Aneurismal subarachnoid hemorrhage (aSAH) is a common disease in the neural system, with high death rate. Our study aimed to explore the clinical effect of external ventricular drainage under intracranial pressure monitoring in the treatment of patients with aSAH and investigate the role along with mechanism of miR-146a-5p in aSAH. Ninety-six aSAH patients were allocated into control group (CG) and study group (SG). The CG was released by lumbar puncture. The SG underwent external ventricular drainage based on intracranial pressure monitoring. The prognosis, daily living ability, neurological function, S100β and NSE (neuron-specific enolase) levels and incidence of complications were monitored. Besides, a rat model of SAH was built to assess the neurobehavioral function, blood-brain barrier permeability, brain water content, neuronal apoptosis as well as inflammation. SAH cell model stimulated by oxyhemoglobin, and cell apoptosis as well as inflammation were measured. Luciferase reporter assay was implemented to explore the interaction between miR-146a-5p and STC1. Results showed higher GOS and BI scores but lower NIHSS scores, S100β and NSE levels and complication rates in SG compared with CG. Additionally, miR-146a-5p presented down-regulation in brain tissues of SAH rat model, and overexpressed miR-146a-5p reduced brain injury along with neuroinflammation in SAH rat model. Oxyhemoglobin-induced nerve cell apoptosis along with inflammation after SAH, and overexpressed miR-146a-5p repressed oxyhemoglobin-induced nerve cell apoptosis along with inflammation. STC1 is the target mRNA of miR-146a-5p, and overexpressed miR-146a-5p represses oxyhemoglobin-induced nerve cell apoptosis along with inflammation via regulating STC1 expression. In conclusion, external ventricular drainage under intracranial pressure monitoring could promote prognosis, promote daily living ability, improve neurological function, reduce S100β protein and NSE levels, and reduce the incidence of complications in patients with aSAH. Meanwhile, miR-146a-5p inhibited early brain injury and neuroinflammation in aSAH via regulating STC1 expression.

Keywords: Aneurismal subarachnoid hemorrhage, Early brain injury, External ventricular drainage, Intracranial pressure monitoring, MiR-146a-5p, STC1

1. Introduction

Aneurismal subarachnoid hemorrhage (aSAH) is mainly caused by aneurismal hemorrhage (aneurismal subarachnoid hemorrhage), often accompanied by nausea, vomiting, meningeal irritation, and severe headaches [1]. About 1/4 of patients with aSAH will have hallucinations, delirium, epilepsy, mild hemiplegia, sensory disorders, etc., in severe cases, cerebral vasospasm, hydrocephalus, rebleeding, convulsions, etc [2]. Therefore, the main objective of clinical treatment of aSAH is to prevent complications and reduce the disability rate and fatality rate.

At present, lumbar puncture release of cerebrospinal fluid is commonly applied in clinical therapy of aSAH. Although it can promote the dilution and release of cerebrospinal bloody fluid to a certain extent, the overall treatment effect is poor and the prognosis of most patients is poor due to the difficulty in mastering the drainage flow of cerebrospinal fluid and the need for repeated lumbar puncture [3]. Recently, with the continuous development and improvement of medical technology, external ventricular drainage under intracranial pressure monitoring has been recognized by clinicians and patients because it has fewer complications and is beneficial to promote the quality of life of aSAH patients [4], but there are few reports on its application effect in patients with aSAH.

SAH is also associated with a variety of complications that can lead to early brain injury (EBI). EBI means direct brain injury along with secondary pathophysiological reac-
tions in 72 h after aSAH, containing elevated intracranial pressure as well as reduced cerebral blood flow, leading to cerebral ischemia [5]. These alterations immediately result in neuronal death, changes in blood-brain barrier (BBB) permeability, along with brain edema. Although efforts have been made in SAH treatment, which has improved the survival rate of patients by 17%, most patients still have varying degrees of neurological and cognitive dysfunction [6]. Hence, a deeper comprehension of the potential molecular mechanisms associated with EBI paves the way for novel prevention methods as well as treatments that may lead to EBI after aSAH.

