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Loss of lncRNA UCA1 ameliorates the injury managed by cerebral ischemia-reperfusion by sponging miR-18a-5p

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Abstract

Introduction: *Acute ischemic stroke (AIS) is a disease with high morbidity and mortality in the clinic. The current experiments aimed to study the effects of UCA1 interfering miR-18a-5p on cerebral ischemia-reperfusion (CI/R).*

Material and methods: *For rat models undergoing middle cerebral artery infarction (MCAO) surgery, the expression of UCA1 and miR-18a-5p was evaluated by qRT-PCR, and underlying function was identified by detecting infarct size, neurological scores, and inflammation. Luciferase report was applied to verify the relationship between UCA1 and miR-18a-5p. In the cell models, the impacts of UCA1 and miR-18a-5p were validated by CCK-8 assay, flow cytometry analysis, and ELISA. In patients with AIS, Pearson correlation was carried out to unveil the association between UCA1 and miR-18a-5p.*

Results: *The expression of UCA1 was at high levels and miR-18a-5p was at low levels in AIS patients. UCA1 knockdown showed a protective role in infarct size, neurofunction, and inflammation via binding miR-18a-5p. MiR-18a-5p participated in the regulation of UCA1 on cell viability, cell apoptosis, lactate dehydrogenase (LDH) levels, and inflammation. In patients with AIS, overexpression of UCA1 and underexpression of miR-18a-5p had a reverse correlation.*

Conclusions: *Elimination of UCA1 was favourable to the recovery of the rat model and cells from CI/R damage by efficaciously sponging miR-18a-5p.*

Key words: *UCA1, cerebral ischemia-reperfusion, inflammation, neurofunction, miR-18a-5p.*

Introduction

Recently, the incidence of acute ischemic stroke (AIS) has been increasing year by year, and its incidence tends to be younger [3]. AIS is mainly caused by cerebral vascular occlusion, which leads to focal neurological deficits [13]. At present, the internationally recognized treatment means is to open occluded blood vessels as soon as possible and reduce ischemic injury of brain tissue [28]. The main treatment methods are thrombolysis and arterial thrombectomy after thrombolysis, but the reperfusion injury to brain tissue after recanalization is one of the main reasons for the poor therapeutic effect [1]. Theoretically, the protection of brain function from reperfusion damage could benefit the recovery of AIS patients [20]. Therefore, exploring the mecha-

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nism of cerebral ischemia-reperfusion injury and studying the methods of controlling and reducing cerebral ischemia-reperfusion injury has become significant.

Long noncoding RNA (lncRNA) is a kind of endogenous RNA with a length longer than 200 nucleotides [27]. LncRNAs may exert their functions by functioning as a sponge of miRNA. MiRNAs are composed of 18-25 nucleotides, which also belong to the non-coding RNA family [6]. As a ceRNA of miRNA, lncRNA interconnects with miRNA and suppresses miRNA expression to reduce the miRNA repressive impacts on targeting mRNA [17]. With the deepening of cerebral ischemia-reperfusion (CI/R) research, the research on the role of lncRNA in brain ischemia-reperfusion (I/R) injury has gradually increased [10]. For instance, lncRNA MALAT1 contributes to the recovery of oxygen-glucose deprivation/reoxygenation (OGD/R)-managed vascular endothelial cells *via* inhibiting apoptotic cells, which shows the probable significance of lncRNA in CI/R [26]. Furthermore, lncRNA-U90926 is abnormally expressed in the mice exposed to CI/R and it might lead to a more aggressive trend of ischemic brain damage [3]. All the above experiments clearly show that miRNA plays an important role in the process of brain I/R injury. Lu *et al*. propose that in Parkinson's disease, the expression of lncRNA urothelial carcinoma associated 1 (UCA1) is elevated and it regulates abnormal apoptosis and viability, which shows that UCA1 participates in the development of brain diseases [18]. In a study published in 2021, UCA1 is abnormally expressed in patients with AIS [21]. Nevertheless, the underlying function behind the UCA1 and AIS is indefinite. The occurrence of cerebral ischemia is accompanied by significant alternations in the expression of many genes, among which microRNA (miRNA) may also participate in the occurrence and progression of this disease [24,30]. MiR-18a-5p is one important component of miRNA regulatory network [23]. Associations between miR-18a-5p and the risks of AIS have previously been studied. It is widely known that atherosclerosis is an inducing factor of AIS. Vegter *et al*. report that an increasing character of atherosclerosis is accompanied by the reduced expression of miR-18a-5p and in the patients of atherosclerotic disease, the expression of miR-18a-5p is significantly lessened [22].

In this study, the expression changes and trends of UCA1 and miR-18a-5p in the middle cerebral artery infarction (MCAO) rat model, OGD/R cell model, and AIS patients. The regulatory relationship between them was further proved. Besides, this investigation provided the function of UCA1 on brain infarct volume, neurological function, and inflammation in the assay of the rat model. Furthermore, the mediated function of miR-18a-5p was investigated.

Material and methods

Animals and artificial transfection

Sprague-Dawley (SD) healthy male rats (Yison BIO, Shanghai, China) were randomly selected as a sham operation group and the rest were given the MCAO operation. 200-250 g adult rats were successfully adapted to the environment after 7 days. This experiment was conducted with the approval of the Animal Ethics Committee of Dongying Hospital of Traditional Chinese Medicine. All institutional and national guidelines for the care and use of laboratory animals were followed.

The short interfering UCA1 (si-UCA1) and its negative control (si-NC) together with miR-18a-5p antagomir and antagomir-NC were synthesized by GenePharma (Shanghai, China), respectively. All these man-made sequences were mixed with Invivofectamine 3.0 reagent (ThermoFisher, Waltham, USA). Three days before the MCAO experiment, the compound was injected into the lateral ventricle separately.

