

Attenuation of Lung Reperfusion Injury by Modified Ventilation and Reperfusion Techniques

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Background: High ventilation and perfusion pressures after lung transplantation may have deleterious effects. We hypothesized that using combined protective approaches for ventilation and perfusion would be optimal for reducing injury and improving function after ischemia-reperfusion.

Methods: Using an isolated, blood-perfused, rabbit lung model, lungs underwent 120 minutes of reperfusion either immediately (Sham) or after 18 hours of cold ischemia (IR). Groups Sham-P and IR-P underwent protective ventilation and reperfusion, and Groups Sham-C and IR-C underwent conventional ventilation and reperfusion. Protective ventilation involved gradually increasing the flow rate during 5 minutes to 1.8 liters/min, and conventional ventilation entailed immediate initiation of flow at 1.8 liters/min. Protective reperfusion involved gradually increasing perfusion during 5 minutes to 60 ml/min, and conventional reperfusion entailed immediate perfusion at 60 ml/min. Two other groups underwent either protective ventilation with conventional perfusion or vice versa. Airway pressure, pulmonary artery pressure, and arterial blood gases were measured throughout reperfusion. Wet/dry weight, highest oxygenation index, and bronchoalveolar lavage (BAL) protein were also measured.

Results: Protective ventilation and perfusion after ischemia (IR-P) resulted in significant improvements in lung function as measured by increased PO_2 and decreased P_{CO_2} , airway pressure, and highest oxygenation index compared with conventional reperfusion (IR-C). Injury was significantly reduced in IR-P lungs as measured by reduced edema (wet/dry weight) and vascular leakage (BAL protein). In most cases, IR-P lungs performed better, with less injury than protective ventilation or perfusion alone.

Conclusions: This protective approach of ventilation and perfusion after ischemia may improve lung function after transplantation, a simple method that could easily be applied clinically. *J Heart Lung Transplant* 2006;25:1467-73. Copyright © 2006 by the International Society for Heart and Lung Transplantation.

Lungs are the most vulnerable solid organs to be transplanted, and even when strict donor selection criteria are used, less than 25% of lungs from multi-organ donors are suitable for transplantation.¹ Despite advancements in organ preservation and perioperative care, reperfusion injury remains a significant cause of early mortality after transplantation.²⁻⁶ In recent years, our understanding of reperfusion injury has increased considerably. It has been shown that cardiac reperfu-

sion injury can be avoided by controlling the composition of the initial reperfusate as well as pressure.⁷ DeCampos et al⁸ demonstrated that rapid initiation of reperfusion leads to pulmonary edema and dysfunction. Progressive reintroduction of blood flow during a 10-minute period was shown to reduce injury and improve function of transplanted lungs,⁹⁻¹¹ presumably because increased shear stress induces endothelial damage.

Although mechanical ventilation is essential for patients undergoing lung transplantation, several studies have shown that mechanical ventilation can produce lung injury de novo.¹² The effect of different modes of ventilation in the early period after transplantation has not been explored clinically. Using a rat lung transplant model, de Perrot et al¹³ demonstrated that ventilation with high tidal volumes and low positive end-expiratory pressure significantly worsened lung function after reperfusion. Our laboratory has shown that hyperoxic ventilation exacerbates lung reperfusion injury, suggesting that the lowest possible tension of inspired oxygen should be used during reperfusion.¹⁴

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These studies suggest that controlled ventilation or perfusion is beneficial after lung ischemia. To date, no study has investigated ventilation and perfusion applied in combination during a post-transplantation reperfusion model. The aim of the current study was to investigate whether a more controlled (protective) method of lung perfusion and ventilation was beneficial. We hypothesized that a combination of controlled ventilation and perfusion after ischemia would result in optimal lung protection.

