Original Article

Dexmedetomidine ameliorates ischemia-induced nerve injury by up-regulating Sox11 expression

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Background: Dexmedetomidine (Dex) is associated with several biological processes. Ischemic stroke has the characteristics of high morbidity and mortality. Herein, we aimed to explore whether Dex ameliorates ischemia-induced injury and determine its mechanism.

Methods: Real-time quantitative polymerase chain reaction (qRT-PCR) and western blotting were used to measure gene and protein expression. Cellular viability and proliferation were assessed by Cell Counting Kit-8 (CCK-8) and 5-ethynyl-2'-deoxyuridine (EdU) assays, respectively. Cell apoptosis was detected by flow cytometry. An oxygen-glucose deprivation/reoxygenation model of SK-N-SH and SH-SY5Y cells was constructed. A middle cerebral artery occlusion (MCAO) model was also built to assess Dex function *in vivo*. Neuronal function was assessed using the Bederson Behavior Score and Longa Behavior Score.

Results: We found that Dex positively and dose-dependently regulated Sox11 expression and prevented damage caused by oxygen-glucose deprivation/reoxygenation (OGD/R), enhancing cell viability and proliferation and reducing apoptosis in SK-N-SH and SH-SY5Y cells. The overexpression of Sox11 antagonized OGD/R-induced SK-N-SH and SH-SY5Y cell apoptosis and promoted cell growth in vitro. Furthermore, cell proliferation was decreased and cell apoptosis was increased after Sox11 knockdown in Dex-treated SK-N-SH and SH-SY5Y cells. We demonstrated that Dex prevented OGD/R-induced cell injury by up-regulating Sox11. Furthermore, we also confirmed that Dex protected rat from ischemia-induced injury in the MCAO model.

Conclusions: The role of Dex in cell viability and survival was verified in this study. Moreover, Dex protected neurons from MCAO-induced injury by up-regulating the expression of Sox11. Our research proposes a potential drug to improve the functional recovery of stroke patients in the clinic.

Keywords: Ischemic stroke; dexmedetomidine (Dex); Sox11; middle cerebral artery occlusion (MCAO); oxygen-glucose deprivation/reoxygenation (OGD/R)

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Introduction

Ischemic stroke causes high levels of mortality and disability worldwide (1), with many stroke patients requiring different levels of long-term assistance, which leads to a significant economic burden (2). Therefore, paying attention to this severe public health problem and the outcome of stroke is crucial. However, few studies have focused on novel therapeutic targets for the prevention and recovery from this devastating disease.

Several drugs have been reported to protect the brain from ischemia-induced injury (3-6). Sodium butyrate was reported to prevent middle cerebral artery occlusion (MCAO)-induced cell apoptosis by activating GPR41/Gβγ/PI3K/Akt signaling, which could be transformed in the clinic (7). Vitexin can suppress neuronal autophagy and prevent MCAO-induced cell injury by activating mechanistic target of rapamycin kinase (mTOR) signaling (6). Furthermore, it has been confirmed that berberine also improves brain injury following ischemia/reperfusion treatment by activating BDNF-TrkB-PI3K/Akt signaling (8). microRNA-153 was also associated with brain injury through the activation of sonic hedgehog signaling molecule (Shh) signaling and angiogenesis, as was reported previously (9). However, it is currently difficult to improve the functional recovery of stroke patients. Therefore, more clinical targets and potential drugs should be explored to improve the prognosis of these patients.

Dexmedetomidine (Dex) is associated with several biological processes; it can inhibit the inflammation of the neuronal system by regulating the transformation of M1/M2 polarization via activation of the mitogen-activated protein kinase kinase/mitogen-activated protein kinase (MEK/ERK) signaling pathway, which might protect neurons from injury (10). Dex was also found to improve cognitive function after surgery compared with the placebo group in an animal model, suggesting that it might protect neurons from injury (11). Moreover, it also significantly inhibited reactive oxygen species (ROS) production and suppressed the Cx43/PKC-α/NOX2/ROS pathway to improve lipopolysaccharide (LPS)-induced inflammation and prevent cell apoptosis (12). In a rat model, Dex was also shown to decrease the apoptosis ratio of cardiomyocytes induced by hydrogen peroxide (H_2O_2) (13). Furthermore, it might also be correlated with the development of cancer by influencing the hypoxia inducible factor (HIF) signaling and the PI3K/Akt pathway (14,15). Also, Dexmedetomidine is a promising agent for off-label use in palliative care (16). Therefore, it seems that Dex plays a vital role in cell survival and apoptosis, especially in the neuronal system. However, whether it is associated with neuronal injury and stroke development remains unclear.

Herein, we confirmed the role of Dex in cell viability and survival. The same results were also observed in the *in vivo* model. Study has shown that Sox11 increases reactivity in peripheral nerve injury with strong regenerative capacity, and played an important role in regulating neuron survival, proliferation and axon growth in the peripheral nervous system (17). And the expression of Sox11 could be modulated by the cell-type specific machinery in transient forebrain ischemia in the rat (18). Additionally, we also observed that Dex protected neurons from MCAO-induced injury by up-regulating SRY-box transcription factor 11 (Sox11) expression. Our research proposes a potential drug to improve the functional recovery of stroke patients in the clinic. We present the following article in accordance with the MDAR and ARRIVE reporting checklists (available at https://atm.amegroups.com/article/view/10.21037/atm-22-6639/rc).

Highlight box

Key findings

- SRY-box transcription factor 11 (Sox11) mediated the effects of dexmedetomidine (Dex) on neuron protection with the oxygen-glucose deprivation/reoxygenation (OGD/R) condition.

What is known and what is new?

- Dex is vital for cell survival and apoptosis, especially in the neuron system.
- However, whether Dex was associated with neuron injury and stroke development is still unclear. Thus, we aimed to explore whether Dex ameliorated ischemia-induced injury and determine its mechanism.

What is the implication, and what should change now?

- The role of Dex in cell viability and survival was verified. Moreover, Dex protected neurons from middle cerebral artery occlusion (MCAO)-induced injury by up-regulating Sox11 expression. Our research proposes a potential drug to improve the functional recovery of stroke patients in the clinic. However, the mechanism of Dex up-regulation of Sox11 remains unclear, and thus, future studies should focus on this problem. Moreover, the influence of Dex on neuronal function via several other targets should also be explored in the future.

