic agents in another article which included similar patient group (Cilli A, et al. Journal of Critical Care 2013).<br>Conclusion :<br>1. Again, lower white blood counts do not reflect reduced inflammation. |

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| **REVIEWER** | ADAMANTIA LIAPIKOU<br>Sotiria Chest Diseases Hospital, Athens, Greece |
| **REVIEW RETURNED** | 28-Jan-2018 |

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| **GENERAL COMMENTS** | Well designed and written ms about a hot-topic in the field of respiratory infections. |

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| **REVIEWER** | Dr Neil John Glassford<br>Austin Hospital, Department of Intensive Care Medicine, Monash University, Department of Epidemiology and Preventative Medicine |
| **REVIEW RETURNED** | 08-Feb-2018 |

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| **GENERAL COMMENTS** | I have no conflicts of interest to declare.<br><br>Peer Review<br><br>1. Yes.<br><br>2. No. While the abstract is adequate as written, it will need to be revised in alignment with changes made to the rest of the document.<br><br>3. No.<br><br>A cross-sectional study would allow the presented hypothesis to be explored and the aims to be fulfilled.<br><br>However, I am uncertain that the authors have performed a true cross sectional study.<br><br>By collecting data at ED admission, RICU admission and through the RICU course, an element of time is added to the design of this study. This is also implied by the use of the APACHE II score, collected at the end of the first 24h in the ICU.<br><br>Cross-sectional studies can only (to my knowledge) demonstrate associations – one of their major limitations is their inability to demonstrate causation due to an absence of a temporal dimension. By introducing this dimension, and by framing the associations demonstrated on regression analysis as predictors of poor outcome, it would seem as though this study should have been analysed as a cohort or case-control study, and I would recommend revising the paper and resubmitting.<br><br>This is particularly important as time acts as a confounder in many |

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| | ways – time to intubation, time to ICU, time to antibiotics, time to sputum sample, etc, and none of these are accounted for in the analysis.<br><br>4. No.<br><br>Additional statistical information is required, as is a decision on the actual design of the study. Further information is required regarding the time course over which data was collected.<br><br>5. Yes<br><br>6. No.<br><br>Outcomes and their derivation should be clearly described in the methods section.<br><br>Additional outcomes such as readmission within 3 months should be considered.<br><br>7. No.<br><br>No information is given regarding the use of the Bonferroni correction for multiple comparisons. Given the number of tests performed, p should be so corrected for comparisons.<br><br>No information is given regarding the conduct of the univariate or multivariate regression and insufficient information is provided regarding the univariate comparisons. Further detail needs to be provided regarding the selection and definition of predictors, inclusion and exclusion, etc. Has it been performed as cross-sectional regression, or linear, implying a temporal component – incorrect in cross-sectional studies?<br><br>Multiple admissions in the same patients are likely to introduce bias. The data should be presented as the first admission for each patient over the time period. At the very least, a sensitivity analysis of first admissions should be included.<br><br>8. No.<br><br>The authors may wish to consider referencing Ho's 2017 Systematic review and Meta-Analysis in Scientific Reports (PMID: 29044160). This is the appropriately aggregated summation of the impact of eosinophilia in COPD, and much of the discussion could be simplified in reference to this. I would also suggest referencing Pavord, et al (N Engl J Med 2017; 377:1613-1629), given that this demonstrates the importance of the eosinophilic phenotype and similarities and differences between the study cohort and Pavord's cohort may have important therapeutic implications for novel drugs.<br><br>9. No.<br><br>Results should be revised in keeping with the study design. If predicting, this requires the adoption of a temporal component to the study and it should be revised accordingly. Otherwise, just present the associations.<br><br>10. No. |

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| | Much of the information in Table S1 should be in table 1. Given that the authors feel they present novel clinical information on their cohort, this should be easily available to the reader.<br><br>Given that a cell count of 300 appears to have no representation in the literature, nor seems to add anything particularly novel to the study. I would suggest framing the rationale for the inclusion of these analyses clearly, or removing them from the main paper and inserting them into the electronic supplemental material.<br><br>Language is important here. A cross-sectional study can only demonstrate associations, not predictors or determinants. Please amend the results sections accordingly.<br><br>Tables 2 and 3 should be a single table presented dependent on the selection of an appropriate study design.<br><br>Was further information regarding the severity of CAP available? Any of the validated scoring systems would offer a useful insight into the relationship between CAP and eosinophilia.<br><br>11. No.<br><br>Key findings should be clarified and presented in the early discussion.<br><br>A lower WCC and PaCO2 do not mean lower levels of inflammation or improved ventilation.<br><br>Given this is retrospective, ventilatory mode, tidal volume, and rate should be available. Indeed, the VT is presented as being available in >75% of patients, but no volume data is offered. Why? CRP is also available.<br><br>Such strong conclusions should be avoided without discussing such supporting or conflicting data. Moreover, a cross-sectional study is the wrong sort of design to be drawing such conclusions.<br><br>The information regarding the importance of 2% eosinophilia should be presented in the introduction.<br><br>12. No.<br><br>An appropriate section describing the limitations of the study should be included in any revised redraft attempted.<br><br>13. While complete, it is for the wrong sort of study.<br><br>14. Yes.<br><br>No concerns.<br><br>15. No.<br><br>This document will require review by a professional editor for minor corrections/tense changes/minor grammar changes.<br><br>Not for further statistical review. |

## VERSION 1 – AUTHOR RESPONSE

Response to Reviewers

Reviewers' Comments to Author

Reviewer: 1
Reviewer Name: Dr. Shakti Shukla
Institution and Country: The University of Newcastle, Australia
Please state any competing interests or state 'None declared': None declared

Please leave your comments for the authors below
The original research paper by Huang et al. entitled "The Clinical Features, Sputum Bacteriology and Treatment Outcomes of Patients with Chronic Obstructive Pulmonary Disease and Community-acquired Pneumonia in An Intensive Care Unit with An Emphasis on Eosinophilia Versus Non-eosinophilia: a retrospective, cross-sectional study" is yet another important study aimed to address whether blood eosinophilia could be a useful marker to predict the treatment outcomes in COPD patients, in particular COPD individuals presenting with life-threatening CAP. The paper is based on robust rationale and backed up by the availability of very useful clinical data. In my opinion, this manuscript could benefit from minor amendments, which are as follows;

1. Abstract (conclusion): Authors should mention the specific patient population to avoid generalisation (COPD patients presenting with CAP requiring mechanical ventilation/ICU admission).

