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Biliary tract external drainage alleviates kidney injury in shock

*Lu Wang, MD, Bing Zhao, MD, PhD, Ying Chen, MD, Li Ma, MD, Er-zhen Chen, MD, PhD, and En-Qiang Mao, MD, PhD**

Department of Emergency Intensive Care Unit, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai, China

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ABSTRACT

Background: Kidney injury is common in hemorrhagic shock (HS). Kidney injury leads to a systemic increase in serum chemokines and cytokines and causes injuries to other vital organs. Our previous studies showed that vitamin C led to organ protection and inflammation inhibitory effects in rat models of HS via induction heme oxygenase-1 (HO-1). We also found that biliary tract external drainage (BTED) increased the expression levels of HO-1 in rat livers. We investigated roles of BTED in kidney injury and its relationship with the HO-1 pathway in HS in this research.

Methods: Rat models of HS were induced by drawing blood from the femoral artery. BTED was performed by inserting a catheter into the bile duct. Thirty-six Sprague–Dawley rats were randomized to sham group; HS group; zinc protoporphyrin IX (Znpp) group; BTED group; BTED + Znpp group, and BTED + bile infusion group. The expression levels of HO-1 in the kidney were analyzed by Western blotting. The expression levels of occludin messenger RNA in the kidney were analyzed by real-time reverse transcription-polymerase chain reaction. The expression levels of occludin in the kidney were analyzed by immunohistochemistry. Histology of renal was performed by hematoxylin and eosin staining.

Results: Occludin messenger RNA and protein levels in the kidney increased markedly after BTED under HS conditions. Renal histopathologic scores decreased significantly after BTED under HS conditions. Znpp significantly inhibited all mentioned effects.

Conclusions: BTED alleviates kidney injury in rats of HS via the HO-1 pathway.

1. Introduction

The kidney is one of the primary ischemic organs in hemorrhagic shock (HS) [1–4]. Fluid resuscitation significantly reduces the morbidity of kidney injury induced by ischemia [5–7]. However, inflammatory cytokines produced in the process of oxidative stress and inflammation responses induced by ischemia–reperfusion also cause kidney injury such as renal tubular damage, inflammation, and vascular endothelial damage [4,8–13]. Kidney injury leads to a systemic increase in serum chemokines and cytokines such as tumor necrosis factor (TNF)-α and then causes injuries to other remote organs [14–20]. Renal insufficiency has been a sensitive marker for poor outcomes in critically ill patients [21,22]. Relieving kidney injury may be an important mechanism to treat HS [3,23,24].

Renal tight junction (TJ) proteins establish cell–cell contact in the intercellular space and are responsible for

* *Corresponding author*. Department of Emergency Intensive Care Unit, Ruijin Hospital, Shanghai Jiao Tong University School of Medicine, Shanghai 200025, China. Tel./fax: +86 21 64370045 665526.
E-mail address: maoeq@yeah.net (E.-Q. Mao).

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paracellular transport of solutes and water and hence in extracellular fluid homeostasis [25,26]. TJ proteins decrease in many kidney diseases [26–28]. Many integral membrane proteins have been identified as components of TJ strands, including occludin and claudins [29–32]. Occludin is a protein present in the filaments that constitute the TJ [26,33]. Occludin is present in epithelial and endothelial cells and absent in cells that lack TJ proteins, such as fibroblasts [33] (Fig. 1). In tubular renal cells, occludin is present at the cell borders [34]. Distribution of occludin results in increases in TJ complexity. The overall distributions of occludin along the nephron agree with the transepithelial electrical resistance reported in the kidney segments. In regions where transepithelial electrical resistance is high, occludin fluorescent signals are located at cell borders, suggesting an intimate relationship between occludin and strength of the TJ [35,36]. Cell-specific disruptions of occludin are observed in renal ischemia–reperfusion injury [37]. Occludin protein content influences renal epithelial paracellular permeability [38]. Rearrangement of occludin causes renal dysfunction [39]. Therefore, occludin was selected as a focus for kidney injury in the present research.

