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Reviewer A

In this retrospective review, Toyoda et al seek to draw a correlation between PGD severity and acute renal injury, and in turn acute renal injury in the peri-transplant period with the severity of chronic kidney disease. This data is compelling in that it is the first to clearly correlate the severity of PGD with AKI, and adds to our growing understanding of mechanisms and outcomes related to post transplant co-morbidities. The authors have presented a well written and coherent manuscript, based on a well characterized transplant registry. I request minor revisions and clarifications.
Methods:

Comment 1:

- The authors should include a description of their approach to use of calcineurin inhibitors post transplant – i.e timing and target trough levels. If other therapies with known nephrotoxicity are routinely used in the early post transplant period, this should be detailed as well.

Reply 1:

- **We appreciate your kind feedback regarding our manuscript. In our protocol, Tacrolimus trough level was used as the target of 8-12 ng/mL for all patients. We included this in the method.**

Change in the text:

- (Page 6, line 22) Tacrolimus trough level was used as the target of 8-12 ng/mL for all patients.

Comment 2:

- PGD Grade: The use of the lowest PFR at any time point within 72 hrs is appropriate by definition, however most studies looking at PGD related outcomes consider PGD grade at 48-72 hrs as a more accurate reflection of this syndrome. I would be interested to see AKI severity in a group of patients with PGD3 at 48 or 72 hrs

Reply 2:

- **This is an excellent point. In our definition, PGD is defined based on ISHLT guideline. Please see the details at method section of PGD grade. As you pointed out, we used the lowest PFR at any time point within 72 hours.**

Change in the text:

- (None)

Comment 3:

- AKI Stage: Please clarify what criteria were used to determine need for renal replacement therapy? If any patients received isolated ultrafiltration, how were they staged?

Reply 3:

- **Thank you so much for bringing this up. An experienced transplant nephrology evaluated the patients and decided the indication of renal replacement therapy based on eGFR, urine output, electrolytes abnormality. We added the sentence referring to this.**

Change in the text:

- (Page 9, line 1 to 2) Indications for renal RRT were determined by an experienced transplant nephrologist based on eGFR, urine output, and electrolyte abnormalities.

Comment 4:

- Patient demographics: Data detailing this cohort's GFR pre-transplant should be incorporated, and correlated to long term CKD outcomes. A current limitation of this study is the absence of this important data.

Reply 4:

- **This is a great point. We included eGFR in table 1. At least the eGFR of patients with AKI stage 0 was no higher than that of patients with other stages.**

Change in the text:

- (None)

Comment 5:

- Page 9, line 21: Is there a threshold value for operative time influencing AKI, or is this a time dependent relationship?

Reply 5:

- **Thank you for pointing this out. Our data showed that AKI group has longer operative time (Table 2.) But other factors are also significantly difference such as blood transfusion during transplant. They are also known risk factors for PGD too. Our scope is relationship between PGD and AKI therefore finding threshold value for operative time influencing AKI is out of scope in this manuscript.**

Change in the text:

- (None)

Comment 6:

- Page 9, line 22: What was higher AKI severity associated with VA ECMO use compared to? Other forms of mechanical support, no support? Was CPB used at all?

Reply 6:

- **Thank you for binging this to our attention. Similar to above question, VA ECMO use could be a risk factor to AKI but other factors are also significantly difference. The VA EVMO use group was compared to the no support group. In our center, we do not use CPB during lung transplant. We added a sentence referring to this in the discussion.**

Change in the text:

- (Page 15, line 22 to page 16, line 2) Central VA-ECMO has been applied in our institution instead of cardiopulmonary bypass (CPB) to reduce PGD based on evidence that ECMO has been shown to reduce intraoperative blood transfusion volume compared with CPB (47).

Comment 7:

- Page 10, line 21: Clarify time frame for survival? Is this also 2 years as noted in the section above?

Reply 7:

- **This is an excellent point. Based on our follow up time, we cut at 2 years in the KM curve. However, hazard ratios were calculated by Cox regression analysis. Therefore, we didn't cut at any point in time.**

Change in the text:

- (None)

Comment 8:

- Page 12, line 17: sentence starting "Moreover, it…" What does it refer to, this reads as an incomplete sentence.

Reply 8:

- **You raise an excellent point. We changed as below.**

Change in the text:

- (Page 14, line 14-16) Common factors post-lung transplantation such as hypoxia (15, 31, 32) and systemic inflammatory cytokine production (33, 34) can exacerbate both lung and kidney dysfunction. Moreover, inflammatory cytokines have been reported to exacerbate PGD and AKI (15, 34).

