doi: 10.4149/gpb_2022058

Protective effects of NF-κB inhibitor and continuous perfusion of pulmonary arteries on pulmonary injury in piglet models of deep hypothermia low flow

Yewei Xie, Rufang Zhang and Jia Li

Department of Pediatric Cardiothoracic Surgery, Shanghai Children's Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai, China

Abstract. Deep hypothermia with low flow perfusion (DHLF) is a common cardiopulmonary bypass (CPB) technique. The associated lung ischemia/reperfusion injury is a major cause of postoperative morbidity and mortality in patients undergoing DHLP; we aimed to investigate the effects of nuclear factor-κB (NF-κB) inhibitor pyrrolidine dithiocarbamate (PDTC) with continuous perfusion of pulmonary arteries (CPP) on DHLF-induced lung injury and the related molecular mechanisms. Twenty-four piglets were randomly divided into the DHLF (control), CPP (with DHLF), or CPP+PDTC (intravenous PDTC before CPP with DHLF) groups. Lung injury was evaluated by respiratory function measurement, lung immunohistochemistry, and serum levels of TNF, IL-8, IL-6, and NF-κB before CPB, at CPB completion, and at 1 h post-CPB. Western blot was used to detect NF-κB protein expression in lung tissues. After CPB, decreased parcial pressure of oxygen (PaO_2) and increased parcial pressure of carbon dioxide ($PaCO_2$) and serum levels of TNF, IL-8, IL-6, and NF-κB were observed in the DHLF group. Both CPP and CPP+PDTC groups showed better indices of lung function, decreased levels of TNF, IL-8, and IL-6, and less severe pulmonary edemas and injuries. PDTC with CPP further improved pulmonary function and mitigated pulmonary injury than did CPP alone. PDTC with CPP better attenuates DHLF-induced lung injury than does CPP alone.

Key words: Deep hypothermia low flow — Continuous perfusion of pulmonary arteries — Cardiopulmonary bypass — NF-κB inhibitor — Inflammation

Abbreviations: ABG, arterial blood gas; CPB, cardiopulmonary bypass; CPP, continuous perfusion of pulmonary arteries; DHLF, deep hypothermia with low-flow perfusion; ELISA, enzyme-linked immunosorbent assay; IKK, IκB kinase; IκBα, phospho-inhibitor of κBα; I/R, ischemia/reperfusion; PDTC, pyrrolidine dithiocarbamate; PMNs, polymorphonuclear leukocytes; T0, end of CPB; T1, 1 hour of post-CPB; W/D, wet-to-dry weight.

Introduction

Deep hypothermia with low flow perfusion (DHLF) is a widely applied cardiopulmonary bypass (CPB) technique in the surgical treatment of congenital heart diseases in infants and neonates (Harten et al. 2012). DHLF provides oxygen supply and blood flow to key organs, and enhances the hypoxia tolerance of cells by decreasing the metabolic rate, thereby protecting organs and suppressing hypoxia-induced cell apoptosis (Yang et al. 2020). The incidence

Correspondence to: Yewei Xie, Department of Pediatric Cardiothoracic Surgery, Shanghai Children's Hospital, School of Medicine, Shanghai Jiao Tong University, No. 355, Luding Road, Putuo district, Shanghai 200062, China
E-mail: xieye_wei@163.com

of lung injury is reportedly extremely high in neonates or infants undergoing DHLF, and it has become a major cause of postoperative morbidity and mortality in these patients (Su et al. 2003; Gaynor et al. 2014).

Continuous perfusion of pulmonary arteries (CPP) protects against pulmonary ischemia/reperfusion (I/R) injury in piglet models of DHLF (Yewei et al. 2013). Pulmonary artery perfusion with oxygenated blood during CPB achieves a significant improvement in postoperative oxygenation in patients undergoing cardiac surgery (Karacalilar et al. 2020). However, because of the limited number of participants, meta-analyses fail to find conclusive evidence on whether CPP is beneficial or harmful during CPB (Buggeskov et al. 2018). Therefore, further studies are required to determine the effects of CPP during CPB.

The nuclear factor-κB (NF-κB) transcription factor is widely accepted as a master regulator of inflammation and immunity, playing a critical role in inflammatory diseases (Mitchell and Carmody 2018). The NF-κB family consists of five members – p50, p52, p65, RelB, and c-Rel – which interact to form isodimers or heterodimers (Napetschnig and Wu 2013). Activation of NF-κB signaling involves both canonical and non-canonical pathways (Hoesel and Schmid 2013). The main mechanism of canonical NF-κB activation is the degradation of phospho-inhibitor of κBα (IκBα). During this process, IκB kinase (IKK) phosphorylates IκBα, contributes to ubiquitination, and finally results in the release and translocation of NF-κB, thereby regulating downstream gene transcription (Peng et al. 2020). The non-canonical NF-κB pathway depends on the activation of the NF-κB2 (P100)/RelB complex, which responds with specificity to a sunset of receptors (B-cell-activating factor (BAFF), BAFF receptor, and receptor activator of NF-κB). NF-κB-inducing kinase (NIK) is a core component of the noncanonical pathway. IKKα is activated by NIK, resulting in p100 phosphorylation. P100 is then processed into its active form, p52, and transported into the nucleus (Peng et al. 2020). Previous studies have reported that NF-κB nuclear translocation induced by ischemia activates pro-inflammatory genes and triggers the release of a large quantity of inflammatory factors, thus exacerbating organ injury (Wang et al. 2018b; Qiu et al. 2019). Emerging evidence has demonstrated that NF-κB plays a key role in the pathogenesis of lung I/R injury (Matsuda et al. 2014; Ye et al. 2018). Moreover, a recent study showed that the NF-κB inhibitor pyrrolidine dithiocarbamate (PDTC) mitigates lung injury due to decompression by suppressing the NF-κB pathway (Wang et al. 2018a). Furthermore, NF-κB reportedly mediates the lung-protective effect of pulmonary artery perfusion with a urinary trypsin inhibitor in an infant piglet model of DHLF (Li et al. 2014). Nonetheless, the effects of NF-κB inhibitors on DHLF-induced lung injury require further investigation.

