Effect of dexmedetomidine up-regulating HIF-1α on intestinal mucosal barrier injury related to sepsis in rats

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Research Article

Keywords: sepsis, dexmedetomidine, HIF-1α, tight junction protein, intestinal mucosal permeability

Posted Date: March 21st, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1416385/v1

Abstract

Dexmedetomidine (DEX), a highly selective α_2 receptor agonist. DEX can inhibit the release of inflammatory factors in septic rats. It can also protect against H_2O_2 injury by up-regulating HIF-1α expression. The aim of this study was to explore whether DEX can up regulate HIF-1α protein level and plays a role in reducing septic intestinal mucosal injury. Twenty-four Sprague Dawley (SD) rats were randomly divided into four groups of 6 rats each: the sham group, sepsis group (subjected to cecal ligation and perforation, CLP), sepsis + dexmedetomidine group (DEX group, 30 μg/kg of DEX by intraperitoneal injection 30 minutes before and 2 hours after CLP), and sepsis + DEX + HIF-1α inhibitor Bay87-2243 (Bay87-2243 group, 30 μg/kg of DEX by intraperitoneal injection 30 minutes before and 2 hours after CLP and 9mg/kg of bay87-2243 by oral administration for 3 days before CLP. The HIF-1α and the tight junction protein (TJs) was detected by Western blot; the plasma concentrations of diamine oxidase (DAO), intestinal fatty acid binding protein (FABP2) and D-lactic acid (D-LAC) were detected by ELISA; the morphological changes of intestinal mucosa were detected by HE staining. DEX significantly increased the expression level of HIF-1α on intestinal mucosa in rats with sepsis injury, thus ameliorated intestinal mucosal pathological injury, reduced Chiu's score, decreased intestinal mucosal permeability, and up-regulated TJs protein expression. Moreover, effect on sepsis induced intestinal mucosal injury of DEX was reversed by HIF-1α inhibitor Bay87-2243. DEX could protect against sepsis-induced intestinal mucosal injury by up-regulating HIF-1α expression in rats.

Background

In recent years, the effect of intestinal dysfunction on sepsis outcome has attracted the attention of many researchers. Intestinal mucosal epithelial cells form a barrier to prevent intestinal bacteria and endotoxin from entering the blood system. Our study showed that early intervention can improve the intestinal mucosal barrier function and prolonged survival time of septic rats (1). Hypoxia inducible factor (HIF-1α) is a key transcription factor for human body to adapt to hypoxic environment and plays an important role in acute hypoxic response (2). Our previous study showed that DMOG, inhibitor of the prolyl-4-hydroxylase, and thus prevents HIF-1α degradation and has a protective effect on septic intestinal mucosal injury, while HIF-1α inhibitor (Bay87-2243) has the opposite effect (3). Dexmedetomidine (DEX) is a selective presynaptic membrane α_2 adrenoceptor agonists (4), with sedative and analgesic effects, are widely used in perioperative period. A small sample clinical study showed that DEX can promote postoperative intestinal function recovery by reducing the permeability of intestinal mucosa in patients with acute intestinal obstruction (5). The combination of DEX pretreatment and post-treatment can more effectively inhibit the release of inflammatory factors in septic rats (6). DEX could protect against H_2O_2 injury by up-regulating HIF-1α expression through activating PI3 K/Akt/mTOR signaling pathway in HK-2 cells (7). Therefore, this study intends to explore whether DEX can up regulate HIF-1α protein level and plays a role in reducing septic intestinal mucosal injury, and provides an experimental basis for the clinical application of DEX in the prevention and treatment of septic intestinal mucosal barrier injury.

Materials And Methods

Animals

Forty-eight SPF male Sprague Dawley (SD) rats weighing 200–250 g and aging 7 ~ 8ws were provided by the experimental animal center of Zhejiang Academy of Medical Sciences (license No. SCXK (Zhe) 2-14-0001). The experiment was completed in full within 20 days. The research protocal was discussed and approved by the Academic Committee and Animal Ethics Committee of Shaoxing People's Hospital (ethical approval No. ZJU2016007). The experiment was completed in the animal experimental center of Shaoxing People's Hospital (SYXK [Zhe] 2017-0007) in strict accordance with the standard operating procedures for the use of experimental animals set by Zhejiang University and the ARRIVE (guidelineshttp://www.nc3rs.org.uk/page.asp?id=1357). All animals were kept for at lease 3 days before experiment for them to adapt to the environment, with well balanced diet during the period. Feeding condition as follows: live in IVC environment with room temperature 20 ± 1 ℃, humidity 50 ~ 60%, 12 h: 12 h light / dark cycle, ventilation 8 ~ 15 times/h.

