Available online at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/burns

Hypertonic saline resuscitation protects against kidney injury induced by severe burns in rats

Chun-Yu Yuan [a,1], Qin-Cheng Wang [a,1], Xu-Lin Chen [a], Qiang Wang [b], Cong-Song Sun [a], Ye-Xiang Sun [a,*], Chun-Hua Wang [a], Ming-Xing Su [b], Hai-Ying Wang [b], Xue-Sheng Wu [c]

[a] Department of Burns, The First Affiliated Hospital of Anhui Medical University, Hefei, Anhui 230022, PR China
[b] Institute of Disease Control and Prevention of the Chinese People's Liberation Army, Beijing 100071, PR China
[c] Department of Emergency Surgery, The First Affiliated Hospital of Anhui Medical University, Hefei, Anhui 230022, PR China

ARTICLE INFO

Article history:
Accepted 13 September 2018

Keywords:
Burns
Hypertonic saline
Kidney
Inflammation
Oxidative stress

ABSTRACT

Background: Proper fluid resuscitation can relieve visceral damage and improve survival in severely burned patients. This study compared the effectiveness of resuscitation with 400 mEq/L hypertonic saline (HS) and sodium lactate Ringer's solution (LR) in rats with kidney injury caused by burn trauma.

Methods: Rats (Sprague-Dawley) underwent burn injury and were randomized into sham, LR, and HS groups. Samples from the kidney were assayed for water content ratio, histopathology, and oxidative stress (superoxide dismutase (SOD) and malondialdehyde (MDA)). Serum sodium, renal function (creatinine and cystatin (Cys)-C), and inflammatory response (tumor necrosis factor (TNF)-α, interleukin (IL)-1β, and high mobility group protein box (HMGB)-1) were also examined as serum markers.

Results: Hypertonic saline resuscitation reduced the renal water content ratio and improved renal histopathology caused by severe burns. This effect was accompanied by reductions in serum creatinine and Cys-C as well as TNF-α, IL-1β, and HMGB1. Serum sodium concentration and SOD activity were increased, whereas MDA content was decreased in the kidney tissue of the HS group.

Conclusions: The data indicate that 400 mEq/L HS solution reduces hyponatremia and renal edema, inhibits the release of inflammatory mediators, and alleviates oxidative stress injury, thus protecting against kidney injury induced by severe burns.

1. Introduction

Severe burns can cause a sharp decline in renal function, leading to acute renal failure and multiple organ dysfunction syndromes, which can increase the mortality rate of burn patients. The frequency of acute kidney injury (AKI) among adults in the early stages after burn injury ranges from 26.6% to 53.3% [1,2]. Timely and appropriate fluid resuscitation is critical for effective burn treatment and can reduce the

* *Corresponding author at*: The First Affiliated Hospital of Anhui Medical University, 218 Jixi Road, Hefei, Anhui, 230022, PR China.
E-mail addresses: sunyexiang@163.com, 744957562@qq.com (Y.-X. Sun).
[1] Chun-Yu Yuan and Qin-Cheng Wang contributed equally to this work.
https://doi.org/10.1016/j.burns.2018.09.017

incidence of post-burn shock and visceral dysfunction and improve survival, especially for patients with more than half of the total body surface area (TBSA) burned [3].

The Parkland-Baxter formula is a frequently adopted clinical guideline for resuscitation during burn shock and can reduce the rate of acute renal failure resulting from hypovolemic shock [4,5]. Lactate Ringer's solution (LR), which has a lower sodium content (130 mEq/L) than extracellular fluid, is frequently used for resuscitation, but excess fluid volume can also cause tissue edema, hyponatremia, abdominal compartment syndrome, and other complications [6,7]. Hypertonic saline solution (HS) can be a substitute for LR, with advantages of lower fluid volume, rapid expansion, and fewer complications than an isotonic solution [8,9]. Hypertonic saline used alone or in combination with colloids and other drugs has anti-inflammatory effects and reduces the incidence of visceral hemorrhagic shock and sepsis in animal models [10,11]. Hypertonic lactate solution (240-300 mEq/L Na^+) has been successfully used to treat burn patients [12]. A recent study [13] showed that 3% HS (513 mEq/L Na^+) was superior to 7.5% HS (1283 mEq/L Na^+) and LR in terms of efficacy and safety in patients with hypovolemic shock. However, a death case, which was related to hypernatremia and hyperosmolar complications following HS resuscitation on severe burn patients was recorded [14]. Another report showed that excessive use of hypertonic saline solution can result in a rapid decrease in urine output when serum sodium is higher than 160 mEq/L or when osmotic pressure is higher than 340 mosm/L [15].

Regarding the advantages and disadvantages of HS solution, the question arises as to whether it is the best choice for burn patients in comparison with LR solution. Therefore, we addressed this in the current study using 400 mEq/L Na^+ HS, where sodium concentration was slightly higher than the Monafo formula computational requirements, in comparison with LR resuscitation. Sodium concentration, renal function, inflammatory mediators, and oxidative stress were examined to validate the effects of higher concentration of HS on the kidney.

2. Materials and methods

2.1. Reagents

NaCl for injection and LR were from Japan Otsuka Pharmaceutical Co. (Tokyo, Japan). Enzyme-linked immunosorbent assay (ELISA) kits for Cys-C, TNF-α, IL-1β, and HMGB 1 were from Applygen Technologies (Beijing, China). Superoxide dismutase (SOD) and malondialdehyde (MDA) kits were obtained from Nanjing Jiancheng Bioengineering Institute (Nanjing, China).