Table 1 qRT-PCR primers

Gene name	Primer sequence (5'-3')	Length
Human-Sox11	F: CGGTCAAGTGCGTGTTTCTG	179 bp
	R: CACTTTGGCGACGTTGTAGC	
Human-GAPDH	F: CTCTGCTCCTCCTGTTCGAC	167 bp
	R: ATGGTGTCTGAGCGATGTGG	
Rat-Sox11	F: TCGTGATTGCAACAAAGGCG	132 bp
	R: TTGTGCCGTCTAAAGGGTCC	
Rat-GAPDH	F: AGAGACAGCCGCATCTTCTT	101 bp
	R: GGTAACCAGGCGTCCGATAC	

qRT-PCR, quantitative real-time polymerase chain reaction; F, forward primer; R, reverse primer.

Methods

MCAO

The MCAO procedure was conducted as previously described (19,20). Sprague-Dawley rats (female, 8 weeks old, 200–220 g) were obtained from the Model Animal Research Center of Nanjing University, China. The rats were placed in an anesthetic chamber and anesthetized with 3% isoflurane coupled with 30% oxygen and 70% nitrous oxide; 1.5% isoflurane was administered for anesthesia maintenance using a facemask. A 6-0 nylon filament coated with silicone rubber (Doccol, Sharon, MA, USA) was then introduced into the external carotid artery. The filament was pushed approximately 9–10 mm along the internal carotid artery to the bifurcation and origin of the middle cerebral artery (MCA) and was kept in place for 1 h.

Dex (Merck, No. 1179333, 100 μmol/kg) was then administered via the tail vein and blood flow to the MCA was restored for 24 h by removal of the filament (21). Control rats received the same surgical procedure but without MCAO. The animals were housed in a temperature-controlled (22±3 ℃) and humidified (60%±5%) environment with a 12-h light/dark cycle. Each group contained eight rats. This animal experiment was approved by the Institutional Animal Ethics Committee of Guangzhou Women and Children's Medical Center (No. S2022-107), in compliance with institutional guidelines for the care and use of animals. A protocol was prepared before the study without registration.

Cell culture

SK-N-SH and SH-SY5Y cells were obtained from the Shanghai Institute of Biochemistry and Cell Biology, Chinese Academy of Sciences (Shanghai, China) and grown in Dulbecco's modified Eagle medium (DMEM, Biosharp, BL308A, Beijing, China) containing 10% fetal bovine serum (FBS), 100 IU/mL penicillin, and 100 μg/mL streptomycin in a 37 ℃/5% CO_2 humidified incubator.

Oxygen-glucose deprivation/reoxygenation (OGD/R) model

The cells were rinsed twice with cold phosphate buffer saline (PBS) and grown in a hypoxic environment (1% O_2, 95% N_2, and 5% CO_2) in oxygen-glucose deprivation (OGD) medium (glucose-free and serum-free DMEM) for 8 h at 37 ℃. Thereafter, they were rapidly reoxygenated in 95% air and 5% CO_2. The cells were then transferred to a standard medium for 24 h.

NA extraction and quantitation

Total RNA was extracted from tissues or cells using a Total RNA Extraction Kit (#17200; AmyJet Scientific, Wuhan, China) according to the provided protocol. RNA concentrations were measured in a Nanodrop 2000 spectrophotometer (Thermo Fisher Scientific). Complementary DNA (cDNA) was synthesized from approximately 1 μg of the total RNA using an Omniscript RT Kit (#205111; Qiagen) followed by quantitative real-time polymerase chain reaction (qRT-PCR) amplification using a TransStart® Green qPCR SuperMix kit (#AQ101-01; TransGen Biotech, Beijing, China). The standard $2^{-\Delta\Delta Ct}$ method was used to calculate the mRNA expression from a minimum of three biological replicates. The primer sequences are listed in *Table 1*.

Western blotting

Total protein was isolated from tissue samples and SK-N-SH and SH-SY5Y cells using radio immunoprecipitation assay (RIPA) buffer, followed by the measurement of protein concentrations with a bicinchoninic acid assay (BCA) kit (Beyotime, Shanghai, China). The protein sample was first electrophoresed for 2 h before being transferred to polyvinylidene fluoride (PVDF) membranes (Millipore, Billerica, MA, USA). TBST with 5% fat-free milk was used to block the non-specific antigens for 1 hour before incubating with primary antibodies [Sox11 (Abcam, ab234996, 1:2,000), GAPDH (Abcam, ab9485, 1:5,000),

BAX (Abcam, ab32503, 1:5,000), BIM (Abcam, ab32158, 1:1,500), MCL-1 (Abcam, ab32087, 1:3,000), and Bcl-2 (Abcam, ab196495, 1:2,000)] and secondary antibodies. The proteins were visualized by enhanced chemiluminescence (ECL) and analyzed using ImageJ (NIH, Bethesda, MD, USA).

Cell viability

Cell viability was assessed by Cell Counting Kit-8 (CCK-8) assays. About 1,500 cells were inoculated in 96-well plates. Dex (1, 10, and 50 μM) was added into wells after 1 day (22). The absorbance was assessed after 2 days using CCK-8 (Dojindo, Japan).

Apoptosis assay

For the apoptosis assay, SK-N-SH and SH-SY5Y cells in six-well plates were incubated with Dex (1, 10, and 50 μM) in the indicated groups for 48 hours. This was followed by incubation with Annexin V-fluorescein isothiocyanate (FITC) and Prodium Iodide (PI) (KeyGEN BioTECH, China) for about 10 mins (as indicated by the instructions) and detection using CytExpert.

5-ethynyl-2'- deoxyuridine (EdU) assay

Cellular proliferation was measured with an EdU assay kit (Ribobio, Guangzhou, China). The cells were digested and inoculated into six-well plates at 10×10^5 per well. Dex (1, 10, and 50 μM) was added into the wells 1 day later. Then, 50 μM of EdU buffer was added into the wells and incubated at 37 ℃ for 2 h as indicated by the instructions. After fixation with 4% formaldehyde for 30 min and permeabilization with 0.1% Triton X-100 for 10 min, the cells were incubated with an EdU solution, and the nuclei were then stained with Hoechst. The cells were examined and imaged by fluorescence microscopy.

Plasmid transfection

Short hairpin RNA (shRNA) for Sox11 knockdown and Sox11 overexpression plasmids pcDNA4.0-Sox11 were obtained from Synbio Technologies (Suzhou, China). The cells were transfected with plasmids expressing shSox11 and cDNA for Sox11 using Lipofectamine 2000 (Life Technologies) for 72 h, as indicated by the instructions. The transfected cells were then selected using 2 mg/mL of puromycin (Gibco; California, USA) for several days.