Response: Thanks for your suggestion. We have indicated that the clinical implications of our findings are applicable to the study population only to avoid inappropriate generalizations in the revised manuscript. (Page 5, lines 10-13 in the marked-up version of manuscript. Page 5, lines 3-5 in the unmarked version of manuscript.)

2. Introduction (paragraph 3): The authors should also mention that blood eosinophilia in stable COPD patients is associated with an increased exacerbation risk, particularly in non-smoking (either ex-smoker/never smoker) patients on maintenance therapies (Price et al., European Respiratory Journal 2014 44: 4416). The majority of participants in this study are indeed either ex-smoker/never smoker.

Response: Thanks for your suggestion. We have mentioned that blood eosinophilia is associated with a higher exacerbation risk in patients with stable COPD in the revised manuscript. However, we cited the article published by Zeiger RS et al. (J Allergy Clin Immunol Pract Published Online First: 15 November 2017. doi: 10.1016/j.jaip.2017.10.004.) rather than that published by Price et al. (European Respiratory Journal 2014 44: 4416) mainly due to the used cut-off value of 300 cells/mm3 for blood eosinophilia in the former which is consistent with our study though these two articles have similar findings. Furthermore, we have not put an emphasis on the non-smoking status of the participants in the revised manuscript because the majority of participants in the former article (Zeiger RS et al. J Allergy Clin Immunol Pract Published Online First: 15 November 2017. doi: 10.1016/j.jaip.2017.10.004.) cited in the present study were current or past smokers that was similar with our study. Instead, we have described that our study enrolled a majority of ex- and current smokers (82.4%) in the revised manuscript. (Page 8, lines 12-14; page 12, lines 12-13 in the marked-up version of manuscript. Page 8, lines 12-14; page 12, lines 9-10 in the unmarked version of manuscript.)

3. I was surprised that approximately half of the participants were not on any maintenance therapy despite majority of the patients were classified as stage II or greater; the authors should discuss the potential clinical implications of this aspect in relation to already published studies exploring the steroid responsiveness in similar population (e.g., Pavord et al., The Lancet Respir Med. 2016:4(9):731–741). Moreover, please discuss the potential implications of these results in context of gender (majority of participants in this study are males).

Response: We agree with your opinions. Published literatures found that elderly patients with COPD may have more adverse effects of maintenance therapies than expected and a preference for the small-volume nebulizers with regard to effectiveness. In addition, poor coordination with either a pressurized metered-dose inhaler or dry powder inhaler is common in the elderly patients with COPD. (Fried TR et al. JAMA 2012;308:1254-63; Restrepo RD et al. Int J Chron Obstruct Pulmon Dis 2008;3:371-84.) These evidences could explain why approximately half of the participants were not on any maintenance therapy despite a majority of the patients were classified as stage II or greater in our study which enrolled participants with an overall mean age of 78.7±8.9 years. We have added this information in the revised manuscript. (Page 12, lines 10-11; page 12, lines 13-15; page 29, lines 12-19 in the marked-up version of manuscript. Page 12, lines 7-8; page 12, lines 10-12; page 23, lines 6-13 in the unmarked version of manuscript.) Moreover, we have not cited the reference by Pavord et al. (The Lancet Respir Med. 2016:4(9):731–741) here as it shows a conclusion that patients with COPD with lower blood eosinophil counts had more pneumonia events than did those with higher counts which seems not to be related to this issue. Instead, we have cited the important article in the paragraph discussing the association between blood eosinophilia and clinical outcomes in patients with COPD in the revised manuscript. (From page 28, line 16 to page 29, line 4 in the marked-up version of manuscript. Page 22, lines 10-17 in the unmarked version of manuscript.)
Our study was composed of only 21 (8.0%) female subjects. Existing evidence shows that sex has a variable impact on the prevalence of eosinophilia and treatment outcomes in patients with COPD due to a combination of both environmental/behavioral factors and genetic/biophysiologic factors, making our findings may not be applicable to female patients with COPD. (Aryal S et al. Int J Chron Obstruct Pulmon Dis 2014;9: 1145-54; DiSantostefano RL et al. Respir Med 2016;112:88-96.) We have added this limitation in the revised manuscript. (Page 28, lines 10-15 in the marked-up version of manuscript. Page 22, lines 4-9 in the unmarked version of manuscript.)

4. Discussion (page 21; line 8-10): The authors mention that the mechanisms linking eosinophilia and adverse clinical outcomes maybe complex, I suggest briefly outlining the possible explanation(s) to better inform the readers. For instance, sputum eosinophilia is considered as a reliable predictor of inhaled corticosteroid responsiveness in COPD patients (Liesker et al., Respir Med 2011; 105: 1853–60).

