Available online at www.sciencedirect.com

SciVerse ScienceDirect

journal homepage: www.JournalofSurgicalResearch.com

The effects of dexmedetomidine on mesenteric arterial occlusion-associated gut ischemia and reperfusion-induced gut and kidney injury in rabbits

Kemal Kılıç, MD,[a] *Volkan Hancı, MD,*[b,*] *Şahbettin Selek, MD,*[c] *Mahmut Sözmen, DVM,*[d] *Nergiz Kiliç, MD,*[e] *Mehmet Çitil, DVM,*[f] *Derya Arslan Yurtlu, MD,*[g] *and B. Serhan Yurtlu, MD*[h,**]

[a] *Department of Surgery, School of Medicine, Kars Kafkas University, Pasacayiri 36100, Kars, Turkey*
[b] *Department of Anesthesiology and Reanimation, Çanakkale Onsekiz Mart University School of Medicine (Formerly Department of Anesthesiology and Reanimation, School of Medicine, Kars Kafkas University), Canakkale, Turkey*
[c] *Department of Biochemistry, Harran University School of Medicine (Formerly Department of Biochemistry, School of Medicine, Kars Kafkas University), Sanliurfa, Turkey*
[d] *Department of Pathology, School of Veterinary, Samsun Ondokuz Mayıs University (Formerly Department of Pathology, School of Veterinary, Kars Kafkas University), Kurupelit, 55139, Samsun, Turkey*
[e] *Department of Obstetrics and Gynecology, School of Medicine, Kars Kafkas University, Kars, Turkey*
[f] *Department of Internal Medicine, School of Veterinary, Kars Kafkas University, Kars, Turkey*
[g] *Clinic of Anesthesiology and Reanimation, Zonguldak Atatürk State Hospital, Zonguldak, Turkey*
[h] *Department of Anesthesiology and Reanimation, Zonguldak Karaelmas University School of Medicine, Zonguldak, Turkey*

ARTICLE INFO

Article history:
Received 15 December 2011
Received in revised form
29 March 2012
Accepted 30 March 2012
Available online 26 April 2012

Keywords:
Experimental mesenteric ischemia
Dexmedetomidine
Renal
Intestinal
Rabbit
Mesenteric artery occlusion

ABSTRACT

Objective: We assessed the antioxidant activity of dexmedetomidine (Dex) administered during the ischemic period in a rabbit model of mesenteric ischemia/reperfusion (I/R) injury using biochemical and histopathological methods.

Methods: A total of 24 male New Zealand white rabbits weighing between 2.5 and 3.0 kg were randomly divided into three groups: the sham group (Group S, $n = 8$), the I/R group (Group I/R, $n = 8$), and the I/R plus Dex treatment group (Group Dex, $n = 8$). In the I/R group, ischemia was achieved with 60 min of mesenteric occlusion. The sham group provided normal basal values. The rabbits in Group I/R were operated to achieve I/R. Group Dex received intravenous Dex 30 min after the commencement of reperfusion (10 μg/kg Dex was infused within 10 min, and then a maintenance dose of 10 μg/kg/h Dex was infused intravenously). For the measurement of tissue malondialdehyde, total antioxidant status, total oxidant status, lipid hydroperoxide levels, superoxide dismutase, catalase, and myeloperoxidase activity levels in the renal tissue samples of animals, the rabbits in each group were sacrificed 3 h after reperfusion. The histopathological examination scores were determined using the intestinal and renal tissues.

* *Corresponding author*. Esentepe Mahallesi, Cumhuriyet Caddesi, Yağmur Apartmanı, Apt No: 30, Daire: 24, Çanakkale, Turkey. Tel.: +90 530 643 32 40.
** *Corresponding author*. Zonguldak Karaelmas Universitesi, Tıp Fakültesi, Zonguldak Karaelmas Universitesi Hastanesi, S Blok Merkezi Ameliyathane, Kat: 3, Esentepe/Kozlu, Zonguldak, Turkey. Tel.: +90 505 570 12 01; fax: +90 372 263 23 83.
E-mail addresses: vhanci@gmail.com (V. Hancı), syurtlu@hotmail.com (B.S. Yurtlu).
0022-4804/$ – see front matter
doi:10.1016/j.jss.2012.03.073

Results: The mean malondialdehyde, total oxidant status, myeloperoxidase, and lipid hydroperoxide levels were significantly higher in Group I/R than in Groups S and Dex ($P < 0.05$). There also were significant decreases in the mean total antioxidant status, catalase, and superoxide dismutase activities in Group I/R compared with Groups S and Dex ($P < 0.05$). The histopathological examination scores of the intestinal and renal tissues were significantly higher in Group I/R compared with Groups S and Dex ($P < 0.05$).
Conclusion: Dex treatment may have biochemical and histopathological benefits by preventing I/R-related cellular damage of intestinal and renal tissues as shown in an experimental mesenteric ischemia model. The preference to use Dex for anesthesia during the mesenteric ischemia procedure may attenuate I/R injury in intestinal and renal tissues.

1. Background

Acute mesenteric arterial occlusion and acute mesenteric ischemia are medical and surgical emergencies that require an immediate diagnosis and intervention [1–4]. Even with a successful diagnosis and medical-surgical therapies, mortality rates are still high [3]. During ischemia/reperfusion (I/R) injury in acute mesenteric arterial occlusion, inflammatory factors are generated, cytotoxic substances are released, enzymes and immune cells are activated in intestinal tissue. Reperfusion injury results from the generation of toxic oxygen free radicals with the return of blood flow to the ischemic tissue [2,4–6]. The identification of experimental agents that can be administered as adjunctive therapy to surgery and rescue the intestinal I/R injury is clinically important [7–13]. However, in the clinical setting, mesenteric arterial occlusion is a surgical emergency, and patients with intestinal ischemia may need anesthesia for urgent surgical intervention. In addition, most of these patients with I/R injury require admission to the critical care unit (CCU). These experimental agents may not be available at the time of the operation or in the CCUs [14]. In these situations, the effects of the anesthetic and sedative agents on I/R injury may become an important issue. Previous studies have evaluated the antioxidant effects of the intravenous (IV) anesthetics, propofol and thiopental, in I/R-induced liver injury [4].

Dexmedetomidine (Dex) is a highly selective and potent alpha-2 adrenergic agonist that is increasingly used as an adjunct in general anesthesia [15]. In addition, it is an effective sedative, anxiolytic, and analgesic agent used in critical care patients requiring mechanical ventilation [16].

Dex can inhibit lipid peroxidation in cell membranes after I/R injury of various tissues [14,16–21]. However, to our knowledge, Dex has not been investigated in the mesenteric I/R injury model. We hypothesized that Dex infusion could have protective effects against ischemia-induced alterations in mesenteric arterial occlusion–reperfusion situations. Therefore, we investigated the antioxidant properties of Dex in a rabbit model of mesenteric I/R injury.

