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Effect of sevoflurane on tissue permeability of lung ischemia–reperfusion injury in rats

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ABSTRACT

Objective: To investigate the effect of sevoflurane on tissue permeability of lung ischemia–reperfusion injury (LIRI) in rats. **Methods:** A total of 45 wistar rats were randomly divided into 3 groups I, II, III. Modified Eppinger method was adopted to establish the rat lung ischemia–reperfusion injury model. Group I served as the control group, group II as ischemia reperfusion group, group III as sevoflurane ischemia–reperfusion group. Blood gas index, lung permeability index (LPI) change, lung tissue pathology change and lung water content were observed and compared between groups of rats at different time points. **Results:** During ischemia reperfusion, all rats kept balance of the MAP during different time points, SPO₂ of group II and III decreased significantly than I group ($P < 0.05$); after reperfusion lung permeability index in Group II and III was higher than the control group significantly ($P < 0.05$), 120 min after reperfusion LPI change and injury of group III was significantly lower than II group ($P < 0.05$); interstitial and alveolar cavity effusion in of group III were lower than that of group II. **Conclusions:** Sevoflurane pretreatment can reduce the lung tissue permeability, and LIRI plays a protective role in LIRI.

1. Introduction

Lung ischemia–reperfusion injury (LIRI) is clinical common in lung transplantation and extracorporeal circulation operation^[1–3]. LIRI can lead to lung dysfunction, and is the leading cause of death after lung transplantation^[4]. Study has showed that^[5], enflurane and sevoflurane drugs play an important protective role in viscera reperfusion. Other studies have suggested^[6] that sevoflurane pretreatment can significantly reduce the lung edema and inflammation induced by tissue endotoxin, and sevoflurane can protect on LIRI. This study aimed to observe the influence of sevoflurane on LIRI rat lung tissue permeability and the

mechanism. We established the rat lung LIRI model, used sevoflurane pretreatment, observe its tissue permeability and the pathological changes, to provide the theory basis for clinical prevention and treatment of LIRI medication.

2. Materials and methods

2.1. Experimental animals

A total of 45 male, clean level, Wistar rats were selected, aged 8 to 10 weeks, weighting (251.3±42.5) g. They were provided by the laboratory animal center, class II, and had free food and water. The experimental process strictly followed “regulations on the administration of experimental animals”.

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2.2. Instrument and reagent

SLIRIecust730 pressure monitor (Siemens Germany); Detax gas monitor (Finland); NOVAbiomedical blood gas analyzer (United States); Optical microscope (BH-2) (Japan). 20% urethane (our center reagent); Sevoflurane (Baxter company, USA).

2.3. Model establishing

Eppinger method was used to establish LIRI model in rats *in vivo*[7]. All rats had anesthesia with intraperitoneal injection of 20% urethane in supine position. The right lower limb groin skin had incision under local anesthesia, and right femoral vein was exposed. Sodium chloride was injected by infusion pump. After neck midline incision, trachea and left carotid artery was separated. Tracheotomy was performed for intubation, and animal breathing machine was used for mechanical ventilation. The parameters were adjusted according to the data. PaCO₂ was maintained at 35 to 45 mmHg. Left carotid artery blood pressure was monitored continuously and blood samples were collected at each time point. Vecuronium bromide amine was infused continuously to maintain anesthesia, and sevoflurane inhalation concentration was adjusted within 1 MAC (2.2%) in intraoperative sevoflurane preconditioning group. Thirty minutes after mechanical ventilation, the bottom left pulmonary ligament was cut off from the fifth rib in left side. Pulmonary hilar was exposed, and 50 U heparin was injected intravenously. Left pulmonary hilar was closed, and lung surface was rinsed with saline. Thoracic cavity was closed by suture, after 45 min blood flow was open.

2.4. Animal groups

A total of 45 Wistar rats were randomly divided into group I, II, III, with 15 in each. group I served as the control group, without block after left pulmonary hilar opening; group II as ischemia-reperfusion group, with perfusion 45 min after left pulmonary hilar blocking; group III as sevoflurane ischemia-reperfusion group, with sevoflurane inhalation for 30 min and with perfusion 45 min after left

pulmonary hilar blocking. There were three time points in each group, ischemia blocking for 45 min, 1 h and 2 h reperfusion. Five rats were sacrificed at each time point.

2.5. Indexes observation

0.5 mL of blood was collected for blood gas analysis at three time points, respectively. Lung wet dry weight ratio was detected[8]. Lung tissue was weighted as wet weight (W), then they were placed in the oven drying to record dry weight (D), to calculate the wet dry /weight ratio (W/D). Lung permeability index (LPI) value was obtained by using the Bradford method[9]. After perfusion, rats were sacrificed. 1 mm left lung tissue was selected, fixed, HE stained to observe changes in lung tissue;

2.6. Statistical analysis

SPSS12.0 statistics software was used to analyze data, measurement data were expressed as mean±SD, $P < 0.05$ was considered as significant difference.