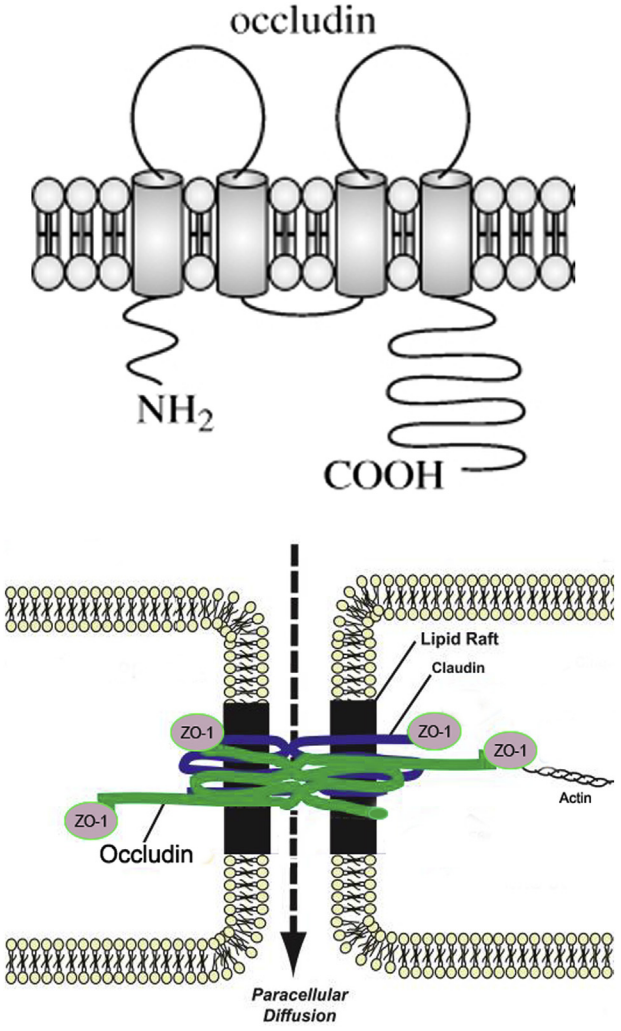

Fig. 1 – The molecular structure and location of occludin. (Color version of figure is available online.)

Organ function in shock patients with severe acute pancreatitis who accept biliary tract external drainage (BTED) rapidly improves in clinical practice. Infection incidence and morbidity of multiple-organ dysfunction syndrome also significantly decrease. Studies in our laboratory demonstrated that BTED decreased proinflammatory cytokine production and relieved tissue damage in the intestine, liver, and lung in rat models of HS [40]. Our studies also showed that vitamin C led to organ protection and inflammation inhibitory effects in rat models of HS via induction heme oxygenase-1 (HO-1) [41,42]. We also found BTED increased the expression levels of HO-1 in rat livers. We investigated roles of BTED in kidney injury and its relationship with the HO-1 pathway in HS in this research.

2. Materials and methods

2.1. *Animal model*

This study was carried out in strict accordance with the guidelines for the care and use of laboratory animals established by the Animal Use and Care Committee of the Shanghai Committee on Animal Care. Animal surgical procedures were approved by the Institutional Animal Care and Use Committee at Shanghai Jiao Tong University, Shanghai, China. Forty-two adult male Sprague–Dawley rats (250–300 g) were purchased from the Experimental Animal Center of Ruijin Hospital.

After a 1-wk adaption period during which food and water were available *ad libitum*, 36 rats were randomly divided into six groups as follows: sham group; HS group; zinc protoporphyrin IX (Znpp) group; BTED group; BTED + Znpp group, and BTED + bile infusion (BI) group. Other rats were used for the collection of normal bile. Rats were fasted overnight before experiments but water was available *ad libitum*. Rats in the Znpp and BTED + Znpp groups were given Znpp, a specific HO-1 inhibitor, (30 mg/kg) via intraperitoneal injection 24 h before the experiments. Rats in the BTED + BI group were anesthetized with sodium pentobarbital (intraperitoneal, 50 mg/kg). Laparotomies were performed after shaving and sterilization. One catheter was placed in the right femoral artery for blood pressure measurement, and another catheter was placed in the left femoral artery for blood withdrawal. Bile duct was exposed long enough for BTED. Rats were subjected to HS by slowly withdrawing blood at a rate of 1 mL/min until a mean arterial pressure of 40 ± 5 mm Hg was achieved. A catheter was inserted into the bile duct for BTED. The distal end of bile duct was ligated. A similar catheter was inserted into the duodenum and fixed for BI. The catheters for BTED and BI were passed through the flank of rats to avoid bile passage into the gut and allow for the external collection and infusion of bile. The abdomen was closed subsequently. A mean arterial pressure of 40 ± 5 mm Hg was maintained for 60 min. Rats were resuscitated with their shed blood and an equal volume of normal saline at the end of the shock period. Rats that were used for collection of normal bile underwent pentobarbital anesthesia, laparotomy, and placement of catheter for external collection of normal bile and suture simultaneously with this experiment. Collected normal bile was slowly infused into the duodenum of rats in the BTED + BI group through the reserved