Lung Injury Rat Model of Cardiopulmonary Bypass with Excellent Survival

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1. Introduction

Acute lung injury is a serious complication of major trauma occurring as a direct consequence of trauma to the lung or, more commonly, arising indirectly as a consequence of trauma elsewhere to the body. Acute respiratory distress syndrome (ARDS) can be associated with various disorders [1-3]. ARDS is a devastating disease, characterized by an edematous reaction in the lungs, leading to defective gas exchange. ARDS frequently develops after trauma, infections or a sequence of such events [4]. Endotoxaemia, or sepsis, is a heterogeneous class of syndromes caused by a systemic inflammatory response to infection. This condition may lead to severe shock and multiple organ failure and is one of the leading causes of death in critically ill patients [5]. Acute lung injury has long been observed in animals and patients with sepsis or endotoxaemia [6,7].

Cardiopulmonary bypass (CPB) is an essential component of conventional cardiac surgery and may be used in many other surgical procedures [8]. Despite excellent improvements, the manifestations of postperfusion syndromes such as clinical signs of pulmonary and renal dysfunction, neurological and gastrointestinal injury, coagulation disorders, and hemolysis, increase in interstitial fluid, and susceptibility to infections have been repeatedly reported [9]. The underlying mechanisms are probably multifactorial, including surgical trauma, anaesthetic effects and muscle paralysis, increase in capillary permeability, and impacts of the CPB apparatus [10].

The purposes of this study were to develop a membrane oxygenator that can improves gas exchange efficiency, and to prove the performance of the membrane oxygenator through animal model test by developing animal model with acute respiratory failure. Vibration method was used to improve the gas exchange rate of the membrane oxygenator. To develop the animal model of inflammatory respiratory failure; LPS (lipopolysacch-aride) was administered directly into the lungs via intratracheal route.

2. Materials and Methods

Male Sprague-Dawley, weighing 450-550 g, were purchased from the Samtako Biokorea. Animals were housed 4 per cage at constant room temperature (22-27°C) with a 12 hours light/12 hours dark cycle. The animals were allowed free access to a standard rat pellet diet and tap water. All procedures in this study were performed in accordance with the National Institutes of Health Guide for the Care.

Animals were randomly divided into four groups. The number of rats in each group was initially more than 9. If death occurred during the observation period, additional experiments were performed to make up the number of rats in each group to 10. In the control group, rats were given an intratrachea of phosphate buffered saline (PBS). Rats in the LPS (Salmonella entritidies; Sigma, St Louis, MO, USA) group followed by i.t. LPS. A number of animals died during the observation period. The survivors were killed with an overdose pentobarbital (100 mg/kg, intravenously (i.v.)). After killing, the arterial blood was taken immediately. The arterial blood was centrifuged at 1,500 rpm for 5 min and supernatants used for blood gas and ion measurements.

The minute CPB circuit comprised a venous reservoir, a specially designed membrane oxygenator, a roller pump, and sterile tubing with an inner diameter of 4 mm for the venous line and of 1.6 mm for the arterial line. The blood was drained from the right atrium via a jugular vein catheter to a 10 ml sterile open reservoir by gravity and siphon. The relatively large venous tube and the high siphon level overcame the resistance of drain flow and induced to a satisfaction venous return. The membrane oxygenator was specially designed with a surface area for gas exchange of 0.05 m^2 and was made of the fiber commonly used in clinical devices. The total assembly dynamic priming volume approximated 4 ml. A rolling pump was used to drive the blood through silicone arterial inflow tubing (1.6 mm) and then to return to the right carotid artery. Body central temperature was monitored with a rectal probe and kept at 36.5–38.3°C by a heat lamp placed above the animal and the CPB equipment.

3. Results

Chest X-ray revealed extensive diffuse infiltration in bilateral lung fields. This result proofed that LPS injection induced ARDS rat. Also, we performed a necropsy on the ARDS rat. Disseminated abscess were found over the area of ARDS rat's lung (Fig. 1).

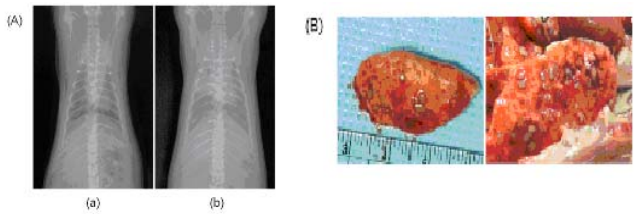

Fig. 1 Chest radiography and necropsy of acute respiratory distress syndrome (ARDS) rat. (A) Chest radiography of rat. (a) Control, (b) LPS induced ARDS model (42 hours), (B) Necopsy of rat: Diffuse abscess over the area of lung

The gas transfer rate is dependent on the resistance of the blood boundary layer, the membrane, and gas boundary layer. Gas transfer depends mainly on the resistance of the blood boundary layer. Therefore, reducing this resistance is important for increasing the gas transfer rate. The maximum

oxygen transfer and CO_2 removal rates occurred at the frequency band of 7 Hz (Fig. 2).

All experiments progressed without incident and all animals survived from the operative process. Neither thrombosis nor haemorrhage occurred in rats with or without CPB. There were two delayed deaths in the CPB group. They presented with respiratory stress and failed to resume normal eating and drinking patterns.