Response: Although there is no easy explanation for the association between eosinophilia and clinical outcomes in patients with COPD, previous studies, together with this present study, showed that patients with COPD with blood eosinophilia requiring hospitalization for severe exacerbations and life-threatening CAP are characterized with better pulmonary function and lower blood leukocyte count and PaCO2. (Kang HS et al. Int J Chron Obstruct Pulmon Dis 2016;11:2467-73; Saltürk C et al. Int J Chron Obstruct Pulmon Dis 2015;10:1837-46) In addition, one previous study found a better treatment response to corticosteroids in the management of exacerbations of COPD. (Bafadhel M et al. Am J Respir Crit Care Med 2012;186:48-55) Together, this information may partly explain the better clinical outcomes in the patients with COPD with blood eosinophilia requiring hospitalization for severe exacerbations and life-threatening CAP. Together with our findings, we have summarized these evidences as the possible explanations for the association between eosinophilia and clinical outcomes in patients with COPD in the revised manuscript to better inform the readers. (Page 29, lines 5-11 in the marked-up version of manuscript. From page 22, line 18 to page 23, line 5 in the unmarked version of manuscript.) The findings by Liesker JJ et al. (Respir Med 2011;105:1853-60)

indicate that sputum eosinophil inflammation can predict exacerbations after withdrawal of inhaled corticosteroids in COPD. Furthermore, several studies found a relationship between eosinophilia and clinical outcomes. (Pavord ID et al. Lancet Respir Med 2016;4:731-41; Ho J et al. Sci Rep 2017;7:13451; Casanova C et al. Eur Respir J 2017;22:50(5)) Together with our findings, we concluded that the eosinophil level in both blood and sputum may be an useful biomarker of clinical outcomes when treating COPD. We also have added this information in the revised manuscript. (From page 28, line 16 to page 29, line 4 in the marked-up version of manuscript. Page 22, lines 10-17 in the unmarked version of manuscript.)

5. Conclusion: It would be worth mentioning how the results of this study should be utilised to further validate the potential of blood eosinophilia in diagnostic/clinical decision making in addition to predicting the clinical outcomes. Moreover, the results should be validated in larger cohorts (especially with a balanced m/f ratios).

Response: Thanks for your suggestion. Indeed, our results have important clinical implications in the management of COPD. Alongside with existing evidence, (Ho J et al. Sci Rep 2017;7:13451; Pavord ID et al. N Engl J Med 2017;377:1613-29) these findings are helpful to clinicians when making treatment decision for the management of COPD, especially when choosing pharmacological and antibiotic therapies. We have added this information in the “Discussion” section rather than the “Conclusion” section of the revised manuscript to better inform the readers how the results of this study should be utilized in clinical practice. (Page 32, lines 7-15 in the marked-up version of manuscript. Page 25, lines 3-9 in the unmarked version of manuscript.) Furthermore, there is a relatively small cohort of patients with a majority of participants being male in our study. Thus, we have mentioned that future studies should enroll a larger cohort with a balanced gender ratio to validate our results in the “Discussion” section of the revised manuscript. (Page 32, lines 15-16 in the marked-up version of manuscript. Page 25, lines 9-10 in the unmarked version of manuscript.)

6. Fig 1. Please correct the spelling of bacterial nomenclature, e.g., Haemophilus influenzae (instead of Hemophilus), Streptococcus pneumoniae (instead of pneumonia)

Response: Thanks for your reminder. We have corrected these spelling errors in the figure 1.

Reviewer: 2
Reviewer Name: Aykut Cilli
Institution and Country: Akdeniz University Hospital Department of Pulmonary Diseases, Antalya-Turkey
Please state any competing interests or state ‘None declared’: none declared

Please leave your comments for the authors below
Wei-Chang et al. reported in their current study that eosinophilia may be associated with better clinical outcomes in patients with COPD and CAP. Here is my comments about their article:
Methods:
1. Authors pointed out that patients with lung cancer were excluded from the study, but in the result section - in table 1- there are 62 patients (19%) with malignancy. Does this mean they included all cancer patients except lung cancer? This must be clearly stated in method section.

Response: Yes, this study included patients with malignancies except for lung cancer. We have mentioned that patients with lung cancer were excluded from this study in the “Methods” section of both the originally submitted version and revised version of manuscripts. Also, we have added this information in Table 1 in the revised manuscript to clearly inform the readers. (Page 10, lines 8-9; Table 1 in page 15 in the marked-up version of manuscript. Page 10, lines 6-7; Table 1 in page 15 in the unmarked version of manuscript.)

2. How were the microbiological samples collected? Trans-bronchial aspiration does not mean sputum! This is called endotracheal aspiraton! If the patients were intubated and recieved mechanical ventilation upon arrival, how could it be possible to collect sputum sample? Authors should not be used “sputum” for endotracheal aspiration.

Response: Thanks for your reminder. We have changed the term “sputum” to “endotracheal aspirate” where appropriate throughout the revised manuscript to reflect the true experimental condition.

Results:

1. Lower white blood count does not mean less inflammation.

Response: We agree with your comment. Thus, we have deleted the incorrect interpretation and described it as “lower white blood count(s)” or “lower leukocyte count(s)” in the revised manuscript. (Page 4, line 16; page 13, line 7; page 13, lines 12-13; page 27, line 8; page 29, line 7 in the marked-up version of manuscript. Page 4, line 16; page 13, lines 1-2; page 13, line 7; page 21, line 5; page 23, line 1 in the unmarked version of manuscript.)

2. Most of the COPD patients (n=170, 53%) were not on any drug treatments for COPD. This is highly interesting finding since all the patients were diagnosed as COPD before admission. Can authors make any comment on this issue?

Response: Thanks for your suggestion. The same comment is also provided by reviewer 1. The response of this interesting finding has been detailed at the point #3 of reviewer 1.

3. How many of the patients were using antibiotic while collecting microbiological samples?

Response: We have re-analyzed the materials and found that, out of 262 participants, 116 (44.3%) used antibiotics while collecting microbiological samples. We have indicated the data in Table 1 in the revised manuscript and in Tables S1-S4 in the revised supplementary information. (Table 1 in page 17 in the marked-up version of manuscript. Table 1 in page 17 in the unmarked version of manuscript. Tables S1-S4 in pages 9-20 in the marked-up version of supplementary information. Tables S1-S4 in pages 6-17 in the unmarked version of supplementary information.)