The current study was designed to study the effects of CPP with the NF-κB inhibitor PDTC on DHLF-induced lung injury in piglet models and to explore the underlying molecular mechanisms. Our study enhances our ability to improve lung-protective strategies against DHLF-induced lung injury in clinical practice.

Materials and Methods

Animals and experimental groups

A total of 24 healthy piglets (2-week-old; 9.5 ± 1.6 kg) were used in this study. Because there was no significant difference in the incidence of congenital heart disease between males and females in the birth population, all experimental animals were randomly selected, and both sexes were acceptable. All animal procedures were approved by the Institutional Animal Care and Use Committee and conformed to the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health (1996).

Anesthesia and monitoring

To maintain deep general anesthesia, atropine (0.01 mg/kg; batch number: 17081774, Xinxiang Changle Pharmaceutical Co., Ltd.), midazolam (0.1 mg/kg; batch number: 20181117, Jiangsu Enhua Pharmaceutical Co., Ltd.), sufentanil (0.5 μg/kg; production batch number: 180161, Impfstoffwerk Dessau-Tornau GmbH), rocuronium (0.6 mg/kg; batch number: R003337, Hameln Pharmaceuticals GmbH), and propofol (2 mg/kg; batch number: 16MI1776, Beijing Fresenius Medical Co., Ltd.) were administered intravenously to all piglets. Tracheal intubation was performed under video laryngoscope guidance. All piglets received pressure-controlled mechanical ventilation at a tidal volume of 6–8 ml/kg, an expiration/inhalation ratio of 2:1, and a partial pressure of end-tidal carbon dioxide of 35–45 mmHg.

The femoral arteries and veins were catheterized to monitor blood pressure, and temperature probes were placed in the rectum and nasopharynx to continuously monitor body temperature. All piglets underwent a median sternotomy to expose their hearts and were administered heparin (3 mg/kg). To initiate CPB, an arterial cannula was inserted into the ascending aorta, and a venous cannula was inserted into the right atrial appendage.

Experimental designs

All piglets were randomly divided into three groups ($n = 8$ for each group): DHLF, CPP, and CPP+PDTC. A timeline of the animal studies is shown in Figure 1.

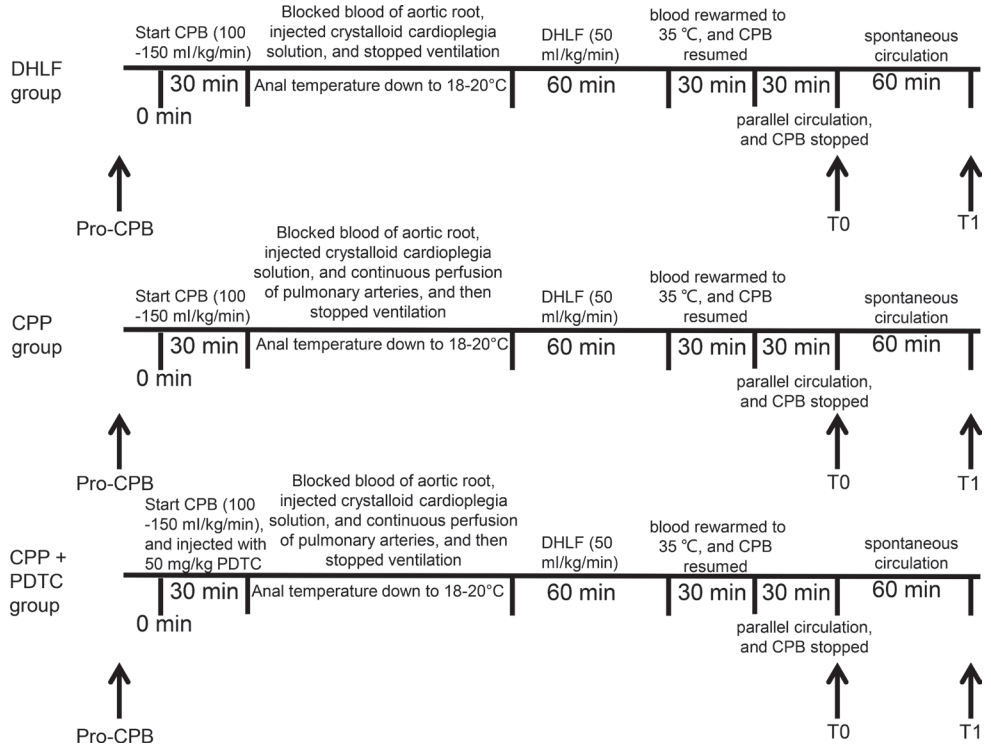

Figure 1. Timelines of the animal studies in the different groups. DHLF, deep hypothermia with low flow perfusion; CPP, continuous perfusion of pulmonary arteries; CPB, cardiopulmonary bypass; PDTC, pyrrolidine dithiocarbamate; Pro-CPB, prior to cardiopulmonary bypass; T0, end of CPB; T1, 1 h post-CPB.

DHLF group: A non-pulsatile systemic CPB flow of 100–150 ml/kg/min was initiated for half an hour. When anal temperature was reduced to 18–20°C, blood flow in the aortic root was blocked, cold crystalloid cardioplegia solution (4°C, 15 ml/kg) for myocardial preservation was injected, and mechanical ventilation was discontinued. After 1 h of DHLF (50 ml/kg/min), the blood was rewarmed to 35°C within half an hour with a flow of 90–120 ml/kg/min, and the blood flow in the aortic root was resumed. The heart was beating normally, and mechanical ventilation was resumed. After half an hour of parallel circulation, CPB was discontinued, and spontaneous circulation was maintained for 1 h.