2. Methods

The study was approved by the Animal Ethics Committee of Kars Kafkas University Medical School. All animals were treated humanly in compliance with the recommendation of the animal care committee of the University and the principles of laboratory animal care (NIH publication No. 85-23, revised 1985). The rabbits were housed in a temperature-controlled room (24 ± 1°C) on a 12 h light–dark cycle and were fed standard rabbit chow and water for 12 h before surgery.

Twenty-four male New Zealand white rabbits weighing between 2.5 and 3.0 kg were randomly divided into three groups of eight each. The sham group (Group S; $n = 8$) served as a control group for normal basal values; Group I/R ($n = 8$) consisted of rabbits with mesenteric I/R procedure via mesenteric artery occlusion alone, and Group Dex ($n = 8$) included rabbits with Dex treatment after the mesenteric I/R procedure.

2.1. Experimental protocol

All animals were fasted overnight and anesthetized by an intramuscular injection of 60 mg/kg ketamine HCl (Ketalar; Parke-Davis, Eczacibasi, Istanbul, Turkey). A heating pad and a heating lamp were used to keep the animals warm. The rabbits were allowed spontaneous breathing during surgery. A catheter was inserted in the dorsal ear vein of each rabbit. Lactated Ringer's solution (LR) (5 mL/kg) was administered intravenously to the rabbits to prevent dehydration. After obtaining blood samples, LR and drug infusions were administered through the same vein. Before midline laparotomy, the rabbit skin was shaved and the abdominal walls were cleaned with 10% povidone iodine. A midline abdominal incision was performed to expose the superior mesenteric artery (SMA), and the rabbits were divided into the three groups. In Group S (sham operated; $n = 8$), the SMA and collateral branches were divided from the celiac axis and the inferior mesenteric artery was isolated but not occluded. Then, the abdominal incision was closed, and the animals were followed for 3 h to simulate the I/R interval in the other groups. The rabbits were administered an IV bolus of LR (2 mL/kg).

In Group I/R ($n = 8$), the SMA was exposed carefully and occluded immediately distal to the aorta with collateral interruption for 60 min with atraumatic microvascular clamps as described elsewhere [6,22,23]. With this procedure, the terminal ileum, cecum, and right colon were rendered ischemic. After 60 min of ischemia, the clamps were removed and a 3-h reperfusion period began. The rabbits were administered an IV bolus of LR (2 mL/kg).

In Group Dex (I/R + Dex-treated group; $n = 8$), the same procedures as described for Group I/R were performed; then,

the rabbits received Dex (10 μg/kg; as Precedex®, 100 μg/mL diluted to a concentration of 25 μg/mL in normal saline) infused within 10 min using a calibrated electronic infusion pump (Infusomat Space, B. Braun Melsungen AG, Melsungen, Germany). Then, maintenance Dex was infused at a rate of 10 μg/kg/h for 30 min after the commencement of the reperfusion period. The dose of Dex (10 μg/kg) was determined from previous experimental I/R studies in which it was shown to improve outcome [14,24–26]. The 30-min waiting duration was chosen based on a previous study [6]. Mesenteric ischemia was confirmed when the mesenteric pulsations stopped and the intestines became pale. Reperfusion was confirmed by the return of pulsatile blood flow through the mesenteric vessels in the intestines that were directly subjected to I/R injury [6].

At the conclusion of each procedure, the rabbits were sacrificed. Terminal ileum and renal samples of animals in each study group were excised for histopathological and biochemical examinations.

2.2. Biochemical examinations

All tissues were washed two times with cold saline solution, placed into glass bottles, labeled, and stored in a deep freezer (−80°C) until processing. The tissues were homogenized in 10 volumes of 150 mM ice-cold KCl using a glass Teflon homogenizer (Ultra Turrax IKA T18 Basic, IKA Labortechnic, Staufen, Germany) for 2 min at 5000 rpm after cutting the tissues into small pieces with scissors. The homogenate was then centrifuged at 5000 g for 15 min, and the supernatant was analyzed.

2.3. Measurement of tissue total antioxidant status

Total antioxidant status (TAS) was determined using an automated colorimetric measurement method [27]. In this method, the hydroxyl radical, the most potent biological radical, is produced by the Fenton reaction and reacts with the colorless substrate o-dianisidine to produce the dianisyl radical, which is bright yellowish-brown in color. On the addition of sample, the oxidative reactions initiated by the hydroxyl radicals in the mixture are suppressed by the antioxidant components of the sample, preventing the color change and thereby providing an effective measure of the total antioxidant capacity of the sample. The assay has excellent precision values, which are lower than 3%. The results are expressed as mmol Trolox equivalent/g protein for intestinal tissue samples.

2.4. Measurement of tissue total oxidant status

Total oxidant status (TOS) was determined using an automated measurement method [28]. The assay was calibrated with hydrogen peroxide, and the results are expressed in Lmol H_2O_2 equivalent/g protein for intestine tissue samples.

2.5. Measurement of tissue malondialdehyde levels

The tissue thiobarbituric acid (TBA)-reactive substance levels were determined by a method based on reaction with TBA at 90°C to 100°C [29]. The precipitate was pelleted by centrifugation, and an aliquot of the supernatant was incubated with an equal volume of 0.67% (wt/vol) TBA in a boiling water bath for 10 min. The results were expressed as nmoles per gram wet tissue, according to the measurements from a standard solution (1,1,3,3-tetramethoxypropane).

2.6. Measurement of tissue lipid hydroperoxides

The tissue lipid hydroperoxide (LOOH) levels were determined by the ferrous ion oxidation-xylenol orange method as previously described [30]. The method is based on a known principle of the oxidation of Fe II to Fe III by LOOH under acidic conditions. The coefficient of variation for the measurement of LOOH levels was 5%. The LOOH levels were expressed as nmol/mg of wet tissue.

2.7. Measurement of tissue superoxide dismutase and catalase activity

Total (Cu–Zn and Mn) superoxide dismutase (SOD) (EC 1.15.1.1) activity was determined according to the method of Sun *et al.* [31]. One unit of SOD was defined as the amount causing 50% inhibition in the nitroblue tetrazolium reduction rate. The activity of SOD was termed as units per milligram protein. Catalase (CAT) activity was assayed in hemolysates of erythrocytes by monitoring the consumption of H_2O_2 at 240 nm, as described by Aebi [32].

2.8. Measurement of tissue myeloperoxidase activity

This method is based on the principal that myeloperoxidase (MPO) activity containing homogenate reduces o-dianisidine dihydrochloride in the presence of H_2O_2, and this reduced product has an absorbance of 460 nm. MPO activity was computed using o-dianisidine extinction quotient. The results were stated as units per gram wet tissue (U/g) [33].