4. Any patients received NIV?

Response: Out of 262 participants, 47 (17.9%) received NIPPV after successful liberation from invasive mechanical ventilation support during the RICU stay. We have indicated the data in Table 1 in the revised manuscript and in Tables S1-S4 in the revised supplementary information. (Table 1 in page 17 in the marked-up version of manuscript. Table 1 in page 17 in the unmarked version of manuscript. Tables S1-S4 in pages 9-20 in the marked-up version of supplementary information. Tables S1-S4 in pages 6-17 in the unmarked version of supplementary information.)

5. Any patients received systemic corticosteroid treatment? If so, any difference between groups?

Response: Out of 259 patients, 230 (88.8%) received systemic corticosteroid treatment and distributed evenly between paired study groups. (Page 13, lines 8-10; Table 1 in page 17 in the marked-up version of manuscript. Page 13, lines 3-5; Table 1 in page 17 in the unmarked version of manuscript. Tables S1-S4 in pages 9-20 in the marked-up version of supplementary information. Tables S1-S4 in pages 6-17 in the unmarked version of supplementary information.)

Discussion:

1. Limitation part must be a separate paragraph and includes low microorganism eradication rate, antibiotic use before admission may mask microbiological etiology, retrospective design, single center, etc…..

Response: Thanks for your excellent comment. We have mentioned the strengths and limitations of this study in a separate paragraph in the “Discussion” section of the revised manuscript. As you mentioned, our study has several limitations that a number of the endotracheal aspirates were collected after antibiotic therapy had been initiated along with the possible use of antibiotics before admission and low microorganism eradication rate in the lower airways of patients with COPD, possibly leading to the low discovery rate of potentially pathogenic microorganisms and the effect on bacterial profiling. Moreover, our study was retrospective in nature, implemented in the respiratory intensive care unit at a single center where medical staff was familiar with the management of COPD, and composed of only 21 (8.0%) female subjects, making our findings be interpreted with caution, especially to undefined groups of patients and outside the respiratory intensive care unit and may not be applicable to female patients with COPD. We have amended and added this information to the revised manuscript to better inform the readers. (From page 6, line 1 to page 7 line 2; from page 27, line 11 to page 28, line 15 in the marked-up version of manuscript. Page 6, lines 1-19; from page 21, line 8 to page 22, line 9 in the unmarked version of manuscript.)

What is the possible underlying mechanism(s) between eosinophil level and clinical outcomes? This must be discussed, at least briefly.

Response: Thanks for your suggestion. The same comment is also provided by reviewer 1. The response has been detailed at the point #4 of reviewer 1.

2. Etiological diagnosis of pneumonia may be difficult in COPD because chronic colonization confounds the interpretation of sputum culture results. S. pneumoniae is the most common cause of CAP in COPD (Cilli A, Curr Infect Dis Rep 2015; 17:444). CAP caused by gram-negative bacilli and P. aeruginosa is an uncommon diagnosis for COPD patients, they are not leading pathogens. P.aeruginosa as a CAP etiology in COPD patients has been shown to be associated with older age (84.9 vs 76.3, $p<0.01$), moderate to severe disease, and those patients who were treated regularly with oral corticosteroid (Pifarre, Respir Med 2007, Ko, Respir Med 2007). Therefore, there is a clear need to be explained by the authors that why is S. pneumoniae is the eight leading etiologic agent in their current study, and why is P. aeruginosa and other gram-negatives are leading pathogens?

Response: Thanks for your brilliant comment. As you mentioned, more infections attributable to P. aeruginosa and gram-negative bacilli are observed in hospitalized patients with COPD and CAP, especially for those who are older, have moderate to severe diseases or receive oral corticosteroids regularly. (Restrepo MI et al. Eur Respir J 2006;28:346-51; Cilli A et al. J Crit Care 2013;28:975-9; Ko FW et al. Respir Med 2008;102:1109-16; Pifarre R et al. Respir Med 2007;101:2139-44) Our study population was characterized by older age and poorer lung function. Together, this may explain why the commonest isolated organisms were P. aeruginosa and other gram-negative bacilli rather than S. pneumoniae in this study. This finding has important clinical implications when choosing antibiotic therapies in patients with COPD and life-threatening CAP. We have added the highlights in the revised manuscript. (From page 30, line 19 to page 31, line 7; page 32, lines 7-15; page 33, lines 5-6 in the marked-up version of manuscript. Page 24, lines 6-13; page 25, lines 3-9; page 25, line 17 in the unmarked version of manuscript.)

3. Authors should also discuss the findings of etiologic agents in another article which included similar patient group (Cilli A, et al. Journal of Critical Care 2013).

Response: Cilli A, et al found that P. aeruginosa was the leading infective pathogen in patients with COPD and CAP requiring admission to the ICU. (Cilli A et al. J Crit Care 2013;28:975-9) This result responds that more infections attributable to P. aeruginosa and gram-negative bacilli are observed in hospitalized patients with COPD and CAP found in our study and that by Restrepo MI et al. (Restrepo MI et al. Eur Respir J 2006;28:346-51). Together with our findings, we have summarized existing evidences and discussed the highlights of causative organisms for CAP in hospitalized patients with COPD and CAP in the revised manuscript. (From page 30, line 19 to page 31, line 7; page 32, lines 7-15; page 33, lines 5-6 in the marked-up version of manuscript. Page 24, lines 6-13; page 25, lines 3-9; page 25, line 17 in the unmarked version of manuscript.)

Conclusion :

1. Again, lower white blood counts do not reflect reduced inflammation.

Response: Thanks for your reminder. We have deleted the inappropriate interpretation in the "Conclusion" section of the revised manuscript. (Page 5, line 8; page 33, line 2 in the marked-up version of manuscript. Page 5, lines 3-5; page 25, lines 14-19 in the unmarked version of manuscript.)