METHODS

Experimental Protocol

Preliminary studies using the isolated rabbit lung model suggested significant differences in physiologic outcomes between ischemic lungs that had undergone certain modes of protective ventilation and perfusion compared with conventional modes. Hence, we chose the modalities that demonstrated these differences and designed a study to test the current hypothesis. An isolated, blood-perfused rabbit lung system (Kent Scientific, Model TIS3862, Litchfield, CT) was used as previously described.¹⁵ All animals were used under approval from the University of Virginia's Institutional Animal Care and Use Committee. All animals received humane care in compliance with the "Principles of Laboratory Animal Care" formulated by the National Society for Medical Research and the "Guide for the Care and Use of Laboratory Animals" prepared by the Institute of Laboratory Animal Resources (NIH Publication No. 86-23, revised 1985).

Harvest Procedure

New Zealand White rabbits of both sexes (3.0–3.5 kg) were randomly assigned to 6 experimental groups. Each animal was anesthetized with intramuscular ketamine (50 mg/kg) and xylazine (5 mg/kg). Intubation was performed through a tracheostomy, and mechanical ventilation (Kent Scientific, Model RSP1002, Litchfield, CT) was instituted with room air at 25 breaths/min.

A median sternotomy and thymectomy were performed. The superior and inferior vena cavae were encircled with ligatures and the pericardium was opened. Both the pulmonary artery (PA) and aorta were similarly encircled. A purse-string suture was placed in the right ventricle, and intravenous heparin was administered (500 U/kg). After the PA was injected with 30 μ g prostaglandin E₁, the vena cavae were ligated to initiate ischemia. The PA was then cannulated through a right ventriculotomy in the center of the purse-string suture, and the right ventricular and PA ligatures were tied to secure the cannula.

After the left ventricle was vented through a left ventriculotomy and the aorta was ligated, 50 ml/kg cold

(4°C) Viaspan (Belzer UW, Barr Laboratories Inc, Pomona, NY) was infused into the PA at 30 cm H₂O. Topical cooling was achieved with cold saline slush. During the PA flush, the left atrium was cannulated through the left ventriculotomy with an outflow catheter. A purse-string suture was placed to secure this cannula. After completion of the PA flush, the inflow cannula was clamped. The lung-heart block was then excised, and the tracheostomy tube was clamped at end-inspiration.

Reperfusion Procedure

After harvest, the lung-heart block was suspended from a force transducer and ventilation was initiated with 100% fraction of inspired oxygen (Model RSP1002 Kent Scientific). Control groups (Sham) underwent 120 minutes of whole-blood perfusion without ischemic storage. Experimental groups (IR) underwent 18 hours of cold ischemic storage, followed by 120 minutes whole-blood reperfusion. For each group, 1 breath of approximately 30 cm H₂O positive end-expiratory pressure was given once per minute during a 5-minute stabilization period to prevent atelectasis. Lungs were volume ventilated at 1.8 liters/min with 3cm H₂O positive end-expiratory pressure at 20 breaths/min. Sham lungs were flushed with 4°C Viaspan, and topical cooling was achieved as described previously. The average ischemic time for sham lungs was 10 to 15 minutes, and all lungs were at the same temperature (4°C) upon initiation of perfusion.

Lung-heart blocks were connected via the PA and outflow catheters to a venous blood reperfusion circuit. New Zealand White rabbits served as fresh venous blood donors. Blood was circulated through a pediatric oxygenator to deoxygenate the blood and add carbon dioxide. Thus, blood flowing through the PA was essentially physiologic for venous blood (P_O₂ = 60 mm Hg/P_{CO}₂ = 60 mm Hg). This permitted arterial blood gas measurements to be made from the left ventricle that reflected physiologic performance of the lungs. The temperature of the lungs and perfused blood were maintained at 37°C throughout reperfusion.

Study Groups

Six groups consisting of 6 lungs each were studied (summarized in Table 1). Two groups (Sham-P and IR-P) underwent protective reperfusion, which involved gradually increasing ventilatory flow rate from 0.8 liter/min (by 0.2 liter/min each minute) over a 5-minute period upon initiation of reperfusion, while keeping peak inspiratory pressure at 15 cm H₂O or lower, to a maximum rate of 1.8 liters/min. The volume remained unchanged for the remainder of the experiment, where the minute ventilation was 810 ml/min. In addition, blood-perfusion rate was gradually increased upon ini-