Reagents and kits

Sevoflurane (Jiangsu Hengrui Pharmaceutical Co., Ltd.), sufentanil citrate injection (Yichang humanwell Pharmaceutical Co., Ltd.), ropivacaine hydrochloride (AstraZeneca Pharmaceutical Co., Ltd.), diamine oxidase (DAO), intestinal type fatty acid binding protein 2 (FABP2) kit (cloud clone, USA), D-lactic acid (D-LAC) kit (abebio, Wuhan). The first antibodies of HIF-1α, ZO-1, occludin, claudin-1, GAPDH (Abcam Company), and BAY87-2243 were provided by MedChemExpress USA. Horseradish peroxidase (HRP)-labeled Goat anti rabbit IgG (second antibody, Jackson Company). SpectraMax Plus, Shanghai Megi molecular Instrument Co., Ltd.; The SPARK Multimode Microplate Reader, Tecan, Switzerland; gel imaging system (ChemiDoc XRS; American Company); Quantity one 4.6 image processing and analysis system (Chemidoc XRS, Biorad, USA).

Establishment of sepsis model rats

SD rats were allowed to adapt to the environment 3 days with free access to water and fasted for 6 hours before the experiment. A cecal ligation and perforation (CLP) model was established as follows: All animals were anesthetized via 5% sevoflurane inhalation and the peritoneal injection of 0.1 μg/kg sufentanil before undergoing surgical procedures, and their body temperatures were maintained at 36–38°C with a heating pad. After the skin had been prepared and disinfected, a 1.5-cm incision was made through the midline. Subsequently, the cecum was ligated with No. 1 silk thread 0.5 cm below the ileocecal valve of the cecum and proximal colon, and 9# needle penetrated the cecum twice and gently squeezed to extrude a small amount of feces from the perforation site. In order to prevent the pinhole from closing, leave No. 1 silk thread and suture the abdominal cavity. Ringer's solution 50 mL/kg was injected subcutaneously for fluid supplementation, and 0.5% ropivacaine 1 mL/kg was injected for postoperative analgesia after CLP. All the animals were allowed to eat and drink freely.

Grouping and modeling

Forty-eight SD rats were randomly divided into the sham group, sepsis group, sepsis + DEX group (DEX group) group and sepsis + DEX + HIF-1 inhibitor Bay87-2243 group (Bay87-2243 group), each of which contained 6 rats. CLP was used to establish a sepsis model. Rats in the DEX group were administered DEX 30 μg/kg by intraperitoneal injection 30 min before and 2 h after CLP, and those in Bay87-2243 group, Bay87-2243, an HIH-1 inhibitor, was orally administered at a dose of 9 mg / kg for 3 days before CLP. Rats in the sham and sepsis groups were were given the same volume of normal saline containing 10% ethanol and 40% Solutol HS-15 by oral gavage or same volume of normal saline by intraperitoneal injection. All reagents were stored at -20 ℃ and dissolved before use.

Specimen collection

and disposal. Under sevoflurane and sufentanil anesthesia, all animals underwent open surgery. A few 1-cm segments of the ileum were taken 2 cm away from the cecum, and some of these segments were used to prepare paraffin blocks for morphological study, while the others were stored at -80 ℃ and used to detect the expression HIF-1α and TJs proteins. Two milliliters of blood was taken from the abdominal aorta and placed in an EDTA anticoagulant tube. One hour later, the blood samples were centrifuged at 3000 R/min and 4 ℃ for 10 min, after which the plasma was separated and stored at -80 ℃ to assess intestinal mucosal permeability markers (DAO, FABP2, and D-LAC). Then, the rats were killed by cervical dislocation.

HE staining was used to detect morphological changes in the intestinal mucosa

After the paraffin-embedded blocks had been sectioned, stained with hematoxylin and erosin (HE) and sealed, the morphological changes in the small intestinal mucosa were observed under a light microscope, and the degree of small intestinal tissue damage was determined by calculating the Chiu's score.

Detection of plasma DAO, FABP2 and D-LAC by enzyme-linked immunosorbent assay (ELISA)

The plasma expression levels of DAO, FABP2, and D-LAC were detected with ELISA kits according to the instructions. The *A* at 450 nm was obtained with a microplate reader. A standard curve was drawn according to the *A*. The DAO, FABP2, D-LAC and inflammatory mediators contents in each sample were calculated, and the average values were taken.

Western blot detection of the expression of HIF-1α and TJs protein and expression in the intestinal mucosa

After treatment, the tissues were washed twice with phosphate-buffered saline (PBS) supplemented with PMSF and RIPA and lysed on ice for 30 minutes. Then, the sample was centrifuged for 15 minutes, and the supernatant was extracted. After quantification by BCA assay, 40 g of total protein was mixed with an equal volume of 2× sample buffer and boiled at 100 ℃ for 3 min for denaturation. The sample was

electrophoresed on a 6 ~ 12% SDS-polyacrylamide gel. The proteins were routinely transferred to a PVDF membrane that was sealed with TBST containing 5% bovine serum albumin at room temperature for 2 hours. Then, the PVDF membrane was washed with TBST 3 times for 5 minutes each. Anti-HIF-1α (1:1000), anti-ZO-1 (1:1000), anti-Occludin (1:1000), anti-Claudin-1 (1:1000), and anti-GAPDH (1:1000) antibodies were added and incubated overnight at 4℃. After being fully washed with TBST, the PVDF membrane was intubated with peroxidase–conjugated affinipure goat anti-rabbit antibody (1:5000) at room temperature for 2 hours, washed 3 times and developed with an ECL luminescence kit. The membrane was photographed with a gel imaging system, and the gray values were analyzed by Image J software.