MicroRNAs (miRNAs) belong to widespread RNA molecules of approximately 21-23 nucleotides in eukaryotes, which represses the translation of mRNA via combining with the 3’-untranslated region (UTR) of the target mRNA [7]. As reported previously, miRNAs are crucial modulators in various neurological diseases. For example, miR-9-5p represses the MMP-stimulated neuron apoptosis in Parkinson’s disease through SCRIB/β-Catenin signaling [8]. MiR-144 promotes microglial autophagy as well as inflammation after intracerebral hemorrhage [9]. Besides, many miRNAs containing miR-706 as well as miR-130b are also discovered to be dysregulated during aSAH pathogenesis [10]. Of note, miR-146a-5p has been documented to be low-expressed in aSAH patients, but its function in aSAH has not been investigated.

In our research, we aimed to explore the clinical impact of external ventricular drainage under intracranial pressure monitoring in the treatment of patients with aSAH and investigate the role along with mechanism of miR-146a-5p in aSAH.

2. Material and methods
2.1. Patient data
Ninety-six aSAH patients who received therapy in our hospital from March 2021 to June 2023 were chosen, followed by dividing into control group (CG) and study group (SG) according to cerebrospinal fluid drainage methods, with 48 cases in each group. The CG contained 28 males together with 20 females. The average age was (45.62±15.42) years, ranging 20-74 years; Subarachnoid hemorrhage Hunt & Hess classification: grade II (16 cases), grade III (18 cases), grade IV (10 cases), grade V (3 cases); Aneurysm location: 6 cases of anterior cerebral artery A1 segment, 14 cases of posterior communicating artery, 28 cases of anterior communicating artery. The SG contained 28 males together with 20 females. The average age was (45.62±15.45) years, ranging 20-74 years; Subarachnoid hemorrhage Hunt & Hess classification: grade II (16 cases), grade III (19 cases), grade IV (11 cases), grade V (2 cases); Aneurysm location: 7 cases of anterior cerebral artery A1 segment, 15 cases of posterior communicating artery, 26 cases of anterior communicating artery. No difference was discovered in general data between 2 groups (P>0.05).

Inclusion criteria: (1) Subarachnoid hemorrhage was confirmed by cranio cerebral CT examination and aneurysm was confirmed by cranio cerebral CT angiography (CTA) examination; (2) Hospital admission within 24 hours of onset; (3) Patients gave informed consent to the study. Exclusion criteria: (1) Patients with coagulopathy; (2) Patients with severe liver, kidney along with other important organ dysfunction; (3) Patients with traumatic subarachnoid hemorrhage; (4) Patients with other types of spontaneous subarachnoid hemorrhage; (5) Hydrocephalus, cerebrovascular spasm and other complications.

2.2. Treatment methods
After admission, patients in 2 groups adopted routine symptomatic treatment containing controlling blood pressure, reducing intracranial pressure, oxygen inhalation, improving microcirculation, anti-inflammatory, correcting water and electrolyte disorders, mild hypothermia therapy, and nutritional cranial nerve.

Patients in the CG were released by lumbar puncture on the basis of conventional symptomatic treatment: lumbar puncture was performed to release cerebrospinal fluid on the second day after admission, once a day, for 8-10 days, cerebrospinal fluid biochemistry and routine examination of cerebrospinal fluid were performed every day until the protein content of cerebrospinal fluid was < 0.4 g/L and the red blood cell count was reduced to <100×10³/L. After puncture, the cerebrospinal fluid was replaced with 0.9% sodium chloride solution.

Patients in the SG underwent external ventricular drainage based on intracranial pressure monitoring on the basis of conventional symptomatic treatment: after clipping the aneurysm, the subarachnoid space was cleaned, intracranial pressure was monitored with an intraventricular intracranial pressure probe once/h, and external ventricular drainage was performed with the frontal Angle as the puncture point. Nimodipine was given to patients with intracranial pressure greater than 25 mm Hg (1 mm Hg=0.133 kPa) to maintain intracranial pressure within the reference range.

Dynamic cranio cerebral CT examination was performed in both groups.