2.2. Animals

Female Sprague-Dawley rats (n=56, weight: 200-250 g) were from the Anhui Medical University Center of Experimental Animals (Hefei, China). Experiments were permitted by the Anhui Medical University Animal Care and Use Committee. Animals were given water and food ad libitum and housed in the laboratory for a week before they were used for experiments.

2.3. Experimental design

Rats were randomly distributed into sham (n=8) and experimental (n=48) groups, and were anesthetized by intraperitoneal injection of 10% chloral hydrate (300 mg/kg) with complete caudal vein catheterization. Rats in the sham group were submerged in 37 °C water without any fluid resuscitation, whereas those in the experimental group were scalded on the back with 98 °C boiling water for 12 s to cause a 30% TBSA burn. The burned rats were divided into two groups (n=24 per group) that were treated with LR and 400 mEq/L HS via tail vein injection. The content of 1 L of HS resuscitation fluid comprised 846 mL of LR with 154 mL of 10% NaCl. The HS resuscitation fluid consisted of Na^+ 400 mEq/L, K^+ 3.4 mEq/L, Ca^{2+} 2.5 mEq/L, and Cl^- 355 mEq/L. Rats in the LR group were resuscitated with LR in accordance with the Parkland formula (4 mL/kg body weight/1% TBSA burn), and 12.5% and 50.0% of the total volume was added before 2 and 8 h, respectively. The fluid applied during the first 24 h was administered based on the volume of hypertonic fluid, which was calculated to give the same sodium load as estimated by the Parkland formula (sodium 0.52 mEq/kg body weight/1% TBSA burn) in the HS group (1.3 mL/kg body weight/1% TBSA burn). Samples were collected 2, 8, and 24 h after resuscitation.

2.4. Determination of renal water content ratio

The rat kidneys were carefully washed with saline, surface moisture was removed, and they were then wet-weighed and placed into a 75 °C oven for 72 h to get the dry weight. Finally, the renal water content ratio was then calculated.

2.5. Histopathological analysis of the kidney tissue

The kidney tissues were embedded in 10% formaldehyde solution followed by dehydration, clearing, paraffin-embedding, and sectioning at a thickness of 3 μm. These sections were stained using hematoxylin and eosin, and the renal pathological morphology was observed under a light microscope (Leiea Co., Germany).

2.6. Measurement of serum sodium, renal function, and inflammation

The concentration of serum sodium and creatinine was measured using an automatic biochemical analyzer (Boehringer Mannheim, Germany). Serum Cys-C, TNF-α, IL-1β, and HMGB1 concentrations were examined by ELISA kits according to the manufacturer's instructions.

2.7. Measurement of oxidative stress

SOD activity and MDA content in the kidney tissue were examined by commercial kits according to the instructions of the manufacturer. Briefly, the tissue was weighed and homogenized in physiological saline at low temperature resulting in 10% (wt/vol) kidney homogenate. The

homogenate was centrifuged at 2500rpm for 10min at 4°C. The concentration of supernatant was diluted to 0.5% with normal saline to detect SOD activity. Another part of the 10% kidney tissue homogenate was measured for MDA content, and the actual MDA content was calculated based on the absorbance at 532nm.

2.8. Statistical analysis

The data are expressed as the mean±SD and were analyzed using SPSS 19.0 statistical software. Statistical analyses were performed using one-way analysis of variance, followed by S-N-K q test. $P<0.05$ was deemed significant.

3. Results

3.1. HS resuscitation protects against hyponatremia caused by severe burns

Serum sodium was measured to compare the two resuscitation fluids associated with hyponatremia in severe burn rats. LR resuscitation significantly decreased serum sodium concentration in the LR group relative to the sham group after burn injury ($P<0.01$, Fig. 1), but this decrease in sodium concentration was prevented in rats receiving HS resuscitation in comparison with the LR group after severe burn injury at each time point ($P<0.01$).

3.2. HS resuscitation reduces renal edema caused by severe burns

The renal water content ratio was calculated to estimate the renal edema between the two treatment groups. The renal water content ratio was increased in both the LR and HS groups after injury relative to that in the sham group and was higher in the LR group ($P<0.01$, Fig. 2). The water content ratio was increased at 2h after burn injury in the two treatment groups, with a peak at 8h, but declined slightly at 24h.

3.3. HS resuscitation improves renal histopathology after severe burn

Rats in the sham group had intact renal tubules as shown in Fig. 3A. Damage to the renal tubules was observed after a 30% TBSA burn in rats. Severe renal tubular injury and interstitial inflammation was demonstrated in the LR group at 8h after burn trauma (Fig. 3B). The deterioration of the renal tubule injury and edema were mitigated by HS treatment after burn (Fig. 3C). Fig. 3D shows the pathological injury score of the renal tubules of burned rats in each group. The renal tubule score in the LR group relative to the sham group after 8h was greatly increased, and it declined with HS resuscitation ($P<0.05$).

3.4. HS resuscitation reduces serum creatinine and Cys-C levels induced by severe burn injury

Compared to the sham group, serum concentrations of creatinine and Cys-C were increased to varying degrees in the LR and HS groups after burn trauma, with lower levels in the latter (Fig. 4A). Serum creatinine levels were similar between the two treatment groups 2h after injury ($P>0.05$) but differed significantly at 8h ($P<0.05$). Serum Cys-C concentrations were increased significantly in the LR group but reduced with HS resuscitation at 2h after injury, and this difference persisted for up to 24h ($P<0.01$, Fig. 4B).