MCAO animal model construction and treatment

The model of transient focal MCAO rat was established in this experiment, which was based on a previous study [7]. 10% chloral hydrate 300 mg/kg was injected intraperitoneally for anaesthesia, and no response of the rat tail was taken as an indication of successful anaesthesia. After successful anaesthesia, the neck hair of rats was removed. The rats were fixed on the operating table in the supine position, and the skin of the neck was disinfected routinely. The skin was cut in the middle of the neck, and the subcutaneous tissue was passively separated. After full separation, the right common carotid artery (CCA), the right external carotid artery (ECA), and the right internal carotid artery (ICA) were exposed. The proximal end of the common carotid artery was completely ligated and a loose knot was made before the bifurcation of the common carotid artery. After blocking for 120 min, the threaded bolt was pulled out and the wound was sutured for 24-h reperfusion. Rats in the sham operation group were treated the same as those in the experimental group except that no thread plugs were inserted. After the operation, the rats were placed in cages covered with clean and dry padding. Their body temperature was kept at about 37°C and they were given enough water. The volume of cerebral infarction and neurological score, including Longa score and Bederson score were evaluated 72 hours after the experiment.

At the end of all *in vitro* trials, rats were anesthetized and fixed by intraperitoneal injection of chloral hydrate (4 ml/kg), and their heads were cut off from

the back. The brains were rinsed with 0.01 mol/l phosphate buffered saline (PBS) and then were quickly frozen in a refrigerator at –34°C for 30 min. The tissues of brain coronal sections were cut every 2 mm and generally cut into 6 pieces. Then, the pieces were placed in 2% TTC away from light for 30 min at 37°C. The area of cerebral infarction was calculated by Image J software.

Cell model and transfection

SH-SY5Y cells (Yiyan Biological Technology, Shanghai, China) were cultured in the DMEM F12 with 5 ml foetal bovine serum and 1 ml double-antibody. All cells were placed in the incubator of 37°C and a humidified air with 5% CO_2. Some cells were incubated in the medium without serum and glucose in a condition without oxygen for 2 hours to establish the OGD/R cell models. Afterward, we substituted the medium with normal medium and cultured cells for 24 hours under a normal air environment. The artificial fragments were transfected into SH-SY5Y cells *via* Lipofectamine 3000 (ThermoFisher Waltham, USA) based on its recommended protocols. After 48-h transfection, cells were collected for subsequent experiments.

Cell viability test

Transfected cells were inoculated in a 96-well plate at a density of 5×10^3 cells/well. Every 24 hours, 20 µl CCK-8 reagent (Beyotime, Shanghai, China) was added to each well. The plate was stored at 37°C for further incubation of 1 hour. The absorption value was detected at 450 nm by a microplate reader.

Flow cytometry analysis

According to the manufacturer's specifications, apoptotic cells were quantified by annexin V-FITC/PI apoptosis detection kit (ThermoFisher, Waltham, USA). Transfected cells were re-suspended in 200 binding buffer, containing 10 µl Annexin V-FITC and 10 µl PI. Immediately after incubation in the dark at room temperature for 30 min, FITC-positive and PI-negative cells were analysed by flow cytometry (BD Bioscience, San Jose, CA).

Detection of inflammation and LDH

The inflammatory states of SD rats or SH-SY5Y cells were reflected by the concentration of tumour necrosis factor α (TNF-α), interleukin (IL)-6, and IL-1β of brain tissues or cell supernatants. The ELISA kit (CUSABIO, Wuhan, China) was used to determine the secretion of inflammatory factors.

The concentration of lactate dehydrogenase (LDH) in the culture supernatant was identified *via* an LDH detection kit (Roche, Basel, Switzerland) based on its procedure. The absorbance data were obtained at 490 nm using a microplate reader.

Luciferase activity assay

The sequences of Wild-type (WT)/mutant (MUT) of UCA1, miR-18a-5p inhibitors, inhibitor NC, miR-18a-5p mimics, and mimic NC were also obtained from GenePharma (Shanghai, China). WT-UCA1 and MUT-UCA1 were cloned into a pmirGLO vector separately. And then, pmirGLO-WT-UCA1 or pmirGLO-WT-UCA1 was co-transfected into SH-SY5Y cells with mimic NC, inhibitor-NC, miR-18a-5p mimic, and miR-18a-5p inhibitor. A dual-luciferase reporter kit (Promega, Madison, USA) was used for measuring the luciferase activity of the cells after 48-h transfection.

Patient collection

A total of 75 AIS patients with a mean age of 53.00 ±4.69 years old, including 42 males and 33 females admitted to the Dongying Hospital of Traditional Chinese Medicine were collected. 30 women and 35 men comprised healthy participants (mean age: 52.29 ±4.37 years old). All AIS patients were identified according to the Chinese guidelines for diagnosis and treatment of acute ischemic stroke of 2018. Inclusion criteria were as follows: 1) unilateral limb or face numbness, weakness, or language disorder; and 2) first onset. Exclusion criteria were as follows: 1) history of mental illness or cognitive dysfunction; 2) abnormal liver and kidney function; 3) systematic inflammatory disorder; 4) malignant tumour; and 5) cerebral haemorrhage. Gender- and age-matched individuals were included in the control group. This study was performed in line with the principles of the Declaration of Helsinki. Approval was granted by the Ethics Committee of Dongying Hospital of Traditional Chinese Medicine. Informed consent was obtained from all individual participants included in the study. Clinical data including age, sex, and body mass index of patients were collected. Venous blood samples were collected for AIS patients within 24-h of admission, and serum samples were stored in a refrigerator at –80°C.