Comment 9:

- Limitations: Only serum Cr was used to measure GFR over time (I presume).

Additionally, no data presented on pre-transplant GFR and its effects on post transplant AKI or CKD.

Reply 9:

- **You bring up a great point. We added GFR in the Table1. There were no significant difference among the four AKI groups.**

Change in the text:

- (None)

Comment 10:

- Figure 2: If possible, may be interesting to superimpose a heat map for HR of death on (A). Presumably, the HR for mortality in AKI 3/PGD 3 is greater than AKI 0/PGD 0. This would be a nice way to illustrate this effect.

Reply 10:

- **Thank you for pointing this out. However, the number of cases was small and significant HR values could not be obtained. As HRs were also varied, we believed we could only provide figures that would be misleading to the reader. Instead, we replaced the figure with a heatmap representation of the number of cases. This allows us to visually capture areas with high case counts.**

Change in the text:

- (Page 25, line 7 to 8) The heatmap shows the number of cases.

Reviewer B

• Overview:

o This is a single institution retrospective study attempting to assess the relationship between severity of primary lung graft dysfunction and acute kidney injury as well as the severity of acute kidney injury and progress to chronic kidney disease. The authors fairly discuss the potential confounding variables in the relationship between PGD and AKI. However, their analysis does not do much to mitigate these potential confounders, making it difficult to understand what the correlation between grade 3 PGD and severe AKI means here. Further obscuring the interpretability of these results is that patients on dialysis pre-operatively were not excluded from the analysis, making it difficult to know whether outcomes in the severe AKI group were related to worse baseline renal function.

• Abstract:

Comment 1:

- This paper doesn't investigate how management of PGD impacts AKI

incidence or vice versa, so the last sentence of the conclusion does not fit.

Reply 1:

- **This is a great point. We deleted this sentence in the abstract conclusion.**

Change in the text:

- (None)

• Introduction:

Comment 2:

- It would strengthen the introduction if the authors discussed why they think stratifying AKI risk by PGD severity matters. We don't have specific interventions for grade 3 PGD that differ from those used in lower grade PGD. Also, if we had evidence there was a causal relationship between PGD and AKI then we should be trying to better under the mechanism(s) by which PGD leads to AKI regardless of whether the relationship between PGD and AKI is moderated by PGD severity.

Reply 2:

- **Thank you for pointing this out. Both PGD and AKI are known risk factor for mortality and link between severity of PGD and AKI are unknown, therefore investigation is very important to improve survival. This is clinical study and investigate mechanism is out of scope in this study as no basic or translational data is available. We mentioned in the introduction: However, the correlation between PGD and AKI severity in the entire lung transplantation cohort remained unclear. If there is a strong connection between PGD and AKI severity, it would be a good rationale to research the mechanism by which PGD leads AKI and to conduct tactics for protecting lung and kidney function.**

Change in the text:

- (None)

Comment 3:

- The writing in the third paragraph should be revised. There are several instances where diction takes away from the clarity of the sentences.

Reply 3:

- **This is a great point. I replaced the word however with therefore.**

Change in the text:

- (Page 6, line 1 to 3) Therefore, the correlation between PGD and AKI severity in the entire lung transplantation cohort remained unclear. If there is a strong

connection between PGD and AKI severity, it would be a good rationale to research the mechanism by which PGD leads AKI.

• Methods:

Comment 4:

- o It would be helpful to know why only transplants between 01/2018 and 01/2022 included. Since you're using medical records, I would imagine that you would have that data available from earlier dates. If so, I could be useful to look at the some of the longer-term outcomes like progression to CKD at greater than just one-year post-transplant. Even if patients no longer follow up in lung transplant clinic beyond that time, it should be evident from the EMR whether they died, went on to/continued to need dialysis, or required further transplantations etc.

Reply 4:

- **Thank you for bringing this up. We are relatively new program and since January 2018, medical record system was significantly changed and collecting details data is not available. Therefore, study period is from January 2018.**

Change in the text:

- (None)

Comment 5:

- Would explain somewhere why you chose to include patients that were on dialysis pre-transplant. It makes it hard to understand whether these patients getting AKIs were just sicker and with poorer renal function to begin with or whether something about either having PGD or being treated for PGD is driving this.