CPP group: CPB flow was established as previously described. When anal temperature was reduced to 18–20°C, blood flow in the aortic root was blocked, and cold crystalloid cardioplegia solution (4°C, 15 ml/kg) was injected for myocardial preservation. The perfusion pump was turned on, and the perfusion fluid (oxygenated blood) from the "Y" style catheter flowed into the pulmonary artery and finally returned to the blood storage bottle through a drainage tube in the left atrium. Mechanical ventilation was discontinued. After 1 h of DHLF (50 ml/kg/min), the blood was rewarmed to 35°C within half an hour with a flow of 90–120 ml/kg/min, and the blood flow in the aortic root was resumed. After half an hour of parallel circulation, CPB was discontinued, and spontaneous circulation was maintained for 1 h.

CPP+PDTC group: the same protocols were used as those in the CPP group except that PDTC (50 mg/kg) (Zhang and Jin 2017; Pan et al. 2018) was intravenously administered to the piglets when CPB was initiated. Spontaneous circulation was maintained for 1 h.

Sample collection and physiologic measurements

The degree of lung injury was evaluated by measurement of respiratory function, immunohistochemical examination of lung tissues, and calculation of the lung wet-to-dry weight (W/D) ratio.

At the time nodes before CPB (Pro-CPB), at the end of CPB (T0), and at 1 h post-CPB (T1), arterial blood (3 ml) was sampled from two piglets (Pro-CPB), three piglets (T0), and three piglets (T1) in the DHLF, CPP, and CPP+PDTC groups, respectively. Some of the blood samples were used for arterial blood gas (ABG) analysis with an Easy Stat ABG analyzer (Medica Corporation, Bedford, USA). The remaining blood samples were used to determine the serum levels of NF-κB, TNF, IL-8, and IL-6 at all three time points using their corresponding enzyme-linked immunosorbent assay

(ELISA) kits according to the manufacturer's instructions. The following ELISA kits were used: NF-κB ELISA kit (human p50, batch number: 10006912-96, Cayman Chemical Company, USA), TNF ELISA kit (batch number: 589201-96, Cayman Chemical Company, USA), IL-8 ELISA kit (batch number: 3560-1H-20, Mabtech, Nacka, Sweden), and IL-6 ELISA kit (batch number: 583361-480, Cayman Chemical Company, USA).

In addition, at each time point, piglets were euthanized by routine air venous embolization or intravenous potassium chloride injection. Lung tissues, including the right and left lower lobes, were collected from the piglets. Among them, a piece of the right lower lobe of the lung tissues (approximately 1 cm^3) was weighed; after drying at 80°C for 48 h, the dry weight of the tissues was also measured to calculate the lung W/D ratio. Furthermore, a part of the left lower lobe of the lung tissues was fixed with 4% formalin, embedded in paraffin, and sectioned. The paraffin-embedded sections were stained with hematoxylin-eosin (HE) and observed under a Nikon Eclipse E100 microscope (Tokyo, Japan). Another part of the left lower lobe was fixed in glutaraldehyde and stained, and the pulmonary ultrastructure was observed under a JEM-ARM200F transmission electron microscope (JEOL, Tokyo, Japan).

Western blot

NF-κB protein expression in lung tissues was measured at the end of the experiment using Western blot. Briefly, lung tissues were lysed using radioimmunoprecipitation assay lysis buffer. The protein samples were separated by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE) and then transferred onto immobilon-P transfer membranes (Millipore, USA). After blocking with 5% skim milk at 37°C for 2 h, the membranes were incubated with a primary antibody against NF-κB (Abcam, Cambridge, USA; batch number: ab195854; 1:2000) or GAPDH (Abcam, Cambridge, USA; batch number: ab181602; 1:3000) at 4°C overnight. Subsequently, the membranes were incubated with a secondary antibody (1:1000, goat anti-rabbit IgG-HRP, Santa Cruz Biotechnology, USA) at 37°C for 2 h. Finally, the membranes were rinsed with Tris buffered saline Tween (TBST), and the protein bands were visualized using enhanced chemiluminescence (Applygen, Beijing, China).

Statistical analyses

In this study, PASS 15.0 software was used to estimate the experimental sample size, and the 3R rules for animal experimentation were strictly followed. All values are expressed as means ± standard deviations. Before statistical analyses, the Shapiro-Wilk test was used to control or assess the normal distribution of all data, and the data were found to be in accordance with the normal distribution. Thereafter, one-way analysis of variance, followed by the Newman-Keuls method, was used to analyze the significant difference between more than two groups. However, for the two groups, Student's *t*-test was used. Statistical significance was set at $p < 0.05$. All statistical analyses were performed using SPSS software (version 19.0; Chicago, IL, USA).

Results

CPP combined with PDTC attenuated the DHLF-induced lung dysfunction

The DHLF, CPP, and CPP+PDTC groups were not significantly different in body weight, heart rate, mean arterial pressure, parcial pressure of oxygen (PaO_2) , and parcial pressure of carbon dioxide ($PaCO_2$) ($p > 0.05$) before CPB. PaO_2 and $PaCO_2$ were measured using ABG analysis at the Pro-CPB,