Quantitative PCR (qPCR)

TRIzol LS reagent was purchased from ThermoFisher (Waltham, USA) and used for the isolation of total RNA from the serum of patients, brain tissue of rats, and SH-SY5Y cells. The reverse transcription experiments were conducted with HyperScript III miRNA 1st Strand cDNA Synthesis Kit (NovaBio, Shanghai, China) for miR-18a-5p and HyperScript III 1st Strand cDNA Synthesis Kit (NovaBio, Shanghai, China) for UCA1. The qPCR was performed by the SYBR Green qPCR Master Mix which was obtained from ThermoFisher

(Waltham, USA). The primer sequences were as follows: UCA1 forward, 5'-CTCTCCTATCTCCCTTCACTGA-3', reverse, 5'-CTTTGGGTTGAGGTTCCTGT-3'; GAPDH forward, 5'-CCACCCATGGCAAATTCCATGGCA-3'; reverse, 5'-TCTAGACGGCAGGTCAGGTCCACC-3'; miR-18a-5p forward, 5'-ACGTAAGGTGCATCTAGTGCAGATA-3', reverse, 5'-GTGCAGGGTCCGAGGT-3'; U6: forward: 5'-GCGCGTCGTGAAGCGTTC-3', reverse: 5'-GTGCAGGGTCCGAGGT-3'. The Ct values were exhibited on the 7500 system and the 2-deltadelta Ct was carried out to identify the final relative expression. U6 and GAPDH were the housekeeping gene of miR-18a-5p and UCA1 respectively.

Statistical analysis

Statistical analysis was made by SPSS and GraphPad software. Comparison between two groups was carried out by Student-*t* test. In variance analysis of more than two groups one-way ANOVA was used. The demographic data collected from AIS patients and healthy individuals were analysed by χ^2 test and Student-*t* test. Pearson analysis was used to evaluate the relationship between miR-18a-5p and UCA1. $P < 0.05$ was statistically significant.

Results

Reduced UCA1 ameliorated the damage elicited by MCAO in SD rats

In order to clarify the expression changes and effects of UCA1 after CI/R injury, we established the MCAO-managed rat model. Compared with the sham group, the expression level of UCA1 in the MCAO group was increased significantly, and the expression of UCA1 in the MCAO + si-UCA1 group was significantly lower than that in the MCAO group (Fig. 1A, $p < 0.001$). The relative volumes of cerebral infarction in the MCAO group were significantly higher than those in the sham group,