2.9. Histopathological examinations

For histopathological examinations, the intestinal tissue samples were fixed in 10% neutral buffered formalin, dehydrated and embedded in paraffin wax, cut into 5-μm sections, and stained with hematoxylin–eosin (H&E). The histological sections were examined with an Olympus BX51 optical microscope (Olympus Optical Co., Osaka, Japan) and photographed. The intestinal lesions were graded in five levels of ischemic injury according the scoring system described originally by Chiu *et al.* [34] (Table 1). An established tubular injury score for the kidneys modified from Houghton *et al.* [35] also was used (Table 2).

2.10. Statistical analyses

The statistical analyses were performed using the Statistical Package for the Social Sciences version 16.0 for Windows (SPSS, Chicago, IL). All data are expressed as the mean ± standard deviation. Mann-Whitney U-tests were used for statistical analyses of data among all groups. $P < 0.05$ was considered significant.

Table 1 – Criteria for scoring intestinal mucosal injury [34].

Grade	Histological appearance
0	No pathological changes
1	Development of subepithelial Gruenhagen's space at the tip of the villus and broadening of villi
2	Extension of the space from the tips to the sides of the villi
3	Increased epithelial lifting and epithelial cell slough from the tips of the villi, preservation of lamina propria
4	Epithelial cell slough from both the tips and the sides of the villi, dilated capillaries, and partial mucosal necrosis of lamina propria
5	Disintegration of the lamina propria, ulceration, and hemorrhage

3. Results

The biochemical and histopathological results are shown in Figs. 1–4.

3.1. *Biochemical results*

Malondialdehyde (MDA), TOS, MPO, and LOOH levels were significantly higher in Group I/R than in Group S in the intestinal and renal tissue samples ($P < 0.05$). MDA, TOS, MPO, and LOOH levels in Group Dex were significantly lower than those in Group I/R in all tissue samples ($P < 0.05$). There was no significant difference between Groups Dex and S for the MDA, TOS, MPO, and LOOH levels in the studied tissue samples ($P > 0.05$; Figs. 1–4).

The TAS, CAT, and SOD activity levels in Group I/R were significantly lower than those in Groups S and Dex in both tissue samples ($P < 0.05$). There was no significant difference between Groups Dex and S for TAS, CAT, and SOD levels in study tissue samples ($P > 0.05$; Figs. 1–4).

3.2. *Histological results*

The intestinal and renal tissue samples were evaluated and the mucosal lesions were graded (Tables 1 and 2) according to the standard grading schemes of Chiu *et al.* [34] and Houghton *et al.* [35], respectively. The histological scores are summarized in Figs. 1–4.

Table 2 – Criteria for grading tubular injury [35].

Grade	Histological appearance
0	Normal histological structure
1	Desquamated tubular epithelial cells in small foci (<1% of total tubule population involved) with presence of focal granulovacuolar epithelial cell degeneration and granular debris in tubular lumina
2	Tubular epithelial cell necrosis and desquamation are prominent and involve 1% to less than 25% of cortical tubules
3	25% to less than 50% of proximal tubules are undergoing necrosis and desquamation with evidence of intact tubules
4	Above 50% of tubular necrosis

3.3. *Results from the intestinal tissue samples*

The intestines of Group S were normal in appearance with a normal histological structure of lamina epithelialis and lamina propria (Fig. 5). The tissue samples of Group I/R showed extended epithelial lifting reaching the base of villi accompanied by epithelial cell degeneration and necrosis with the presence of cellular debris in the intestinal lumen (Fig. 6). The lamina propria was infiltrated by mononuclear cells with evidence of hiperemia and dilated capillaries. Group Dex (Fig. 7) revealed minimal histological alterations in most areas; the findings were comparable with sham-operated animals. Only partial separation and mild subepithelial edema were observed in the histological examination of Group Dex. The comparisons of the grading scores (mean ± standard error of measurement) between the groups are shown in Figs. 1 and 2. The intestinal wall injury was significantly more severe in Groups I/R and Dex compared with Group S ($P < 0.05$). In Group Dex, the intestinal injury score was significantly lower than that in Group I/R ($P < 0.05$; Figs. 1 and 2).

3.4. *Results from the kidney tissue samples*

The kidneys of Group S showed clear H&E staining along the basement membranes of tubular epithelial cells and glomeruli without any signs of necrosis (Fig. 8). However, sections of the kidneys showed prominent changes in the tubular epithelial cells after I/R-induced intestinal injury (Group I/R). These alterations were characterized by extensive tubular epithelial necrosis with desquamation of the tubular epithelium into the tubular lumen. Some tubules showed complete loss of the lining cells; others showed single-cell necrosis with nuclear pyknosis and cytoplasmic eosinophilia. Some tubules were dilated, leading to increased amounts of protein in the tubular lumen. The changes were more pronounced in the inner cortex and outer medulla (Fig. 9).

Although the damage persisted in Group Dex, the kidneys showed a significant decrease in the amount of sloughed epithelial cells and tubular necrosis present in the outer medulla compared with the kidneys obtained from Group I/R (Fig. 10).

The tubular injury score was significantly higher in Groups I/R and Dex compared with Group S ($P < 0.05$). In Group Dex, the tubular injury score was significantly lower than that of Group I/R ($P < 0.05$; Figs. 3 and 4).

4. Discussion

Mesenteric ischemia is a medical and surgical emergency [1,2]. After reperfusion (reoxygenation), toxic oxygen free radicals are generated with the return of blood flow after ischemia. These free radicals react with lipids in the cell membrane and initiate the lipid peroxidation process, which is known to be responsible for the I/R injury. Identifying

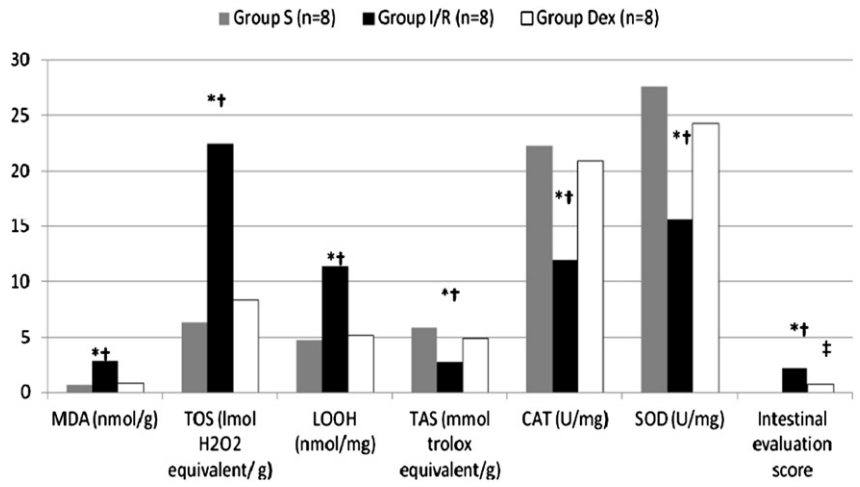

Fig. 1 – Tissue MDA, TOS, TAS, CAT, SOD, and LOOH levels and histopathological evaluation scores in terminal ileum tissue of the study groups. *P < 0.05; Groups S and I/R, Mann-Whitney U test. †P < 0.05; Groups DEX and I/R, Mann-Whitney U test. ‡P < 0.05; Groups S and DEX, Mann-Whitney U test.