Table 1. Summary of Experimental Groups

Group	Ischemia	Ventilation ^a	Perfusion ^b
Sham-C	None	Conventional	Conventional
Sham-P	None	Protective	Protective
IR-C	18 hrs	Conventional	Conventional
IR-P	18 hrs	Protective	Protective
IR-PC	18 hrs	Protective	Conventional
IR-CP	18 hrs	Conventional	Protective

Sham-C, immediate conventional ventilation; Sham-P, immediate protective ventilation; IR-C, conventional ventilation and reperfusion; IR-P, protected ventilation; IR-PC, protective ventilation and conventional perfusion; IR-CP, conventional ventilation and protective perfusion.

^aProtective ventilation involved gradually increasing flow rate to 1.8 liter/min over the initial 5 minutes of reperfusion, whereas conventional ventilation involved initiating reperfusion at the standard ventilation rate of 1.8 liter/minute regardless of peak inspiratory pressure.

^bProtective perfusion involved gradually increasing perfusion rate to 60 ml/minute over the initial 5 minute reperfusion whereas conventional perfusion involved initiating reperfusion at the standard rate of 60ml/min. See Methods for more details.

tiation of reperfusion from 10 ml/min, by 10-ml/min increments, during a 5-minute period to the standard rate of 60 ml/min.

Two groups (Sham-C and IR-C) underwent conventional reperfusion, which involved initiating ventilation at a flow rate of 1.8 liters/min (tidal volume, 12 ml/kg) regardless of peak inspiratory pressure, and perfusion was initiated at the standard rate of 60 ml/min. The remaining 2 groups (IR-CP and IR-PC) underwent reperfusion with either protective ventilation or protective perfusion. Here, Group IR-PC underwent protective ventilation and conventional perfusion whereas Group IR-CP underwent conventional ventilation and protective perfusion. The peak inspiratory pressures during the first 5 minutes of reperfusion for all groups were 15 cm H₂O or less.

Physiologic Variables

Continuous recordings of PA pressure and mean airway pressure were collected by a dynamic data acquisition program (DASYLab, DASYTEC, USA, Bedford, NH). Venous blood samples were collected for blood gas analysis (Bayer 348 Analyzer, Bayer Corp, E Walpole, MA) after 15, 30, 60, and 120 minutes of reperfusion.

Lung Wet/Dry Weight

Lung wet/dry weight was used as an indicator of pulmonary edema. After reperfusion, samples of right lung tissue were blotted, weighed, and desiccated under vacuum at 55°C until a stable weight was achieved. The wet and dry weights were then used to calculate the wet/dry weight ratio.

Bronchoalveolar Lavage Protein Concentration

Bronchoalveolar (BAL) fluid was obtained at the end of reperfusion. Saline (30 ml) was used to intratracheally flush the left lung 3 times. The recovered fluid was then centrifuged (model 5804R, Eppendorf North America, Westbury, NY) for 10 minutes at 4°C. Protein concentration in the supernatant was measured by using the BCA Protein Assay (Pierce Biotechnology, Inc, Rockford, IL) and the BioMate 3 spectrophotometer (Thermo Electron Corp, Waltham, MA).

Statistical Analysis

Statistical analysis was performed using an analysis of variance for each time point and variable. Tukey's multiple comparison analysis was used to determine which groups were significantly different when comparing the Sham and IR Groups. Dunnett's *t*-test using the IR-C Group as a control was used to determine which groups were significantly different for the experimental analysis. A value of *p* ≤ 0.05 was used to indicate a significant difference. Measurements are reported as mean ± standard error of the mean.