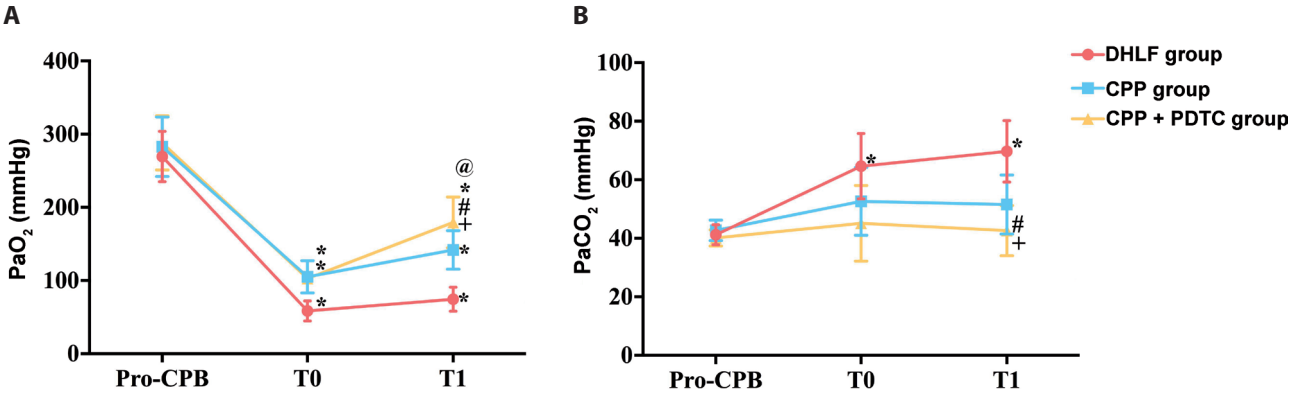

Figure 2. Effects of CPP and PDTC on DHLF-induced lung dysfunction. The content of PaO_2 (**A**) and $PaCO_2$ (**B**) at three time points in the different groups. * $p < 0.05$ *vs.* Pro-CPB group; $^{\#}$ $p < 0.05$ *vs.* DHLF group; $^{@}$ $p < 0.05$ *vs.* CPP group; $^{+}$ $p < 0.05$ *vs.* in group T0. PaO_2, arterial partial pressure of oxygen; $PaCO_2$, arterial partial pressure of carbon dioxide. For other abbreviations, see Figure 1.

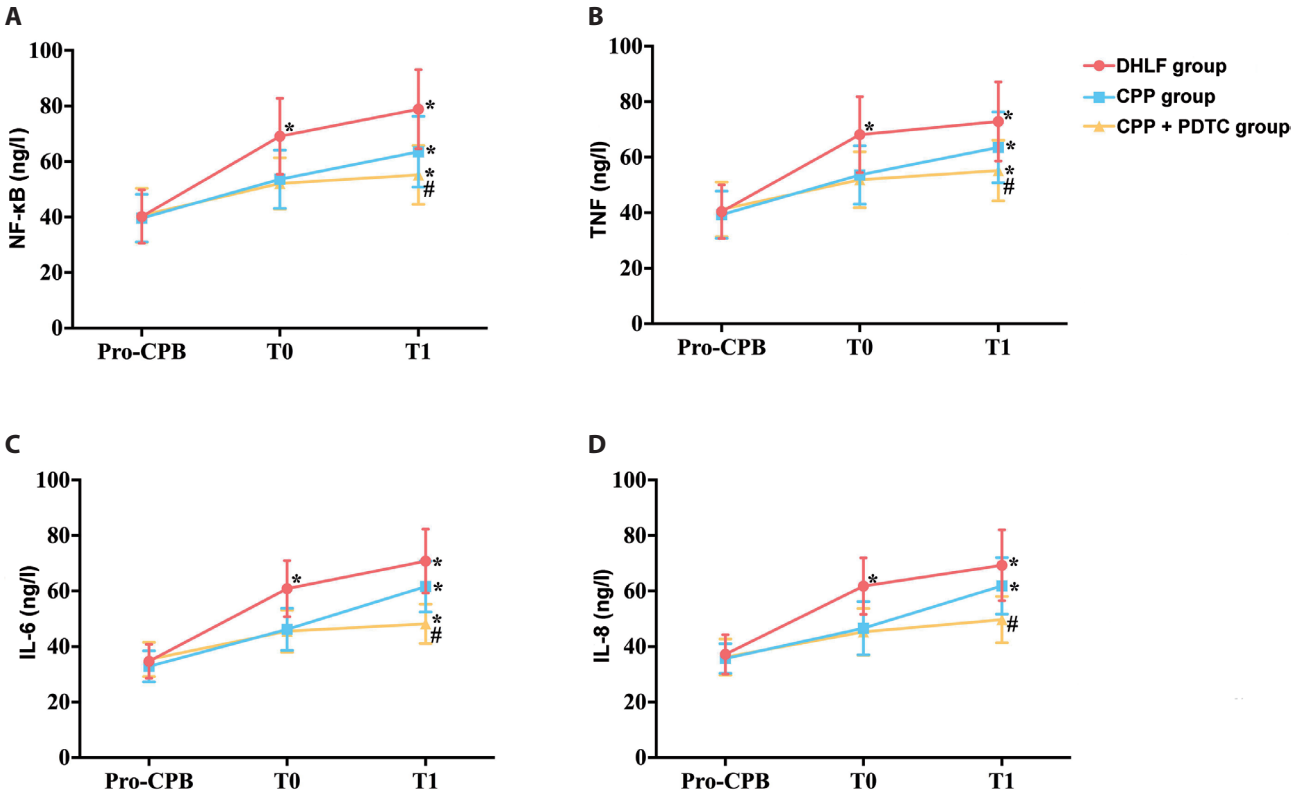

Figure 3. Effects of CPP and PDTC on DHLF-induced release of inflammatory factors. Serum levels of NF-κB (**A**), TNF (**B**), IL-6 (**C**), and IL-8 (**D**) at three time points in the different groups. * $p < 0.05$ *vs.* Pro-CPB group; # $p < 0.05$ *vs.* DHLF group; @ $p < 0.05$ *vs.* CPP group; + $p < 0.05$ *vs.* in group T0. For abbreviations, see Figure 1.

T0, and T1 time points. As shown in Figure 2, each group had significantly decreased PaO_2 and increased $PaCO_2$ at T0 and T1 than at Pro-CPB ($p < 0.05$), suggesting that the piglet models of DHLF were successfully established. Moreover, at T1, both CPP and CPP+PDTC groups had considerably elevated PaO_2 and decreased $PaCO_2$ in comparison with the DHLF group ($p < 0.05$), indicating that both CPP and PDTC with CPP could alleviate DHLF-induced lung dysfunction. Furthermore, PaO_2 was further improved in the CPP+PDTC group that in the CPP group ($p < 0.05$) at T1; however, the difference in $PaCO_2$ levels was not significant between the CPP and CPP+PDTC groups ($p > 0.05$, Figs. 2A,B). This reveals that the combined use of PDTC and CPP better alleviates DHLF-induced lung dysfunction than does CPP alone.