therapeutic measures for alleviating the I/R injury and preserving cell integrity under other forms of oxidative stress would potentially be useful [4–13]. A number of chemicals and drugs, including oxygen radical scavengers, such as melatonin [7], ascorbic acid [8], L-carnitine [9], n-acetylcysteine [10], L-arginine [11], alpha-tocopherol [12], and resveratrol [13], have been successfully used to reduce I/R injury in animal models of mesenteric arterial occlusion, but few of them are currently used clinically because of severe adverse side effects [14]. In addition, these chemicals and drugs may not be available at the time of the operation [14,16]. In this situation, the effect of anesthetic or sedative agent that can already be used in anesthesia becomes an important issue for the reduction of I/R injury [4,14,16]. Kaplan *et al.* [4] studied the protective effects of propofol and thiopental in gut I/R-induced liver injury. Kaplan *et al.* demonstrated that propofol effectively stabilizes MDA levels and decreases the tissue injury scores of the liver and gut. This group concluded that propofol might offer advantages by inhibiting lipid peroxidation and inflammatory cytokine production in an animal model of gut I/R-induced liver injury [4]. Similarly, Vasileiou *et al.* [36] determined that using propofol for anesthesia efficiently prevents intestinal I/R-induced lung injury. The effects of ketamine on intestinal I/R injury have also been studied [37], and researchers have concluded that ketamine protects the intestine against intestinal I/R injury [37]. Similar findings with ketamine were observed by Cámara *et al.* [38].

Dex can inhibit lipid peroxidation in cell membranes after I/R injury of various tissues [14,16–21,25,39–47]. Previous studies have reported that Dex was effective in protecting rats and rabbits from complete or incomplete cerebral ischemia [19,20,43–46]. In our previous studies, we demonstrated that Dex mitigates reperfusion injury, and it has neuroprotective effects in spinal cord injury [25] and subarachnoid hemorrhage [42] models. It also was reported that Dex alters gene expression induced by cerebral ischemia and prevents late ischemic neural death [48]. Engelhard *et al.* [19] have shown in a rat cerebral I/R model that the apoptotic processes occurring after reperfusion can be prevented by Dex, and Dex increases the concentrations of the anti-apoptotic proteins Bcl-2 and Mdm-2. Furthermore, previous experimental studies have reported that Dex has a cardioprotective effect in an isolated rat heart global ischemia model, which was mediated by alpha-2 adrenergic stimulation [18]. Dex decreased myocardial oxygen demand and reduced oxygen deficiency in ischemic myocardium [49]. Willigers *et al.* [50] demonstrated that the sympatholytic effects of Dex decrease myocardial lactate release and, therefore, minimize emergence-related myocardial ischemia.

In our previous study, we showed that Dex treatments have potential biochemical and histopathological benefits by preventing I/R-related cellular damage in an experimental testicular torsion model in rats [14]. These findings

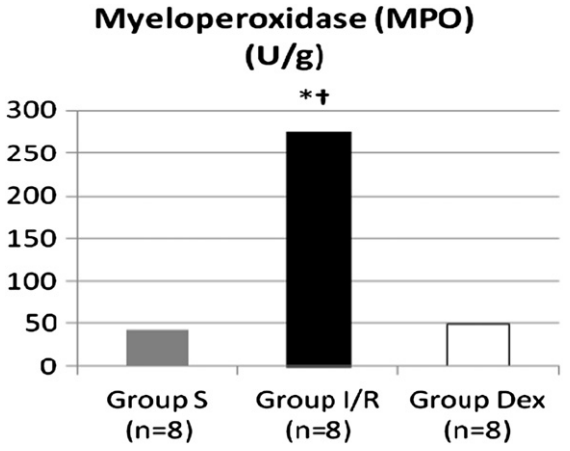

Fig. 2 – Tissue MPO levels in terminal ileum tissue of the study groups. *P < 0.05; Groups S and I/R, Mann-Whitney U test. †P < 0.05; Groups DEX and I/R, Mann-Whitney U test.

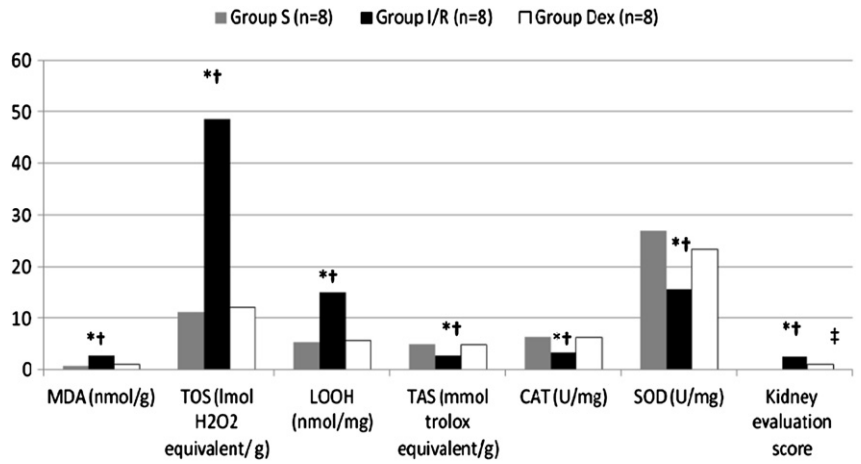

Fig. 3 – Tissue MDA, TOS, TAS, CAT, SOD, and LOOH levels and histopathological evaluation scores in renal tissue of the study groups.*P < 0.05; Groups S and I/R, Mann-Whitney U test. †P < 0.05; Groups DEX and I/R, Mann-Whitney U test. ‡P < 0.05; Groups S and DEX, Mann-Whitney U test.

were confirmed in a clinical human study, which demonstrated that Dex decreases I/R injury induced by tourniquet use [17].

However, there is no evidence in the published literature regarding the administration of Dex in an animal model of mesenteric I/R. We have now demonstrated that Dex could mitigate I/R-related cellular damage in intestinal and renal tissues of rabbits.