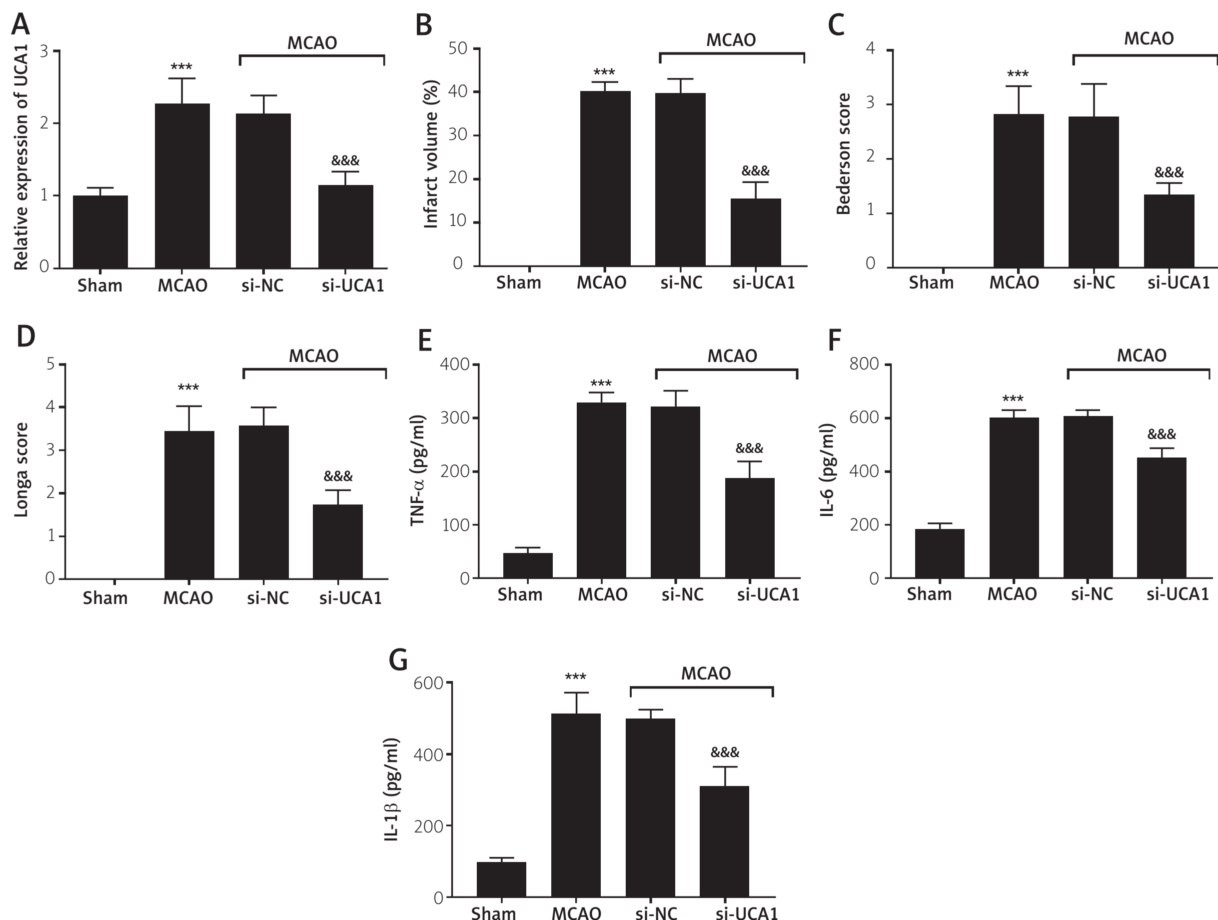

Fig. 1. Protective roles of UCA1 knockdown in SD rat models induced by the MACO operation. **A**) The outcome of qPCR; **B**) Silenced UCA1 restricted the increased infarct size caused by MACO. The function of UCA1 on neurofunctional scores (**C**, **D**) and inflammation (**E**, **F**). $^{***}p < 0.001$, compared with the sham group; $^{\&\&\&}p < 0.001$, compared with the MCAO group.

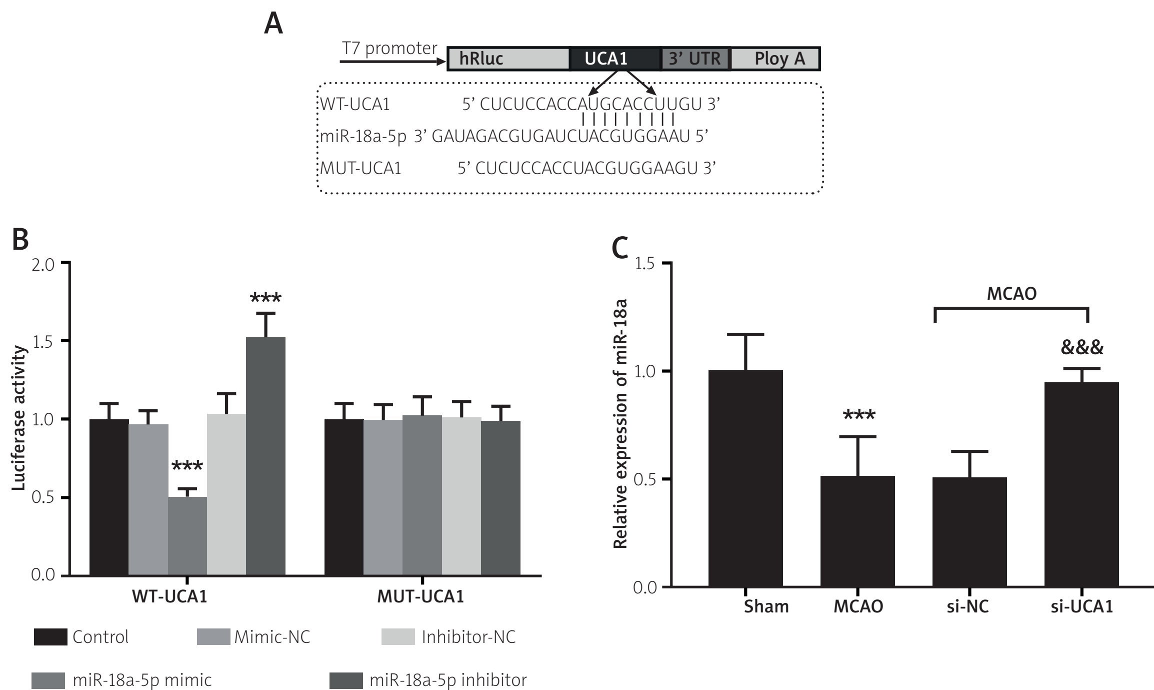

Fig. 2. A) The exhibition of complementary sites between UCA1 and miR-18a-5p. **B**) Luciferase assay confirmed the relationship between UCA1 and miR-18a-5p. **C**) MiR-18a-5p expression was reduced in the MCAO group and loss of UCA1 reversed the trend. ***$p < 0.001$, compared with the sham group; &&&$p < 0.001$, compared with the MCAO group.

while the volume in the MCAO + si-UCA1 group was significantly reduced when compared with the MCAO group (Fig. 1B, $p < 0.001$). The Bederson score and the Longa score of the MCAO group were significantly higher than those of the sham group, while silenced UCA1 reduced the abnormal Bederson score and the Longa score of the MCAO group (Fig. 1C, D, $p < 0.001$). Furthermore, the inflammatory disorders emerged in the MCAO group and loss of UCA1 recovered the inflammatory state of the MCAO group (Fig. 1E-G, $p < 0.001$).

MiR-18a-5p serves as a ceRNA of UCA1

Further analysis of the potential mechanism of UCA1 was evaluated by the elucidation of its target gene. The findings of Figure 2A demonstrated the complementary sites between miR-18a-5p and UCA1. The target relationship between miR-18a-5p and UCA1 was identified by the result of luciferase activity. The result showed that overexpression of miR-18a-5p lessened the luciferase activity and reduced miR-18a-5p facilitated the relative luciferase activity (Fig. 2B, $p < 0.001$). Besides, the expression of miR-18a-5p was significantly lower in the MCAO group when compared to that in the sham group, and the expression of miR-18a-5p in the MCAO + si-UCA1 group was significantly higher than that in the MCAO group (Fig. 2C, $p < 0.001$).