Reply 5:

- **This is an excellent point. We included history of dialysis but at the time of lung transplant with relatively maintain GFR. At the suggestion of another reviewer, we decided to include preoperative eGFR in Table 1. The results indicate that patients with more severe AKI do not have lower preoperative eGFR.**

Change in the text:

- (None)

Comment 6:

- For the relationship between PGD grade and AKI severity, Spearman's test is

not really that informative since there are many potential confounders between PGD and AKI. If possible, it would be more useful to perform a logistic regression model with AKI as the outcome (perhaps Grade 3 vs none/Grade 1 or 2) so you could at least account for some of the confounding variables.

Reply 6:

- **You bring up a great point. We think Spearman's test can show the casual link between severity of PGD and AKI which are in the Figure 2. We added logistic regression model to supplemental table.**

Change in the text:

- (Page 11, line 13 to page 12, line 2) Comparing the AKI stage 0-2 and AKI stage 3 groups, the univariate logistic analysis showed that preoperative dialysis and ECMO use, high LAS, COVID-19-associated ARDS, high total bilirubin level, and female donors were more common in the AKI stage 3 group (dialysis use, odds ratio [OR]=7.76; ECMO use, OR=5.69; LAS, OR=1.03; COVID-associated ARDS, OR=2.87; high total bilirubin level, OR=1.80; and female donors, OR=2.76) (Supplemental Table 1). Similarly, patients in the AKI stage 3 group had longer operative times, higher transfusion and intraoperative VA-ECMO use (operative times, OR=1.49; packed red blood cells, OR=1.13; and intraoperative VA-ECMO use, OR=2.96). Furthermore, PGD grades 1 to 3 and PGD grade 3 were significantly higher in AKI stage 3 than in AKI stage 0-2 (PGD grades 1 to 3, OR=5.94; and PGD grade 3, OR=32.6). The ventilator connections, and length of intensive care unit and hospital stays were also longer in the AKI stage 3 group (ventilator connections, OR=1.05; intensive care unit stay, OR=1.05; and hospital stay, OR=1.05) (Supplemental Table 1).

• Results:

Comment 7:

- 25% of the AKI stage 3 cohort was on dialysis preoperatively. Do you know how many of these patients had grade 3 PGD? If you exclude patients that were on HD pre-transplant was there still higher post-op dialysis and dialysis at discharge in the stage 3 AKI group?

Reply 7:

- **You raise an excellent point. We did not exclude those patients in our analysis.**

Change in the text:

- (None)

Comment 8:

- Similarly, does the relationship between PGD grade and AKI severity persist if you exclude patients that were on dialysis pre-op?

Reply 8:

- **Thank you for binging this to our attention. Please see above answer.**

Change in the text:

- (None)

Comment 9:

- For Figure 4, since the progression of CKD categories seems to occur similarly independent of the AKI stage, is the correlation between AKI stage and GFR category of CKD just due to worse baseline renal function in the AKI stage 3 cohort?

Reply 9:

- **This is a great point. As shown in Figure 3, there is a strong correlation between AKI stage and GFR category of CKD at any point in time. These correlations were not only due to AKI stage 3 cohort. Renal function may deteriorate in all patients after lung transplantation.**

Change in the text:

- (None)

• Discussion:

Comment 10:

- Again it would helpful to explain why you think PGD severity would matter here.

Reply 10:

- **Thank you for pointing this out. Both PGD and AKI are known risk factor for mortality and link between severity of PGD and AKI are unknown, therefore investigation is very important to improve survival. This is clinical study and investigate mechanism is out of scope in this study as no basic or translational data is available. We mentioned in the introduction: However, the correlation between PGD and AKI severity in the entire lung transplantation cohort remained unclear. If there is a strong connection between PGD and AKI severity, it would be a good rationale to research the mechanism by which PGD leads AKI and to conduct tactics for protecting lung and kidney function.**

Change in the text:

- (None)

Comment 11:

- The first sentence in the second paragraph would be better stated as something like “Common factors post lung transplant such as hypoxia and systemic inflammatory cytokine production can similarly exacerbate both lung and kidney dysfunction.”

Reply 11:

- **Thank you for bringing this up. We changed following your suggestion.**

Change in the text:

- (Page 14, line 14-15) Common factors post-lung transplantation such as hypoxia (15, 31, 32) and systemic inflammatory cytokine production (33, 34) can exacerbate both lung and kidney dysfunction.