CPP combined with PDTC reduced the DHLF-induced release of inflammatory factors

In the DHLF group, the serum levels of NF-κB, TNF, IL-8, and IL-6 were significantly higher at T1 than at Pro-CPB ($p < 0.05$, Fig. 3A–D), suggesting that DHLF induced a considerable release of inflammatory factors. Moreover, at T1, the levels of NF-κB, TNF, IL-8, and IL-6 were markedly increased in the CPP ($p < 0.05$) and CPP+PDTC groups ($p < 0.05$) than in the DHLF group (Fig. 3A–D). Furthermore, TNF was remarkably decreased in the CPP+PDTC group than in the CPP group ($p < 0.05$, Fig. 3B); however, the differences in serum levels of NF-κB, IL-8, and IL-6 were not significant between the two groups ($p > 0.05$, Fig. 3A,C,D).

NF-κB protein expression in the lung tissues was detected at T1 by Western blot. Compared with the DHLF group, NF-κB protein was considerably downregulated in the CPP group and further downregulated in the CPP+PDTC group (Fig. 4).

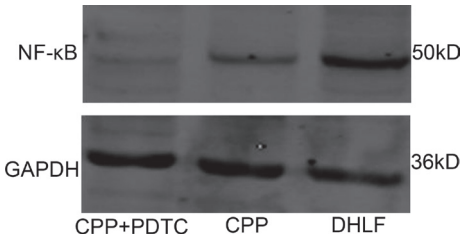

Figure 4. Protein expression of NF-κB in the different groups after CPP by Western blot. For abbreviations, see Figure 1.

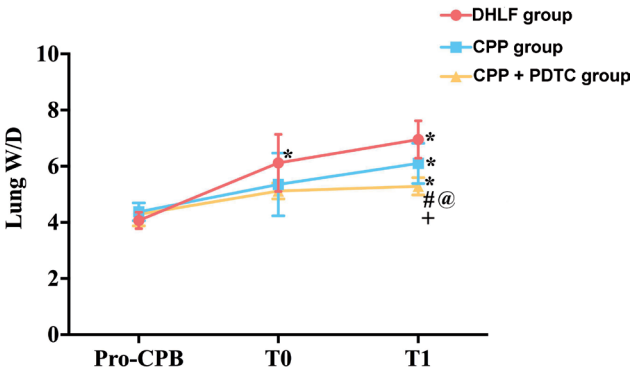

Figure 5. Changes of lung wet-to-dry weight (W/D) ratio at three timepoints in the different groups. * $p < 0.05$ *vs.* Pro-CPB; # $p < 0.05$ *vs.* DHLF group; @ $p < 0.05$ *vs.* CPP group; + $p < 0.05$ *vs.* in group T0. For abbreviations, see Figure 1.

CPP combined with PDTC mitigated the DHLF-induced pulmonary edema and injury

The DHLF piglets had a significantly higher lung W/D ratio at T1 than at Pro-CPB ($p < 0.05$, Fig. 5). Furthermore, at T1, both CPP and CPP with PDTC caused a considerable decrease in the lung W/D ratio ($p < 0.05$), and the extent of the decrease caused by CPP combined with PDTC was much greater ($p < 0.05$, Fig. 5).

Histopathological changes in the pulmonary tissues were evaluated by HE staining under a light microscope and a transmission electron microscope. Pulmonary edema and injury was severe in the DHLF group, moderate in the CPP group, and slight in the CPP+PDTC group (Fig. 6).

Discussion

DHLF-induced pulmonary injury is a common postoperative complication of CPB, which increases hospital stay and postoperative mortality in infants and neonates with congenital heart disease (Axelrod et al. 2016). To our knowledge, our study is the first to determine whether CPP and NF-κB inhibitor are effective pulmonary protection strategies to relieve DHLF-induced pulmonary injury. Pulmonary injury resulting from DHLF is characterized by an uncontrolled inflammatory response involving complicated interactions between various inflammatory cells, transcription factors, cytokines, and inflammatory mediators (Laubach and Sharma 2016; Hou et al. 2018; Wang et al. 2018c). In our study, lower PaO_2 and higher $PaCO_2$ and serum levels of NF-κB, TNF, IL-8, and IL-6 were observed in the piglet models of DHLF, suggesting that our infant piglet models successfully simulated clinical practice.

Direct contact of blood with the CPB circuit induces pulmonary inflammation and activates polymorphonuclear cells, primarily polymorphonuclear leukocytes (PMNs), that can release various inflammatory cytokines, such as TNF, IL-6, and IL-8 (Su et al. 2003). These cytokines increase capillary permeability, thereby accelerating lung tissue edema and injury (Takashima et al. 2014). Moreover, PMNs accumulated in the lung tissue release elastase and oxygen free radicals, resulting in swelling and degeneration of pulmonary vascular