MiR-18a-5p mediated the function of UCA1 in MCAO models

The mediated impacts of UCA1 were further explored in the MACO-stimulated rat models. Compared with the sham group, the expression of miR-18a-5p in the MCAO group decreased significantly, and the expression level of miR-18a-5p decreased significantly in the MCAO + miR-18a-5p antagomir group when compared with the MCAO group (Fig. 3A, $p < 0.01$). As shown in Figure 3B, the expression of miR-18a-5p was enhanced in the MCAO + si-UCA1 group and the interference of miR-18a-5p reduced this aberrant expression trend ($p < 0.001$). The effects of silenced UCA1 on infarct volume and neurological function were reversed by the downregulation of miR-18a-5p (Fig. 3C-E, $p < 0.001$). Moreover, the elimination of UCA1 acted as a suppressor on inflammation of MCAO models, however, the intervention of miR-18a-5p reversed this influence (Fig. 3F-H, $p < 0.001$).

MiR-18a-5p mediated the function of UCA1 in OGD/R cell models

In vitro, cell models of CI/R were established by the OGD/R experiments and the significance of UCA1 and

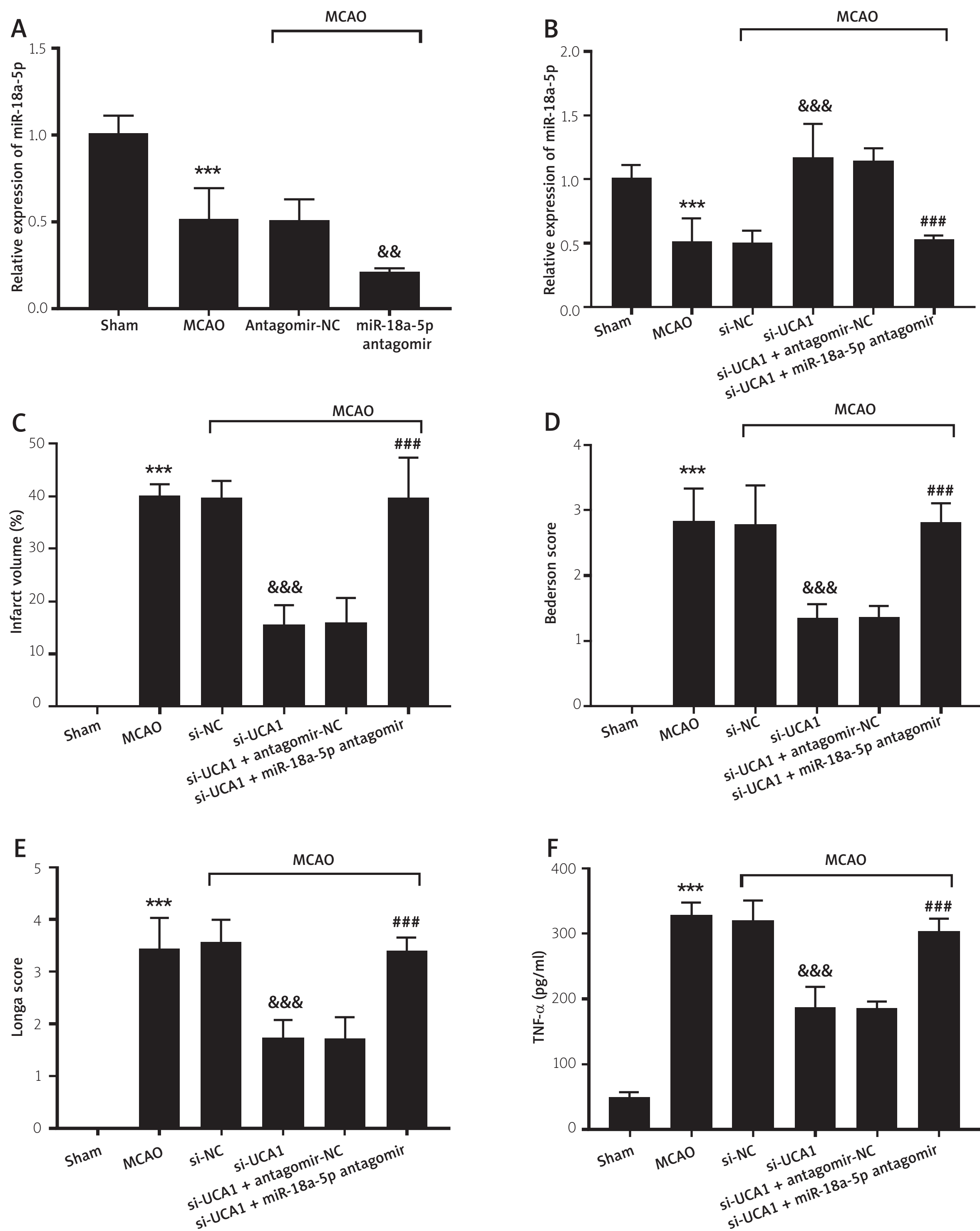

Fig. 3. The function of miR-18a-5p in the SD rat models induced by MACO treatment. **A, B**) The results of transfection of miR-18a-5p antagomir and co-transfection of si-UCA1 and miR-18a-5p. The mediated function of miR-18a-5p on infarct size (**C**), neurofunctional situations (**D, E**) and inflammation (**F-H**). $^{***}p < 0.001$, compared with the control group; $^{\&\&\&}p < 0.001$, compared with the MCAO group; $^{\#\#\#}p < 0.001$, compared with the si-UCA1 group.