Assessment of neurological deficits

Neurological deficits were examined using the Longa and Bederson scores. The rats were evaluated 24 h after awakening from anesthesia and were scored as follows: 0 points, no symptoms of damage; 1 point, unable to extend contralateral forepaw completely; 2 points, circling to the opposite side; 3 points, collapsing to the opposite side; 4 points, unable to walk spontaneously; and 5 points, unconscious.

Statistical analysis

All experiments were performed three times and all of the data were expressed as means ± standard deviation (SD). Analysis was performed using SPSS 25.0 (IBM Corp., NY, USA) and graphics were constructed using GraphPad Prism 8.0 (La Jolla, CA, USA). *T*-tests were used to evaluate differences between groups, and multiple group comparisons were analyzed with one-way ANOVA, and P values <0.05 were considered significant.

Results

Dex positively regulates the expression of Sox11 under OGD/R conditions

Given that Sox11 is indispensable for neuronal survival, neurite outgrowth, axon regeneration, and neuron fate diversity (23-25), we first verified whether DEX treatment influenced Sox11 expression. As expected, the OGD/R conditions markedly reduced both the mRNA and protein levels of Sox11 (*Figure 1A-1C*). The cells were then incubated with varying concentrations of Dex (1, 10, and 50 μM). As shown in *Figure 1A-1C*, Dex treatment significantly elevated the levels of SOX in cells under OGD/R conditions (*Figure 1A-1C*). Thus, we demonstrated Dex positively regulates Sox11 expression under OGD/R conditions.

Dex protects cells from OGD/R-induced damage

Next, we examined the influence of Dex on cell survival. As expected, the OGD/R conditions notably inhibited cell viability, as indicated by the CCK-8 assay. However, Dex dose-dependently ameliorated the OGD/R-induced effects on cell survival (*Figure 2A*). As depicted by the EdU

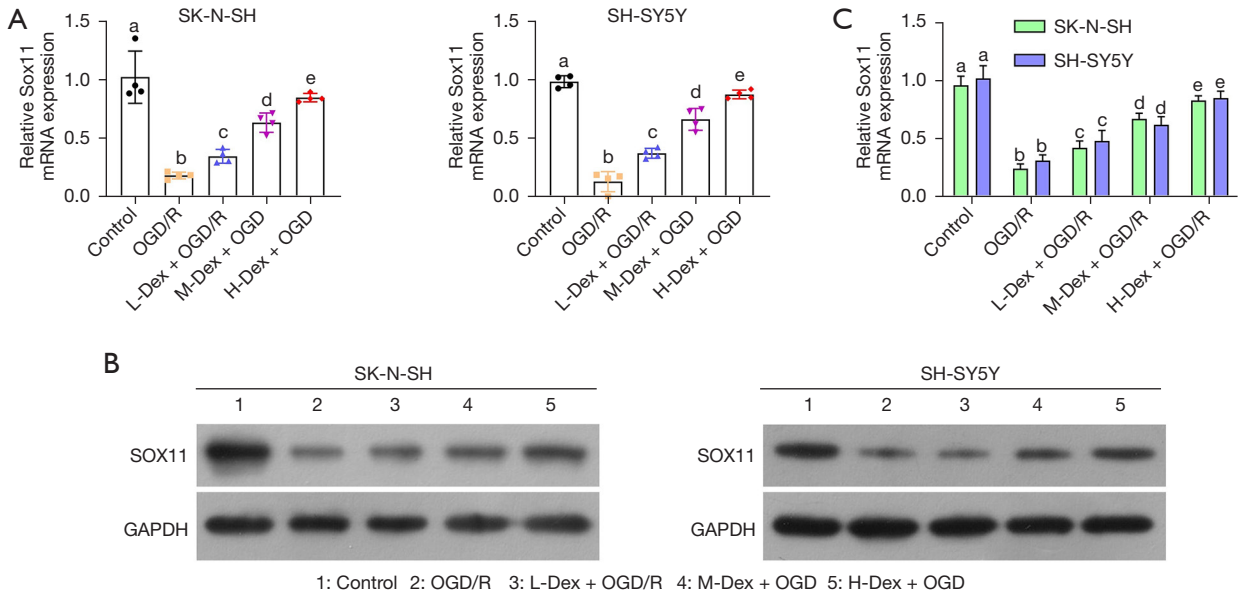

Figure 1 Dex positively regulates the expression of Sox11 under OGD/R conditions. (A) The relative Sox11 mRNA expressions in the different groups measured by qRT-PCR. (B) Sox11 protein levels evaluated by western blotting. (C) Quantitation of Sox11 protein levels in the different groups. For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. OGD/R, oxygen-glucose deprivation/reoxygenation; Dex, dexmedetomidine; qRT-PCR, quantitative real-time polymerase chain reaction.