3.5. HS resuscitation reduces serum TNF-α, IL-1β, and HMGB1 concentrations following burn injury

A large number of inflammatory factors were released into the blood after the injury. Serum levels of TNF-α and IL-1β began to increase in the two treatment groups at 2h and peaked at 8h before declining slightly at 24h. Compared to those in the LR group, the serum TNF-α and IL-1β levels were decreased at 2h in the HS group ($P<0.01$, Fig. 5A and B), and this effect persisted for up to 24h. The serum HMGB1 concentration was unaltered 2h after injury relative to that in the sham group but increased at 8 and 24h after injury. HMGB1 levels were similar between the two treatment groups 2h after injury but differed significantly at 8 and 24h ($P<0.01$, Fig. 5C).

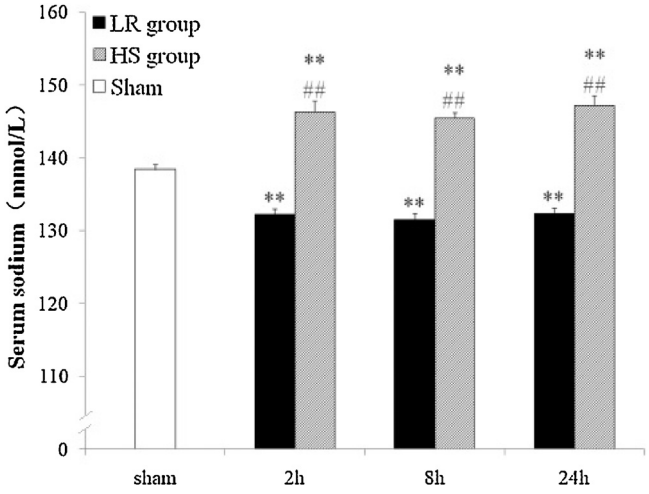

Fig. 1 – Effect of hypertonic saline resuscitation (400mEq/L) on serum sodium induced by 30% TBSA full-thickness burn. $^{**}P<0.01$, vs. sham group; $^{\#\#}P<0.01$, vs. LR group.

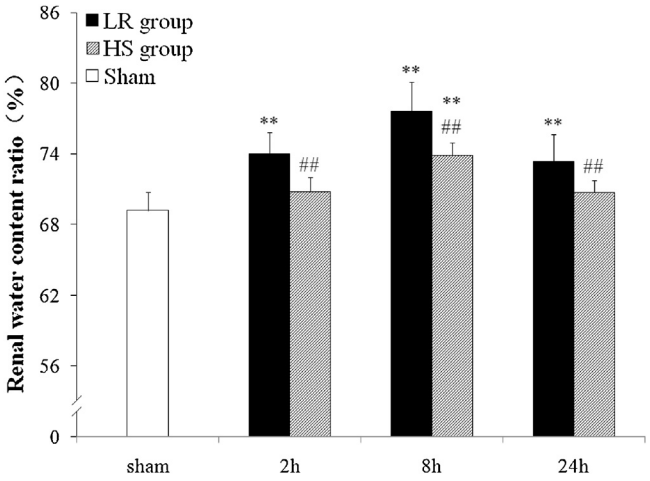

Fig. 2 – Effect of hypertonic saline resuscitation (400 mEq/L) on renal water content ratio induced by 30% TBSA full-thickness burn. $^{**}P<0.01$, vs. sham group; $^{\#\#}P<0.01$, vs. LR group.