Comment 12:

- Overall the discussion is fair is does not overstate the studies findings.

Reply 12:

- **Thank you for your comment.**

Change in the text:

- (None)

Reviewer C

Comment 1:

- The study is interesting, although it suffers from several methodological limitation. The following comments are intended for your consideration:
- 1. Firstly, it is retrospective observational study and all the limitations of a retrospective observational study design apply.
- 2. The measurements of PaO2/FiO2 are not correct in those recipients without PaO2. ISHLT consensus recommends to measure PaO2/FiO2 with PaO2 no with saturation, it invalidates the sample.

Reply 1:

- **Thank you so much for bringing this up. Our definition of PGD is based on ISHLT guideline. If PaO2/FiO2 is available, we used it.**

Change in the text:

- (None)

Reviewer D

This study examined the association between primary graft dysfunction (PGD) and acute kidney injury (AKI) in lung transplant patients, as well as the impact of AKI on

chronic kidney disease (CKD) progression over time. The retrospective review of a lung transplantation database included 206 patients, and the results showed a significant correlation between PGD grading and AKI staging, as well as between AKI staging and CKD progression. The study highlights the importance of well-balanced respiratory management and renal protection strategies in the early postoperative period.

My impression is this is a single-center study that examines the association between PGD and AKI. They have found that these correlates.

Comment 1:

- The question is what is the difference between patients with PGD who have AKI and those who do not? The second half of the article is not consistent with the title, as it shows the relationship between kidney failure and prognosis, and it is difficult to understand whether the author wants to discuss the relationship between PGD and AKI or renal failure.

Reply 1:

- **You bring up a great point. First of all, we would like to discuss the link between severity of PGD and AKI, and then discuss long term outcomes about kidney function leading to CKD or not.**

Change in the text:

- (None)

Reviewer E

In this manuscript Toyoda et al. investigate the effect of primary graft dysfunction in lung transplantation on the development of AKI and also assess propensity of lung transplant AKI patients to develop chronic kidney disease (CKD). Lung transplantation-induced AKI, which occurs mostly within the first 24hours after transplant, is a significant clinical topic due to its high prevalence and negative protracted effects on morbidity and mortality of lung transplant patients. How lung transplantation induces AKI is essentially unknown.

The investigators examined 206 lung transplant recipients in a retrospective observational single-center study and correlated degree of primary graft dysfunction (PGD) with the degree of AKI. PGD was determined by clinical observations within 72hours of transplant, including pulmonary edema on chest radiograph, low FIO2/PaO2 ratio, use of extracorporeal membrane oxygenation.
AKI was staged using KDIGO criteria, considering degree of serum creatinine increase

and need for renal replacement therapy. CKD was also staged by KDIGO.
Chronic lung allograft dysfunction (CLAD) was defined as a persistent decline (≥20%) in the measured forced expiratory volume in 1 second from the post-transplantation baseline.

As noted by the authors, previous studies have shown that lung transplant can induce AKI of different severity, and that the increasing degree of AKI severity directly correlates with reduced 5-year survival (e.g. Nephrol Dial Transplant (2014) 29: 1702–1709 doi: 10.1093/ndt/gfu226).
In this manuscript the authors show that the degree of severity of PGD positively correlates with the degree of severity of AKI; and also that the severity of AKI predicts progression to CKD. The authors state that one previous study focusing on lung transplanted cystic fibrosis patients also showed that severity of PGD is correlated to severity of subsequent AKI after transplant (Scaravilli V, Merrino A, Bichi F, et al. Longitudinal assessment of renal function after lung transplantation for cystic fibrosis: transition from post-operative acute kidney injury to acute kidney disease and chronic kidney failure. J Nephrol. 2022;35(7):1885-93.).

In the view of this reviewer the findings reported in this manuscript only incrementally improve our current understanding of this clinical problem. Nonetheless, the work reports solid findings and corroborates previous data by others in a new single center patient cohort.

I have a few suggestions and comments that may help improve the manuscript:

Comment 1:

- 1. The authors state that "… the impact of AKI severity on CKD risk and the temporal evolution of CKD grades has not been well investigated (21)." This may be true in the lung transplant population, but the effect of degree of AKI on the development of subsequent CKD has been studied in detail in kidney patients (e.g. Chawla et al. Kidney international 2011). This should at least be included in the discussion.