The mechanism of the protective effect of Dex against I/R injury induced by mesenteric artery occlusion is not clear. Previous studies have reported that the anti-ischemic effect of Dex may be associated with inhibition of ischemia-induced excess noradrenalin secretion via presynaptic alpha adrenoreceptors [20,43]. Dex can prevent the potential destructive effects of free-oxygen radical production by preventing the effects of noradrenalin on these presynaptic alpha adrenoreceptors [43,46].

It is known that tissue I/R injury activates families of protein kinases that regulate the expression of pro-inflammatory genes. The resulting products include enzymes (induced-nitric oxide [NO] synthase [iNOS], phospholipase A2, and cyclooxygenase), cytokines (Tumor necrosis factor-α [α], Interleukin-1 [IL-1], and Interleukin-6 [IL-6]), and adhesion molecules [2,51]. In previous studies, Dex administration decreased the production of TNF-α in ischemic hippocampal tissue [52] and the TNF-α and IL-6 concentrations of endotoxin-exposed rats [53]. We also demonstrated the same reduction in TNF-α and IL-6 levels in an experimental spinal cord injury [26]. These experimental data were supported with clinical studies demonstrating decreased TNF-α and IL-6 levels in critically ill sepsis [54] or postoperative major surgery [55] patients. Therefore, these anti-inflammatory effects of Dex can also be responsible for the prevention of mesenteric artery occlusion-induced I/R injury.

Among the free radicals produced during the I/R process, NO is very important. The excess production of NO through iNOS contributes to the pathophysiology of I/R in the intestine [2,56,57]. Previously, we demonstrated that Dex decreases inducible NO synthase levels in an experimental testicular torsion model in rats [14]. This antagonist effect of Dex may be another possible explanation for the protective effect of Dex against I/R injury in this study.

In addition, our results showed that Dex treatment significantly reduced renal injury compared with Group I/R. In an experimental study, Kocoglu *et al.* [21] demonstrated that Dex can reduce the renal injury caused by I/R of the kidney and concluded that Dex may be useful in enhancing the tolerance of the kidney against ischemia. Similarly, Gu *et al.* [58] showed that pre- or posttreatment with Dex provided cytoprotection and improved tubular architecture and function after renal ischemia. However, Billings *et al.* [59] determined that the alpha(2)-adrenergic receptor agonists Dex and clonidine protect mice against radiocontrast-induced nephropathy by preserving outer medullary renal blood flow. In another study, Marangoni *et al.* [60] evaluated the effects of Dex on renal function and histology after acute hemorrhage in rats and concluded that Dex results in a better renal function but higher tubular dilation scores after acute hemorrhage. Similarly, Villela *et al.* [61] investigated the

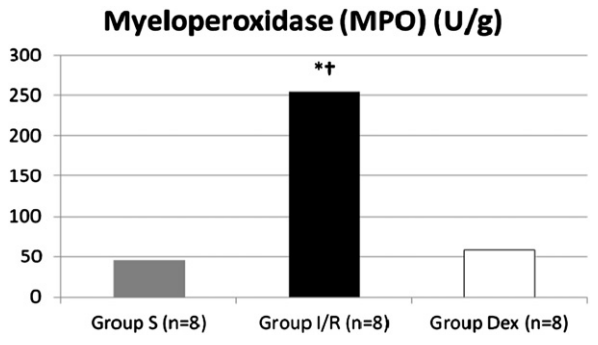

Fig. 4 – Tissue MPO levels in renal tissue of the study groups. *P < 0.05; Groups S and I/R, Mann-Whitney U test. †P < 0.05; Groups DEX and I/R, Mann-Whitney U test.

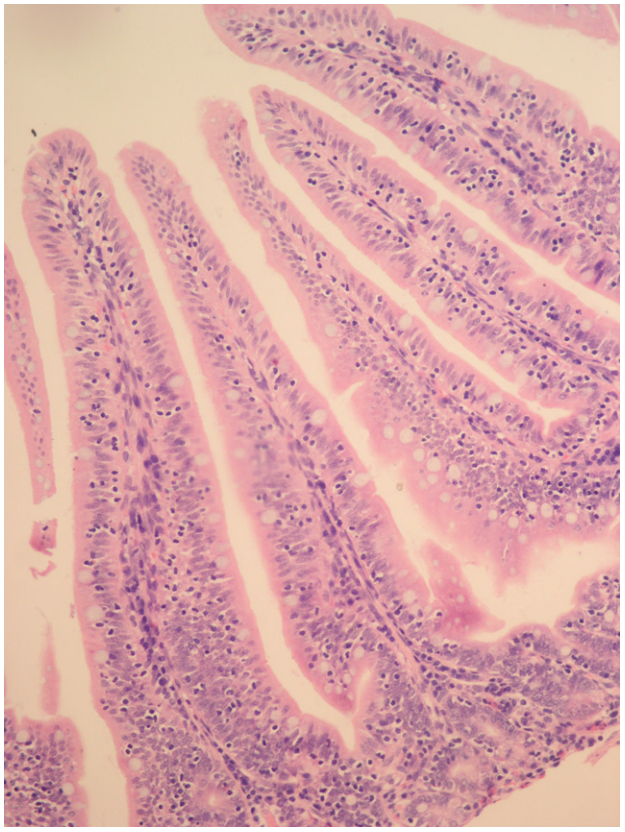

Fig. 5 – Normal intestinal mucosa and general aspect of intestinal villi and crypts from Group S. The sections were stained with H&E, ×20. (Color version of figure is available online.)

effects of Dex on the renal system and on vasopressin plasma levels in anesthetized dogs and found that low doses of Dex can protect kidneys during ischemia by inhibiting

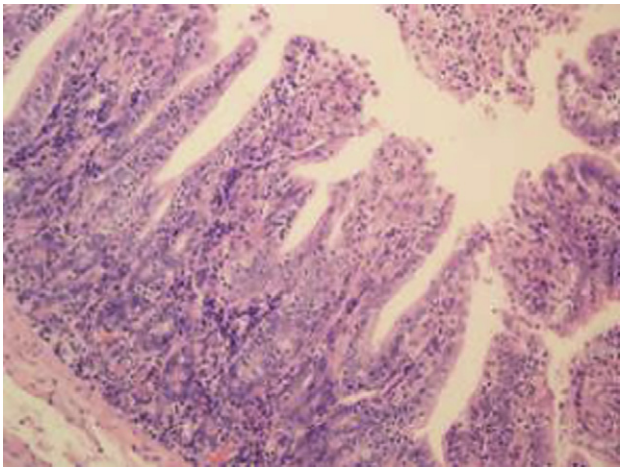

Fig. 6 – A representative tissue section from Group I/R depicting epithelial cell lifting extending toward the base of villi and epithelial cell slough from the tips of the villi. The lamina propria is preserved; grading score 3. The sections were stained with H&E, ×20. (Color version of figure is available online.)