Statistics

The data were analyzed with GraphPad prism (version 7.0). All experimental data are expressed as the mean (SD). One-way ANOVA was used to analyze the differences among groups. For data with a homogeneous distribution, LSD was used for pairwise comparison. For data with an inhomogeneous distribution, the Dunnett T3 test was used. $p < 0.05$ was considered statistically significant.

Results

Comparison of histopathological changes in intestinal mucosa between groups in rats

Sepsis appeared 6 hours after CLP, as observed through clutching, decreased activity, mucus secretion, the presence of erect hair, etc. Behavioral changes in the rats were observed and recorded every 4 hours after CLP. At 24 hours after CLP, one mouse in the Bay87-2243 group and sepsis group died respectively, but the other animals survived. Intestinal villi in sepsis group were disordered and exfoliated, and lamina propria hyperplasia was obvious, and a significantly increased Chiu's score ($p < 0.05$, Fig. 1). In the DEX group, the boundary of intestinal villi was clearer than that in sepsis group, the villi structure was complete, and the hyperplasia of lamina propria was obvious, and the Chiu's score was significently lower than that in the sepsis group ($p < 0.05$, Fig. 1). In the Bay 87-2243 group, the boundary of intestinal villi was disordered and fell off, and the lamina propria disintegrated, and and the Chiu's score was significantly increased compared to that in the DEX group ($p < 0.05$, Fig. 1).

Comparison of HIF-1 and TJS expression levels in intestinal mucosa between groups in rats

Compared with that in the sham group, the expression level of HIF-1α was significantly increased in the sepsis group, and the expression levels of ZO-1, occludin and claudin-1 were significantly decreased (Fig. 2; $p < 0.05$). Compared with the sepsis group, the DEX group exhibited significantly higher expression levels of HIF-1α and ZO-1, occludin and Claudin-1 in the intestinal mucosa (Fig. 2; $p < 0.05$). Furthermore, the expression level of HIF-1α was lower in the Bay87-2243 group compared with the DEX group, and the

protein expression levels of ZO-1, occludin and claudin-1 protein were significantly decreased compared to those in the DEX group (Fig. 2; $p < 0.05$).

Comparison of the plasma concentrations of DAO, FABP2, D-LAC between groups in rats

Compared with those in the sham group, the levels of DAO, FABP2, and D-LAC in the sepsis group were significantly increased. Additionally, compared with those in the sepsis group, the levels of DAO, FABP2, D-LAC in DEX were significantly decreased (Fig. 3; $p < 0.05$) in the DEX group, however, the levels of DAO, FABP2, D-LAC the Bay87-2243 group were increased in Bay 87-2243, when compared with DEX group (Fig. 3; $p < 0.05$).

Discussion

Sepsis is an organ dysfunction caused by the imbalance of the body's response to infection (8). In sepsis, the intestine, the first organ involved, was known as both the “initiating organ” and the "target organ" of sepsis (9). The intestinal mucosal barrier, composed of a monolayer of intestinal epithelial cells linked by TJs and a mucus layer mainly secreted by goblet cells, is the first line of defense against pathogenic and non pathogenic microorganisms (10). TJs proteins play an important role in maintaining intercellular connections and the cell barrier. TJs protein occludin is involved in adhesion and maintains the physical properties of the intestinal tract. Claudin-1 forms an ion-selective channel in cells that affects permeability to intercellular substances (11) while the TJs protein ZO-1 binds a variety of cytoskeletal proteins and plays a role in supporting the cytoskeleton (12). In this study, the expressions of the TJs proteins ZO-1, occludin and claudin-1 in the intestinal mucosa were decreased in septic rats, and the pathological morphology of the intestinal mucosa was destroyed, suggesting that sepsis led to the destruction of the morphology of intestinal mucosa, which were similar to our previous research results (3).

Upon intestinal mucosal injury and cells shedding, DAO and FABP2, which are mainly expressed in intestinal mucosal cells, are rapidly released into the blood circulation; thus they can be used as specific markers of intestinal mucosal injury (13). D-LAC is a metabolite of intestinal bacteria and when the permeability of the intestinal mucosa increases, D-LAC enters the blood, and because mammals cannot metabolize D-LAC, an increase in blood D-LAC concentration indirectly reflects an increase in intestinal permeability (13). In present study, the above mentioned plasma markers were increased in septic rats, compared to sham rats, suggesting that sepsis can lead to intestinal mucosal injury and increased intestinal permeability.