Reviewer: 3
Reviewer Name: ADAMANTIA LIAPIKOU
Institution and Country: Sotiria Chest Diseases Hospital, Athens, Greec
Please state any competing interests or state 'None declared': None declared

Please leave your comments for the authors below
Well designed and written ms about a hot-topic in the field of respiratory infections.

Response: We appreciate your positive feedback.

Reviewer: 4
Reviewer Name: Dr Neil John Glassford
Institution and Country: Austin Hospital, Department of Intensive Care Medicine; Monash University, Department of Epidemiology and Preventative Medicine
Please state any competing interests or state 'None declared': None declared

Please leave your comments for the authors below
I have no conflicts of interest to declare.

Peer Review

1. Yes.

2. No. While the abstract is adequate as written, it will need to be revised in alignment with changes made to the rest of the document.

Response: Thanks for your suggestion. We have re-designed and turned the study into a retrospective case-control study. Also, we have modified the abstract accordingly in the revised manuscript. (From page 4, line 1 to page 5, line 13 in the marked-up version of manuscript. From page 4, line 1 to page 5, line 5 in the unmarked version of manuscript.)

3. No.

A cross-sectional study would allow the presented hypothesis to be explored and the aims to be fulfilled.

However, I am uncertain that the authors have performed a true cross sectional study.

By collecting data at ED admission, RICU admission and through the RICU course, an element of time is added to the design of this study. This is also implied by the use of the APACHE II score, collected at the end of the first 24h in the ICU.

Cross-sectional studies can only (to my knowledge) demonstrate associations – one of their major limitations is their inability to demonstrate causation due to an absence of a temporal dimension. By introducing this dimension, and by framing the associations demonstrated on regression analysis as predictors of poor outcome, it would seem as though this study should have been analysed as a cohort or case-control study, and I would recommend revising the paper and resubmitting.

This is particularly important as time acts as a confounder in many ways – time to intubation, time to ICU, time to antibiotics, time to sputum sample, etc, and none of these are accounted for in the analysis.

Response: Thanks for your brilliant comment. We agree with your opinion and have re-designed the study as a retrospective case-control study. Thus, we have amended the sections of TITLE, ABSTRACT, METHODS, RESULTS, and DISCUSSION in the revised manuscript as well as the Supplementary Information of our revised manuscript accordingly. (Page 1, line 4; from page 4, line 1 to page 5, line 13; page 9, line 12; from page 10, line 16 to page 11, line 6; from page 21, line 1 to page 27, line 10 in the marked-up version of manuscript. Page 1, line 4; from page 4, line 1 to page 5, line 5; page 9, line 11; from page 10, line 14 to page 11, line 4; from page 18, line 10 to page 21, line 7 in the unmarked version of manuscript. Page 1, line 6; page 2, lines 2-3; pages 7-20 in the marked-up version of supplementary information. Page 1, line 6; page 2, lines 1-2; pages 6-17 in the unmarked version of supplementary information.)

4. No.

Additional statistical information is required, as is a decision on the actual design of the study. Further information is required regarding the time course over which data was collected.

Response: We have modified the section of “Statistical analysis” based on the re-designed retrospective case-control study in nature in the revised manuscript. (Page 5, lines 7-14 in the marked-up version of supplementary information. Page 5, lines 5-12 in the unmarked version of supplementary information.)

5. Yes

6. No.

Outcomes and their derivation should be clearly described in the methods section.

Additional outcomes such as readmission within 3 months should be considered.

Response: Four previously validated (using a similar study population) adverse treatment outcomes, including RICU length of stay >14 days, failed weaning, death, and readmission arising from respiratory diseases within 3 months were considered to analyze the association between blood eosinophilia and clinical outcomes in our study. (Breen D et al. Thorax 2002;57:29-33; Menzies R et

al. Chest 1989;95:398-405; Chu CM et al. Thorax 2004;59:1020-5) We have cited the references of the derivation of studied treatment outcomes in the revised manuscript. Furthermore, we have added the clinical outcome of readmission arising from respiratory diseases within 3 months as the reviewer suggested in the revised manuscript and supplementary information. (Page 11, lines 2-5; Table 2 in pages 22-23 in the marked-up version of manuscript. From page 10, line 18 to page 11, line 3; Table 2 in pages 19-20 in the unmarked version of manuscript. From page 2, line 25 to page 3, line 3; pages 7-20 in the marked-up version of supplementary information. From page 2, line 24 to page 3, line 2; pages 6-17 in the unmarked version of supplementary information)

7. No.

No information is given regarding the use of the Bonferroni correction for multiple comparisons. Given the number of tests performed, p should be so corrected for comparisons.

Response: Bonferroni correction is a method for correction for Type 1 error rates associated with multiple comparisons. (Kao LS et al. J Surg Res. 2008; 144: 158–170) As a result, independent t test/ chi-square test rather than Bonferroni correction were used for assessment between two groups (case-control) in the present study. (Page 5, lines 7-9 in the marked-up version of supplementary information. Page 5, lines 5-7 in the unmarked version of supplementary information)

No information is given regarding the conduct of the univariate or multivariate regression and insufficient information is provided regarding the univariate comparisons. Further detail needs to be provided regarding the selection and definition of predictors, inclusion and exclusion, etc. Has it been performed as cross-sectional regression, or linear, implying a temporal component – incorrect in cross-sectional studies?