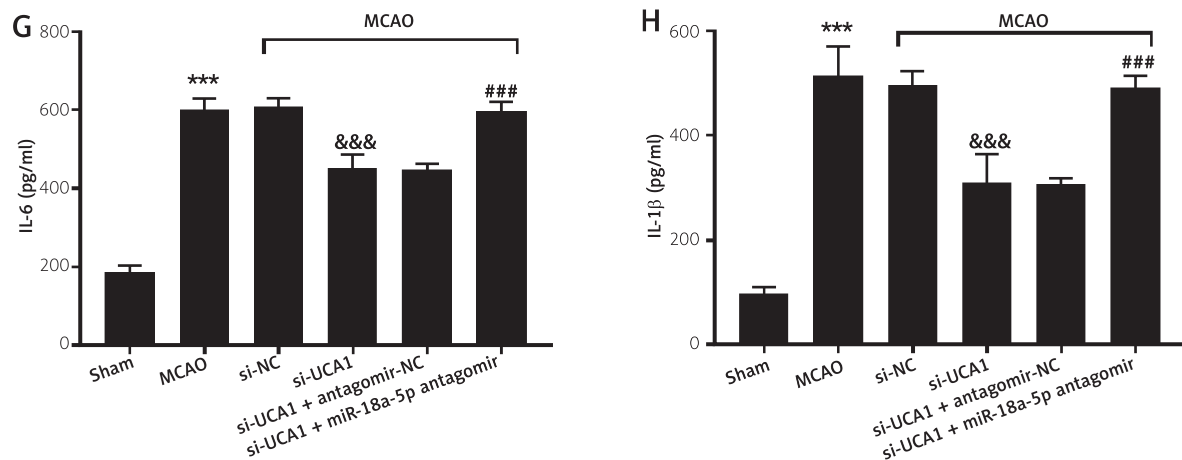

Fig. 3. Cont. The function of miR-18a-5p in the SD rat models induced by MACO treatment. The mediated function of miR-18a-5p on infarct size (**C**), neurofunctional situations (**D, E**) and inflammation (**F-H**). $^{***}p < 0.001$, compared with the control group; $^{\&\&\&}p < 0.001$, compared with the MCAO group; $^{\#\#\#}p < 0.001$, compared with the si-UCA1 group.

miR-18a-5p was validated. The UCA1 expression was obviously raised in the OGD/R group and knockdown of UCA1 inhibited the overexpression of UCA1 in the OGD/R-induced cells (Fig. 4A, $p < 0.001$). The expression of miR-18a-5p was declined in the OGD/R-steered cells and absence of miR-18a-5p facilitated the underexpression of miR-18a-5p (Fig. 4B, $p < 0.01$). What is more, the expression of miR-18a-5p was augmented in the OGD/R + si-UCA1 group when compared to the OGD/R group and inhibited miR-18a-5p reversed this trend (Fig. 4C, $p < 0.001$).

In addition, the roles of UCA1 and the co-regulation relationship of UCA1 and miR-18a-5p were also elucidated. In the observation of Figure 4D, the cell viability was lower in the OGD/R group than in the control group and repressed UCA1 expression attenuated cell viability of OGD/R cells ($p < 0.001$). Likewise, the co-transfection of si-UCA1 and miR-18a-5p inhibitor reversed the function of silenced UCA1 on cell viability (Fig. 4D, $p < 0.001$). The apoptotic cells in the OGD/R group were increased significantly and reduced UCA1 expression inhibited the abundance of cell apoptosis (Fig. 4E, $p < 0.001$). The release of LDH and inflammatory indicators was aggravated in the OGD/R group relative to the control group and disruption of UCA1 reversed this trend (Fig. 4F, G, $p < 0.001$). Additionally, the exhibition of Figure 4E-G also propounded that co-transfection of si-UCA1 and miR-18a-5p inhibitor moderated the impacts of UCA1 on apoptosis, leakage of LDH, and inflammation ($p < 0.01$).

Expression of UCA1 and miR-18a-5p in AIS patients

For further identification of UCA1 expression in AIS, we collected AIS patients and detected the expression of UCA1 and miR-18a-5p. As substantiated in Figure 5A, the expression of UCA1 was highly expressed in the serum samples of AIS patients relative to the healthy control (HC) group ($p < 0.001$). Conspicuously, the relative levels of serum miR-18a-5p in the AIS group were lower than that in the control group (Fig. 5B, $p < 0.001$). The results of Pearson analysis certified that the levels of miR-18a-5p were associated with the expression of UCA1 (Fig. 5C, $r = –0.7143$, $p < 0.001$).

Discussion/Conclusion

Ischemic stroke is the most common type of stroke, which is characterized by the sudden interruption of blood circulation in the brain region [25]. At present, intravenous thrombolysis and interventional embolectomy have become effective treatments for ischemic stroke to restore blood supply quickly [8]. However, this management will lead to CI/R injury and aggravate further brain injury and dysfunction. Therefore, clarifying the potential molecular mechanism of brain I/R injury is helpful to improve the functional recovery after CI/R injury.

CI/R injury is an intricate pathophysiological process, which is regulated by many elements [4]. LncRNA has been proved to play a regulatory role in death caused by brain I/R injury [2]. In the current project, the expression of UCA1 was augmented in the MCAO-treated SD rats, which elucidated that the operation of MACO might contribute to the abundance of UCA1. The findings also revealed that MACO might be a trigger of increased infarct size, raised Bederson score, Longa score, and abnormal inflammatory deficit, indicating the damage of MACO on normal brain function. Furthermore,