DEX is one of the commonly used drugs during the perioperative period of clinical anesthesia. When DEX combined anesthesia is used in patients with acute intestinal obstruction, it can reduce the intestinal mucosal permeability, shorten the anal exhaust time and shorten the postoperative hospital stay.[5)] A multicenter clinical study showed that DEX combined anesthesia promoted the recovery of intestinal

function (14). Another study showed that DEX had a protective effect on intestinal ischemia-reperfusion injury in rats by reducing intestinal microcirculation dysfunction, reducing mucosal cell apoptosis and tight junction injury, and maintaining the integrity of intestinal cell structure (15). In present study, the combined injection of DEX in septic rats can improve the pathological damage of intestinal mucosa, reduce the permeability of intestinal mucosa and up-regulate the expression of intestinal mucosal structural protein TJs. Therefore, it has a protective effect on septic intestinal mucosal injury.

In mice with colitis, AKB-4924, through prolyl hydroxylase (PHD) inhibition enhanced intestinal mucosal barrier function, but no protective effect on the intestinal mucosa was observed in HIF-1α- deficient mice, indicating that HIF-1α in the intestinal mucosa is a target of AKB-4924-mediated protection (16). Intestinal HIF-1α knockout in mice aggravated alcohol-induced destruction of the intestinal mucosal barrier (17). Knockout and overexpression experiments showed that HIF-1α plays an essential role in regulating claudin-1 expression at the gene level, revealing that claudin-1 may be an important target gene of HIF-1α. The abnormal expression of Claudin-1 in the HIF-1α-deficient intestinal mucosa led to an abnormal structure of TJ structure (18). Our previous study showed that HIF-1α improved intestinal mucosal injury in sepsis by alleviating the inflammatory response of sepsis and inhibiting the level of oxidative stress (3). Another study showed DEX could reduce endotoxin-caused oxidative stress injury to macrophages, improve mitochondrial function and inhibit mitochondrial apoptosis, and the mechanism may be related to up-regulating the expression of HIF-1α in mice (19). DEX treatment can maintain the dynamic balance of mitochondrial fusion/division by regulating HIF-1a/HO-1 signaling pathway, so as to improve endotoxin induced acute lung injury (20). In this study, after DEX treatment in septic rats, the expression of HIF-1α in intestinal mucosa up-regulated, thus alleviated the intestinal mucosal injury, but the HIF-1α inhibitor, Bay87-2243 counteracted the protective effect of DEX on intestinal mucosa. The results suggested that DEX can alleviate intestinal mucosal injury in septic rats by up regulating the expression of HIF-1α.

CLP model, a fast, simple, cheap and reproducible method, is the "gold standard" of sepsis model, which is characterized by being closer to the pathophysiological process of sepsis in human (21). Previous study showed that the inflammatory cytokines and multi organ function of septic rats changed significantly 24 hours after CLP. Therefore, 24 hours after CLP was selected as the exposure time in this study (22). The distribution half-life of DEX in rats after a single intravenous infusion is about 2 min, and the terminal half-life is 57 min (23); Martens *et al* (24) compared the protective effects of DEX administration at different times on sepsis-induced lung injury. The results showed that the effects of DEX pre-treatment and post-treatment on inflammatory factors, anti-inflammatory factors and JAK/STAT signal pathway were better than those of simple pretreatment or post-treatment. Therefore, this study adopted the combined scheme of DEX pre- and post-processing.

The results of this study provide a preliminary animal experimental basis for the possible protective effect of DEX on intestinal injury in sepsis through HIF-1α pathway, and provide the clues for further research. The pathophysiological mechanism of intestinal injury in sepsis is complicated, and moreover, this study also has some limitations. To further clarify the protective effect of DEX/HIF-1α on sepsis

associated intestinal mucosal barrier injury, HIF-1α knockout mice will be selected for further experiment. The specific pathway DEX/HIF-1α on sepsis intestinal mucosal barrier function needs to be further clarified.

Declarations

ACKNOWLEDGMENTS

This study was supported by Zhejiang Provincial Department of Science and Technology Fund (Grant No LY21H150001), Zhejiang Provincial Health Committee Fund (Grant No. 2020KY329), and Shaoxing Science and Technology Bureau Fund (Grant No. 2020A13014).

AVAILABILITY OF DATA AND MATERIALS

The datasets generated and/or analyzed during the current study are available from the corresponding author upon reasonable request.

AUTHORS' CONTRIBUTIONS

XL performed the experiments and made substantial contributions to the manuscript writing. BA performed the experiments and analyzed the data. LY and SZ made substantial contributions to conception and design and guided the experiments. HS designed the experiments. All the authors have read and approved the final manuscript. LJ and HS confirm the authenticity of the raw data.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

Not applicable.