Reply 1:

- **Thank you for binging this to our attention. We added this to our manuscript.**

Change in the text:

- (Page 6, line 8-10) However, while the impact of the degree of AKI on subsequent CKD risk and CKD grade over time has been studied in patients with kidney disease, it has not been studied much in the field of lung

transplantation.

Comment 2:

- 2. The authors state that "As no studies have investigated the molecular epidemiology of AKI after lung transplantation, it remains to be seen whether these very high rates of clinical grade concordance reflect a novel molecular mechanism of renal injury." It is true that very few mechanistic studies have been performed in pre-clinical models, but lung ischemia-reperfusion injury is a possible mediator of downstream kidney injury. Lung IRI being one important part of lung transplantation (the other being immunology). One study shows that warm ischemia-reperfusion via pulmonary hilar clamping in rats results in higher plasma pro-inflammatory cytokine levels and worse structural kidney damage on electron microscopy, compared to sham controls (Xiang et al. 2017). To our knowledge, no pre-clinical models of lung transplantation have been used to date to investigate lung-kidney crosstalk after lung transplantation. The lung ischemia-reperfusion injury findings could be discussed in the manuscript. The occurrence of AKI usually within 24hours of lung transplant suggests that tissue injury of the lung (as after lung IRI) plays a significant role.
- Xiang, B.-Q., Gao, H., Luo, Z.-Y., Fang, Z.-X. & Wang, W.-T. [Effect of dexmedetomidine on renal injury induced by lung ischemia/reperfusion in mice]. Zhongguo Ying Yong Sheng Li Xue Za Zhi Zhongguo Yingyong Shenglixue Zazhi Chin J Appl Physiology 33, 380–384 (2017).

Reply 2:

- **You raise an excellent point. We added this to our manuscript.**

Change in the text:

- (Page 14, line 16-19) One study showed that warm ischemic reperfusion with pulmonary portal clamping in rats resulted in higher plasma inflammatory cytokine levels and worsened structural kidney injury on electron microscopy compared to sham controls (35).

Comment 3:

- 3. "Both lungs and kidneys have common exacerbating factors, such as hypoxia (15, 29, 30) and exacerbation of inflammatory cytokines (31, 32), due to lung transplantation. Moreover, it has been reported to exacerbate PGD and AKI (15, 32)."
- It is unclear what is referred to in the second sentence….

Reply 3:

- **This is a great point. Please see above answer.**

Change in the text:

- (Page 14, line 14-16) Common factors post-lung transplantation such as hypoxia (15, 31, 32) and systemic inflammatory cytokine production (33, 34) can exacerbate both lung and kidney dysfunction. Moreover, inflammatory cytokines have been reported to exacerbate PGD and AKI (15, 34).

Comment 4:

- 4. Injurious ventilation can cause AKI, and low tidal volume ventilation improves mortality in ARDS patients. The effect of mechanical ventilation during the lung transplant operation and immediately afterwards could be an important factor in the development of AKI. Mechanical ventilation and its effects on the kidney have been studied in mice.
- These points could be included into the discussion. If there is sufficient data, it could be interesting to include an analysis that correlates mechanical ventilation settings and ventilation time to the occurrence of AKI. This would add a new dimension to the data not previously reported to my knowledge.
- Si, M. K. H. et al. Inhibition of poly (adenosine diphosphate-ribose) polymerase attenuates lung-kidney crosstalk induced by intratracheal lipopolysaccharide instillation in rats. Respir Res 14, 126 (2013).
- Kapoor, K., Singla, E., Sahu, B. & Naura, A. S. PARP inhibitor, olaparib ameliorates acute lung and kidney injury upon intratracheal administration of LPS in mice. Mol Cell Biochem 400, 153–162 (2015).
- Hegeman, M. A. et al. Ventilator-induced endothelial activation and inflammation in the lung and distal organs. Crit Care 13, R182 (2009).
- Amato, M. B. P. et al. Effect of a Protective-Ventilation Strategy on Mortality in the Acute Respiratory Distress Syndrome. New Engl J Medicine 338, 347–354 (1998).

Reply 4:

- **Thank you for pointing this out. We added them in our reference.**

Change in the text:

- (Page 14, line 23 to page 15, line 2) Injurious ventilation can cause AKI, and low-tidal-volume ventilation improves mortality in patients with ARDS. The effect of mechanical ventilation during the lung transplant operation and immediately afterwards could be an important factor in the development of AKI (38-40).

Comment 5:

- 5. Discussion could include what methods could be used or have been used to minimize PGD.