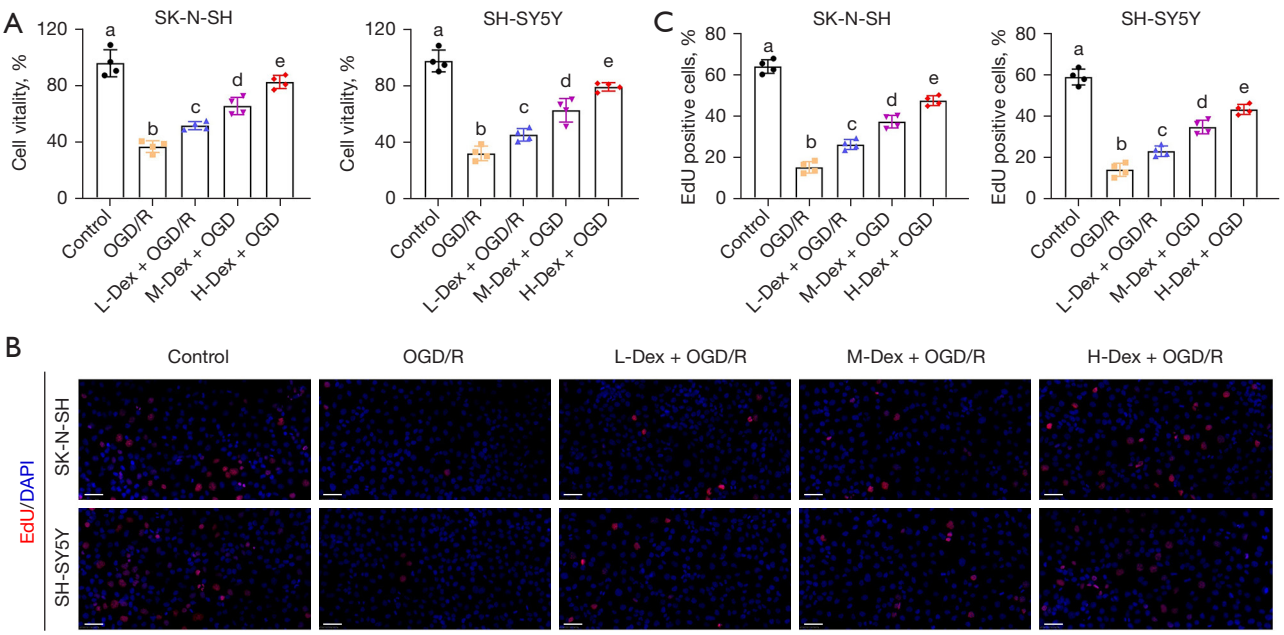

Figure 2 Dex rescues the reduced viability and proliferation of cells after OGD/R. (A) Viability measured by CCK-8 assays. (B) Cell proliferation measured by EdU staining assay (scale bar, 50 μm). (C) Quantitative analysis of the EdU assay results for the different groups. For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. OGD/R, oxygen-glucose deprivation/reoxygenation; Dex, dexmedetomidine; CCK-8, Cell Counting Kit-8; EdU, 5-ethynyl-2'-deoxyuridine; DAPI, 2-(4-amidinophenyl)-6-indolecarbamidine dihydrochloride.

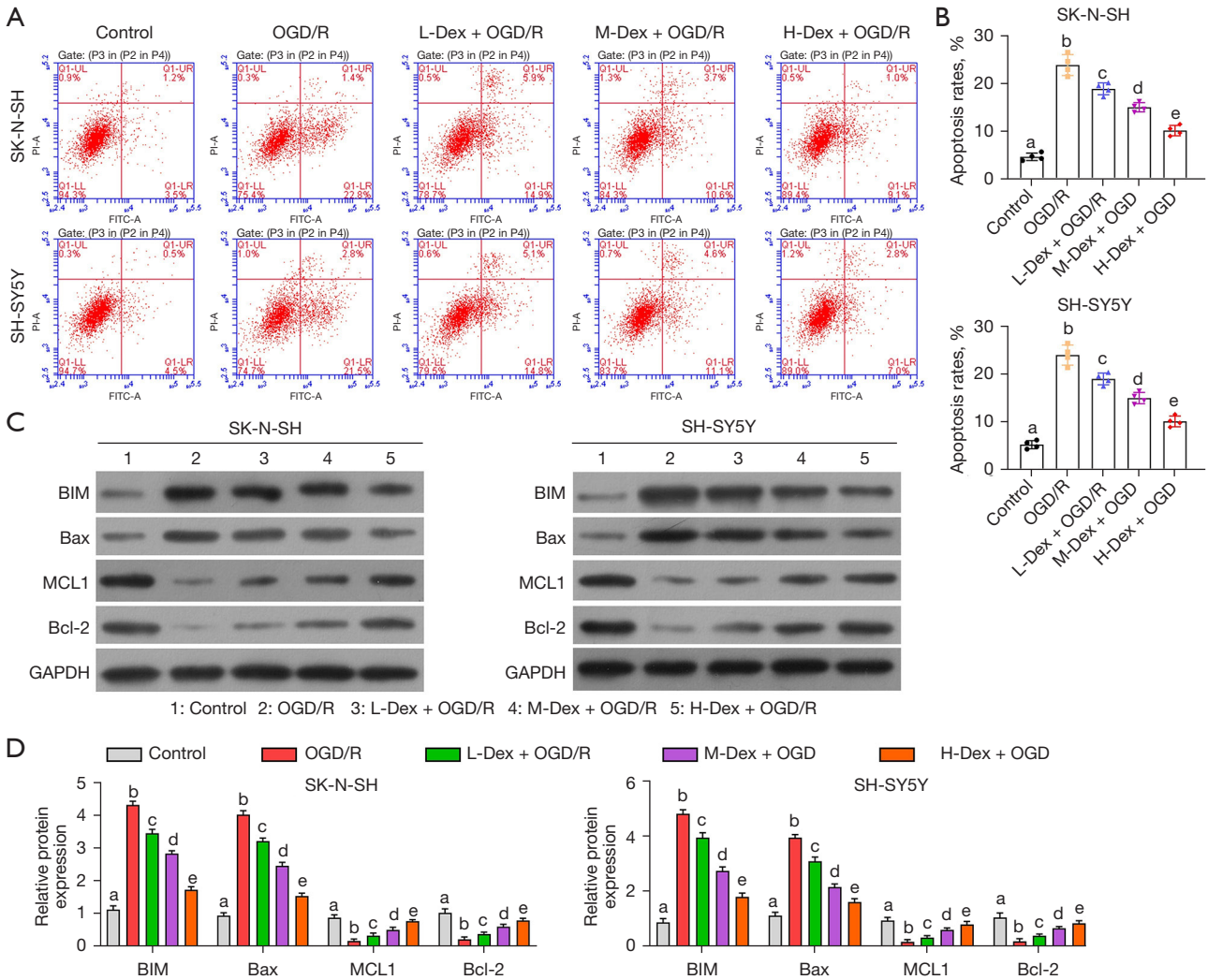

Figure 3 Dex ameliorated cellular apoptosis induced by OGD/R. (A) Apoptosis ratios determined by flow cytometry in the different groups. (B) Quantitation of apoptosis measured by flow cytometry. (C) Levels of apoptosis-related proteins shown by western blotting. (D) Quantitative analysis of apoptosis-related protein levels. For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. OGD/R, oxygen-glucose deprivation/reoxygenation; Dex, dexmedetomidine; FITC, fluorescein isothiocyanate; PI, propidium iodide.

assay, the proliferation of SK-N-SH and SH-SY5Y cells was markedly reduced after OGD/R (*Figure 2B,2C*), while Dex treatment counteracted these effects and enhanced cell proliferation. Similarly, OGD/R significantly induced apoptosis, which was dose-dependently reversed by Dex treatment (*Figure 3A,3B*).