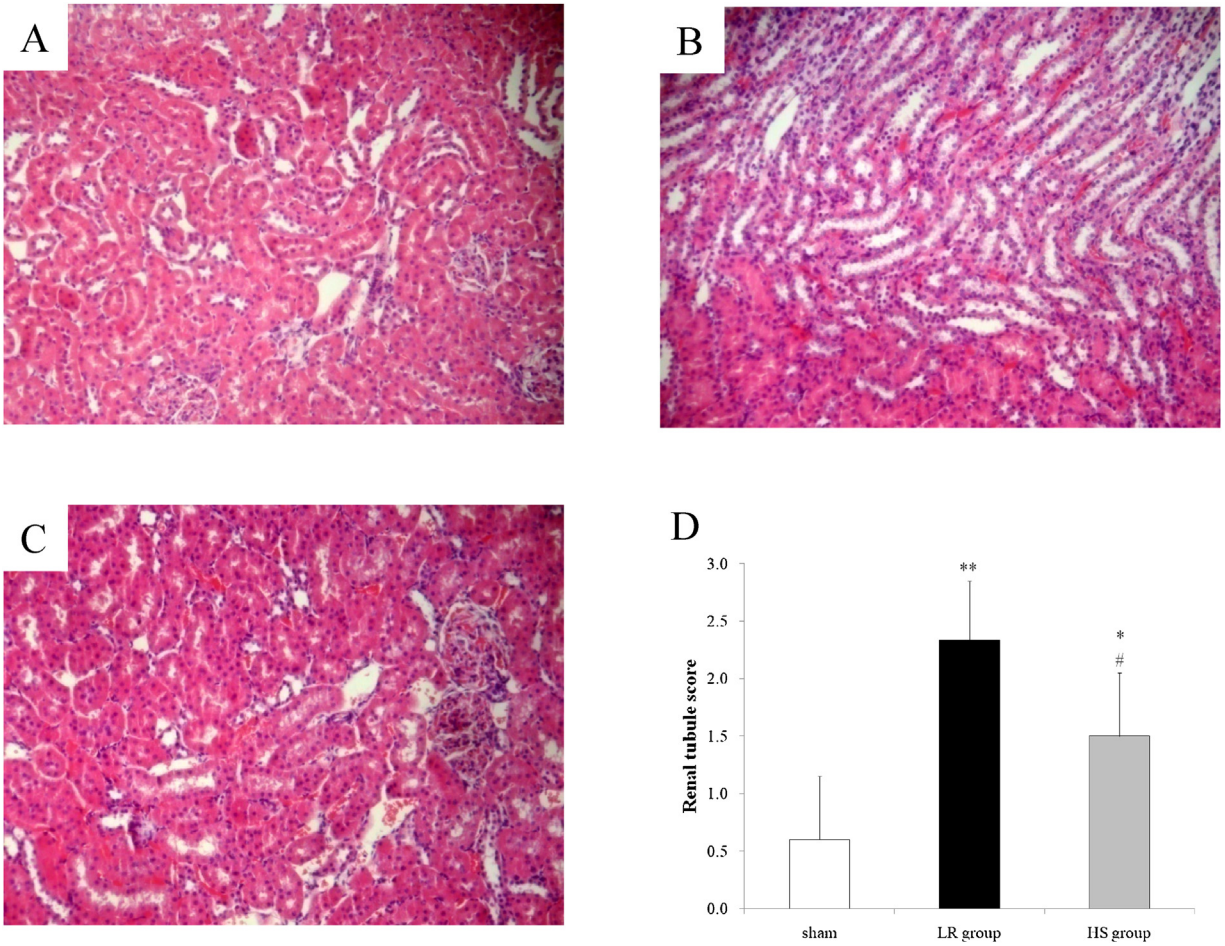

Fig. 3 – Effects of hypertonic saline resuscitation (400 mEq/L) on renal histopathology at 8 h after severe burn (100×). A: sham group. B: LR group. C: HS group. D: renal tubule score in each group. $^{*}P<0.05$, $^{**}P<0.01$, vs. sham group; $^{\#}P<0.05$, vs. LR group.

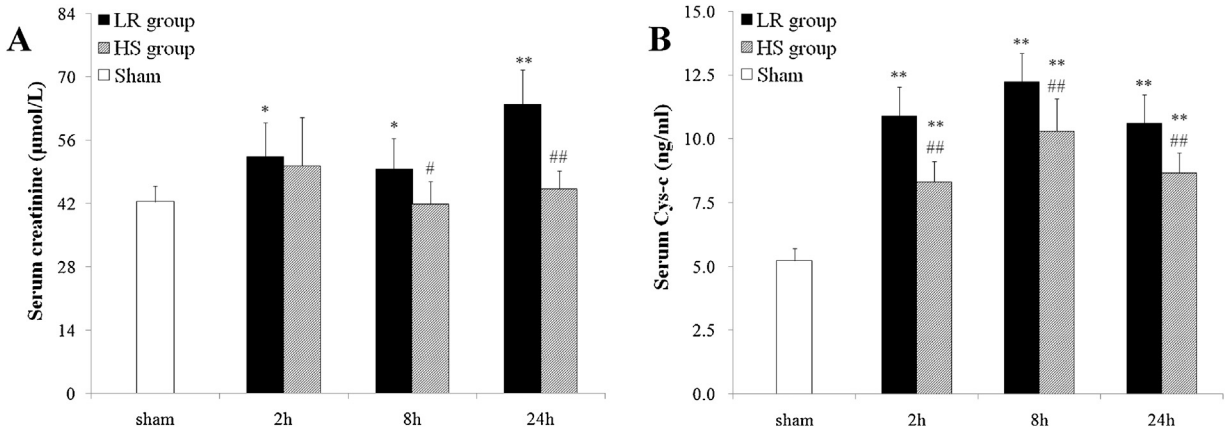

Fig. 4 – Effect of hypertonic saline resuscitation (400 mEq/L) on changes in renal function induced by 30% TBSA full-thickness burn. $^{*}P<0.05$, $^{**}P<0.01$, vs. sham group; $^{\#}P<0.05$, $^{\#\#}P<0.01$, vs. LR group.

3.6. *HS resuscitation increases SOD activity and reduces MDA accumulation in kidney following burn injury*

The SOD activity in the kidney began to decline in the two treatment groups 2 h after burn injury relative to the sham group and then increased at 8 h before declining once more at 24 h. The SOD activity was reduced with HS resuscitation compared to LR at each time point ($P<0.05$, Fig. 6A). The kidney MDA content in the LR group was significantly increased relative to the sham group after injury ($P<0.05$, Fig. 6B). The rats receiving HS resuscitation prevented these increases at 2, 8, and 24 h ($P<0.05$).