RESULTS

Arterial Blood Gas

During reperfusion, arterial Po₂ was reduced at all times after ischemia, with significant reductions at 30 and 60 minutes (IR-C vs Sham-C, *p* < 0.01; Figure 1). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in significantly higher Po₂ compared with IR-C at all times (*p* < 0.02); levels which were comparable

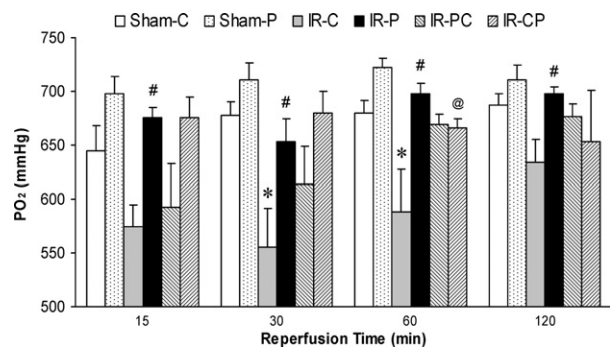


Figure 1. Mean arterial oxygenation (Po₂, mm Hg) during reperfusion. The Po₂ in Group IR-C was reduced at all times, with significant reductions at 30 and 60 minutes (**p* < 0.01 vs Sham-C). Group IR-P had significantly improved Po₂ at all times (#*p* < 0.02 vs IR-C). IR-P usually had higher Po₂ than either protective ventilation (IR-PC) or perfusion (IR-CP) alone (@*p* = 0.049 vs IR-P). Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

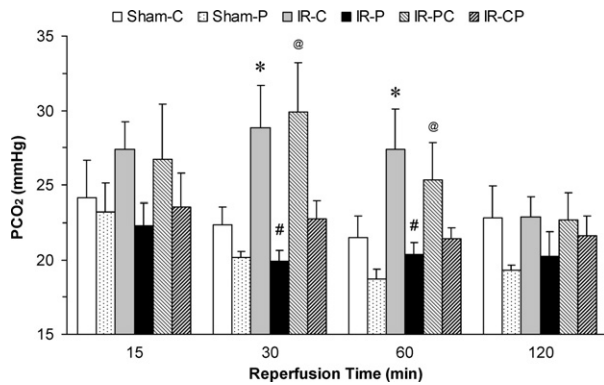


Figure 2. Mean arterial Pco₂ (mm Hg) during reperfusion. The Pco₂ was elevated in Group IR-C at all times after ischemia (**p* < 0.04 vs Sham-C). Group IR-P had reduced Pco₂ at all times, with significant reductions at 30 and 60 minutes (#*p* < 0.02 vs IR-C). IR-P had lower Pco₂ at all times compared with protective ventilation (IR-PC) or perfusion (IR-CP) (@*p* < 0.05 vs IR-P). Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and conventional perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

with the Sham Group (Figure 1). No significant differences were observed between the Sham Groups, although Sham-P consistently had higher Po₂ than Sham-C. When compared with the protective ventilation (IR-PC) or protective perfusion (IR-CP) groups, IR-P usually had higher Po₂, which reached statistical significance at 60 minutes (IR-PC vs IR-P, *p* = 0.049, Figure 1).

During reperfusion, arterial Pco₂ was elevated at all times after ischemia, with significant increases at 30 and 60 minutes (IR-C vs Sham-C, *p* < 0.04) (Figure 2). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in reduced Pco₂ compared with IR-C at all times, which reached significance at 30 and 60 minutes (*p* < 0.02, Figure 2); levels which were comparable with the Sham Group. No significant differences were observed between the Sham Groups, although Sham-P consistently had a lower Pco₂ than Sham-C. When compared with the protective ventilation (IR-PC) or protective perfusion (IR-CP) groups, IR-P had lower Pco₂ at all times and was significant at 30 and 60 minutes (IR-PC vs IR-P, *p* < 0.05, Figure 2).

Mean Airway Pressure

During reperfusion, mean airway pressure was significantly increased at 15, 30, and 60 minutes after ischemia (IR-C vs Sham-C, *p* < 0.05; Figure 3). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in significantly reduced mean airway pressure in IR-P compared with IR-C throughout reperfusion (*p* < 0.01, Figure 3); levels which were compa-