3. Results

3.1. PaCO₂ change

There was no statistical difference in PaCO₂ basic value between three groups ($P > 0.05$); PaCO₂ decreased significantly in II, III group 60, 120 min after reperfusion ($P < 0.05$), and compared with group I the differences was statistically significant ($P < 0.05$); decrease in Group II at each time point was more significant than that of group III, but there was no statistically significant difference ($P > 0.05$) (Table 1).

3.2. Lung W/D detection

There was no statistical difference of rats W/D between three groups ($P > 0.05$). 60, 120 min after reperfusion lung W/D in group I had no significant change compared with that at ischemia time ($P > 0.05$); 60, 120 min after reperfusion W/D of Group II and III was significantly increased compared with

Table 1

PaCO₂ change of three groups at each time point (mmHg).

Group	Basic value	45 min after ischemia	1 h after reperfusion	2 h after reperfusion
Group I	101.06±22.34	103.11±22.05	95.23±13.14	98.61±10.23
Group II	101.34±26.65	99.93±21.34	48.59±7.61*	45.86±7.09*
Group III	103.21±24.31	104.89±25.21	61.07±5.78*	57.27±3.38*

*Compared with Group I $P < 0.05$.

Table 2

Lung W/D.

Group	Basic value	45 min after ischemia	1 h after reperfusion	2 h after reperfusion
Group I	4.41±0.36	4.20±0.20	4.31±0.24	4.31±0.24
Group II	4.34±0.26	5.22±0.11	5.67±0.42*	5.67±0.42*
Group III	4.20±0.21	4.88±0.08	5.29±0.12* [△]	5.29±0.12* [△]

*Compared with Group I $P<0.05$; [△]Compared with Group II $P<0.05$.

that of group I ($P<0.05$). 60, 120 min after reperfusion W/D of Group II was significantly increased compared with that of group III ($P<0.05$) (Table 2).

3.3. LPI

120 min after reperfusion LPI of group II [(2.73±0.09)%] and III [(1.56±0.18)%] were significantly higher than group I [(0.73±0.13)%] ($P<0.05$), and the difference was significant different between group III and group II ($P<0.05$).

3.4. Pathological observation

Group I showed normal lung tissue structure, a small amount of accidental effusion in alveolar space, capillary without congestion; group II showed lung capillary expansion congestion, edema of broadening alveolar interval, infiltrated interstitial inflammatory cell, exudated red blood cells and inflammatory cells from alveolar lumen. Lung tissue structural damage was serious reperfusion after 120 min; group III showed that lung tissue injury was reduced significantly than group II, interstitial and alveolar cavity effusion reduced as shown in Figure 1.



Figure 1. Pathological observation (HE×400).

4. Discussion

LIRI tends to occur in cardiopulmonary resuscitation, extracorporeal circulation, lung transplantation, and other circumstances. The pathogenesis is not yet clear. Some scholars reported that it is related with many factors, and acute LIRI is important factors affecting postoperative recovery^[10–13]. After rebuilding blood supply, lung tissue

damage is aggravating^[14]. Therefore, the effective prevention and control measure is hot spot of clinical research. Studies have shown that^[15–17], pretreatment with drugs can significantly reduce the degree of lung injury. This study Eppinger method is adopted to establish the LIRI rats model in vivo. Pathological changes of lung tissue were obvious, and lung capillary hyperemia and expansion were obvious after reperfusion. Alveolar cavities erythrocyte effused in great quantities, followed with interstitial edema and inflammatory cells infiltration, showing LIRI model was successfully established.

Sevoflurane inhalation anesthesia is common. Studies have shown that^[18–20], it can improve the lung injury caused by endotoxin, alleviate endotoxin induced lung edema and inflammatory cell infiltration, without influence on regular inhalation of lung tissue. In this study, PaCO₂ of group II decreased significantly after infusion. It may be related to increasing capillary permeability after lung ischemia reperfusion, which lead to pulmonary interstitial edema, blood flow to the ventilation/dysfunction ratio^[21–23]; In Group III after sevoflurane pretreatment, PaCO₂ was increased slightly, showing that LIRI has certain protective effect. LPI is the main indicators for lung tissue protein permeability. In this study, LPI of group II after reperfusion was increased most significantly. LPI in group III decreased significantly pretreatment than II group after sevoflurane inhalation. It also shows that sevoflurane pretreatment can decrease the LPI rising degree. The lung D/W ratio in group III was lower than that in group II, the result changes were positively related to the level of the LPI changes, showing sevoflurane pretreatment can reduce pulmonary vascular permeability after reperfusion. The lung tissue can play a protective role. HE staining showed that, tissue damage degree in the group III was significantly lighter than group II, which confirmed protection effect of sevoflurane pretreatment on LIRI.

According to the results of this study, pretreatment with sevoflurane can improve tissue permeability of lung ischemia–reperfusion injury in rats, to protective LIRI lung tissue.

Conflict of interest statement

We declare that we have no conflict of interest.

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