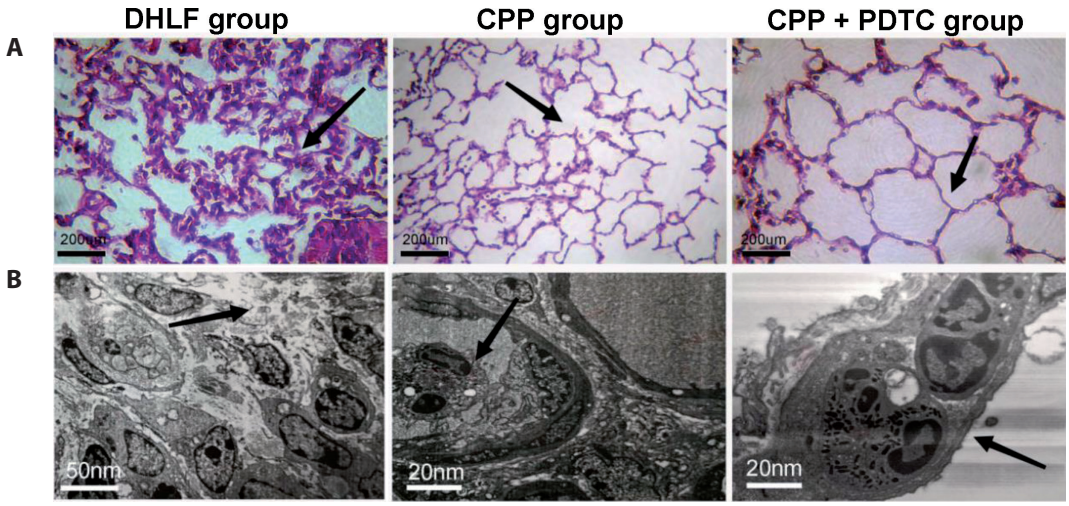

Figure 6. Effects of CPP and PDTC on the histopathological presentation of pulmonary tissue of piglets after CPB. **A.** Morphological presentation of pulmonary tissues in the different group using a light microscope. The arrow in the CPP group represents the partially destroyed cell wall. The arrow in the CPP+PDTC group indicates the complete structure of cells and inter-cells. **B.** Pulmonary ultrastructure of the pulmonary tissues in the different groups. The arrow in the DHLF group means shattered nuclei. The arrow in the CPP group represents occasional apoptotic bodies. The arrow in the CPP+PDTC group indicates an unbroken blood-air barrier. For abbreviations, see Figure 1.

endothelial cells and pulmonary interstitial edema (Freitas et al. 2016). Furthermore, PMNs have been demonstrated to be key effector cells in the pathogenesis of lung I/R injury (Rebetz et al. 2018). In the present study, accumulation of inflammatory cells, primarily PMNs, inside and outside the alveolar cavity and rupture of alveolar septa were found in the DHLF group. These findings confirmed the critical role of PMNs in DHLF-induced pulmonary injury.

Pulmonary I/R injury exacerbates pulmonary inflammation and has been recognized as a crucial initiator of post-DHLF lung injury (Salameh et al. 2017). A growing body of evidence suggests that pulmonary perfusion provides better oxygenation, relieves ischemia, and diminishes post-CPB lung injury (Yewei et al. 2013; Maltesen et al. 2018; Karacalilar et al. 2020). Our study showed that CPP increased PaO_2 and decreased $PaCO_2$ and serum levels of NF-κB, TNF, IL-8, and IL-6, and abated pulmonary edema and injury in piglet models of DHLF at 1 h post-CPB, corroborating these reports. This indicates that CPP improves oxygenation and attenuate inflammation, thereby reducing lung injury.

The NF-κB signaling pathway plays a vital role, and NF-κB is a central mediator in the inflammatory response (Huang et al. 2019; Wang et al. 2019). Following IκB degradation via IKK phosphorylation, NF-κB is activated, and it translocates from the cytoplasm to the nucleus, thereby activating the transcription of target genes involved in inflammation (Pordanjani and Hosseinimehr 2016). NF-κB has been implicated in regulating the production of pro-inflammatory cytokines, including TNF, IL-6, and IL-8, in acute lung injury, which can further activate NF-κB (Zhu et al. 2018). Inhibition of the NF-κB pathway by calcitriol counteracts inflammation and pulmonary edema induced by seawater aspiration (Zhang and Jin 2017). NF-κB inhibitor PDTC inhibits activation of NF-κB in the cytoplasm, and it has been demonstrated to exert protective effects against lung injury (Wang et al. 2011; Francioli et al. 2017; Wang et al. 2018a). In agreement with these studies, we found that compared with CPP alone, intravenous administration of PDTC along with maintenance of CPP further increased PaO_2, decreased $PaCO_2$, and attenuated regional hemorrhage of lung tissue, alveolar edema, and inflammatory cell infiltration in infant piglet models of DHLF. Moreover, the combination of PDTC and CPP appeared to have a greater anti-inflammatory effect, as evidenced by the lower serum levels of TNF, IL-8, IL-6, and NF-κB in infant piglet models of DHLF at 1 h post-CPB. These results collectively suggest that the combined application of PDTC and CPP confers a better lung-protective effect against DHLF-induced lung injury than does CPP alone by improving oxygenation of lung tissue and suppressing NF-κB protein expression and the inflammatory response.

This study had some limitations that should not be neglected. Our study only focused on lung injury and dysfunction during the 3-h perioperative period of DHLF. Alterations of PMNs in lung tissues and serum levels of TNF, IL-8, IL-6, and NF-κB should be monitored for a longer time following DHLF to further investigate the molecular mechanisms underlying the lung-protective effect of PDTC and CPP. In addition to these inflammatory factors, additional *in vivo* and *in vitro* experiments should be performed to further uncover detailed molecular mechanisms. In addition, larger randomized clinical trials are needed to validate the lung-protective activity of PDTC and CPP before their clinical use can be considered. The combined effects of PDTC and CPP in DHLF lung injury, as well as the translocation of active NF-κB to the nucleus and subsequent downstream genes, require further investigation. Additionally, this study focused on the roles of PDTC, an inhibitor of NF-κB, in the DHLF lung injury model. Further experiments are needed to explore the effects of PDTC as an antioxidant and an inhibitor of IκBα phosphorylation in this model.

Conclusion

In conclusion, our study revealed that PDTC administration together with CPP was more effective in protecting pulmonary function against DHLF-CPB-induced lung injury than was CPP alone. The mechanisms may involve improved oxygenation and inhibition of the inflammatory response and NF-κB signaling pathway. Our findings suggest that the combined use of PDTC and CPP could be a novel preventative and therapeutic strategy for DHLF-induced lung injury in infants and neonates undergoing cardiac surgery.