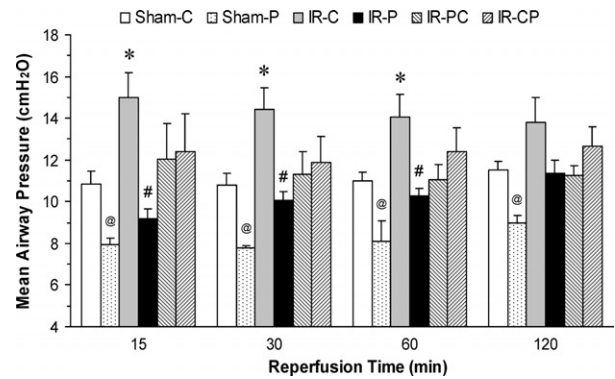


Figure 3. Mean airway pressure (cm H₂O) during reperfusion. Mean airway pressure in Group IR-C was increased after ischemia at all times (**p* < 0.05 vs Sham-C). Group IR-P had improved mean airway pressure at all times, with significant reductions at 15, 30, and 60 minutes (#*p* < 0.01 vs IR-C). Sham-P had significantly lower mean airway pressures than the Sham-C Group throughout reperfusion (@*p* < 0.04 vs Sham-C). Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and conventional perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

table with the Sham Group. When compared with Sham-C, mean airway pressure was significantly reduced in Sham-P at all times (*p* < 0.04). When compared with the protective ventilation (IR-PC) or protective perfusion (IR-CP) groups, IR-P had equal or lower mean airway pressure at all times; however, this did not reach significance.

Mean Pulmonary Artery Pressure

No significant differences in mean PA pressure were observed among all groups throughout reperfusion

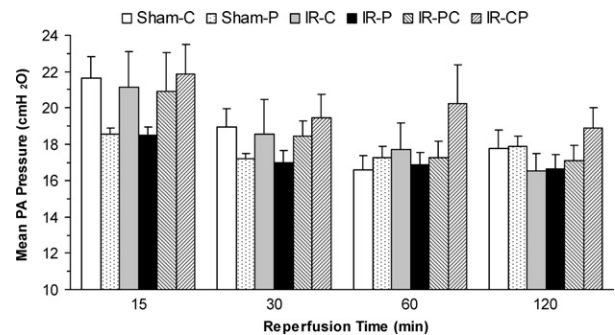


Figure 4. Mean pulmonary artery pressure (cm H₂O) throughout reperfusion did not significantly differ between groups. Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and conventional perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

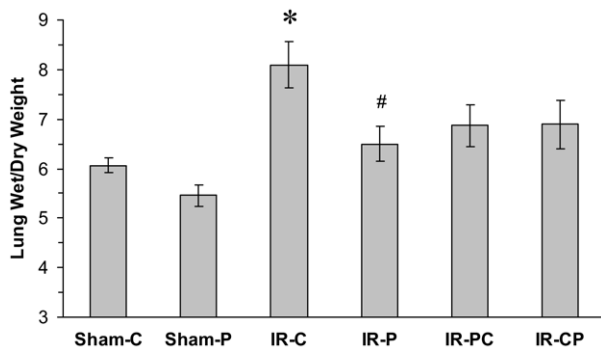


Figure 5. Lung wet/dry weight after ischemia–reperfusion. Ischemia–reperfusion (IR-C) resulted in increased wet/dry weight, as expected ($*p = 0.001$ vs Sham-C). Reperfusion using protective ventilation and perfusion (IR-P) resulted in significantly reduced wet/dry weight ($\#p = 0.035$ vs IR-C). Groups IR-PC and IR-CP also had reduced wet/dry weight, which was not significant. Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

(Figure 4). However, combined protective ventilation and perfusion (Sham-P and IR-P) did result in the lowest PA pressure at the earlier times (15 and 30 minutes).

Lung Wet/Dry Weight

As expected, lung wet/dry weight, an indicator of edema, was significantly increased after ischemia–reperfusion (IR-C vs Sham-C, $p = 0.001$; Figure 5). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in a significantly reduced wet/dry weight compared with IR-C ($p = 0.035$; Figure 5). The IR-PC and IR-CP Groups also had reduced wet/dry weight; however, this was not significant compared with IR-C. The difference between the Sham Groups was not significant.