2.3. Observation indicators
Glasgow Prognostic Scale (GOS) was used to evaluate the prognosis 6 months after surgery [11]. The total score was 1-5, with 1 being death. The higher score indicated better prognosis.

Barthel index (BI) was adopted to evaluate the daily living ability of 2 groups before and 6 months after surgery [12], with the total score ranging 0-100. The higher score represented better daily living ability.

National Institutes of Health Stroke Scale (NIHSS) was implemented to evaluate the neurological defects of 2 groups before and 6 months after surgery [13]. The lower the score, the better the neurological function was.

3 mL of fasting venous blood was gathered before as well as 6 months after surgery, and serum was acquired after centrifugation. S100β protein and (neuron-specific enolase) NSE levels were determined by enzyme-linked immunosorbent assay.

The incidence of complications, including infection, cerebral infarction, epilepsy, and hydrocephalus were compared between 2 groups during hospitalization.

2.4. SAH model in rats
Adult male Sprague-Dawley (SD) rats, weighing 230 ~ 335 g, were placed in a specific pathogen-free animal laboratory with constant temperature and humidity, free access to food and water, and a light and dark cycle of 12 h/12 h. This experiment was approved by the Animal Ethics Committee of the hospital.

The rat SAH model was established using the method
described in the literature. The operation was as follows: the rats were anesthetized with 3% isoflurane and subjected to mechanical ventilation. A sharp 4-0 monofilar nylon suture was inserted from the external carotid artery into the internal carotid artery. Then a further 3 mm was advanced to puncture the blood vessel, the nylon sutures were pulled out, the external carotid artery was lapped to restore blood flow, and the wound was stitched to allow the rat to recover on a heated blanket. After 1 h of SAH modeling, SAH-stimulated rats received injections with miR-146a-5p agomir and agomir NC in stereotactic ventricle. Healthy rats underwent the same operation without perforating the blood vessels and were set as the sham group.

2.5. Stereotactic lateral ventricular injection
MiR-146a-5p agomir and agomir NC were purchased from Shanghai GenePharma (Shanghai, China). One hour following SAH model was established, 2.5% isoflurane-anesthetized rats were fixed on Stoelting animal stereotactic apparatus. Afterwards, MiR-146a-5p agomir or agomir NC (0.5 μL/min) was injected with a 10 μL syringe at 3.5 mm submeningeal, 1.5 mm outside the right ventricle (right), and 0.9 mm posterior to the right ventricle, using the fontanel as a reference point. Next, the needle was slowly pulled out, the pores were sealed with bone wax, the scalp was sutured, and an electric blanket was used to keep warm before anesthesia.

2.6. Neurobehavioral tests
Neurobehavioral tests were performed 24 h after SAH and evaluated via an improved Garcia score method [14]. The neurobehavioral test was administered by 2 independent researchers, and average scores were calculated.

2.7. Morphological observation of rat brain
The rats were sacrificed, and the brain tissue could be isolated, dehydrated, embedded, followed by cutting into 5 μm slices, and HE staining was implemented to observe the histopathology and pathology of rat brain under microscope.

2.8. Cerebral water content determination
The brain tissues of the rats were gathered immediately after euthanasia, followed by separating into right hemisphere, left hemisphere, cerebellum, as well as brainstem. They weigh the above parts (recorded to be wet weight) and place them in an oven at 105°C for 3 consecutive days. Then weigh the dry brain, called dry weight; Brain water content = [(wet weight - dry weight)/(wet weight)]×100%.

2.9. Blood-brain barrier (BBB) test
The BBB permeability was assessed by Evans Blue exudation. Evans Blue dye (2%, 5 mL/kg) was injected through the right femoral vein, and normal saline was injected through the cardiac vein until the drainage was colorless. Afterwards, the brain tissue was separated, milled, and centrifuged, followed by obtaining the supernatant. The same amount of ethanol trichloroacetic acid was added to the sample for incubation at 4°C overnight. The absorbance of the sample could be measured by UV-VIS spectrophotometer at 620 nm. The EB solution standard curve was drawn with 50 ~ 1000 ng/mL Ethidium bromide (EB) standard solution. The absorbance value of the sample was compared with the EB standard curve to calculate the EB level of the sample.