Funding. This study was supported by Shanghai Medical Guidance and Technology Support Project (No. 17411969000).

Conflict of interest. The authors declared they have no conflict of interest.

Availability of data and material. The data that support the findings of this study are available in Science Data Bank at https://www.scidb.cn/datalist, DOI number 10.11922/sciencedb.01544.

Authors' contributions. YX carried out the conception and design of the research, analysis, and interpretation of data, performed the statistical analysis, participated in obtaining funding and drafted the manuscript. JL participated in the acquisition of data. RZ participated in the design of the study and revision of manuscript for important intellectual content. All authors read and approved the final manuscript.

References

Axelrod DM, Chock VY, Reddy VM (2016): Management of the preterm infant with congenital heart disease. Clin. Perinatol. **43,** 157-171

https://doi.org/10.1016/j.clp.2015.11.011
Buggeskov KB, Grønlykke L, Risom EC, Wei ML, Wetterslev J (2018): Pulmonary artery perfusion versus no perfusion during cardiopulmonary bypass for open heart surgery in adults. Cochrane Database Syst. Rev. **2,** Cd011098 https://doi.org/10.1002/14651858.CD011098.pub2
Francioli C, Wang X, Parapanov R, Abdelnour E, Lugrin J, Gronchi F, Perentes J, Eckert P, Ris H-B, Piquilloud L, et al. (2017): Pyrrolidine dithiocarbamate administered during ex-vivo lung perfusion promotes rehabilitation of injured donor rat lungs obtained after prolonged warm ischemia. PLoS One **12,** e0173916 https://doi.org/10.1371/journal.pone.0173916
Freitas CR, Sa Malbouisson LM, Benicio A, Negri EM, Bini FM, Massoco CO, Otsuki DA, Vidal Melo MF, Carvalho Carmonaet MJ (2016): Lung perfusion and ventilation during cardiopulmonary bypass reduces early structural damage to pulmonary parenchyma. Anesth. Analg. **122,** 943-952 https://doi.org/10.1213/ANE.0000000000001118
Gaynor JW, Kim DS, Arrington CB, Atz AM, Bellinger DC, Burt AA, Ghanayem NS, Jacobs JP, Lee TM, Lewis AB, et al. (2014): Validation of association of the apolipoprotein E ε2 allele with neurodevelopmental dysfunction after cardiac surgery in neonates and infants. J. Thorac. Cardiovasc. Surg. **148,** 2560-2568 https://doi.org/10.1016/j.jtcvs.2014.07.052
Harten AEV, Scheeren TWL, Absalom AR (2012): A review of postoperative cognitive dysfunction and neuroinflammation associated with cardiac surgery and anaesthesia. Anaesthesia **67,** 280-293 https://doi.org/10.1111/j.1365-2044.2011.07008.x
Hoesel B, Schmid JA (2013): The complexity of NF-kappaB signaling in inflammation and cancer. Mol. Cancer **12,** 86 https://doi.org/10.1186/1476-4598-12-86
Hou L, Yang Z, Wang Z, Zhang X, Zhao Y, Yang H, Zheng B, Tian W, Wang S, He Z, et al. (2018): NLRP3/ASC-mediated alveolar macrophage pyroptosis enhances HMGB1 secretion in acute lung injury induced by cardiopulmonary bypass. Lab. Invest. **98,** 1052-1064 https://doi.org/10.1038/s41374-018-0073-0
Huang XT, Liu W, Zhou Y, Hao C-X, Zhou Y, Zhang C-Y, Sun C-C, Luo Z-Q, Tang S-Y (2019): Dihydroartemisinin attenuates lipopolysaccharide-induced acute lung injury in mice by suppressing NF-κB signaling in an Nrf2-dependent manner. Int. J. Mol. Med. **44,** 2213-2222 https://doi.org/10.3892/ijmm.2019.4387
Karacalilar M, Onan I, Onan B, Sen O, Gonca S, Solakoglu S, Yeniterzi M (2020): Effects of pulmonary perfusion during cardiopulmonary bypass on lung functions after cardiac operation. J. Card. Surg. **35,** 2469-2476 https://doi.org/10.1111/jocs.14934
Laubach VE, Sharma AK (2016): Mechanisms of lung ischemia-reperfusion injury. Curr. Opin. Organ Transplant. **21,** 246-252 https://doi.org/10.1097/MOT.0000000000000304
Li W, Wu X, Yan F, Liu J, Tang Y, Ma K, Li S (2014): Effects of pulmonary artery perfusion with urinary trypsin inhibitor as a lung protective strategy under hypothermic low-flow cardiopulmonary bypass in an infant piglet model. Perfusion **29,** 434-442 https://doi.org/10.1177/0267659113517286
Maltesen R, Buggeskov K, Andersen C, Plovsing R, Wimmer R, Ravn H, Rasmussen B (2018): Lung protection strategies during cardiopulmonary bypass affect the composition of bronchoalveolar fluid and lung tissue in cardiac surgery patients. Metabolites **8,** 15 https://doi.org/10.3390/metabo8040054
Matsuda A, Yang W-L, Jacob A, Aziz M, Matsuo S, Matsutani T, Uchida E, Wang P (2014): FK866, a visfatin inhibitor, protects against acute lung injury after intestinal ischemia-reperfusion in mice via NF-κB pathway. Ann. Surg. **259,** 1007-1017 https://doi.org/10.1097/SLA.0000000000000329
Mitchell JP, Carmody RJ (2018): NF-κB and the transcriptional control of inflammation. Int. Rev. Cell. Mol. Biol. **335,** 41-84 https://doi.org/10.1016/bs.ircmb.2017.07.007
Napetschnig J, Wu H (2013): Molecular basis of NF-kappaB signaling. Annu. Rev. Biophys. **42,** 443-468 https://doi.org/10.1146/annurev-biophys-083012-130338
Pan WZ, Du J, Zhang LY, Ma JH (2018): The roles of NF-kB in the development of lung injury after one-lung ventilation. Eur. Rev. Med. Pharmacol. Sci. **22,** 7414-7422
Peng C, Ouyang Y, Lu N, Li N (2020): The NF-kappaB signaling pathway, the microbiota, and gastrointestinal tumorigenesis: recent advances. Front. Immunol. **11,** 1387 https://doi.org/10.3389/fimmu.2020.01387
Pordanjani SM, Hosseinimehr SJ (2016): The role of NF-kB inhibitors in cell response to radiation. Curr. Med. Chem. **23,** 3951-3963 https://doi.org/10.2174/0929867323666160824162718
Qiu Y, Wu Y, Zhao H, Sun H, Gao S (2019): Maresin 1 mitigates renal ischemia/reperfusion injury in mice via inhibition of the TLR4/MAPK/NF-κB pathways and activation of the Nrf2 pathway. Drug Des. Devel. Ther. **13,** 739-745 https://doi.org/10.2147/DDDT.S188654
Rebetz J, Semple JW, Kapur R (2018): The pathogenic involvement of neutrophils in acute respiratory distress syndrome and transfusion-related acute lung injury. Transfus. Med. Hemother. **45,** 290-298 https://doi.org/10.1159/000492950
Salameh A, Greimann W, Vollroth M, Dhein S, Bahramsoltani M, Dahnert I (2017): Lung protection in cardio-pulmonary bypass. J. Physiol. Pharmacol. **68,** 99-116
Su ZK, Sun Y, Yang YM, Zhang HB, Xu ZW (2003): Lung function after deep hypothermic cardiopulmonary bypass in infants. Asian Cardiovasc. Thorac. Ann. **11,** 328-331 https://doi.org/10.1177/021849230301100412
Takashima K, Matsushima M, Hashimoto K, Nose H, Sato M, Hashimoto N, Hasegawa Y, Kawabe T (2014): Protective effects of intratracheally administered quercetin on lipopolysaccharide-induced acute lung injury. Respir. Res. **15,** 150 https://doi.org/10.1186/s12931-014-0150-x
Wang HT, Fang Y-Q, You P, Bao X-C, Li K-C, Yao J, Li C, Yuanet H-R (2018a): PDTC ameliorates decompression induced-lung injury caused by unsafe fast buoyancy ascent escape via inhibition of NF-κB pathway. Undersea Hyperb. Med. **45,** 351-362 https://doi.org/10.22462/05.06.2018.10
Wang L, Ma H, Xue Y, Shi H, Ma T, Cui X (2018b): Berberine inhibits the ischemia-reperfusion injury induced inflamma-