Bronchoalveolar Lavage Protein Concentration

As expected, BAL protein concentration, an indicator of vascular leakage, was significantly increased after ischemia–reperfusion (IR-C vs Sham-C, $p = 0.001$; Figure 6). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in significantly reduced BAL protein compared with IR-C ($p = 0.05$; Figure 6). The IR-PC and IR-CP Groups also had reduced BAL protein; however, this was not significant compared with IR-C. The difference between the Sham Groups was not significant.

Highest Oxygenation Index

After lung transplantation, an elevation of the highest oxygenation index (HOI), calculated as [(mean airway

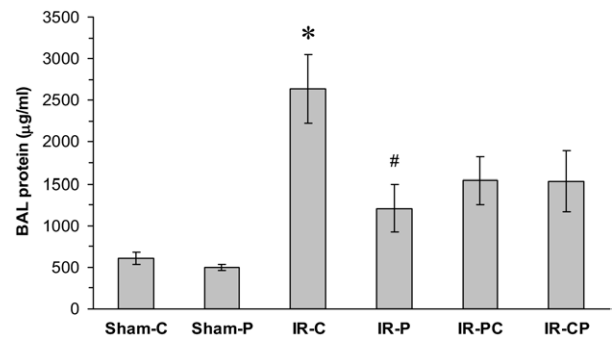


Figure 6. Bronchoalveolar lavage (BAL) protein concentration ($\mu\text{g/ml}$) after ischemia–reperfusion. Ischemia–reperfusion (IR-C) resulted in increased BAL protein, as expected ($*p = 0.001$ vs Sham-C). Reperfusion using protective ventilation and perfusion (IR-P) resulted in significantly reduced BAL protein ($\#p = 0.05$ vs IR-C). Groups IR-PC and IR-CP also had reduced BAL protein, which was not significant. Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

pressure \times percent of inspired oxygen)/partial pressure of arterial oxygen], can be an early predictor of severe respiratory failure requiring acute intervention.¹⁶ In support of this, the HOI after ischemia–reperfusion was significantly elevated (IR-C vs Sham-C, $p = 0.003$; Figure 7). Reperfusion using combined protective ventilation and perfusion (IR-P) resulted in significantly reduced HOI vs the IR-C Group ($p = 0.0016$; Figure 7). Protec-

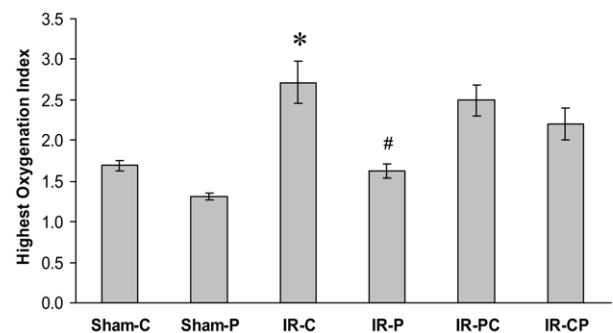


Figure 7. Highest oxygenation index (HOI) after ischemia–reperfusion. Ischemia–reperfusion (IR-C) resulted in increased HOI, indicative of respiratory failure ($*p = 0.003$ vs Sham-C). Reperfusion using protective ventilation and perfusion (IR-P) resulted in significantly reduced HOI ($\#p = 0.0016$ vs IR-C). Groups IR-PC and IR-CP had insignificant reductions in HOI. Range bars show the standard error of the mean. Sham-C, non-ischemic, conventional ventilation and perfusion; Sham-P, non-ischemic, protective ventilation and perfusion; IR-C, ischemic, conventional ventilation and perfusion; IR-P, ischemic, protective ventilation and perfusion; IR-PC, ischemic, protective ventilation and conventional perfusion; IR-CP, ischemic, conventional ventilation and protective perfusion.

tive ventilation (IR-PC) or protective perfusion (IR-CP) alone did not result in significant reductions in HOI. Although HOI was lower in Sham-P vs Sham-C, this was not significant ($p = 0.39$).