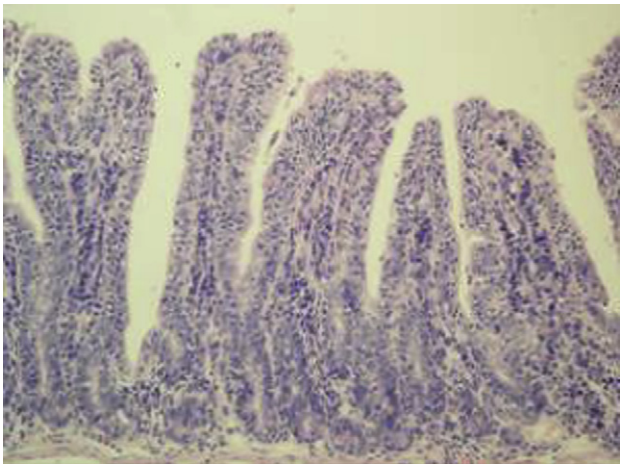

Fig. 7 – A representative tissue section of Dex-treated intestine showing histological changes consistent with sham-operated animals. The sections were stained with H&E, ×20. (Color version of figure is available online.)

vasopressin secretion and leading to aqueous diuresis. Our results in the present study are in accordance with the previous studies for the protective effects of Dex on renal tissue. However, Curtis *et al.* [62] noted that ketamine plus I/R injury was damaging to rat kidneys, according to histological changes, and Dex may not have completely protected the kidneys from these injuries.

Dex may cause bradycardia and hypotension at clinically relevant doses [41,63]; however, the optimal dose that does not produce adverse effects is not known. Although there are studies suggesting that low doses (3–6.5 μg/kg) are beneficial for the maximal inhibition of delayed neuronal death [24,25,52], several other studies have demonstrated neuroprotective effects at only higher doses (up to 100 μg/kg)

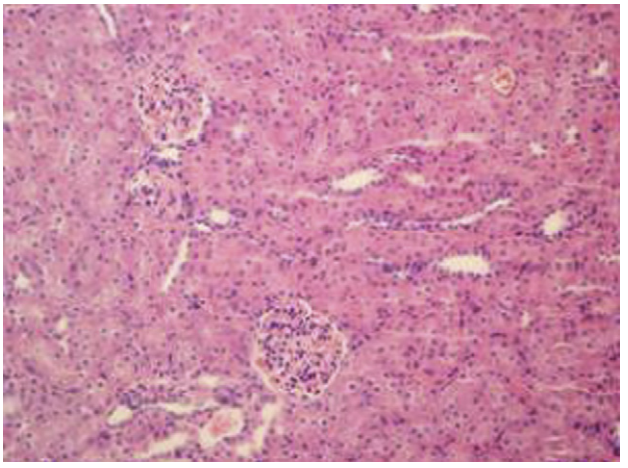

Fig. 8 – The renal histology of a sham-operated kidney showing normal glomeruli surrounded by Bowman's capsule and proximal and distal convoluted tubules. The sections were stained with H&E, ×20. (Color version of figure is available online.)

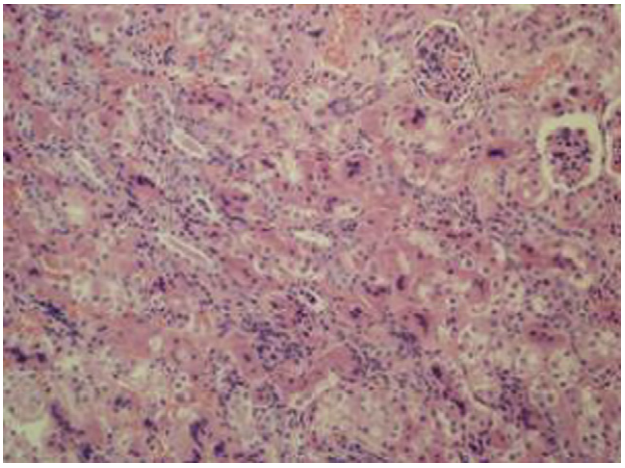

Fig. 9 – A tissue section of Group I/R showing wide-spread tubular necrosis and tubular epithelial cells shed into the tubular lumen. A dilated Bowman's space because of glomerular tuft contraction and the presence of congested vessels adjacent to tubules were observed. Some of the proximal tubule cells are degenerated and have vacuolated cytoplasm. The sections were stained with H&E, ×20. (Color version of figure is available online.)

[19,41–43,45]. In a study by Jolkkonen *et al.* [43], it was found that 9 μg/kg Dex was neuroprotective without significant side effects. In this study, we chose a 10-μg/kg dose because it was presumed that lower doses might be ineffective and higher doses would cause adverse effects [14,24–26].

Although our findings suggest that Dex is a promising anesthetic in cases of gut I/R-induced gut and kidney injury, our study had limitations. First, because it was impossible to perform this type of surgical intervention in rabbits without anesthesia, we did not have a negative control group [4]. Second, ketamine, which was the main anesthetic in our study, may have influenced the results by mitigating I/R injury and affecting our biochemical and histopathological results [37,38]. However, the ketamine effect on I/R injury is limited and dose dependant; ketamine only diminishes I/R injury-related changes in the intestine rather than abolishing cellular damage. Furthermore, this effect of ketamine did not have a protective role in our sham-operated group. Third, because of our limited laboratory facilities, we did not evaluate the possible benefits of Dex with more specific measurements, such as tissue nitrotyrosine and poly (adenosine di phosphate [ADP]-ribose) polymerase levels or immunohistochemical staining for apoptosis protease activating factor 1 antibody, iNOS, or endothelial NO synthase [4,14].

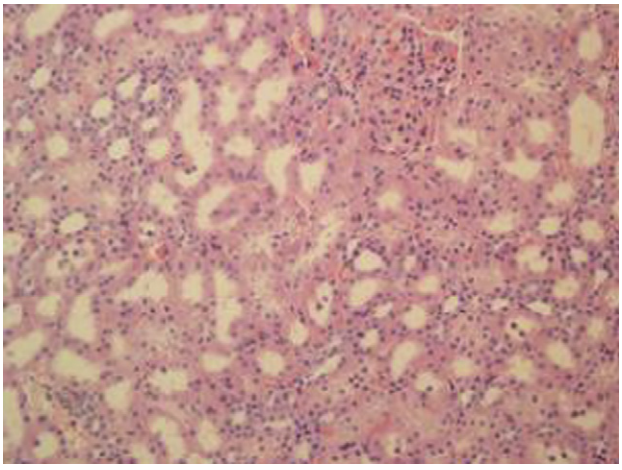

Fig. 10 – A section of kidney from a Dex-treated rabbit showing dilated proximal tubules, a decreased number of epithelial cell shedding and vacuolar degeneration. The sections were stained with H&E, ×40. (Color version of figure is available online.)