2.10. RT-qPCR
Total RNA was extracted from harvested cortex tissues by help of RNAiso Plus (Takara Bio, Dalian, China). PrimeScript RT Master Mix (Takara, Japan) was implemented for the reverse transcription of RNA to cDNA. RT-qPCR was implemented using Hieff® qPCR SYBR Green Master Mix (Yeasen, China). The 2-ΔΔCt method was adopted to identify the gene relative expression levels.

2.11. TUNEL
The brain tissue was fixed, dehydrated, and impregnated with wax for embedding. The tissues were sliced 4 μm thick, roasted at 60°C for 3 h, and dewaxed. According to the Apo Alert TUNEL cell apoptosis detection kit instructions (Biosciences Clontech, USA), the sections were finally sealed with a sealing solution including anti-fluorescence quencher, and the images could be observed via a fluorescence microscope to calculate the proportion of apoptotic nerve cells.

2.12. ELISA
The brain tissue samples were ground and placed in a centrifuge tube, homogenized in the bath with a high-speed homogenizer, centrifuged, and the supernatant was extracted following the kit instructions. ELISA was adopted to detect the concentrations of IL-6, IL-1β as well as TNF-α.

2.13. Cell culture and in vitro SAH model
Murine HT22 cells were cultivated in Dulbecco’s modified Eagle medium (DEME) which contained 10% fetal bovine serum along with 1% penicillin/streptomycin at 37°C with CO₂. Oxyhemoglobin (5 μM) was added into the culture medium for 24 h to stimulate SAH damage in vitro.

2.14. Cell transfection
MiR-146a-5p mimics and NC mimics, along with pcDNA3.1-STC1 and pcDNA3.1 were purchased from GenePharma (Shanghai, China). Cell transfection was implemented using Lipofectamine 2000 (Thermo Fisher, USA).

2.15. CCK-8
Cells were planted in 96-well plates, and then CCK-8 reagent (10 μL) mixed with 90 μL of complete medium was added to each well for 1.5 h of cultivation. The optical density was tested at 450 nm by means of a microplate reader.

2.16. Flow cytometry
Cells were digested, collected, washed, and resuspended. Then, 5 μL Annexin V-FITC, as well as 5 μL Propidium Iodide, were added into cells for reaction in the dark for 15 min at room temperature. Cell apoptosis was analyzed by flow cytometry.

2.17. Luciferase reporter assay
Cells were planted into 12-well plates, followed by co-transfection with luciferase plasmids of STC1 3’UTR-WT/MUT in the pmirGlo vector (GenePharma, China) or miR-146a-5p mimics/NC mimics for 24 h. The Dual-
Luciferase Reporter Assay System (Promega, USA) was implemented to assess the luciferase activity.

2.18. Statistical analysis
Statistical analysis was implemented with GraphPad Prism. All experiments were performed more than 3 times. Data that satisfied normality and homogeneity of variance were exhibited as the mean ± standard deviation (SD) and analyzed with student t-test or one-way analysis of variance (ANOVA) followed by multiple comparisons using Tukey’s post hoc analysis. Statistical difference was defined as P<0.05.

3. Results

3.1. Clinical effect of external ventricular drainage under intracranial pressure monitoring in the treatment of patients with aSAH
Six months after surgery, the GOS score of the SG presented higher when compared with the CG (P<0.01, Fig. 1A). Before surgery, no difference was discovered in BI score between 2 groups (P>0.05). Six months after surgery, the BI score was elevated in 2 groups, and that in the SG presented elevation when compared with the CG (P<0.05, Fig. 1B). Before surgery, no difference was discovered in NIHSS score between 2 groups (P>0.05). Six months after surgery, the NIHSS score was lessened in 2 groups, and that in the SG presented lower when compared with the CG (P<0.05, Fig. 1C). Before surgery, no difference was discovered in S100β and NSE levels between 2 groups (P>0.05). Six months after surgery, the S100β and NSE levels were lessened in 2 groups, and those in the SG presented reduction when compared with the CG (P<0.05, Fig. 1D-E). Table 1 displayed that the incidence of complications in the SG presented reduction when compared with the CG (P<0.05).