Reply 5:

- **Thank you for bringing this up. In our center, we avoid using CPB during procedure and minimize blood transfusion. We added the sentence referring this in the discussion section.**

Change in the text:

- (Page 15, line 22 to page 16, line 2) Central VA-ECMO has been applied in our institution instead of cardiopulmonary bypass (CPB) to reduce PGD based on evidence that ECMO has been shown to reduce intraoperative blood transfusion volume compared with CPB (47).

Reviewer F

In this study Toyoda et al, retrospectively evaluate their lung transplant database and compare the association between primary graft dysfunction and acute kidney injury after lung transplantation. Although the sample size is relatively small, I find the study clinically relevant and well-written. However, I have critical concerns with regard to how the statistical analysis was carried out, especially the use of Person correlation between categorical variables, there are other tests that would be more appropriate for example Person chi-square, and Cramer's V test.

Comment 1:

- I suggest expanding on how PGD was grated, was the PGD score determined once during the first 72h after transplant? please clarify this in the manuscript.

Reply 1:

- **This is an excellent point. Our definition of PGD is based on ISHLT guideline, which are written in the method section. The lowest PaO2/FiO2 ratio within 72 hours after lung transplantation was used.**

Change in the text:

- (None)

Comment 2:

- In table 1 the term "bilateral" means double lung transplant.? if yes I suggest to the authors exclude the single transplant cases or adjust the analysis accounting for this variable, because the distribution of "bilateral" cases is not equally distributed among the groups.

Reply 2:

- **Thank you so much for bringing this up. That is correct bilateral means double lung transplant. The aim of this study is to assess association of severity of PGD and AKI, and long-term outcome of kidney function.**

Therefore, single lung transplants were included.

Change in the text:

- (None)

Comment 3:

- In table 2, what is the difference between the terms "GPD" and "PGD grade 3"? Here I suggest to the authors include all the PGD scores for performing the comparison (PGD 0, PGD1, PGD2, PGD3)

Reply 3:

- **You bring up a great point. PGD means PGD grades 1 to 3. We clarify it.**

Change in the text:

- (None)

Comment 4:

- With regards to CLAD was there a specific cut-off time for identifying CLAD? or was this any time after the lung transplant? please clarify this in the manuscript.

Reply 4:

- **Thank you for binging this to our attention. Our definition of CLAD is based on ISHLT guideline, which are in the method section. CLAD was confirmed when a persistent decline (≥20%) in the measured forced expiratory volume in 1 second (FEV1) from the post-transplantation baseline that persisted for three months after the first value was taken. We added some words referring them.**

Change in the text:

- (Page 9, line 13-16) The ISHLT defined CLAD as a persistent decline (≥20%) in the measured forced expiratory volume in 1 second (FEV1) from the post-transplantation baseline that persisted for three months after the first value was taken (29). Patients with ≤3 total FEV1 measurements, precluding CLAD diagnosis, were excluded.

Comment 5:

- Figure 4-a, I suggest to the author adjust the linear mixed model by CKD risk factors. Additionally, considering the focus of this study will be relevant to compare AKI and PGD, similar to figure 4-B.

Reply 5:

- **You raise an excellent point. Similar to above answer, the aim of this study is to assess association of severity of PGD and AKI, and long term outcome of kidney function. Therefore single lung transplant were included.**

Therefore, relation between AKI stage and CKD stage is important. As CKD is also known risk factor for late mortality after lung transplant.

Change in the text:

- (None)

Reviewer G

In this retrospective study, Toyoda T et al investigated the association/correlation between PGD and acute kidney injury after lung transplantation. Patients with PGD showed a higher prevalence of severe acute kidney in patients with PGD. Acute Kidney Injury correlated then with GFR at follow-up. The manuscript ist well-written but I have the following remarks:

Comment 1:

- 1) The finding that patients with PGD have a higher prevalence of acute kidney injury after lung transplantation is expected. Negative fluid balance is a primary therapy for PGD, and in my opinion, the main cause of worsening renal function. This aspect should be more stressed in the discussion (more than some non-proven inflammatory mechanisms...).

Reply 1:

- **This is a great point. Please see above answer. There are multiple potential factors causing AKI after lung transplant. We added several new references based on different reviewer suggestions.**

Change in the text:

- (None)

Comment 2:

- 2) The authors should perform a Cox multivariable analysis for mortality. Just a univariable Cox analysis is not enough to prove a statistically and clinically significant association.