We then evaluated the apoptosis-associated proteins in each group. BIM and Bax were notably increased by OGD/R treatment. Furthermore, the different concentrations of Dex decreased BIM and Bax levels compared to OGD/R-only treatment (*Figure 3C,3D*). Moreover, MCL1 and Bcl-2 levels were notably reduced by OGD/R treatment, and were increased by the different concentrations of Dex, as compared to the OGD/R group (*Figure 3C,3D*). Taken together, these results confirmed that Dex prevented OGD/R-induced injury.

Sox11 rescues OGD/R-induced cell damage

To determine the function of Sox11 in cell protection,

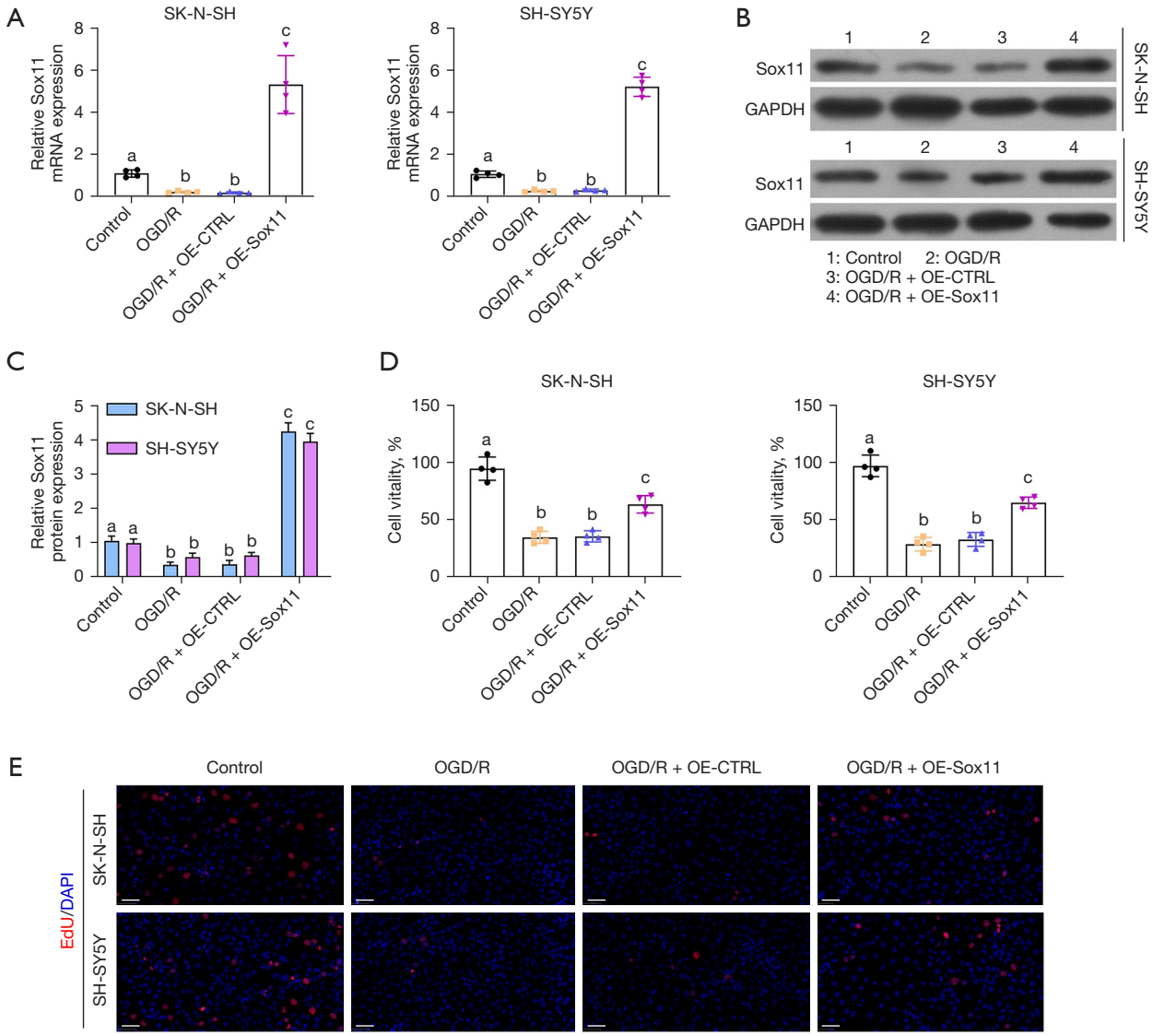

Figure 4 Sox11 ameliorates reduced cellular viability and proliferation induced by OGD/R. (A) Verification of Sox11 overexpression, as shown by qRT-PCR. (B) Verification of Sox11 overexpression, as shown by western blotting. (C) Quantitation of Sox11 protein levels in the different groups. (D) Cell viabilities in the different groups, as shown by the CCK-8 assays. (E) Cell proliferation in the different groups, shown by the EdU staining assay (scale bar, 50 μm). For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. OGD/R, oxygen-glucose deprivation/reoxygenation; qRT-PCR, quantitative real-time polymerase chain reaction; CCK-8, Cell Counting Kit-8; OE, overexpression; EdU, 5-ethynyl-2'-deoxyuridine; DAPI, 2-(4-amidinophenyl)-6-indolecarbamidine dihydrochloride.

we overexpressed Sox11 in OGD/R-treated cells, and the efficacy of Sox11 overexpression was confirmed (*Figure 4A-4C*). We then evaluated cell viability and proliferation. OGD/R treatment notably inhibited cell viability. However, Sox11 overexpression counteracted the effects of OGD/R on cell survival (*Figure 4D*). The EdU assay indicated that cell proliferation was significantly inhibited with OGD/R. Meanwhile, Sox11 overexpression promoted cell proliferation under OGD/R conditions (*Figure 4E* and Figure S1).