3.2. Overexpressed miR-146a-5p reduces brain injury and neuroinflammation in SAH rat model
In the SAH group, the Garcia score could be decreased, the BBB permeability was increased, and the brain water content was enhanced (Fig. 2A-C). HE staining was used to assess the brain damage in rats. In the sham group, the cortical structure was intact, the neuronal morphology was normal, the nucleus was intact and blue spherical, the cytoplasm was light stained, and there was no neuronal damage. In the SAH group, the cortical structure of rats was disorganized and loose, the number of neurons was lessened, as well as the pathological changes such as nuclear pyretic contraction and deep staining were observed, and the brain tissue and neurons were damaged (Fig. 2D). Subsequently, the cerebral cortex tissue was sliced and TUNEL staining was implemented to detect neuronal apoptosis. The number of TUNEL-positive cells was elevated in the SAH group (Fig. 2E). ELISA unveiled that IL-1β, IL-6 along with TNF-α levels presented higher in the SAH group (Fig. 2F). In addition, miR-146a-5p presented down-regulation in the SAH group (Fig. 2G).

First, miR-146a-5p agomir was transfected into the

![Fig. 1. Clinical effect of external ventricular drainage under intracranial pressure monitoring in the treatment of patients with aSAH. (A) GOS score. (B) Barthel index. (C) NIHSS score. (D-E) S100β and NSE levels. **P<0.01, compared with before treatment. *P<0.01, compared with control group.](image1)

![Fig. 2. Overexpressed miR-146a-5p reduces brain injury and neuroinflammation in SAH rat model. (A) Neural function in SAH rat model. (B) BBB permeability in SAH rat model. (C) Brain water content in SAH rat model. (D) HE staining of SAH rat brain tissues. (E) TUNEL staining of apoptosis in SAH rat model. (F) ELISA assessed IL-1β, IL-6 and TNF-α levels in SAH rat model. (G) RT-qPCR examined miR-146a-5p expression in SAH rat model. (H) RT-qPCR examined miR-146a-5p overexpression efficiency in SAH rat models. (I) Effect of miR-146a-5p overexpression on neural function. (J) Effect of miR-146a-5p overexpression on BBB permeability. (K) Brain water content after miR-146a-5p elevation. (L) HE staining of SAH rat brain tissues. (M) TUNEL staining of apoptosis in SAH rat model after miR-146a-5p elevation. (N) ELISA assessed inflammation in SAH rat model after miR-146a-5p overexpression. **P<0.01.](image2)
3.3. Overexpressed miR-146a-5p represses oxyhemoglobin-stimulated nerve cell apoptosis along with inflammation

It was observed in CCK-8 assay that oxyhemoglobin treatment apparently reduced HT22 cells viability (Fig. 3A). Meanwhile, flow cytometry analysis indicated that oxyhemoglobin treatment apparently elevated HT22 cells apoptosis (Fig. 3B). As for inflammation, oxyhemoglobin treatment enhanced IL-1β, IL-6 along with TNF-α levels in HT22 cells (Fig. 3C). RT-qPCR results displayed that oxyhemoglobin could reduce miR-146a-5p expression in HT22 cells (Fig. 3D). Thus, we elevated miR-146a-5p expression in HT22 cells to assess the impacts of miR-146a-5p overexpression on oxyhemoglobin-stimulated nerve cell apoptosis along with inflammation (Fig. 3E). As displayed in Fig. 3F-H, miR-146a-5p overexpression definitely elevated the viability as well as repressed apoptosis and inflammation in oxyhemoglobin-induced HT22 cells.

3.4. STC1 is the target mRNA of miR-146a-5p

Through microT, RNA22 and Targetscan databases, STC1 was predicted to have binding sites with miR-146a-5p (Fig. 4A), and the binding sites between them were displayed in Fig. 4B. Through luciferase report assay, we proved that the luciferase activity of STC1 3’UTR-WT could be weakened after miR-146a-5p overexpression, but the luciferase activity of STC1 3’UTR-MUT was not changed (Fig. 4C). Besides, we discovered that oxyhemoglobin treatment could elevate STC1 expression in HT22 cells (Fig. 4D). More importantly, STC1 expression could be repressed in oxyhemoglobin-treated HT22 cells after miR-146a-5p overexpression (Fig. 4E).