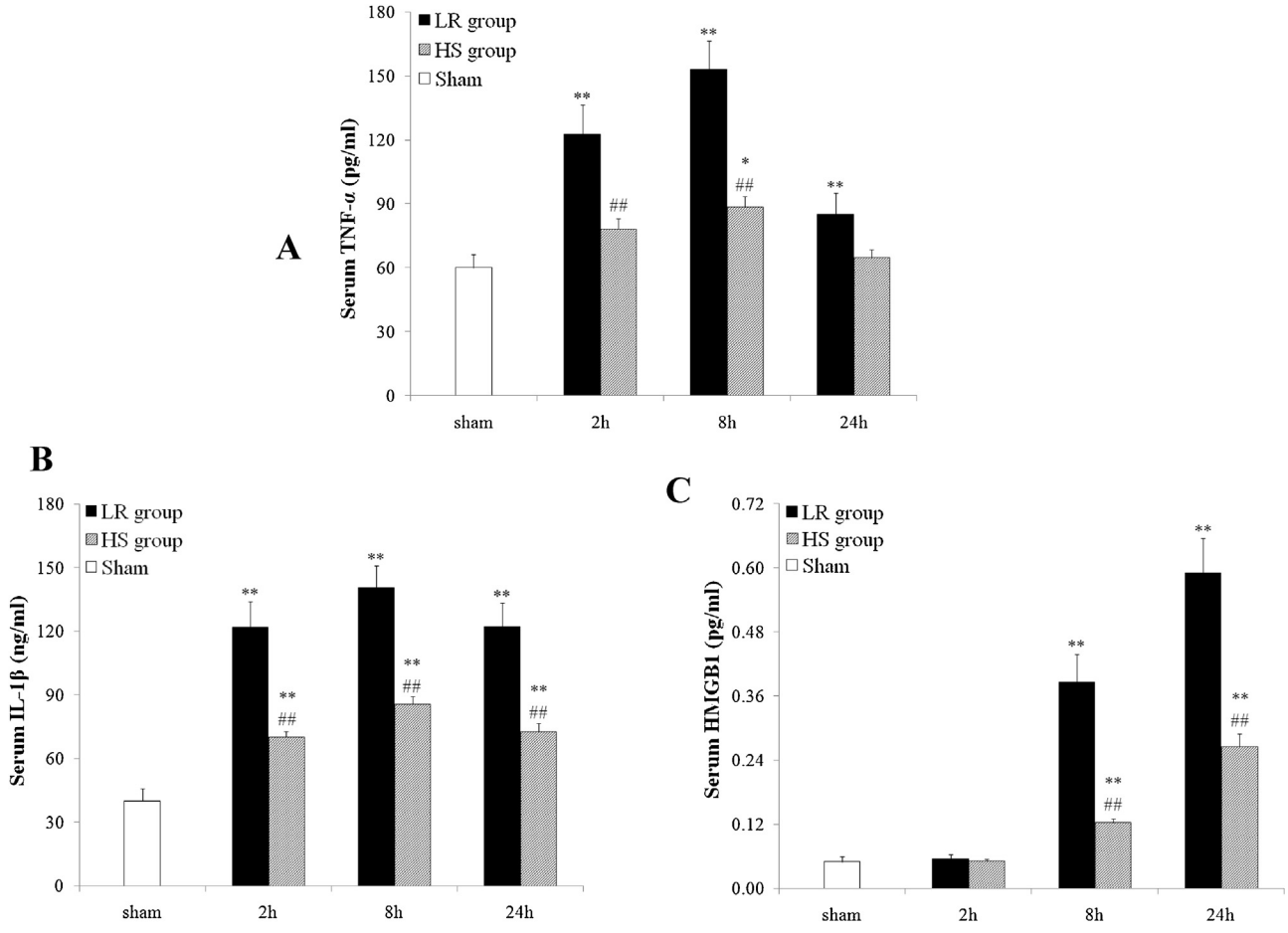

Fig. 5 – Effect of hypertonic saline resuscitation (400 mEq/L) on inflammatory response induced by 30% TBSA full-thickness burn. $^{*}P<0.05$, $^{**}P<0.01$, vs. sham group; $^{\#\#}P<0.01$, vs. LR group.

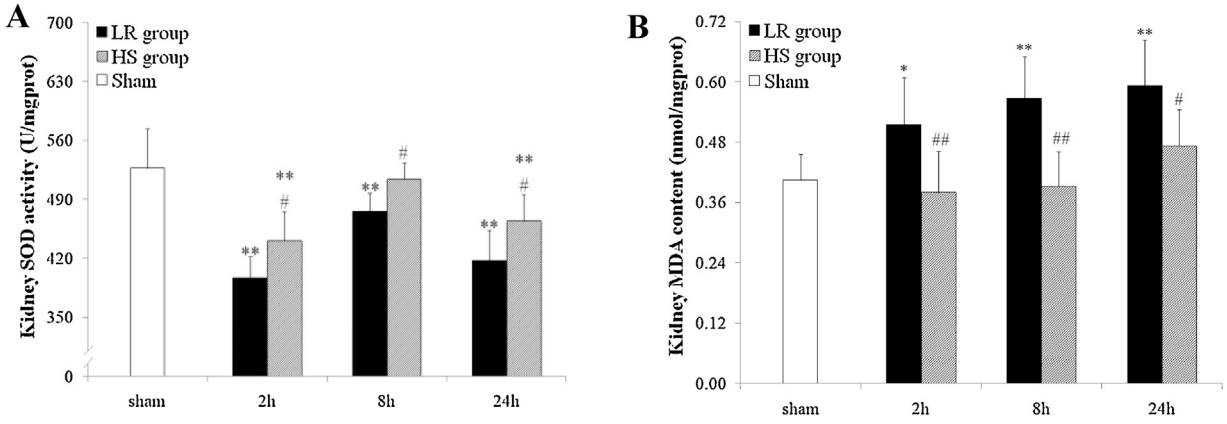

Fig. 6 – Effect of hypertonic saline resuscitation (400 mEq/L) on oxidative stress induced by 30% TBSA full-thickness burn. $^{*}P<0.05$, $^{**}P<0.01$, vs. sham group; $^{\#}P<0.05$, $^{\#\#}P<0.01$, vs. LR group.