CONFLICT OF INTEREST

The authors declare that they have no competing interests.

References

1. Zhu Y, Wang Y, Teng W, Shan Y, Yi S, Zhu S, Li Y. Role of aquaporin-3 in intestinal injury induced by sepsis. *Biol Pharm Bull.* (2019) 42: 1641-50. doi: 10.1248/bpb.b19-00073
2. Gojkovic M, Cunha PP, Darmasaputra GS, Barbieri L, Rundqvist H, Veliça P, Johnson RS. Oxygen-mediated suppression of $CD8^+$ T cell proliferation by macrophages: role of pharmacological inhibitors of HIF degradation. *Front Immunol*. (2021) 12:12:633586. doi: 10.3389/fimmu.2021.633586
3. He R, Teng WB, Yao LX, Shan Y, Zhu SM, Li YH. Protective effect of hypoxia inducible factor-1α on intestinal mucosal barrier in sepsis. *Chin J Clin. Pharmacol*. (2021) 26: 264-270 (Chinese). doi:10.12092/j.issn.1009-2501.2021.03.004

4. Yu X, Franks NP, Wisden W. Sleep and sedative states induced by targeting the histamine and noradrenergic systems. *Front Neural Circuits*. (2018) 12:4. doi: 10.3389/fncir.2018.00004
5. Zheng XH, He R, Ding QN, Wnag YL, Li YH. Effect of anesthetic factor on intestinal barrier function in patients with acute intestinal obstruction: dexmedetomidine-based anesthesia. *Chin J Anesthesiol*. (2020) 40: 395-8 (Chinese). doi:10.3760/cma.j.cn131073.20191217.00403
6. Martens EC, Neumann M, Desai MS. Interactions of commensal and pathogenic microorganisms with the intestinal mucosal barrier. *Nat Rev Microbiol*. (2018) 16:457-70. doi: 10.1038/s41579-018-0036-x.
7. Zhang JB, Wang XQ, Qiu XD, Ruan L, Huang HS. Protection effect of dexmedomidine against HO injury by up-regulating HIF-1α in human renal tubular apithelial cells. *Shiyong Yixue Zazhi*. (2016) 32: 1084-7 (Chinese). doi: CNKI:SUN:SYYZ.0.2016-07-016
8. Singer M, Deutschman CS, Seymour CW, Shankar-Hari M, Annane D, Bauer M, et al. The Third International Consensus Definitions for Sepsis and Septic Shock (Sepsis-3). *JAMA*. (2016) 315:801-10. doi: 10.1001/jama.2016.0287.
9. Meng M, Klingensmith NJ, Coopersmith CM. New insights into the gut as the driver of critical illness and organ failure. *Curr Opin Crit Care*. (2017) 23:143-8. doi: 10.1097/MCC.0000000000000386.
10. Glover LE, Lee JS, Colgan SP. Oxygen metabolism and barrier regulation in the intestinal mucosa. J Clin Invest. (2016) 126: 3680-8. doi: 10.1172/JCI84429
11. Donaldson GP, Lee SM, Mazmanian SK. Gut biogeography of the bacterial microbiota. *Nat Rev Microbiol*. (2016) 14: 20-32. doi: 10.1038/nrmicro3552
12. Pawłowska B, Sobieszczańska BM. Intestinal epithelial barrier: the target for pathogenic Escherichia coli. *Adv Clin Exp Med*. (2017) 26:1437-45. doi: 10.17219/acem/64883
13. Montagnana M, Danese E, Lippi G. Biochemical markers of acute intestinal ischemia: possibilities and limitations. *Ann Transl Med*. (2018) 6: 341. doi: 10.21037/atm.2018.07.22
14. Nie Y, Tu W, Shen X, Yu W, Yu Y, Song X, et al. Dexmedetomidine added to sufentanil patient-controlled intravenous analgesia relieves the postoperative pain after cesarean delivery: A prospective randomized controlled multicenter study. *Sci Rep*. (2018) 8: 9952. doi: 10.1038/s41598-018-27619-3
15. Zhang XK, Zhou XP, Zhang Q, Zhu F. The preventive effects of dexmedetomidine against intestinal ischemia-reperfusion injury in Wistar rats. Iran. J. *Basic Med Sci*.(2015) 18: 604-9. PMID: 26221485
16. Keely S, Campbell EL, Baird AW, Hansbro PM, Shalwitz RA, Kotsakis A, et al. Contribution of epithelial innate immunity to systemic protection afforded by prolyl hydroxylase inhibition in murine colitis. *Mucosal Immunol.* (2014) 7: 114-23. doi: 10.1038/mi.2013.29
17. Shao T, Zhao C, Li F, Gu Z, Liu L, Zhang L, et al. Intestinal HIF-1α deletion exacerbates alcoholic liver disease by inducing intestinal dysbiosis and barrier dysfunction. *J Hepatol.* (2018) 69: 886-95. doi: 10.1016/j.jhep.2018.05.021
18. Saeedi BJ, Kao DJ, Kitzenberg DA, Dobrinskikh E, Schwisow KD, Masterson JC, et al. HIF-dependent regulation of claudin-1 is central to intestinal epithelial tight junction integrity. *Mol Biol Cell*. (2015)