3.5. Overexpressed miR-146a-5p represses oxyhemoglobin-stimulated nerve cell apoptosis along with inflammation via regulating STC1 expression

We elevated STC1 expression in oxyhemoglobin-treated HT22 cells (Fig. 5A), followed by performing rescue assays to certify whether overexpressed miR-146a-5p represses oxyhemoglobin-stimulated nerve cell apoptosis along with inflammation via regulating STC1 expression. It was discovered in Fig. 5B-D that the elevated viability as
well as the lessened apoptosis and inflammation caused by miR-146a-5p elevation in oxyhemoglobin-treated HT22 cells could be offset after co-transfection of pcDNA3.1-STC1.

4. Discussion
The results of epidemiological investigation have shown that the annual prevalence of aSAH is 10.6/100,000, accounting for 22% of all cerebrovascular lesions and 80% of spontaneous subarachnoid hemorrhage [15]. Cerebrovascular malformation, abnormal vascular network disease of skull base, vasculitis, and connective tissue diseases are risk factors for sSAH, while EBI is a risk factor for death of sSAH patients [16]. Subarachnoid hemorrhage can lead to cerebral edema and metabolic disorders of cerebral microvascular endothelial cells, and then destroy neuronal skeleton and nuclear contraction, leading to neuronal swelling and even apoptosis [17]. At the same time, blood flow into the subarachnoid space through the ruptured blood vessels can lead to the secretion of vasoconstrictor substances such as prostaglandin, vascular endothelin, catecholamine, and then cause smooth muscle tonic contraction and aggravate cerebral vasospasm [18]. Therefore, early removal of hemorrhagic cerebrospinal fluid is essential in decreasing the incidence of cerebral vasospasm along with improving the prognosis of patients with aSAH.

Release of cerebrospinal fluid by lumbar puncture is a common method to remove the hemorrhagic cerebrospinal fluid, which can improve the circulation of cerebrospinal fluid by reducing the stimulation of hemorrhagic cerebrospinal fluid. However, excessive drainage of cerebrospinal fluid can lead to ventricle shrinkage, brain hernia, brain tissue retraction, etc., resulting in bridge vein rupture and hemorrhage and intracranial hemotoma. Recent literatures have shown that the mortality rate of aSAH patients with recurrent aneurysm rupture is about 70%, among which the duration of intracranial hypertension and the degree of intracranial pressure increase are risk factors for the death of aSAH patients with recurrent aneurysm rupture, mainly related to the continuous increase of intracranial pressure leading to consciousness disturbance and brain hernia [19]. Therefore, effectively reducing cerebral edema and improving cerebral perfusion pressure have crucial clinical significance in declining the risk of death in aSAH patients.

External ventricular drainage under intracranial pressure monitoring is a new hemorrhagic cerebrospinal fluid drainage method. Intracranial pressure monitoring is helpful in guiding clinicians to accurately assess the degree of craniocerebral injury and early detection of intracranial space, which is conducive to reducing secondary brain injury and improving the prognosis of patients. The placement of intracranial intraventricular pressure probe is conducive to dynamic monitoring of intracranial pressure changes, effectively avoiding increased blood viscosity, renal function injury, and electrolyte disturbance caused by blind application of dehydrating agents, shortening the placement time of drainage tube and reducing the risk of intracranial infection [20]. Intracranial pressure monitoring can determine the drainage flow according to intracranial pressure, which can effectively avoid rebleeding caused by intracranial pressure fluctuation [21].

In our study, we explored the clinical effect of external ventricular drainage under intracranial pressure monitoring in the treatment of patients with aSAH, and discovered that in contrast to the CG, the GOS score of the SG presented elevation, the BI score in the SG presented higher, the NIHSS score in the SG presented reduction, and the incidence of complications in the SG presented reduction, suggesting that external ventricular drainage under intracranial pressure monitoring could promote prognosis, promote daily living ability, improve neurologic function and repress the incidence of complications of aSAH patients, which was in accordance with previous report [22].