Reply 2:

- **Thank you for pointing this out. PGD, AKI, some thrombosis complication, CLAD and post-discharge hemodialysis introduction shown in the univariate Cox analysis are clearly confounding factors. We do not believe that it is worthwhile to perform a multivariate Cox analysis to find independent factors directly related to mortality.**
- **The purpose of this univariate Cox analysis is not to prove an independent association between AKI and mortality, but to show that AKI is a**

complication with a high number of cases and a high HR, making it a potentially attractive therapeutic target.

Change in the text:

- (None)

Comment 3:

- 3) In the methods, the authors should briefly describe how they treat lung-transplanted patients which developed PGD, especially what concerns fluid balance and use of hemodialysis.

Reply 3:

- **Thank you for bringing this up. In our center, once severe PGD was developed, maintain negative fluid balance, low tidal volume ventilation to protect new allograft. Experienced transplant nephrologist evaluated patients and decide indication of hemodialysis. We added the sentence referring the indication for RRT.**

Change in the text:

- (Page 9, line 1 to 2) Indications for RRT were determined by an experienced transplant nephrologist based on eGFR, urine output, and electrolyte abnormalities.

Comment 4:

- 4) In the methods, it is not really clear if the authors used the PGD score at 0, 24, 48 or 72 hours after transplantation. Or if they simply considered the presence of PGD at any time after transplantation.

Reply 4:

- **This is an excellent point. Our definition of PGD is based on ISHLT guideline and within 72 hours. We wrote that the lowest PaO2/FiO2 ratio within 72 hours after lung transplantation was used.**

Change in the text:

- (None)

Reviewer H

This retrospective, single center study analyzed the association between primary graft dysfunction grading and acute kidney injury staging, and the impact of AKI on subsequent changes to chronic kidney disease, in a cohort of 206 patients who underwent lung transplantation between January 2018 and June 2022.

The study showed a significant correlation between PGD grade and AKI stage

($p<0.001$), and a significant correlation between AKI stage and chronic kidney disease at 3, 6, 9 and 12 months after lung transplantation ($p<0.001$).
This study brings interesting and new information about the relationship between PGD and AKI. The strong correlation observed between PGD grade and AKI stage highlights the key importance of a meticulous fluid management in the perioperative period. However, the retrospective design of the study, and the relatively small size of the cohort, limits its generalizability.
Some aspects of this manuscript have, to my point of view, to be modified before considering it for publication, including adding some clinical data. Moreover, care must be taken in interpreting the results.

Major comments

Comment 1:

- Interpretation of the results: even a strong correlation has been observed between PGD grade and AKI stage, a cause-effect relationship between these two events cannot be inferred. A positive effect of PGD grade reduction on renal function can also not be deduced. Some sentences in the manuscript have, to my point of view, to be modified (page 4: lines 13-15; page 6: lines 2-4 ; page 12: lines 10-11 ; page 13: lines 7-8)

Reply 1:

- **We modified those sentence.**

Change in the text:

- (Page 4, line 13-14) Our study provides a clinical basis for speculating the potential causal relationship between PGD and AKI.
- (Page 6, line 2-3) If there is a strong connection between PGD and AKI severity, it would be a good rationale to research the mechanism by which PGD leads AKI.
- (Page 14, line 8-9) Our study provides a clinical basis for speculating the potential causal relationship between PGD and AKI.
- (Page 13, line 7-8) Strategies to reduce PGD grade might be advantageous not only for lowering the stage of AKI but also for maintaining and managing renal function during the chronic phase of the disease. → Deleted

Comment 2:

- The delay of AKI occurrence in the post operative period is not described in the results and could provide interesting information about the relationship between the two events.

- I think there is a mistake in Table 1 or Table 2: numbers and percentages of recipients with RRT before and after surgery are the same.

Reply 2:

- **Thank you so much for bringing this up. We corrected the number.**

Change in the text:

- (None)

Comment 3:

- Why the authors did not exclude recipients who were dependent from hemodialysis before surgery?

Reply 3:

- **You bring up a great point. We included those patients as this is a history of hemodialysis and at the time of lung transplant, GFR was maintained.**

Change in the text:

- (None)

Minor comments

- Methods section:

Comment 4:

- I suggest to add more information about the perioperative management of the recipients in the study center (describe the center protocols for haemodynamic management before and after surgery, for ECMO support and weaning, for immunosuppression regiment).