26: 2252-62. doi: 10.1091/mbc.E14-07-1194

19. Mao X, Chen HG, Yan MY, Feng JC, Wang GL, Xie KL, et al. Effect of dexmedetomidine on expression of hypoxia-inducible factor-1α during endotoxin-caused apoptosis in macrophages of mice. *Chin J Anesthesiol*. (2018) 38: 1505-8 (Chinese). doi: 10.3760/cma.j.issn.0254-1416.2018.12.026
20. Shi J, Yu T, Song K, Du S, He S, Hu X, Li X, et al. Dexmedetomidine ameliorates endotoxin-induced acute lung injury in vivo and in vitro by preserving mitochondrial dynamic equilibrium through the HIF-1a/HO-1 signaling pathway. *Redox Biol*. (2021) 41: 101954. doi: 10.1016/j.redox.2021.101954
21. Buras JA, Holzmann B, Sitkovsky M. Animal models of sepsis: setting the stage. *Nat Rev Drug Discov*. (2005).4: 854-65. doi: 10.1038/nrd1854
22. Weber GF, Chousterman BG, He S, Fenn AM, Nairz M, Anzai A, et al. Interleukin-3 amplifies acute inflammation and is a potential therapeutic target in sepsis. *Science*. (2015) 347: 1260-5. doi: 10.1126/science.aaa4268
23. Bol CJJG, Danhof M, Stanski DR, Mandema JW. Pharmacokinetic-pharmacodynamic characterization of the cardiovascular, hypnotic, EEG and ventilatory responses to dexmedetomidine in the rat. J Pharmacol Exp Ther. (1997) 283: 1051-8.