4. Discussion

Disruption of the sodium-ATPase pump induced by severe burn results in sodium ions inflow, which causes the transfer of water along the osmotic gradient into the cell, thereby leading to hyponatremia [16]. Excessive fluid, with sodium content below the normal blood sodium level (142 mEq/L Na^+), entering the body fluid can aggravate hyponatremia and tissue cell edema. Therefore, the best method for quickly correcting hyponatremia and restoring the extracellular sodium concentration during the shock stage of burn is infusion with hypertonic saline. Hypertonic resuscitation with mild sodium concentration has been shown to successfully prevent hyponatremia caused by LR solution after burn injury [17]. In this study, 400 mEq/L HS abolished the incidence of hyponatremia after severe burn, and the total sodium depletion did not increase, indicating that the serum sodium concentration was less than 160 mEq/L.

The kidney is the most important excretory organ in the human body, which plays a vital role in maintaining the balance of water, electrolytes and acid-base, and the stability of the internal environment. Burns larger than 20% TBSA can lead to considerable loss of extracellular fluid and plasma albumin, which can induce hypovolemic shock and hypoalbuminemia; moreover, larger burn areas are associated with an increased risk of ARF [18,19]. Our previous study found that the use of hypertonic saline in severely burned rats can significantly reduce lung and intestinal edema [17,20]. HS has been confirmed to reduce brain edema caused by traumatic brain injury, thereby preventing secondary brain damage [21]. In this study, HS resuscitation (400 mEq/L Na^+) successfully decreased renal water content relative to LR, indicating that HS can effectively reduce renal edema caused by burn trauma. This was confirmed by our histopathological analysis, which revealed that renal tubular epithelial cell edema and renal tubular injury were improved by HS resuscitation (Table 1).

Serum creatinine is excreted through glomerular filtration into the urine, whereas the glomerulus provides the only means of removing Cys-C from the circulation. The levels of both molecules are markers of renal function reflecting glomerular filtration function [22,23]. In our study, 400 mEq/L HS reversed the increases in serum creatinine and Cys-C levels to a greater extent than LR in rats with burns covering 30% TBSA. The results were consistent with a previous study in which 7.5% HS alleviated renal dysfunction in a rat model of ischemia/reperfusion [24]. Other studies have reported that HS reduced the serum Cys-C level after coronary artery bypass grafting, thereby reducing the risk of AKI [25]. Thus, HS may preserve renal function after trauma by abrogating the reduction in GFR. The similar serum creatinine concentrations in the two treatment groups 2h after burn injury suggests that the creatinine level is not a sufficiently sensitive biomarker for early diagnosis of kidney injury, and that slight renal impairment does not trigger significant changes. In comparison, Cys-C is a more sensitive biomarker for early or mild kidney injury [26]. Indeed, we observed that serum Cys-C levels were significantly increased 2h after injury in both the treatment groups.

Various proinflammatory factors and cytokines are released following burn trauma that can induce systemic inflammation and cause organ damage. Kupffer cells are the primary source of TNF-α and IL-1β, which are associated with inflammation in the early stages of burns [27,28]. In our study, TNF-α and IL-1β concentrations were reduced by HS compared to LR treatment, suggesting that HS may reduce inflammation induced by burn injury. This is in agreement with a study demonstrating that treatment with 10% HS

Table 1 – Mc Whinnie's score of renal tubule injury.

Score	Representation
0	Normal structures
1	Tubular epithelial swelling, nuclear condensation, loss of the brush border with up to one third of the tubular profile exhibiting nuclear loss
2	More than one-third but less than two-thirds of the tubular profile exhibiting nuclear loss
3	Over two-thirds of the tubular profile exhibiting nuclear loss

reduced Na/K/C1 co-transporter 1 expression by suppressing the release of TNF-α and IL-1β by brain microglia, which in turn alleviated brain edema [29]. HMGB1, as an extracellular nuclear factor, regulates inflammation after burn injury [30]. Burns have been shown to increase plasma HMGB1 level, which affects immune function in burn patients [31]. In this study, the two treatment groups had similar serum HMGB1 concentrations 2h after burn injury. The increase in HMGB1 levels 8h after burn injury suggests that it is an important mediator of inflammation at later post-injury stages [32].

Oxidative stress induces inflammation and influences cellular metabolism, and is closely related to many pathophysiological processes including burn trauma [33]. SOD is an endogenous antioxidant that prevents oxygen free radical-induced injury by converting intracellular anions of oxygen (O_2^-) to less reactive hydrogen peroxide [34]. MDA is one of the most important products of membrane lipid peroxidation that can have cytotoxic effects on the kidney following burn trauma [35]. Reactive oxygen species-mediated oxidative stress and inflammation in kidney tissue can result in AKI in the early stages of severe burn injury [36]. As reported by a previous study [37], hypertonic saline/hydroxyethyl starch resuscitation reduced the consumption of SOD and prevented MDA accumulation in lung tissue, which prevented lung reperfusion-induced damage during hemorrhagic shock in rats. Our results showed that 400mEq/L HS has similar effects on the kidney tissue of rats with burn injury. Thus, HS resuscitation may reduce oxidative stress and protect against kidney damage caused by burn injury.