In conclusion, Dex administration during mesenteric arterial occlusion decreases I/R-related cellular damage. A preference for Dex as an anesthetic during the reperfusion procedure or after sedation during mechanical ventilation in the CCU has the potential to attenuate reperfusion injury. The clinical significance of these preliminary findings should be further investigated in human subjects.

Acknowledgments

Financial support: None.
Conflict of interest: None.

REFERENCES

[1] Sreedharan S, Tan YM, Tan SG, et al. Clinical spectrum and surgical management of acute mesenteric ischaemia in Singapore. Singapore Med J 2007;48:319.
[2] Cerqueira NF, Hussni CA, Yoshida WB. Pathophysiology of mesenteric ischemia/reperfusion: A review. Acta Cir Bras 2005;20:336.
[3] Stamatakos M, Stefanaki C, Mastrokalos D, et al. Mesenteric ischemia: Still a deadly puzzle for the medical community. Tohoku J Exp Med 2008;216:197.
[4] Kaplan N, Yagmurdur H, Kilinc K, et al. The protective effects of intravenous anesthetics and verapamil in gut ischemia/reperfusion-induced liver injury. Anesth Analg 2007;105:1371.
[5] Teke Z, Kabay B, Ozden A, et al. Effects of tempol, a membrane-permeable radical scavenger, on local and remote organ injuries caused by intestinal ischemia/reperfusion in rats. J Surg Res 2008;149:259.
[6] Kabay B, Teke Z, Aytekin FO, et al. Pyrrolidine dithiocarbamate reduces lung injury caused by mesenteric ischemia/reperfusion in a rat model. World J Surg 2007;31:1707.
[7] Ozacmak VH, Sayan H, Arslan SO, et al. Protective effect of melatonin on contractile activity and oxidative injury induced by ischemia and reperfusion of rat ileum. Life Sci 2005;76:1575.
[8] Higa OH, Parra ER, Ab'Saber AM, et al. Protective effects of ascorbic acid pretreatment in a rat model of intestinal ischemia-reperfusion injury: A histomorphometric study. Clinics (Sao Paulo) 2007;62:315.
[9] Yuan Y, Guo H, Zhang Y, et al. Protective effects of L-carnitine on intestinal ischemia/reperfusion injury in a rat model. J Clin Med Res 2011;3:78.
[10] Ayvaz S, Aksu B, Inan M, et al. The effects of N-acetylcysteine on intestinal ischemia/reperfusion injury in rats. Saudi Med J 2009;30:24.