Figures

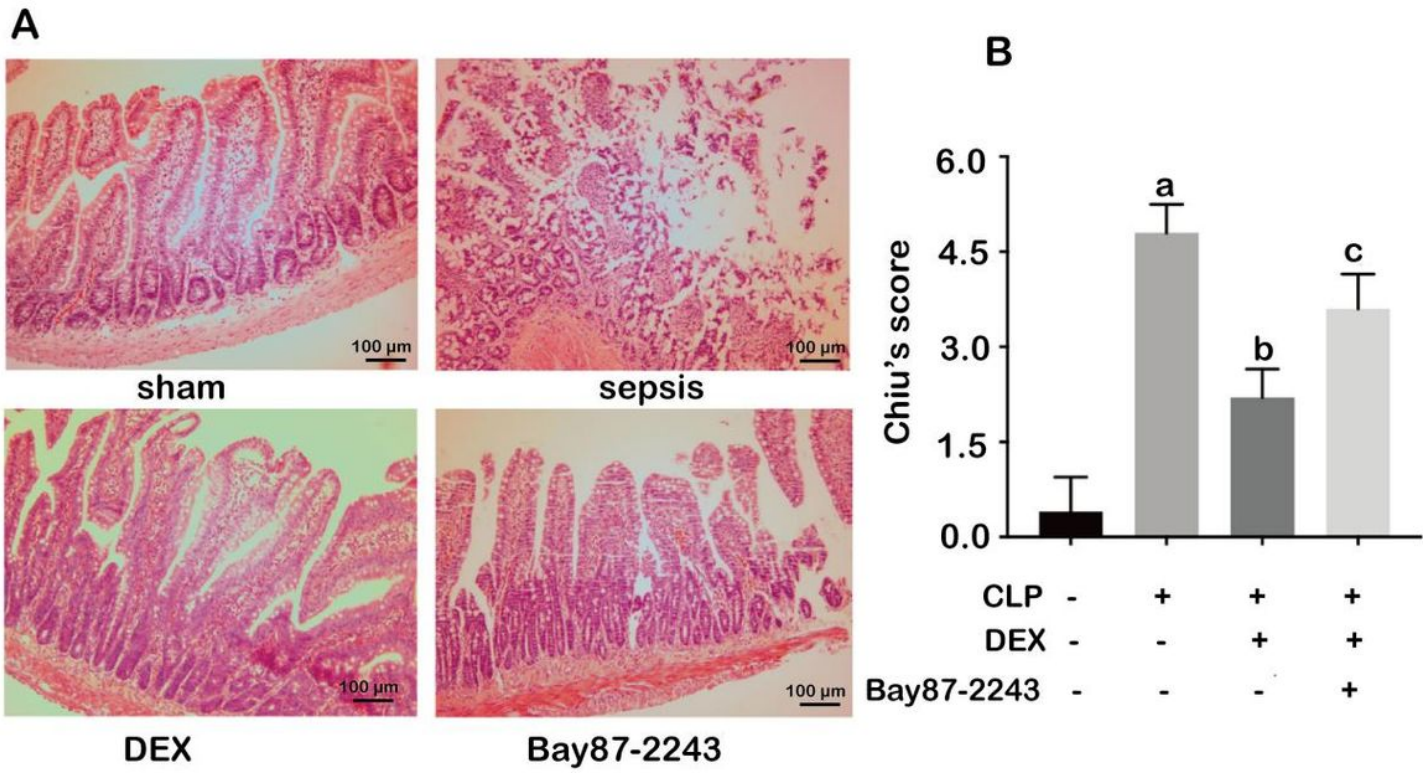

Figure 1

Comparison of histopathological changes in intestinal mucosa between groups in rats (×200) ($\bar{x}$ ± s, n = 24)

A: The morphological changes in intestinal mucosa in each group; B: the changes of Chiu's score in each group, and the statistical data were from four different representative experimental results. [a]$p < 0.05$, compared with sham group; [b]$p < 0.05$, compared with sepsis group; [c]$p < 0.05$, compared with DEX group.

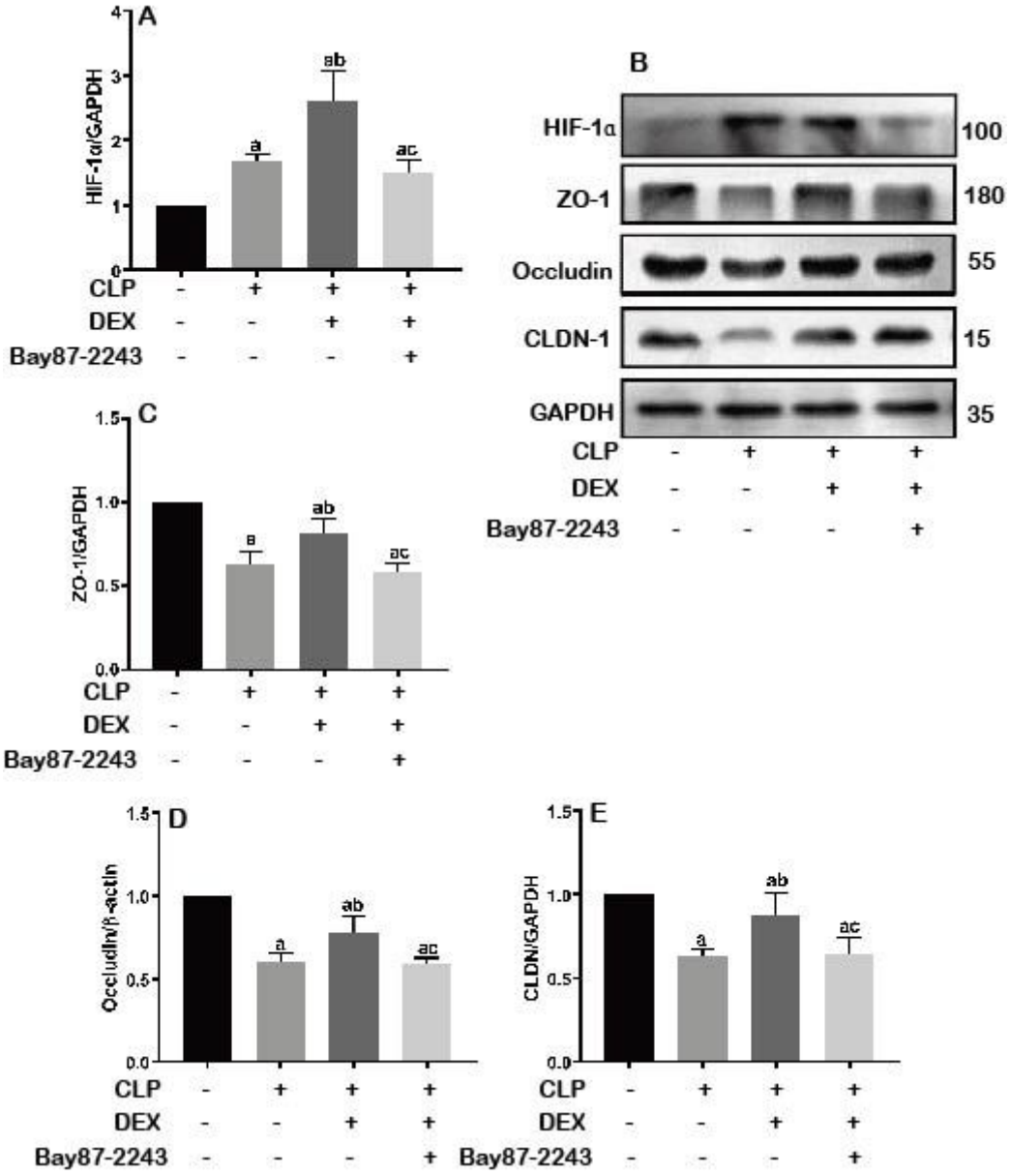

Figure 2

Comparison of HIF-1α and TJS expression levels in intestinal mucosa between groups in rats ($\bar{x} \pm s$, n = 24).