Dubose et al. found that traumatic patients resuscitated with 5% HS (856mEq/L Na^+) showed only transient hypernatremia, with no adverse sequelae for 3 days; this indicates that the concentration of the hypertonic saline used for this study was within a safe concentration range [38]. Another study reported that 3% HS had similar effects when used for resuscitation in comparison with 7.5% HS and LR, but led to a lower occurrence of renal failure, coagulopathy, and pulmonary edema [13]. However, Saeedi et al. showed that 3% HS accelerated death caused by uncontrolled hemorrhagic shock in goats, possibly due to the fact that HS can alter the blood coagulation system and accelerate bleeding [39]. Huang et al. reported that using 290mEq/L HS for severe burn patients was associated with hypernatremia and higher risk of renal failure [14], which may be due to the fact that HS is not suitable for resuscitation in patients with a large area burned (39.2±2.7% TBSA, inhalation injury 41.5%). Therefore, more research is necessary to prove the safety and effectiveness of hypertonic saline solution in clinical therapeutics.

Our study has some limitations. The single treatment concentration may be less satisfying, indicating that different concentrations of hypertonic resuscitation fluid should be evaluated to find the best therapeutic effect. Moreover, to better evaluate the therapeutic effect of HS, the survival rate of burned rats should be observed, and we hope to do this in a future study using a large animal-burned model.

In conclusion, the present study indicated that 400mEq/L HS solution is superior to LR for fluid resuscitation following burn trauma. Furthermore, HS can prevent hyponatremia and alleviate renal edema caused by burns, and protect renal function by reducing the release of inflammatory mediators and suppressing oxidative stress injury.

Conflict of interest

None.

Acknowledgments

We thank the laboratory technicians at ANHUI MED UNIV for their assistance with sample testing. This work was supported by the National Natural Science Foundation of China (grant numbers 81372050 and 81272092).

REFERENCES

[1] Palmieri T, Lavrentieva A, Greenhalgh DG. Acute kidney injury in critically ill burn patients. Risk factors, progression and impact on mortality. Burns 2010;36(2):205–11.
[2] Coca SG, Bauling P, Schifftner T, Howard CS, Teitelbaum I, Parikh CR. Contribution of acute kidney injury toward morbidity and mortality in burns: a contemporary analysis. Am J Kidney Dis 2007;49(4):517–23.
[3] Samuel JC, Campbell EL, Mjuweni S, Muyco AP, Cairns BA, Charles AG. The epidemiology, management, outcomes and areas for improvement of burn care in central Malawi: an observational study. J Int Med Res 2011;39(3):73–9.
[4] Greenhalgh DG. Burn resuscitation. J Burn Care Res 2007;28 (4):555–65.
[5] Benicke M, Perbix W, Lefering R, Knam F, Ipaktchi KR, Tannapfel A, et al. New multifactorial burn resuscitation formula offers superior predictive reliability in comparison to established algorithms. Burns 2009;35(1):30–5.
[6] Hayek S, Ibrahim A, Abu Sittah G, Atiyeh B. Burn resuscitation: is it straightforward or a challenge? Ann Burns Fire Disasters 2011;24(1):17–21.
[7] Santry HP, Alam HB. Fluid resuscitation: past, present, and the future. Shock 2010;33(3):229–41.
[8] Griswold JA, Anglin BL, Love Jr. RT, Scott-Conner C. Hypertonic saline resuscitation: efficacy in a community-based burn unit. South Med J 1991;84(6):692–6.
[9] Zhao JX, Wang B, You GX, Wang Y, Chen G, Wang Q, et al. Hypertonic saline dextran ameliorates organ damage in beagle hemorrhagic shock. PLoS One 2015;10(8):e0136012.
[10] Urbano J, Lopez-Herce J, Solana MJ, Del Castillo J, Botran M, Bellon JM. Comparison of normal saline, hypertonic saline and hypertonic saline colloid resuscitation fluids in an infant animal model of hypovolemic shock. Resuscitation 2012;83 (9):1159–65.
[11] Theobaldo MC, Barbeiro HV, Barbeiro DF, Petroni R, Soriano FG. Hypertonic saline solution reduces the inflammatory response in endotoxemic rats. Clinics (Sao Paulo) 2012;67 (12):1463–8.
[12] Monafo WW, Chuntrasakul C, Ayvazian VH. Hypertonic sodium solutions in the treatment of burn shock. Am J Surg 1973;126(6):778–83.
[13] Han J, Ren HQ, Zhao QB, Wu YL, Qiao ZY. Comparison of 3% and 7.5% hypertonic saline in resuscitation after traumatic hypovolemic shock. Shock 2015;43(3):244–9.
Huang PP, Stucky FS, Dimick AR, Treat RC, Bessey PQ, Rue LW, et al. Hypertonic sodium resuscitation is associated with