Cell apoptosis was also detected by the apoptosis assay. Similarly, OGD/R significantly induced cell apoptosis,

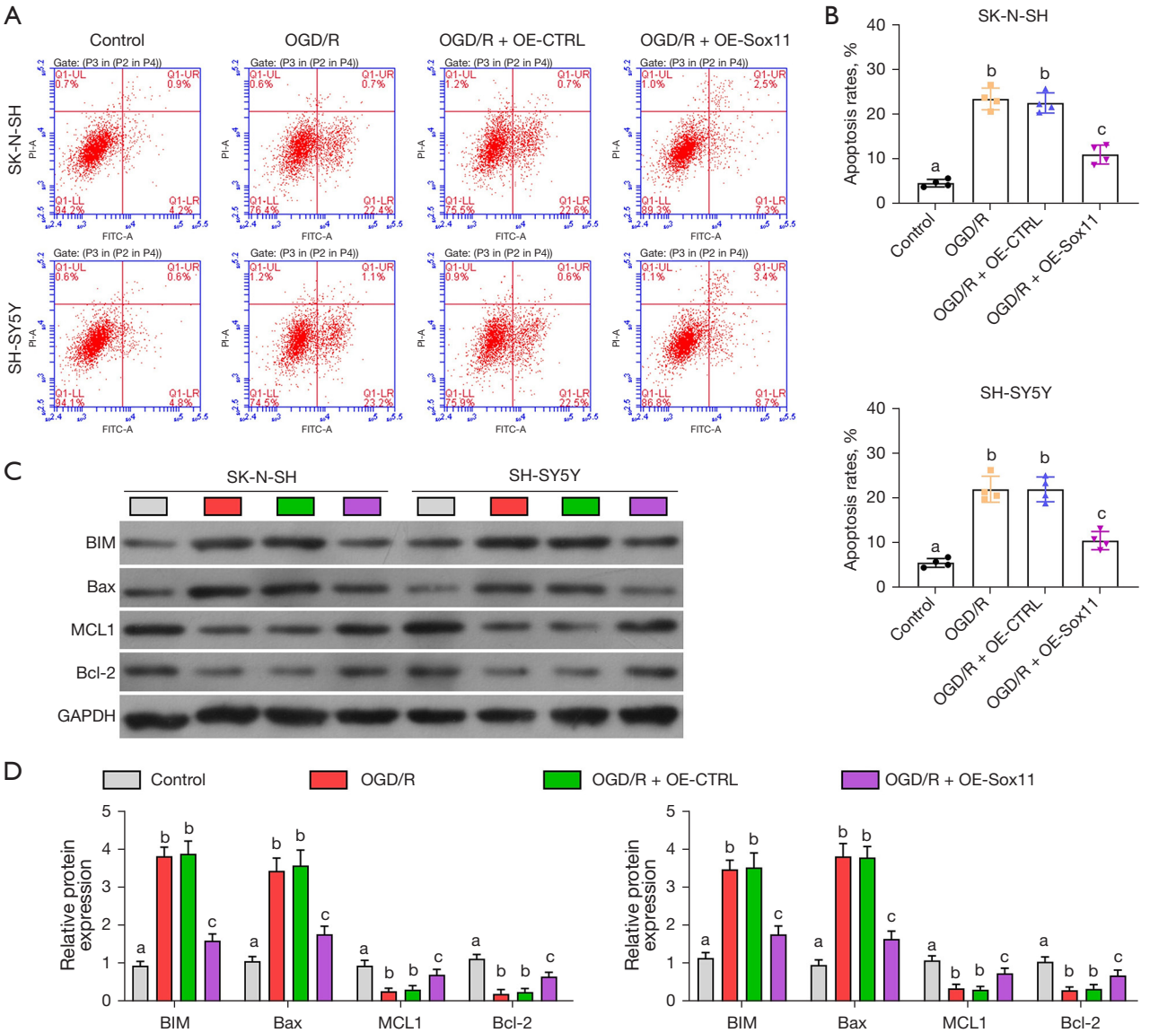

Figure 5 Sox11 reduces apoptosis induced by OGD/R. (A) Apoptosis ratios for the different groups, as measured by flow cytometry. (B) Quantitation of apoptosis in the different groups, as measured by flow cytometry. (C) Apoptosis-related proteins in the different groups, as shown by western blotting. (D) Quantitation of apoptosis-related in the different groups. For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. OGD/R, oxygen-glucose deprivation/reoxygenation; OE, overexpression; FITC, fluorescein isothiocyanate; PI, propidium iodide; CTRL, control.

while Sox11 overexpression notably rescued the apoptosis ratio under OGD/R conditions (*Figure 5A*,*5B*). We then evaluated apoptosis-associated proteins in each group. As expected, BIM and Bax were notably increased by OGD/R treatment, and were decreased by Sox11 overexpression, as compared to the OGD/R group (*Figure 5C*,*5D*). Additionally, the expressions of MCL1 and Bcl-2 were notably decreased by OGD/R treatment, and were increased by Sox11 overexpression, as compared to the OGD/R group (*Figure 5C*,*5D*). Therefore, Sox11 rescues cells from OGD/R-induced damage.