Response: As we have turned this study into a retrospective case-control study in nature, multivariate logistic regression models were used to analyze associated factors for various in-RICU adverse outcomes if they were significant in univariate analysis. Factors selected for univariate analysis included the cutoff values for blood eosinophilia used in this study (> 2% versus ≤ 2% and > 300 cells/μL versus ≤ 300 cells/μL), all of the factors in Table 1, and the types of bacteriology of endotracheal aspirates. We have mentioned this information in the revised manuscript. (Page 5, lines 9-13 in the marked-up version of supplementary information. Page 5, lines 7-10 in the unmarked version of supplementary information. Page 21, lines 7-11 in the marked-up version of manuscript. Page 18, lines 11-14 in the unmarked version of manuscript.)

Multiple admissions in the same patients are likely to introduce bias. The data should be presented as the first admission for each patient over the time period. At the very least, a sensitivity analysis of first admissions should be included.

Response: We agree with your comment that multiple admissions in the same patient are likely to introduce bias. Thus, only the first admission was included for each patient who had multiple RICU admissions that fulfilled all the inclusion and exclusion criteria during the study period. We have amended the inclusion and exclusion criteria as well as the patient enrollment flow chart in the revised manuscript. Finally, a total of 262 patients were enrolled in the analysis. The sections of both RESULTS and DISCUSSION have been modified accordingly. (Page 10, lines 9-11; from page 12, line 8 to page 33, line 7 in the marked-up version of manuscript. Page 10, lines 8-10; from page 12, line 5 to page 25, line 19 in the unmarked version of manuscript. Pages 6-20 in the marked-up version of supplementary information. Pages 5-17 in the unmarked version of supplementary information)

8. No.

The authors may wish to consider referencing Ho's 2017 Systematic review and Meta-Analysis in Scientific Reports (PMID: 29044160). This is the appropriately aggregated summation of the impact of eosinophilia in COPD, and much of the discussion could be simplified in reference to this. I would also suggest referencing Pavord, et al (N Engl J Med 2017; 377:1613-1629), given that this demonstrates the importance of the eosinophilic phenotype and similarities and differences between the study cohort and Pavord's cohort may have important therapeutic implications for novel drugs.

Response: Thanks for your suggestion. Since these two important articles demonstrate that blood eosinophilia is associated with better outcomes in terms of reduced length of hospital stay and may have important therapeutic implications for steroidal and bronchodilator therapies as well as biologic therapies (Mepolizumab), this information, together with our findings that patients with COPD and blood eosinophilia using a cut-off value of 2% complicated with CAP requiring invasive mechanical ventilation and admission to an ICU had superior in-ICU treatment outcomes in terms of shorter ICU length of stay and that by Liesker JJ et al. indicating that sputum eosinophilia is an independent predictor for a COPD exacerbation after ICS withdrawal, suggests that eosinophil level in both blood and sputum may be an useful biomarker of clinical outcomes in the management of COPD as well as blood eosinophilia may be predictive of favorable response to biological and steroidal and bronchodilator therapies in patients with stable COPD. Therefore, we have cited these two references, summarized all the information, and added the highlights in the revised manuscript. (From page 28, line 16 to page 29, line 4; page 32, lines 7-15 in the marked-up version of manuscript. Page 22, lines 10-17; page 25, lines 3-9 in the unmarked version of manuscript.)

9. No.

Results should be revised in keeping with the study design. If predicting, this requires the adoption of a temporal component to the study and it should be revised accordingly. Otherwise, just present the associations.

Response: We have re-designed the study as a retrospective case-control study and presented just the associations between blood eosinophil levels and in-RICU adverse outcomes in the section of RESULTS of the revised manuscript. (Page 21, lines 1-14; Table 2 in pages 22-23; page 27, lines 7-10 in the marked-up version of manuscript. Page 18, lines 10-17; Table 2 in pages 19-20; page 21, lines 4-7 in the unmarked version of manuscript.)

10. No.

Much of the information in Table S1 should be in table 1. Given that the authors feel they present novel clinical information on their cohort, this should be easily available to the reader.

Response: Thanks for your suggestion. We have merged Table 1 and Table S1 of the originally submitted version of manuscript and added several important parameters (e.g. CURB-65 scores, Use of antibiotics while microbiological sampling, Ventilator settings including ventilator mode, tidal volume, and respiratory rate, and Use of NIPPV after successful liberation from IMV support during the RICU stay) in the Table 1 of the revised version of manuscript, making it better inform to readers. (Pages 14-17 both in the marked-up version of manuscript and in the unmarked version of manuscript.)

Given that a cell count of 300 appears to have no representation in the literature, nor seems to add anything particularly novel to the study. I would suggest framing the rationale for the inclusion of these analyses clearly, or removing them from the main paper and inserting them into the electronic supplemental material.

Response: Numerous references using a cut-off value of whether 2% or 300 cells/μL for blood eosinophilia show significant clinical implications for blood eosinophilia in patients with COPD. (Pascoe S et al. Lancet Respir Med 2015;3:435-42; Bafadhel M et al. Am J Respir Crit Care Med 2012;186:48-55; Bafadhel M et al. Chest 2016;150:320-8; Kang HS et al. Int J Chron Obstruct Pulmon Dis 2016;11:2467-73; Saltürk C et al. Int J Chron Obstruct Pulmon Dis 2015;10:1837-46; Vedel-Krogh S et al. Am J Respir Crit Care Med 2016;193:965-74; Zeiger RS et al. J Allergy Clin Immunol Pract Published Online First: 15 November 2017. doi: 10.1016/j.jaip.2017.10.004) Therefore, an eosinophil level greater than whether 2% or 300 cells/μL appears to be representative to blood eosinophilia and we have indicated the clinical implications of blood eosinophilia when using a threshold of whether 2% or 300 cells/μL in the section of INTRODUCTION as well as implemented analyses using both 2% and 300 cells/μL as a threshold in the revised manuscript. (Page 8, lines 12-16; page 9, lines 2-5; Table 1 in pages 14-17 in the marked-up version of manuscript. Page 8, lines 12-16; page 9, lines 2-5; Table 1 in pages 14-17 in the unmarked version of manuscript.)