Reply 4:

- **Thank you for binging this to our attention. We added that information in the method section.**

Change in the text:

- (Page 9, line 18 to page 10, line 17)
- Indication of ECMO

  Patients with respiratory failure were considered for VV-ECMO if they failed to achieve satisfactory gas exchange (PaO2>55 mmHg, Oxygen saturations >88%, pH>7.2, with plateau pressures less than 35) despite lung protective mechanical ventilation and recruitment maneuvers with or without neuromuscular blockade. The decision to cannulate was made by a multidisciplinary ECMO team. All patients were cannulated by thoracic surgeons.

  VV-ECMO management

Patients did not receive continuous anticoagulation unless there was a specific indication, such as Deep Venous Thrombosis (DVT) or Pulmonary Embolism (PE), and there was no monitoring of bleeding parameters such as the Activated Clotting Time or activated Partial Thromboplastin Time, consistent with our prior study. (30) All patients who were not receiving continuous systemic anticoagulation received 5,000 U subcutaneous unfractionated heparin every 8 hours as a prophylaxis dose to prevent deep venous thrombosis. VV-ECMO flow was maintained at a minimum of 3.0-3.5L/min, consistent with our recent reports, to reduce thrombotic complications in the ECMO circuit. (30) Transfusions were administered if any of the following criteria were met: Platelets <50,000/mL, Hemoglobin <7 g/dL, or hemodynamic instability in the setting of active blood loss. Different cannulation strategies (Internal jugular vein – femoral vein cannulation vs ProtekDuo® cannulation (CardiacAssist Inc., Pittsburgh, PA, USA)) were used in patients depending on surgeon preference. The VV-ECMO circuit included a Quadrox-iD adult (7.0) oxygenator (MAQUET Holding B.V. & Co. KG, Germany) and Rotaflow pump (MAQUET Holding B.V. & Co. KG, Germany). All components of the ECMO circuit had a heparin coating except for the cannulas.

Comment 5:

- What was the postoperative delay for AKI recording?

Reply 5:

- **You raise an excellent point. There was no delay in this study.**

Change in the text:

- (None)

- Discussion:

Comment 6:

- page 13, line 5: I suggest to add a reference

Reply 6:

- **We added some of the reference in the discussion based on other reviewer comments.**

Change in the text:

- (None)

Comment 7:

- page 14: I suggest to add in the limitation section, that the retrospective design of the study limits the capacity to rightly classify KDIGO stages, because it does not allow for diureses record

Reply 7:

- **This is a great point. We added those sentence in the discussion.**

Change in the text:

- (Page 16, line 8-13) The relatively small sample size of this study, due to the single institution, limits its generalizability. In addition, accurate diuresis records were not available retrospectively, which limited the ability to correctly classify KDIGO AKI stages. Furthermore, only the GFR category of the KDIGO diagnostic criteria for chronic renal failure was used, because albuminuria was not measured during our standardized follow-up. Therefore, it may not accurately reflect the severity of chronic renal failure.

Comment 8:

- Table 1 and 2: I suggest to add a fourth column entitled « all cohort », in order to improve the readability of the results

Reply 8:

- **Thank you for pointing this out. We added them in table 1 and 2.**

Change in the text:

- (None)

Comment 9:

- Table 1: If possible, I suggest to add more information about donors (PaO2/FiO2 ratio before organ donation, duration of mechanical ventilation, pneumonia…)

Reply 9:

- **Thank you for bringing this up. Unfortunately, we do not have those specific data.**

Change in the text:

- (None)

Comment 10:

- Table 1: I suggest to remove preoperative INR which is not clinically pertinent

Reply 10:

- **This is an excellent point. We removed it.**

Change in the text:

- (None)

Comment 11:

- Table 1: I suggest to specify aetiology of ARDS (COVID-19?)

Reply 11:

- **Thank you so much for bringing this up. We specified it.**

Change in the text:

- (None)

Comment 12:

- Table 2: If possible, I suggest to add more clinical information about haemodynamic status during and after surgery (Cathecholamine use, dosage and duration, vascular filling, nephrotoxic agent administration

Reply 12:

- **Thank you so much for bringing this up. Unfortunately, we do not have those specific data.**

Change in the text:

- (None)

Comment 13:

- Table 2: How do the authors explain the high rate of digital ischemia in the KDIGO 3 group?

Reply 13:

- **You bring up a great point. We think that is due to small number of sample size.**

Change in the text:

- (None)