DISCUSSION

This study demonstrates that the ventilation and perfusion strategies used during acute reperfusion can have a major effect on subsequent lung function. We have shown that combined protective ventilation and perfusion strategies can significantly diminish ischemia-reperfusion injury. These strategies could easily be applied clinically to improve outcomes in transplant recipients. One example of another method that could be easily applied in the clinical setting was demonstrated by Wittwer et al,¹⁷ where it was shown that the endobronchial application of iloprost to donor lungs significantly improved function after ischemia. Another study by Markart et al¹⁸ showed that a brief episode of ischemic preconditioning before ischemia may also provide protection.

Several studies have shown that reduced reperfusion pressure is beneficial after lung transplantation.^{7,19,20} Although it remains uncertain why higher reperfusion pressures cause increased damage after ischemia, considerable evidence suggests that it is primarily due to endothelial cell injury²¹⁻²³ causing attachment of leukocytes, which in turn leads to further endothelial cell damage.^{21,24-26} The ideal length of controlled reperfusion, as well as optimal pressure, remains uncertain, but these data suggest that the first few minutes of reperfusion are critical and that by using a more controlled mode of reperfusion, subsequent tissue edema, leukocyte adherence, and cellular injury can be minimized.²⁰ The same can be said for high ventilation pressure. One study demonstrated that transplanted lungs ventilated with conventional mechanical ventilation had a significant increase in elastase with higher cytokine concentrations and greater pathologic changes compared with those with minimal stress ventilation.¹⁵

Our laboratory has historically used Viaspan preservation solution in the isolated rabbit lung model. We realize that Perfadex (Vitrolife, Inc., Englewood, CO, USA) is more commonly being used today clinically. Because all lungs in this study were flushed with the same solution (Viaspan), direct comparisons between these groups hold true. The potassium concentrations do differ between these 2 solutions (Perfadex has 6 mmol/liter and Viaspan has 125 mmol/liter), and thus, we cannot eliminate the possibility that the use of Perfadex could lead to different results. However, we now use Perfadex in the isolated rabbit lung model and find that it results in similar lung physiology as observed in the control groups of the present study, and thus we

feel that our results and conclusions would differ little if Perfadex were used.

To our knowledge, this study is the first to investigate the combination of minimal stress ventilation and controlled reperfusion on lung function after ischemia. According to the physiologic variables studied, the ischemic lungs that underwent protective ventilation and reperfusion (IR-P) performed better than the ischemic lungs that underwent conventional protocols (IR-C), as demonstrated by improved PO_2 , PCO_2 , mean airway pressure, and HOI. Although evaluation of lung histology revealed no differences between the ischemic groups (data not shown), evidence of reduced injury in ischemic lungs that underwent combined protective ventilation and reperfusion was demonstrated by decreased wet/dry weight and BAL protein. The isolated rabbit lung model used in this study results in a relatively mild injury, which is probably why PCO_2 levels did not exceed 30 mm Hg. This is also probably why PA pressures were not significantly elevated by ischemia. However, as shown in Figure 4, PA pressures were consistently lower in the protective groups (Sham-P and IR-P) compared with the conventional groups (Sham-C and IR-C) at 15 and 30 minutes reperfusion.

Although each of the separate protective approaches for ventilation (IR-PC) or perfusion (IR-CP) often provided some protection to the lung after ischemia, in most cases this was less protective than combined protective ventilation and perfusion (IR-P). The most significant differences noted were that wet/dry weight, BAL protein, and HOI were not significantly improved by the separate protective approaches (IR-PC or IR-CP), whereas combined protective ventilation and perfusion (IR-P) did significantly improve these variables. These results suggest that controlled ventilation and perfusion can substantially attenuate injury and improve function after lung ischemia-reperfusion and that the combination of protective ventilation and perfusion is optimal.

CONCLUSION

This study shows that combined protective ventilation and perfusion after ischemia decreases injury, improves function, and is superior to protective ventilation or protective perfusion alone. This is a relatively simple strategy that does not require significant alterations to current surgical techniques used in the operating room. The number of patients on the waiting list for lung transplantation is constantly increasing, but less than 25% of donor lungs are currently used. Hence, the development of new strategies to improve the quality of lungs could have a tremendous impact on the number of transplants performed. The approach described in this study may not only improve post-operative recovery but

may also allow the procurement of less than perfect lungs for transplantation, thus increasing the donor pool.

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