Language is important here. A cross-sectional study can only demonstrate associations, not predictors or determinants. Please amend the results sections accordingly.

Response: Thanks for your suggestion. We have turned the study into a retrospective case-control study in nature and presented the associations between blood eosinophil levels and in-RICU adverse outcomes accordingly in the section of RESULTS of the revised manuscript. (Page 21, lines 1-14; Table 2 in pages 22-23; page 27, lines 7-10 in the marked-up version of manuscript. Page 18, lines 10-17; Table 2 in pages 19-20; page 21, lines 4-7 in the unmarked version of manuscript.)

Tables 2 and 3 should be a single table presented dependent on the selection of an appropriate study design.

Response: Thanks for your suggestion. Based on the retrospective case-control study in nature, we have provided the amended univariate analysis results in Table S1-S4 in the revised version of supplementary information and multivariate logistic regression analysis results in Table 2 in the revised manuscript. (Pages 9-20 in the marked-up version of supplementary information. Pages 6-17 in the unmarked version of supplementary information. Pages 22-23 in the marked-up version of manuscript. Pages 19-20 in the unmarked version of manuscript.)

Was further information regarding the severity of CAP available? Any of the validated scoring systems would offer a useful insight into the relationship between CAP and eosinophilia.

Response: We have added CURB-65 scores in the revised manuscript. However, it showed that the CURB-65 scores distributed evenly between the eosinophilia group and the non-eosinophilia group and were not associated with any in-ICU adverse treatment outcomes. (Table 1 in pages 14-17; Table 2 in pages 22-23 in the marked-up version of manuscript. Table 1 in pages 14-17; Table 2 in pages 19-20 in the unmarked version of manuscript. Page 2, lines 13-15; pages 9-20 in the marked-up version of supplementary information. Page 2, lines 12-14; pages 6-17 in the unmarked version of supplementary information.)

11. No.

Key findings should be clarified and presented in the early discussion.

Response: We have presented the main findings of our study in the first paragraph of the section of DISCUSSION in the revised manuscript. (Page 27, lines 7-10 in the marked-up version of manuscript. Page 21, lines 4-7 in the unmarked version of manuscript.)

A lower WCC and PaCO2 do not mean lower levels of inflammation or improved ventilation.

Response: We agree with your comment. The PaCO2 level showed no significant difference between the eosinophilia group and the non-eosinophilia group after re-designing and re-analyzing our study. Thus, we have deleted the incorrect interpretation for the PaCO2 level. Furthermore, we have amended and described the results of WBC as “lower white blood count(s)” or “lower leukocyte count(s)” when comparing between the eosinophilia group and the non-eosinophilia group in the revised manuscript. (Page 4, line 16; page 13, line 7; page 13, lines 12-13; page 27, line 8; page 29, line 7 in the marked-up version of manuscript. Page 4, line 16; page 13, lines 1-2; page 13, line 7; page 21, line 5; page 23, line 1 in the unmarked version of manuscript.)

Given this is retrospective, ventilatory mode, tidal volume, and rate should be available. Indeed, the VT is presented as being available in >75% of patients, but no volume data is offered. Why? CRP is also available.

Response: We have added these important parameters in the Table 1 of the revised manuscript, making it more informative to readers. (Table 1 in page 17 both in the marked-up version of manuscript and in the unmarked version of manuscript.)

Such strong conclusions should be avoided without discussing such supporting or conflicting data. Moreover, a cross-sectional study is the wrong sort of design to be drawing such conclusions.

Response: In the revised manuscript, we concluded that, regardless of whether 2% or 300 cells/μL blood eosinophil level was used as a threshold, the eosinophilia group had distinct characteristics than the non-eosinophilia group, while 2% rather than 300 cells/μL was associated with clinical outcomes in such study population. Moreover, such population had a distinct bacterial profiling for the causative organisms of CAP. Together, these data should be considered in the management of patients with COPD and CAP requiring IMV and ICU admission. We have re-written the conclusion based on the results available when using a retrospective case-control study design, making it supported by the data presented. (From page 32, line 19 to page 33, line 7 in the marked-up version of manuscript. Page 25, lines 13-19 in the unmarked version of manuscript.)

The information regarding the importance of 2% eosinophilia should be presented in the introduction.

Response: Numerous studies have reported that, when using whether 2% or 300 cells/μL as a threshold, blood eosinophilia is associated with a higher exacerbation risk in patients with stable COPD as well as an association between peripheral blood eosinophilia and a reduced future risk of exacerbations in patients with stable COPD and better outcomes in patients with exacerbations of COPD following treatment with inhaled and systemic corticosteroids. (Pascoe S et al. Lancet Respir Med 2015;3:435-42; Bafadhel M et al. Am J Respir Crit Care Med 2012;186:48-55; Bafadhel M et al. Chest 2016;150:320-8; Kang HS et al. Int J Chron Obstruct Pulmon Dis 2016;11:2467-73; Saltürk C et al. Int J Chron Obstruct Pulmon Dis 2015;10:1837-46; Vedel-Krogh S et al. Am J Respir Crit Care Med 2016;193:965-74; Zeiger RS et al. J Allergy Clin Immunol Pract Published Online First: 15 November 2017. doi: 10.1016/j.jaip.2017.10.004)This information indicates that blood eosinophilia using a threshold of whether 2% or 300 cells/μL has important clinical implications in the management of COPD. Therefore, we have added the information regarding the importance of both 2% and 300 cells/μL as a threshold for blood eosinophilia in patients with COPD in the section of INTRODUCTION in the revised manuscript. (Page 8, lines 12-16 both in the marked-up version of manuscript and in the unmarked version of manuscript.)

12. No.

An appropriate section describing the limitations of the study should be included in any revised redraft attempted.