[11] Taha MO, Miranda-Ferreira R, Fagundes AL, et al. Effects of L-nitro-arginine methyl ester, an inhibitor of nitric oxide biosynthesis, on intestinal ischemia/reperfusion injury in rabbits. Transplant Proc 2010;42:457.
[12] Yağmurdur MC, Ozdemir A, Ozenç A, et al. The effects of alpha - tocopherol and verapamil on mucosal functions after gut ischemia / reperfusion. Turk J Gastroenterol 2003;14:26.
[13] Petrat F, de Groot H. Protection against severe intestinal ischemia/reperfusion injury in rats by intravenous resveratrol. J Surg Res 2011;167:e145.
[14] Hanci V, Erol B, Bektaş S, et al. Effect of dexmedetomidine on testicular torsion/detorsion damage in rats. Urol Int 2010; 84:105.
[15] Paris A, Tonner P. Dexmedetomidine in anaesthesia. Curr Opin Anesthesiol 2005;18:412.
[16] İnci F, Doğan IV, Eti Z, et al. The effects of dexmedetomidine infusion on the formation of reactive oxygen species during mesenteric ischemia-reperfusion injury in rats. Marmara Medical Journal 2007;20:154.
[17] Yagmurdur H, Ozcan N, Dokumaci F, et al. Dexmedetomidine reduces the ischemia-reperfusion injury markers during upper extremity surgery with tourniquet. J Hand Surg Am 2008;33:941.
[18] Okada H, Kurita T, Mochizuki T, et al. The cardioprotective effect of dexmedetomidine on global ischaemia in isolated rat hearts. Resuscitation 2007;74:538.
[19] Engelhard K, Werner C, Eberspächer E, et al. The effect of the α 2-agonist dexmedetomidine and the N-methyl-d-aspartate antagonist S+-ketamine on the expression of apoptosis-regulating proteins after incomplete cerebral ischemia and reperfusion in rats. Anesth Analg 2003;96:524.
[20] Kuhmonen J, Pokornı J, Miettinen R, et al. Neuroprotective effects of dexmedetomidine in the gerbil hippocampus after transient global ischemia. Anesthesiology 1997; 87:371.
[21] Kocoglu H, Ozturk H, Ozturk H, et al. Effect of dexmedetomidine on ischemia-reperfusion injury in rat kidney: A histopathologic study. Ren Fail 2009;31:70.
[22] Dundar ZD, Cander B, Gul M, et al. Serum ischemia-modified albumin levels in an experimental acute mesenteric ischemia model. Acad Emerg Med 2010;17:1233.
[23] Kuzu MA, Tanik A, Kale IT, et al. Effect of ischemia/reperfusion as a systemic phenomenon on anastomotic healing in the left colon. World J Surg 2000;24:990.
[24] Hanci V, Karakaya K, Yurtlu S, et al. Effects of dexmedetomidine pretreatment on bupivacaine cardiotoxicity in rats. Reg Anesth Pain Med 2009;34:565.
[25] Gul S, Hanci V, Bahadir B, et al. The effectiveness of dexmedetomidine in experimental spinal cord injury compared to methylprednisolone in rats. J Clin Neurosci 2010;17:490.
[26] Can M, Gul S, Bektas S, et al. Effects of dexmedetomidine or methylprednisolone on inflammatory responses in spinal cord injury. Acta Anaesthesiol Scand 2009;53:1068.
[27] Erel O. A novel automated method to measure total antioxidant response against potent free radical reactions. Clin Biochem 2004;37:112.
[28] Erel O. A new automated colorimetric method for measuring total oxidant status. Clin Biochem 2005;38:1103.
[29] Conti M, Morand PC, Levillain P, et al. Improved fluorimetric determination of malondialdehyde. Clin Chem 1991;37:1273.
[30] Arab K, Steghens JP. Plasma lipid hydroperoxides measurement by an automated xylenol orange method. Anal Biochem 2004;325:158.
[31] Sun Y, Oberley LW, Ying L. A simple method for clinical assay of superoxide dismutase. Clin Chem 1988;34:497.
[32] Aebi H. Catalase in vitro. Meth Enzymol 1984;105:21.
[33] Bradley PP, Preibat D, Christerser RD, et al. Measurement of cutaneous inflammation: Estimation of neutrophil content with an enzyme marker. J Invest Dermatol 1982;78:206.
[34] Chiu CJ, McArdle AH, Brown R, et al. Intestinal mucosal lesion in low-flow states. I. A morphological, hemodynamic, and metabolic reappraisal. Arch Surg 1970;101:478.
[35] Houghton DC, Plamp CE 3rd, DeFehr JM, et al. Gentamicin and tobramycin nephrotoxicity. A morphologic and functional comparison in the rat. Am J Pathol 1978;93:137.
[36] Vasileiou I, Kalimeris K, Nomikos T, et al. Propofol prevents lung injury following intestinal ischemia-reperfusion. J Surg Res 2012;172:146.
[37] Guzmán-De La Garza FJ, Cámara-Lemarroy CR, Ballesteros-Elizondo RG, et al. Ketamine reduces intestinal injury and inflammatory cell infiltration after ischemia/reperfusion in rats. Surg Today 2010;40:1055.
[38] Cámara CR, Guzmán FJ, Barrera EA, et al. Ketamine anesthesia reduces intestinal ischemia/reperfusion injury in rats. World J Gastroenterol 2008;14:5192.
[39] Hoy SM, Keating GM. Dexmedetomidine: A review of its use for sedation in mechanically ventilated patients in an intensive care setting and for procedural sedation. Drugs 2011;71:1481.
[40] Riker RR, Shehabi Y, Bokesch PM, et al. SEDCOM (Safety and Efficacy of Dexmedetomidine Compared With Midazolam) Study Group. Dexmedetomidine vs midazolam for sedation of critically ill patients: A randomized trial. JAMA 2009; 301:489.
[41] Hancı V, Yurdakan G, Yurtlu S, et al. Protective effect of dexmedetomidine in a rat model of α-naphthylthiourea-induced acute lung injury. J Surg Res; 2012. doi:10.1016/j.jss.2012.02.027. online on 8 March.
[42] Ayoglu H, Gul S, Hanci V, et al. The effects of dexmedetomidine dosage on cerebral vasospasm in a rat subarachnoid haemorrhage model. J Clin Neurosci 2010;17:770.
[43] Jolkkonen J, Puurunen K, Koistinaho J, et al. Neuroprotection by the alpha2-adrenoceptor agonist, dexmedetomidine, in rat focal cerebral ischemia. Eur J Pharmacol 1999;372:31.
[44] Kuhmonen J, Haapalinna A, Sivenius J. Effects of dexmedetomidine after transient and permanent occlusion of the middle cerebral artery in the rat. J Neural Transm 2001; 108:261.
[45] Maier C, Steinberg GK, Sun GH, et al. Neuroprotection by the alpha 2-adrenoreceptor agonist dexmedetomidine in a focal model of cerebral ischemia. Anesthesiology 1993;79:306.
[46] Hoffman WE, Kochs E, Werner C, et al. Dexmedetomidine improves neurologic outcome from incomplete ischemia in the rat. Anesthesiology 1991;75:328.
[47] Yang CL, Tsai PS, Huang CJ. Effects of dexmedetomidine on regulating pulmonary inflammation in a rat model of ventilator-induced lung injury. Acta Anaesthesiol Taiwan 2008;46:151.
[48] Wittner M, Sivenius J, Koistinaho J. Alpha2-Adrenoreceptor agonist, dexmedetomidine, alters acute gene expression after global ischemia in gerbils. Neurosci Lett 1997;232:75.
[49] Roekaerts PM, Prinzen FW, De Lange S. Beneficial effects of dexmedetomidine on ischaemic myocardium of anaesthetized dogs. Br J Anaesth 1996;77:427.
[50] Willigers HM, Prinzen FW, Roekaerts PM, et al. Dexmedetomidine decreases perioperative myocardial lactate release in dogs. Anesth Analg 2003;96:657.
[51] Attuwaybi BO, Kozar RA, Moore-Olufemi SD, et al. Heme oxygenase-1 induction by hemin protects against gut ischemia/reperfusion injury. J Surg Res 2004;118:53.
[52] Eser O, Fidan H, Sahin O, et al. The influence of dexmedetomidine on ischemic rat hippocampus. Brain Res 2008;1218:250.

[53] Taniguchi T, Kurita A, Kobayashi K, et al. Dose- and time-related effects of dexmedetomidine on mortality and inflammatory responses to endotoxin-induced shock in rats. J Anesth 2008;22:221.
[54] Memis D, Hekimoglu S, Vatan I, et al. Effects of midazolam and dexmedetomidine on inflammatory responses and gastric intramucosal pH to sepsis, in critically ill patients. Br J Anaesth 2007;98:550.
[55] Venn RM, Bryant A, Hall GM, et al. Effects of dexmedetomidine on adrenocortical function, and cardiovascular, endocrine and inflammatory responses in post-operative patients needing sedation in the intensive care unit. Br J Anaesth 2001;86:650.
[56] Cuzzocrea S, Chatterjee P, Mazzon E, et al. Role of induced nitric oxide in the initiation of the inflammatory response after postischemic injury. Shock 2002;18:169.
[57] Montalto MC, Hart ML, Jordan JE, et al. Role for complement in mediating intestinal nitric oxide synthase-2 and superoxide dismutase expression. Am J Phy 2003; 285:G197.
[58] Gu J, Sun P, Zhao H, et al. Dexmedetomidine provides renoprotection against ischemia-reperfusion injury in mice. Crit Care 2011;15:R153.
[59] Billings FT 4th, Chen SW, Kim M, et al. alpha2-Adrenergic agonists protect against radiocontrast-induced nephropathy in mice. Am J Physiol Renal Physiol 2008;295:F741.
[60] Marangoni MA, Hausch A, Vianna PT, et al. Renal function and histology after acute hemorrhage in rats under dexmedetomidine action. Acta Cir Bras 2007;22:291.
[61] Villela NR, do Nascimento Júnior P, de Carvalho LR, et al. Effects of dexmedetomidine on renal system and on vasopressin plasma levels. Experimental study in dogs. Rev Bras Anestesiol 2005;55:429.
[62] Curtis FG, Vianna PT, Viero RM, et al. Dexmedetomidine and S(+)-ketamine in ischemia and reperfusion injury in the rat kidney. Acta Cir Bras 2011;26:202.
[63] Hanci V, Erdoğan G, Okyay RD, et al. Effects of fentanyl-lidocaine-propofol and dexmedetomidine-lidocaine-propofol on tracheal intubation without use of muscle relaxants. Kaohsiung J Med Sci 2010;26:244.