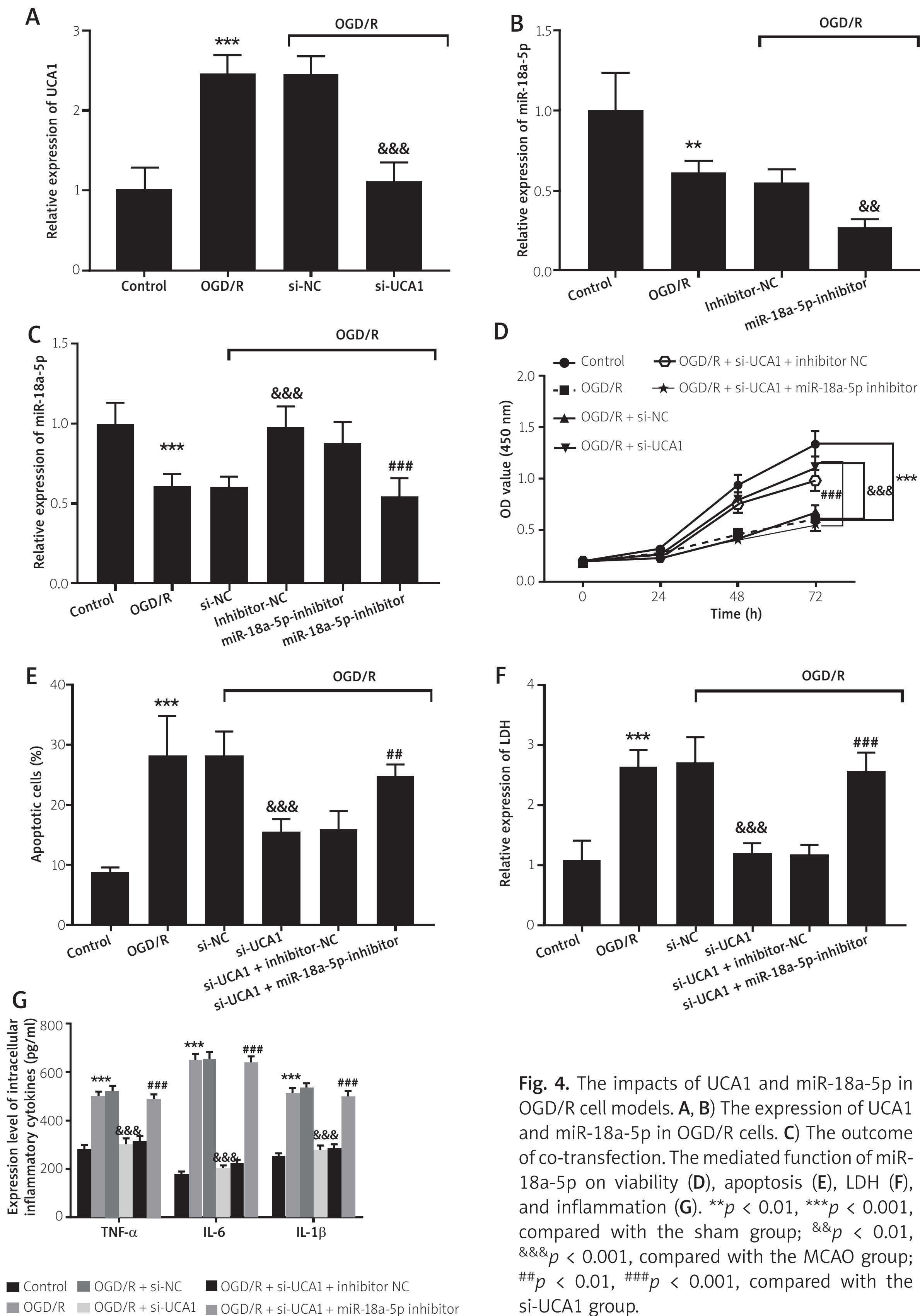

Fig. 4. The impacts of UCA1 and miR-18a-5p in OGD/R cell models. **A**, **B**) The expression of UCA1 and miR-18a-5p in OGD/R cells. **C**) The outcome of co-transfection. The mediated function of miR-18a-5p on viability (**D**), apoptosis (**E**), LDH (**F**), and inflammation (**G**). $^{**}p$ < 0.01, $^{***}p$ < 0.001, compared with the sham group; $^{\&\&}p$ < 0.01, $^{\&\&\&}p$ < 0.001, compared with the MCAO group; $^{\#\#}p$ < 0.01, $^{\#\#\#}p$ < 0.001, compared with the si-UCA1 group.