Dex prevents OGD/R-mediated damage by up-regulating Sox11

To verify whether Dex reduced OGD/R-mediated damage

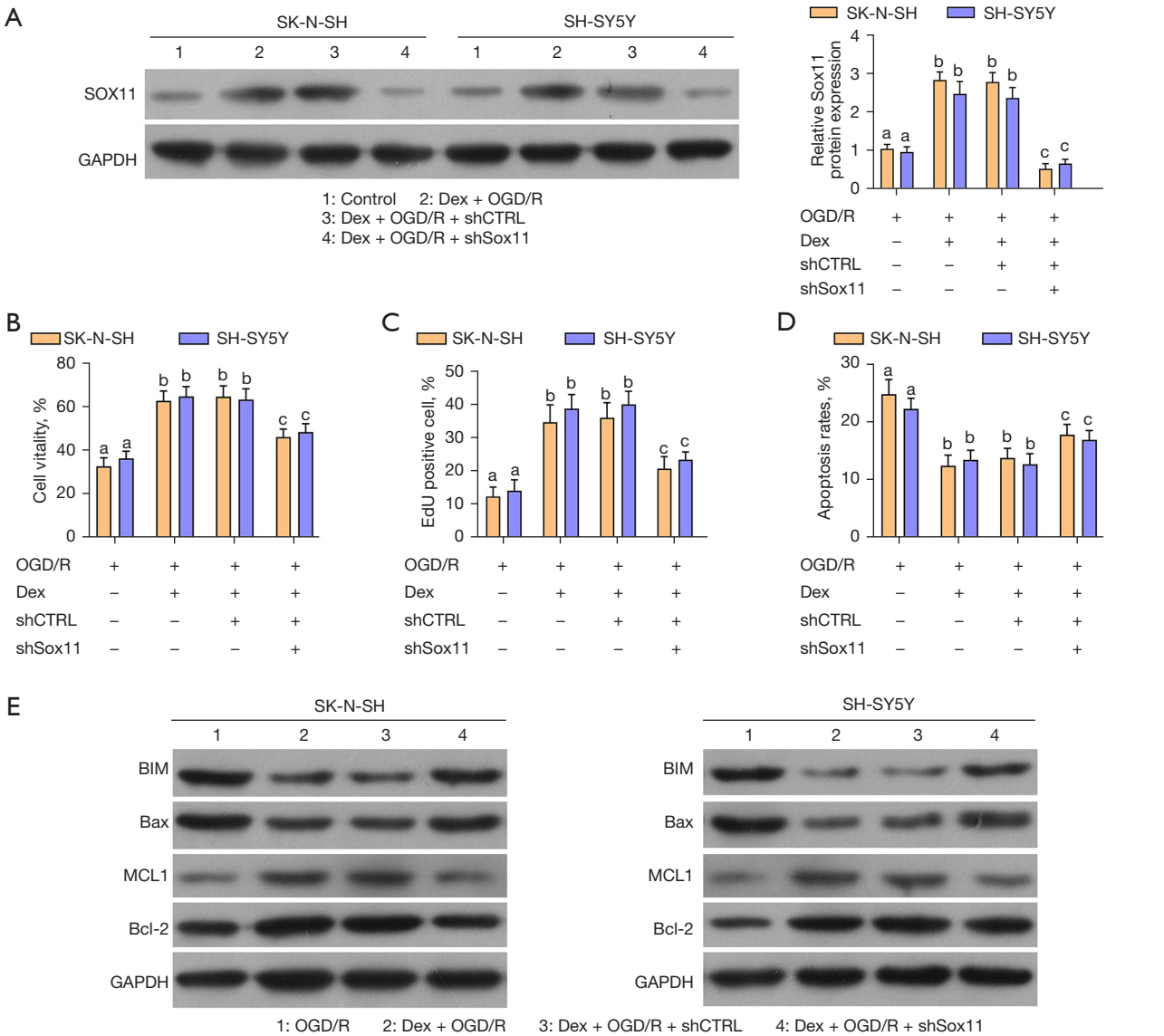

Figure 6 Dex ameliorates OGD/R-mediated cell damage via up-regulation of Sox11. (A) The efficacy of Sox11 knockdown verified by western blotting. (B) Cell viability in the different groups, as determined by CCK-8 assays. (C) Quantitation of proliferation in the different groups, as determined by EdU assays. (D) Quantitation of apoptosis in the different groups, as measured by flow cytometry. (E) Levels of apoptosis-related proteins in the different groups, as shown by western blotting. For statistical analysis, different letters showed significant differences between groups in SH-N-SH or SH-SY5Y cells. Dex, dexmedetomidine; OGD/R, oxygen-glucose deprivation/reoxygenation; CCK-8, Cell Counting Kit-8; EdU, 5-ethynyl-2'-deoxyuridine; sh, short hairpin RNA; CTRL, control.

via the promotion of Sox11 expression, Sox11 was knocked down in the OGD/R-pretreated cells, which were also treated with Dex. The efficacy of Sox11 knockdown was also confirmed (*Figure 6A*). We then evaluated cell viability, proliferation, and apoptosis. Dex treatment notably promoted cell viability under OGD/R conditions. However, Sox11 knockdown rescued the effects of Dex on cell survival (*Figure 6B*). The EdU assay indicated that cell proliferation was notably promoted with Dex treatment. Meanwhile, Sox11 knockdown inhibited cell proliferation with Dex and OGD/R treatment (*Figure 6C*). Similarly, Dex also significantly decreased cell apoptosis. Moreover, Sox11

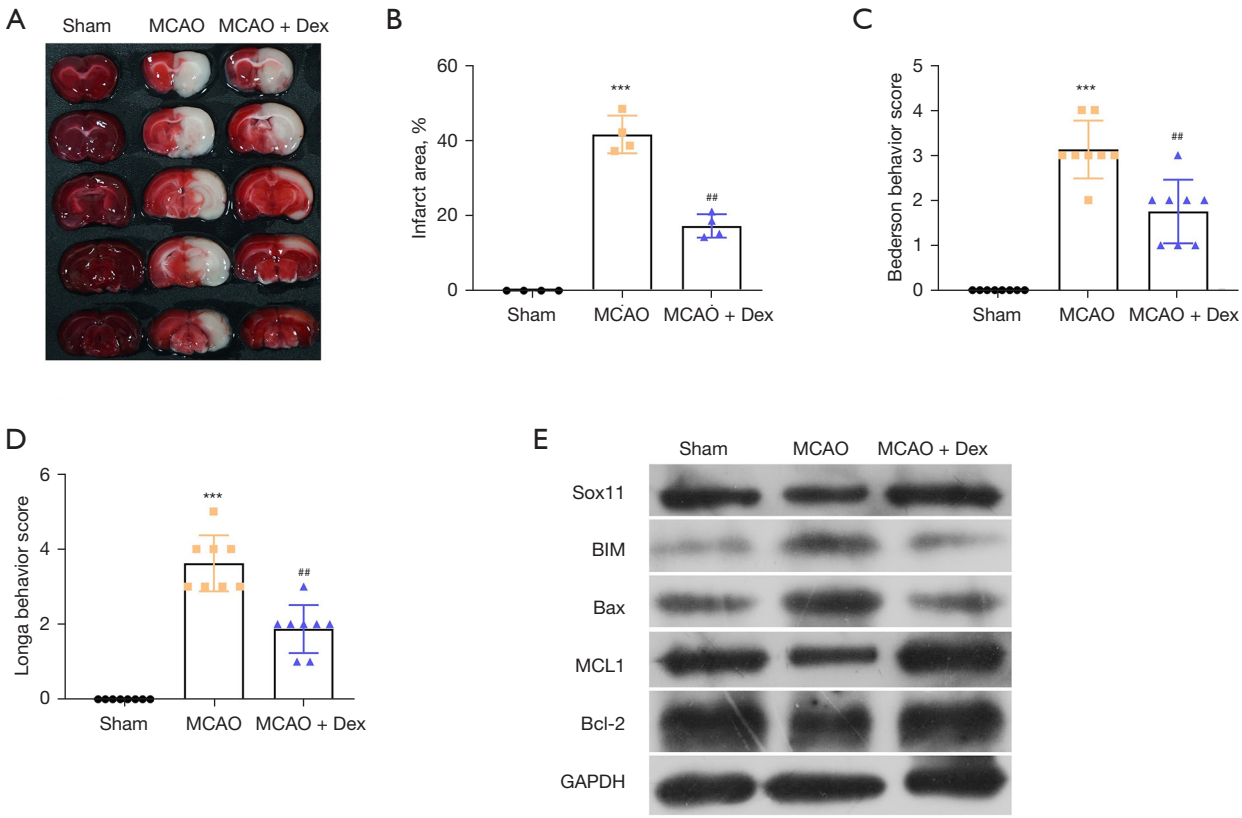

Figure 7 Dex protects rat from ischemia-induced injury in the MCAO model. (A) Representative images of rat brains after MCAO for the different groups. (B) Infarct areas in the rat brains. (C) Bederson behavioral scores for the different groups. (D) Longa behavioral scores for the different groups. (E) Levels of apoptosis-associated proteins in the brain tissue of the different groups, as shown by western blotting. ***, P<0.001 *vs.* sham group; and ##, P<0.01 *vs.* MCAO group. Dex, dexmedetomidine; MCAO, middle cerebral artery occlusion.

knockdown notably rescued the apoptosis ratio under Dex treatment (*Figure 6D*).