tory response and apoptosis of myocardial cells through the phosphoinositide 3-kinase/RAC-α serine/threonine-protein kinase and nuclear factor-κB signaling pathways. Exp. Ther. Med. **15,** 1225-1232
https://doi.org/10.3892/etm.2017.5575

Wang M-L, Wei C-H, Wang W-D, Wang J-S, Zhang J, Wang J-J (2018c): Melatonin attenuates lung ischaemia-reperfusion injury via inhibition of oxidative stress and inflammation. Interact. Cardiovasc. Thorac. Surg. **26,** 761-767
https://doi.org/10.1093/icvts/ivx440

Wang M, Liu T, Wang D, Zheng Y, Wang X, He J (2011): Therapeutic effects of pyrrolidine dithiocarbamate on acute lung injury in rabbits. J. Transl. Med. **9,** 61
https://doi.org/10.1186/1479-5876-9-61

Wang T, Jiang L, Wei X, Dong Z, Liu B, Zhao J, Wang L, Xie P, Wang Y, Zhou S (2019): Inhibition of miR-221 alleviates LPS-induced acute lung injury via inactivation of SOCS1/NF-κB signaling pathway. Cell Cycle **18,** 1893-1907
https://doi.org/10.1080/15384101.2019.1632136

Yang Y, Wang J, Peng W, Shu Y, Mo X (2020): Cytochrome P450 epoxygenase 2J2 protects against lung ischemia/reperfusion injury by activating the P13K/Akt/GSK-3-β/NF-kB signaling pathway during deep hypothermic low flow in mice. J. Surg. Res. **253,** 8-17
https://doi.org/10.1016/j.jss.2019.12.052

Ye Y, Shan Y, Bao C, Hu Y, Wang L (2018): Ginsenoside Rg1 protects against hind-limb ischemia reperfusion induced lung injury via NF-κB/COX-2 signaling pathway. Int. Immunopharmacol. **60,** 96-103
https://doi.org/10.1016/j.intimp.2018.04.040

Yewei X, Liya D, Jinghao Z, Rufang Z, Li S (2013): Study of the mechanism of pulmonary protection strategy on pulmonary injury with deep hypothermia low flow. Eur. Rev. Med. Pharmacol. Sci. **17,** 879-885

Zhang H, Liu Y, Wang LK, Wei N (2017): Pyrrolidine dithiocarbamate alleviates the anti-tuberculosis drug-induced liver injury through JAK2/STAT3 signaling pathway: An experimental study. Asian Pac. J. Trop. Med. **10,** 520-523
https://doi.org/10.1016/j.apjtm.2017.05.010

Zhang M, Jin F (2017): 1α,25-Dihydroxyvitamin D3 ameliorates seawater aspiration-induced lung injury by inhibiting the translocation of NF-κB and RhoA. Inflammation **40,** 832-839
https://doi.org/10.1007/s10753-017-0527-3

Zhu WD, Xu J, Zhang M, Zhu TM, Zhang YH, Sun K (2018): MicroRNA-21 inhibits lipopolysaccharide-induced acute lung injury by targeting nuclear factor-κB. Exp. Ther. Med. **16,** 4616-4622

Received: August 25, 2022
Final version accepted: November 8, 2022