[a]$p < 0.05$, compared with sham group; [b]$p < 0.05$, compared with sepsis group; [c]$p < 0.05$, compared with DEX group.

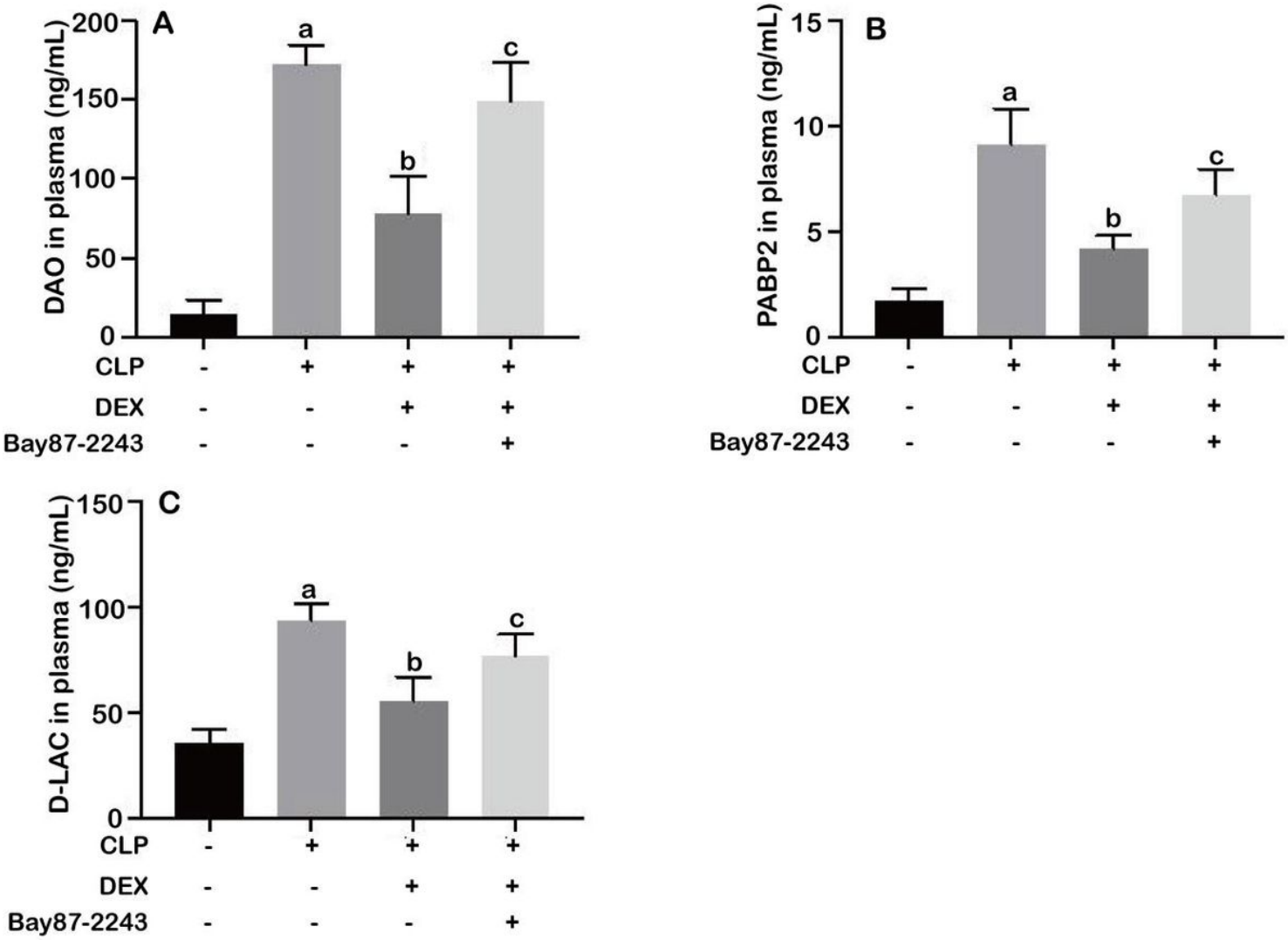

Figure 3

Comparison of the plasma concentration of DAO (A), FABP2 (B), D-LAC (C) between groups in rats ($\bar{x}$ ± s, n = 24).

[a]*p*< 0.05, compared with sham group; [b]*p*< 0.05, compared with sepsis group; [c]*p*< 0.05, compared with DEX group.