We then evaluated apoptosis-associated proteins in each group. As expected, BIM and Bax were notably decreased by Dex treatment. Furthermore, Sox11 knockdown increased BIM and Bax expression comparing with Dex treatment in OGD/R. In addition, MCL1 and Bcl-2 expression were notably up-regulated by Dex treatment. Sox11 knockdown reduced the levels of MCL1 and Bcl-2 compared to those in the Dex group (*Figure 6E*). These results indicate that Dex improved OGD/R-induced cell damage by up-regulating Sox11.

Dex protected rats from ischemia-induced injury in the MCAO model

We also explored the effects of Dex *in vivo*. As expected, MCAO significantly induced ischemia and injury to the rat brains, as shown by a larger infarct area and higher Bederson and Longa behavior scores (*Figure 7A-7D*). However, Dex treatment notably protected rat from ischemia-induced injury compared with the MCAO group (*Figure 7A-7D*). The apoptosis-associated proteins were also detected. The expressions of BIM and Bax were markedly increased in the MCAO group. Furthermore, Dex decreased the BIM and Bax levels compared to those in the MCAO group (*Figure 7E*). Additionally, MCL1 and Bcl-2 levels were notably reduced in the MCAO group, and were increased by Dex treatment, as compared to the MCAO group (*Figure 7E*). Together, these results highlight that Dex prevented ischemia-induced injury in the MCAO model.

Discussion

Ischemic stroke leads to high levels of both mortality and

disability worldwide (1). When acute ischemic reperfusion occurs, neurons are damaged by inflammation. However, anti-inflammation therapies were not transformed in the clinic (26). In this study, we confirmed that Dex treatment enhanced cell viability and proliferation and reduced apoptosis. The same results were also observed in the *in vivo* model. Additionally, we also observed that Dex protected neurons from MCAO-induced injury by up-regulating Sox11 expression.

Ischemic stroke can cause neuronal apoptosis (27-30). Therefore, anti-apoptosis treatment could be a promising method of improving stroke-induced damage. Using a rat MCAO model, a previous study proved that delayed recanalization efficiently protected neurons from apoptosis mediated by HGF/c-Met/STAT3/Bcl-2 signaling (31). Long OPA1 mitochondrial dynamin like GTPase (L-OPA1) restoration also suppresses neuronal apoptosis induced by ischemic stroke by regulating mitochondrial function (32). It was reported that γ-glutamylcysteine suppressed neuronal apoptosis by decreasing ROS production as well as glutathione balance (33). Herein, we found that Dex significantly inhibited neuronal apoptosis and promoted cell survival and proliferation. These results are meaningful for patients suffering from stroke and neuronal damage.

As previously reported, Dex has several targets (34-36). Studies have confirmed that Dex regulates c-Fos/NLRP3/caspase-1 cascades, ERK signaling, AKT signaling, and the miR-330-3p/ULK1 axis (34-37). It can also regulate several miRNAs to modulate cell apoptosis, drug resistance, and cancer metastasis (38). In adipogenesis, Dex directly up-regulates miR-155 and promotes the differentiation of 3T3-L1 preadipocytes (39). FOXO1 is another target of Dex in MC3T3-E1 cells, which was identified by bioinformatics analysis (40). In our study, we identified that Sox11 is a new target of Dex; Dex positively regulated Sox11 in neuronal cells to maintain cell survival and inhibit apoptosis.

Sox11 is vital for cell survival. It has been reported to regulate neuronal survival, is a key regulator of sensory neuron development (41), and is also a regulator of cancer stem cells (42). Sox11 overexpression also improves angiotensin II receptor type 2 (AT2) mechanical injury and could represent a target for treating ventilator-induced lung injury (43). A previous study showed that Sox11 overexpression augmented the excitation of dentate gyrus (DG) granule cells and reduced the levels of various potassium channel subunits, suggesting that Sox11 activity is vital for modulating the plasticity of DG neurons (44). Our results indicated that Sox11 was important for neuronal survival under OGD/R conditions and protected against stroke-induced injury. Furthermore, Sox11 mediated the neuronal protective effects of Dex under OGD/R conditions.

Our study had several limitations that should be noted. Firstly, the mechanism of Dex up-regulation of Sox11 is still unclear, and thus, future studies should focus on elucidating this mechanism. Secondly, Dex might influence neuronal function via several other targets, and we will explore this in the future. Then, the mechanism we clarified in the study was should be confirmed in the rat model. Finally, ischemic reperfusion also could induce inflammation, whether Dex could inhibit the inflammation of the neuronal system, and the effect of Dex on the cytokines expression need to be further confirmed.

Conclusions

In this study, we verified the role of Dex in cell viability and survival. Moreover, Dex protected neurons from MCAO-induced injury by up-regulating Sox11 expression. Our research proposed a potential drug to improve the functional recovery of stroke patients in the clinic.

Acknowledgments

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Footnote

Reporting Checklist: The authors have completed the MDAR and ARRIVE reporting checklists. Available at https://atm.amegroups.com/article/view/10.21037/atm-22-6639/rc

Data Sharing Statement: Available at https://atm.amegroups.com/article/view/10.21037/atm-22-6639/dss

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://atm.amegroups.com/article/view/10.21037/atm-22-6639/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work, including ensuring that any questions related to the accuracy or integrity of any part of the work have been appropriately investigated and resolved. This animal experiment was approved by the Institutional Animal Ethics Committee of Guangzhou Women and Children's Medical

Center (No. S2022-107), in compliance with institutional guidelines for the care and use of animals.

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