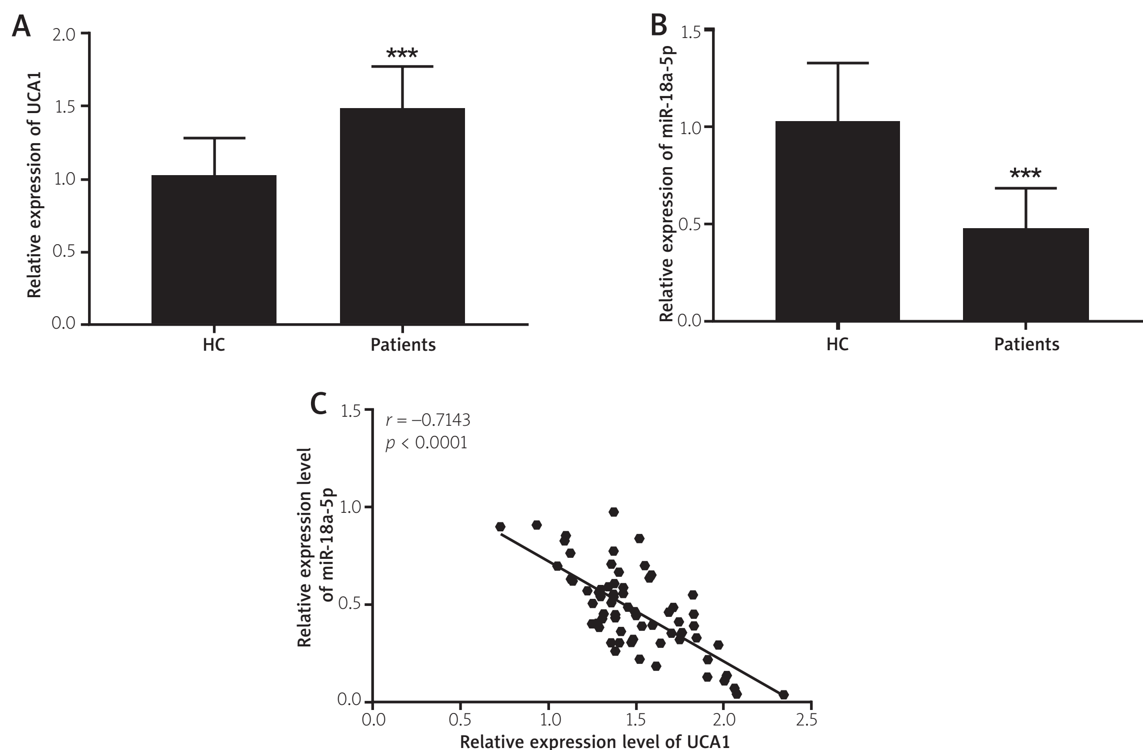

Fig. 5. The levels of UCA1 (**A**) and miR-18a-5p (**B**) in AIS patients. **C**) The correlation between UCA1 and miR-18a-5p. ***$p < 0.001$, compared with the HC group.

the declined UCA1 expression reversed the influence of MCAO on infarct volume, neurological deficits, and inflammation, reflecting protective roles of the silenced UCA1. Recently, several experiments on UCA1 relative to neurological disorders and inflammation have been unveiled. In a study of AIS patients, the alternation of expression of UCA1 was elevated with the increased release of IL-17 and IL-6, which was further verified by our investigation [21].

Presently, more and more researches on miRNA are emergent. To clarify the regulatory mechanisms of UCA1, miR-18a-5p was identified as a downstream transcription factor of UCA1. This deduction was further manifested by the outcome of luciferase report. Furthermore, miR-18a-5p was lowly expressed in the MCAO models and downregulation of UCA1 regulated the levels of miR-18a-5p reversely. A project concerning hypertension-induced heart failure provides a perspective that miR-18a-5p ameliorates the dysfunction of the heart engendered by hypertension, which displays the roles of miR-18a-5p on cardiac protection [12]. The overexpression of miR-18a-5p exerts a protective role in OGD/R-stimulated cells *via* facilitating cell viability and decreasing the apoptotic cells [19]. These previous researches identify the possible influence of miR-18a-5p on the CI/R damage.

Further analysis on the mediated function of miR-18a-5p was confirmed in the MCAO rat model. The regulation of miR-18a-5p was constructed by the injection of miR-18a-5p antagomir, and the result showed that miR-18a-5p antagomir made successful accommodation of miR-18a-5p levels. In proliferative diabetic retinopathy and diabetic cardiac fibrosis, the abnormal change of miR-18a-5p expression was associated with the development of complications of diabetes [9, 11]. In patients with metabolic syndrome, the miR-18a-5p was lessened relative to control subjects, and its levels correlate with the expression of IL-6 [14]. In this present investigation, the lowly expressed miR-18a-5p restricted the depressed infarct size, Bederson score, and Longa score led by UCA1 loss, which documented that the beneficial impacts of diminished UCA1 on CI/R injury were regulated by miR-18a-5p. Especially, the mediated influence of miR-18a-5p was concluded by the finding that the inflammatory disorders were mitigated by the downregulation of UCA1 and aggravated by the co-transfection of miR-18a-5p antagomir and si-UCA1. In addition, the expression of UCA1 is elevated in the mouse model of Parkinson's disease and the detection of SH-SY5Y cells indicates UCA1 disruption improves the percent of viable cells and mitigates the dead cells [18]. Besides, UCA1 can attenuate the harm steered by

hypoxia/reoxygenation of cardiac cells *via* binding miR-143 [5]. In our study, we found that the expression of UCA1 was raised in the OGD/R cells and silenced UCA1 protected cells against OGD/R damage via expediting cell viability, repressing cell apoptosis, alleviating LDH release, and hindering inflammation. Furthermore, the advantageous character of silenced UCA1 was reversed by the interference of miR-18a-5p.

Most importantly, the expression of UCA1 and miR-18a-5p was clarified in the AIS patients. The demonstration displayed highly expressed UCA1 and lowly expressed miR-18a-5p in AIS patients. And then, the interconnection between UCA1 and miR-18a-5p was manifested. In a systemic lupus erythematosus, the levels of UCA1 were enhanced in the plasma of patients [15]. Jiang *et al*. performed a study on acute myocardial infarction and they identified the abnormally expressed levels of miR-18a-5p in target patients [16]. Besides, the expression of miR-18a-5p is verified as downregulated in several types of heart failure, including ischemic cardiomyopathy [29]. These mentioned researches also provide supplementary evidence to our conclusion.

Taken together, silencing UCA1 could improve CI/R injury by reducing infarct size, neurofunctional scores, and inflammation *via* sponging miR-18a-5p. Additionally, miR-18a-5p, as a ceRNA of UCA1, contributed to the amelioration of OGD/R harm on cells by inhibiting apoptosis, LDH, and inflammatory factor levels, and motivating cell viability. Besides, in AIS patients, the levels of UCA1 were upregulated and the expression of miR-18a-5p was downregulated. The expression of UCA1 was correlated with miR-18a-5p expression. This investigation provided insight into fundamental pathophysiology of AIS and might contribute to further analysing comprehensive methods for diagnosis and treatment of AIS.

Disclosure

The authors report no conflict of interest.

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