[14] renal failure and death. Ann Surg 1995;221(5)543–54 discussion 54–57.
[15] Herndon DN. Total burn care. 4th ed. London, United Kingdom: Saunders/ElSevier; 2012.
[16] Pham TN, Cancio LC, Gibran NS. American Burn Association practice guidelines burn shock resuscitation. J Burn Care Res 2008;29(1):257–66.
[17] Sun YX, Wu XS, Gao Z, Wang F, Liu S, Chen XL. Effect of 200 mEq/L Na+ hypertonic saline resuscitation on systemic inflammatory response and oxidative stress in severely burned rats. J Surg Res 2013;185(2):477–84.
[18] Holliday MA. Extracellular fluid and its proteins: dehydration, shock, and recovery. Pediatr Nephrol 1999;13(9):989–95.
[19] Kim GH, Oh KH, Yoon JW, Koo JW, Kim HJ, Chae DW, et al. Impact of burn size and initial serum albumin level on acute renal failure occurring in major burn. Am J Nephrol 2003;23 (1):55–60.
[20] Sun YX, Han LN, Gao Z, Wu XS, Zhou M, Wang F, et al. 200 mM hypertonic saline resuscitation attenuates intestinal injury and inhibits p38 signaling in rats after severe burn trauma. Burns 2017;43(8):1693–701.
[21] Rockswold GL, Solid CA, Paredes-Andrade E, Rockswold SB, Jancik JT, Quickel RR. Hypertonic saline and its effect on intracranial pressure, cerebral perfusion pressure, and brain tissue oxygen. Neurosurgery 2009;65(6)1035–41 discussion 41–42.
[22] Herrero-Morin JD, Malaga S, Fernandez N, Rey C, Dieguez MA, Solis G, et al. Cystatin C and bet-microglobulin: markers of glomerular filtration in critically ill children. Crit Care 200711 (3) R59.
[23] Ortuno-Anderiz F, Cabello-Clotet N, Vidart-Simon N, Postigo-Hernandez C, Domingo-Marin S, Sanchez-Garcia M. Cystatin C as an early marker of acute kidney injury in septic shock. Rev Clin Esp 2015;215(2):83–90.
[24] Brito MV, Yasojima EY, Percario S, Ribeiro RFJ, Cavalcante LC, Monteiro AM, et al. Effects of hypertonic saline solution associated to remote ischemic perconditioning in kidney ischemia/reperfusion injury in rats. Acta Cir Bras 2017;32 (3):211–8.
[25] Yousefshahi F, Bashirzadeh M, Abdollahi M, Mojtahedzadeh M, Salehiomran A, Jalali A, et al. Effect of hypertonic saline infusion versus normal saline on serum NGAL and cystatin C levels in patients undergoing coronary artery bypass graft. J Tehran Heart Cent 2013;8(1):21–7.
[26] Alharazy SM, Kong N, Saidin R, Gafor AH, Maskon O, Mohd M, et al. Serum neutrophil gelatinase-associated lipocalin and cystatin C are early biomarkers of contrast-induced nephropathy after coronary angiography in patients with chronic kidney disease. Angiology 2014;65(5):436–42.
[27] Chen XL, Xia ZF, Wei D, Han S, Ben DF, Wang GQ. Role of p38 mitogen-activated protein kinase in Kupffer cell secretion of the proinflammatory cytokines after burn trauma. Burns 2003;29(6):533–9.
[28] Jeschke MG, Barrow RE, Herndon DN. Extended hypermetabolic response of the liver in severely burned pediatric patients. Arch Surg 2004;139(6):641–7.
[29] Huang LQ, Zhu GF, Deng YY, Jiang WQ, Fang M, Chen CB, et al. Hypertonic saline alleviates cerebral edema by inhibiting microglia-derived TNF-alpha and IL-1beta-induced Na-K-Cl Cotransporter up-regulation. J Neuroinflammation 2014;11:102.
[30] Han SJ, Min HJ, Yoon SC, Ko EA, Park SJ, Yoon JH, et al. HMGB1 in the pathogenesis of ultraviolet-induced ocular surface inflammation. Cell Death Dis 2015;6:e1863.
[31] Lantos J, Foldi V, Roth E, Weber G, Bogar L, Csontos C. Burn trauma induces early HMGB1 release in patients: its correlation with cytokines. Shock 2010;33(6):562–7.
[32] Chen XL, Sun L, Guo F, Wang F, Liu S, Liang X, et al. High-mobility group box-1 induces proinflammatory cytokines production of Kupffer cells through TLRs-dependent signaling pathway after burn injury. PLoS One 2012;7(11):e50668.
[33] Szczesny B, Brunyanszki A, Ahmad A, Olah G, Porter C, Toliver-Kinsky T, et al. Time-dependent and organ-specific changes in mitochondrial function, mitochondrial DNA integrity, oxidative stress and mononuclear cell infiltration in a mouse model of burn injury. PLoS One 2015;10(12):e0143730.
[34] Çevik Ö, Oba R, Macit Ç, Çetinel Ş, Kaya ÖTÇ, Şener E, et al. Lycopene inhibits caspase-3 activity and reduces oxidative organ damage in a rat model of thermal injury. Burns 2012;38 (6):861–71.
[35] Ozguc H, Tokyay R, Kahveci N, Serdar Z, Gur ES. Hypertonic saline dextran alleviates hepatic injury in hypovolemic rats undergoing porta hepatis occlusion. Shock 2003;19(4):383–7.
[36] Feng Y, Liu Y, Wang L, Cai X, Wang D, Wu K, et al. Sustained oxidative stress causes late acute renal failure via duplex regulation on p38 MAPK and Akt phosphorylation in severely burned rats. PLoS One 2013;8(1):e54593.
[37] Du Z, Jia H, Liu J, Zhao X, Xu W. Effects of three hydrogen-rich liquids on hemorrhagic shock in rats. J Surg Res 2015;193 (1):377–82.
[38] Dubose JJ, Kobayashi L, Lozornio A, Teixeira P, Inaba K, Lam L, et al. Clinical experience using 5% hypertonic saline as a safe alternative fluid for use in trauma. J Trauma 2010;68(5):1172–7.
[39] Saeedi M, Hajiseyedjavadi H, Vahdati SS, Eslami V, Mokhtarpour M, Momeni M, et al. Hypertonic saline, normal saline or neither: which is best for uncontrolled hemorrhagic shock? An experimental study in goats. Ulus Travma Acil Cerrahi Derg 2013;19(6):500–6.