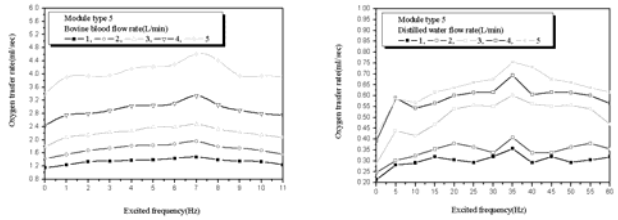

Fig. 2 Relationships between the oxygen transfer rate with the various excited frequencies.

Fig. 3 shows the physiologic data from the CPB and Sham rats over the study period. The results of blood gas analysis at different times from both groups all fell within our acceptable range and represented well gas exchange effects of the minioxygenator. In the CPB group, CPB and post-CPB PaO_2 values were significantly lower than baseline, whereas $PaCO_2$ remained stable. In the Sham group, there were no changes in PaO_2 and $PaCO_2$. HCT had a significant decrease in the CPB group during the perfusion process but had no change in the Sham group. This hemodilution followed by a return to nearly baseline values was due to the nature of CPB. As time passed, PaO_2 returned gradually, autonomous respiration restarted, and the animals recovered.

Fig. 4 shows that gas exchange efficiency could be increased using membrane oxygenator and in the same time the use of antioxidants could prolong the life of the subject with acute respiratory failure up to 5 days.

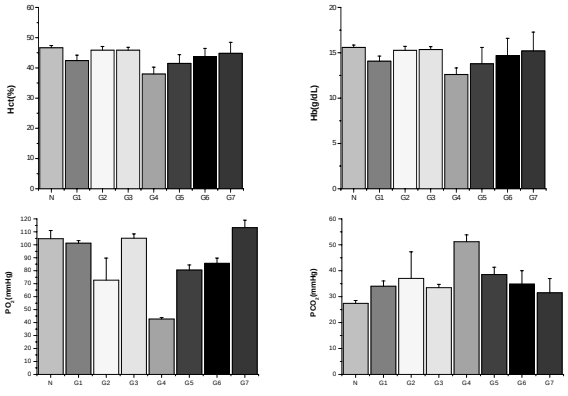

Fig. 3 The results of blood gas analysis in the rats. N : Normal group, G1 : Phosphate buffer saline group, G2 : Taurine+LPS group, G3 : Taurine+LPS+DEX group, G4 LPS group, G5 : Oxygenator treat group(0Hz), G6 : Oxygenator treat group(7Hz), G7 : Taurine+LPS+DEX +Oxygenator treat group

4. Discussion

In this study gas exchange in membrane oxygenator using vibration method was improved, and maximum gas exchange efficiency was attributed at 7Hz. Chest X-ray result of the rat on the third day after LPS administration indicated to the respiratory failure was occurred by the expansion of lung volume and all rat died on 5th day showing ARDS. Lung inflammation was confirmed by histopathological study where defuse infiltration of reactive cells was present. Experimental results showed that gas exchange efficiency could be increased using membrane oxygenator and in the same time the use of antioxidants could prolong the life of the subject with acute respiratory failure up to 5 days.

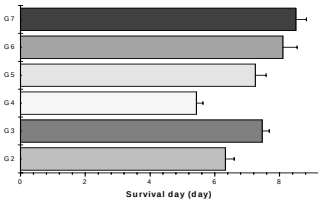

Fig. 4 Survival day in the rats. N : Normal group, G1 : Phosphate buffer saline group, G2 : Taurine+LPS group, G3 : Taurine+LPS+DEX group, G4 LPS group, G5 : Oxygenator treat group(0Hz), G6 : Oxygenator treat group(7Hz), G7 : Taurine+LPS+DEX +Oxygenator treat group

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References

1. Kim, G. B., Hong, C. U., Kwon, T. K., “Vibration characteristics of piezoelectric lead zirconate titanate by fluid flow in intravascular oxygenator”, Jpn J Appl Phys, **45(4B)**, 3811-3817. 2006.
2. Kim, G. B. , Hong, C. U., Kwon, T. K., “Design of an intravenous oxygenstor”, J Artif Organs, **9**, 34-41, 2006.
3. Kim, G. B., Kim, S. J., Hong, C. U., Kwon, T. K., Kim, N. G., “Enhancement of oxygen transfer in hollow fiber membrane by the vibration method”, Korean J Chem Eng, **22(4)**, 521-527, 2005.
4. Hasleton, P. S., “Adult respiratory distress syndrome: A review. Histopathology”, **7**, 307-332, 1983.
5. Angus, D. C., Linde-Zwirble, W. T., Lidicker, J., Clermont, G., Carcillo, J., Pinsky, M. R., “Epidemiology of severe sepsis in the United States: Analysis of incidence, outcome, and associated costs of care”, Crit Care Med, **29**, 1303-1310, 2001.
6. Brigham, K. L., Meyrick, B., “Endotoxin and lung injury. Am Rev Respir Dis”, **133**, 913-927, 1986.
7. Cope, D. K., Grimbert, F., Downey, J. M., Taylor, A. E., “Pulmonary capillary pressure: A review”, Crit Care Med, **20**, 1043-1056, 1992;.
8. Palanzo, D. A., “Perfusion safety: past, present, and future”, J Cardiothorac Vasc Anesth, **11**, 383, 1997.
9. Paparella, D., Yau, T. M., Young, E., “Cardiopulmonary bypass induced inflammation: pathophysiology and treatment”, An update Eur J Cardiothorac Surg, **21**, 232, 2002.
10. Levy, J. H., Tanaka, K. A., “Inflammatory response to cardiopulmonary bypass”, Ann Thorac Surg, **75**, S715, 2003.