After the occurrence of chronic hydrocephalus after aSAH, the neurons of the body are damaged, and NSE enters the blood through the injured BBB, resulting in the increase of NSE level [23]. S100β protein is produced by astrocytes and oligodendrocytes. When the BBB is damaged, S100β protein is released by the injured cells [24]. Our study also indicated that after treatment, the S100β and NSE levels in the SG presented reduction when compared with the CG, suggesting that external ventricular drainage under intracranial pressure monitoring could improve BBB permeability in patients with aSAH.

Some research has found that miRNA is closely linked to neuropathology and can be used as a possible biomarker for EBI therapy after SAH [25]. The potential of miR-146a-5p in human malignancies has been extensively studied [26]. Moreover, it has been documented that miR-146a-5p relieves neuropathic pain via repressing TRAF6 signaling in the spinal cord [27]. Notably, this study was the first to study the potential of miR-146a-5p in rat and cellular models of SAH. It was discovered that miR-146a-5p presented down-regulation. The causes of EBI within 72 h after SAH include abnormal cerebral blood flow regulation, disorders of cerebral metabolism, damage to the BBB, inflammatory factors, and thrombosis pathways [28]. Neuronal apoptosis is considered to be the key event of EBI after SAH, and is the cause of neurological injury in SAH patients. In addition, physiological abnormalities during EBI trigger an inflammatory cascade leading to the destruction of the BBB and neuronal degeneration [29]. The expression levels of inflammatory factors containing IL-1β, IL-6 as well as TNF-α in SAH patients are up-regulated, which can accelerate brain injury by elevating apoptosis pathway [30].

In our study, we established a rat model of SAH as well as SAH cell model induced by oxyhemoglobin and discovered that miR-146a-5p presented down-regulation in brain tissues of SAH rat model and oxyhemoglobin-treated HT22 cells. Besides, our study revealed that overexpressed miR-146a-5p reduced brain injury as well as neuroinflammation in SAH rat model and hindered oxyhemoglobin-induced nerve cell apoptosis along with inflammation, suggesting that miR-146a-5p exerted a protective function in SAH, which was similar to a study proposed by Mai et al. [31].

Stanniocalcin-1 (STC1) belongs to a homodimeric glycoprotein hormone [32] and is expressed in diverse normal organs containing the skin, lung, ovary, along with cervix, [33]. Studies have discovered that STC1 plays a role in neurological diseases, including glioma as well as ischemic stroke [34]. Notably, it has been reported that serum STC1 concentration presents elevation in aSAH patients [35-37]. In our research, we discovered that STC1 was
proved to be the target gene of miR-146a-5p, and further rescue assays indicated that overexpressed miR-146a-5p hindered oxyhemoglobin-stimulated nerve cell apoptosis along with inflammation via regulating STC1 expression.

5. Conclusion
In conclusion, our study demonstrates that external ventricular drainage under intracranial pressure monitoring can promote prognosis, promote daily living ability, improve neurological function, reduce S100β protein and NSE levels and reduce the incidence of complications of patients with aSAH. Meanwhile, miR-146a-5p inhibits EBI and neuroinflammation in aSAH via regulating STC1 expression.

Conflict of Interests
The authors declare no competing interests.

Consent for publications
The author read and approved the final manuscript for publication.

Ethics approval and consent to participate
We have received approval from the Ethics Committee of Tianjin Huanhu Hospital and Tianjin Fourth Central Hospital.

Informed Consent
We have received informed consent from the Ethics Committee of Tianjin Huanhu Hospital and Tianjin Fourth Central Hospital.

Availability of data and material
If you have any additional questions about the study’s original contributions, please contact the corresponding author.

Authors’ contributions
LJ contributed to the study conception and design. Experimental operation, data collection and analysis were performed by WC and SJ. The first draft of the manuscript was written by WC. All authors commented on previous versions of the manuscript.

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miR-146a-5p/STC1 axis in inhibiting early brain injury