Response: Thanks for your reminder. Our study has several limitations that a number of the endotracheal aspirates were collected after antibiotic therapy had been initiated along with the possible use of antibiotics before admission and low microorganism eradication rate in the lower airways of patients with COPD, possibly leading to the low discovery rate of potentially pathogenic microorganisms and the effect on bacterial profiling. Moreover, our study was retrospective in nature, implemented in the respiratory intensive care unit at a single center where medical staff was familiar with the management of COPD, and composed of only 21 (8.0%) female subjects, making our findings be interpreted with caution, especially to undefined groups of patients and outside the respiratory intensive care unit and may not be applicable to female patients with COPD. We have mentioned the limitations of this study in a separate paragraph in the “Discussion” section of the revised manuscript, making it better inform the readers. (From page 27, line 11 to page 28, line 15 in the marked-up version of manuscript. From page 21, line 8 to page 22, line 9 in the unmarked version of manuscript.)

13. While complete, it is for the wrong sort of study.

Response: Thanks for your comment. We have re-designed and turned the study into a retrospective case-control study. Also, we have modified the content of our manuscript accordingly. (Page 1, line 4; from page 4, line 1 to page 5, line 13; page 9, line 12; from page 10, line 16 to page 11, line 6; from page 21, line 1 to page 27, line 10 in the marked-up version of manuscript. Page 1, line 4; from page 4, line 1 to page 5, line 5; page 9, line 11; from page 10, line 14 to page 11, line 4; from page 18, line 10 to page 21, line 7 in the unmarked version of manuscript. Page 1, line 6; page 2, lines 2-3; pages 7-20 in the marked-up version of supplementary information. Page 1, line 6; page 2, lines 1-2; pages 6-17 in the unmarked version of supplementary information.)

14. Yes.

No concerns.

15. No.

This document will require review by a professional editor for minor corrections/tense changes/minor grammar changes.

Response: Thanks for your suggestion. The editorial assistance of the revised manuscript is provided by a professional editor who is a native English speaker.

**VERSION 2 – REVIEW**

| | |
|---|---|
| **REVIEWER** | Dr NJ Glassford<br>Royal Melbourne Hospital, Melbourne Health. Critical Care Research, Department of Epidemiology and Preventative Medicine, School of Public Health and Preventative Medicine, Monash University, Australia |
| **REVIEW RETURNED** | 29-Mar-2018 |

| | |
|---|---|
| **GENERAL COMMENTS** | Thank you for asking me to review this manuscript once more. |

| | |
|---|---|
| | The authors are to be commended on a fine job in incorporating so many changes from the reviewers.<br><br>I have several recommendations that would require revision before I could recommend accepting this paper for publication.<br><br>6. Please define the outcomes of interest early and clearly. Identify primary outcomes and secondary outcomes.<br><br>7. Insufficient information provided regarding the conduct of MRA.<br><br>Please see: Quality of reporting of multivariable logistic regression models in Chinese clinical medical journals. Zhang YY1, Zhou XB, Wang QZ, Zhu XY. Medicine (Baltimore). 2017 May;96(21):e6972.<br><br>A review of two journals found that articles using multivariable logistic regression frequently did not report commonly recommended assumptions. Ottenbacher KJ, Ottenbacher HR, Tooth L, Ostir GV. J Clin Epidemiol. 2004 Nov;57(11):1147-52. Review. PMID: 15567630<br><br>An appraisal of multivariable logistic models in the pulmonary and critical care literature. Moss M, Wellman DA, Cotsonis GA. Chest. 2003 Mar;123(3):923-8. PMID: 12628895<br><br>15. Still requires editorial review for grammatical error. |

## VERSION 2 – AUTHOR RESPONSE

Reviewer(s)' Comments to Author:

Reviewer: 4

Reviewer Name: Dr NJ Glassford

Institution and Country: Royal Melbourne Hospital, Melbourne Health.

Critical Care Research, Department of Epidemiology and Preventative Medicine, School of Public Health and Preventative Medicine, Monash University, Australia

Please state any competing interests or state ‘None declared’: None Declared

Please leave your comments for the authors below

Thank you for asking me to review this manuscript once more.

The authors are to be commended on a fine job in incorporating so many changes from the reviewers.

I have several recommendations that would require revision before I could recommend accepting this paper for publication.

6. Please define the outcomes of interest early and clearly. Identify primary outcomes and secondary outcomes.

Response: The study's primary and secondary aims of interest have been clarified early and clearly in the "METHODS" section in the revised manuscript. (Page 9, lines 11-18 in both the marked-up version of manuscript and the unmarked version of manuscript.)

7. Insufficient information provided regarding the conduct of MRA.

Please see: Quality of reporting of multivariable logistic regression models in Chinese clinical medical journals. Zhang YY1, Zhou XB, Wang QZ, Zhu XY. Medicine (Baltimore). 2017 May;96(21):e6972.

A review of two journals found that articles using multivariable logistic regression frequently did not report commonly recommended assumptions. Ottenbacher KJ, Ottenbacher HR, Tooth L, Ostir GV. J Clin Epidemiol. 2004 Nov;57(11):1147-52. Review. PMID: 15567630

An appraisal of multivariable logistic models in the pulmonary and critical care literature. Moss M, Wellman DA, Cotsonis GA. Chest. 2003 Mar;123(3):923-8. PMID: 12628895

Response: Thanks for your excellent suggestion. The detailed information on the multivariate logistic regression analyses regarding the treatment outcomes of interest in the study has been provided in the footnote of Table 2 in the revised manuscript. (Pages 19-20 in both the marked-up version of manuscript and the unmarked version of manuscript.)

15. Still requires editorial review for grammatical error.

Response: To make it fluent to the readers, the English editorial services of the revised manuscript have been done by ATS Medical Editing.

**VERSION 3 – REVIEW**